UNIVERZITA KARLOVA V PRAZE

Přírodovědecká fakulta

Katedra fyziologie živočichů a člověka

Bakalářská práce

Gliové buňky a jejich role v průběhu Alzheimerovy choroby

Glial cells and their role in Alzheimer's disease

Barbora Eliášová

Školitelka: Ing. Miroslava Anděrová, CSc.

Praha, 2013

Prohlášení:

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

V Praze, 06.05.2013

Podpis

Poďakovanie:

Chcela by som poďakovať hlavne svojej školiteľke Ing. Miroslave Anděrovej, CSc. za jej pomoc, ochotu a trpezlivosť, a takisto svojej rodine a priateľom za ich neustálu podporu a cenné rady.

Abstrakt

Alzheimerova choroba je neurodegeneratívne ochorenie, postihujúce hlavne starších ľudí. Postihuje ich pamäť, schopnosť rozprávať, učiť sa či rozhodovať sa. Tie sa postupne zhoršujú až ich pacient úplne stráca. Alzheimer je najčastejšia forma demencie vo svete, no dodnes nie je známy liek, ktorý by ho úplne vyliečil. Hlavným dôvodom je, že ešte stále nie sú úplne objasnené všetky mechanizmy a dôvody, prečo k tomuto ochoreniu dochádza. Okrem postupného odumierania neurónov vplyvom hromadenia proteínu β amyloidu a hyperfosforylovaného tau proteínu hrajú v priebehu Alzheimerovej choroby dôležitú úlohu aj gliové bunky centrálnej nervovej sústavy. Astrocyty, mikroglie, oligodendrocyty a nedávno objavené synantocyty zabezpečujú rôznorodé funkcie, ktoré sú životne dôležité pre správne fungovanie mozgu a ich poškodenie môže byť smrteľné. Pri neurodegenratívnych ochoreniach mozgu ako je Alzheimer sú schopné pomáhať a zmierňovať ochorenie, no často naopak priebeh choroby zhoršujú nesprávnym fungovaním alebo stratou jednej či viacerých funkcií.

Kľúčové slová: Alzheimerova choroba, β amyloid, tau proteín, astrocyty, oligodendrocyty, mikroglie, synantocyty

Abstract

Alzheimer's disease is a neurodegenerative disorder, affecting mostly elderly people. It causes memory impairment and modifies the ability to talk, learn and make decisions. These are gradually getting worse until the patient loses them completely. Alzheimer's is the most common form of dementia worldwide, however until these days there is no cure. The main reason for this is that mechanisms and causes of this disease are still not utterly understood. Besides the neurodegeneration caused by aggregation of β amyloid protein and hyperphosphorylated tau protein, glial cells of central nervous system play also important role in the Alzheimer's disease. Astrocytes, microglia, oligodendrocytes and recently discovered synantocytes ensure various functions necessary for correct functioning of the brain and damage of these cells can be fatal. During a neurodegenerative disorder such as Alzheimer's, they are able to improve the course of the disease but also do the contrary and aggravate it by malfunctioning or losing one or even more of their functions.

Key words: Alzheimer's disease, β amyloid, tau protein, astrocytes, microglia, oligodendrocytes, synantocytes

Contents

Shortcut List

AD	Alzheimer's disease
ADL	activities of daily living
ALdh1L1	aldehyde dehydrogenase 1 family, member L1
ApoE	apolipoprotein E
Aß	ß amyloid
CNS	central nervous system
GFAP	glial fibrillary acidic protein
GSK-3	glycogen synthase kinase 3
hAPP	human amyloid precursor protein
MRI	magnetic resonance imaging
NFT	neurofibrillary tangle
OL	oligodendrocyte
PET	positron emission tomography
PHF	paired helical filaments
ROS	reactive oxygen species
3×Tg-AD	triple transgenic Alzheimer's disease model

Introduction

With the increasing life expectancy of men, more and more diseases affecting our body and mind emerge. People are able to enjoy their lives longer, but an older age is also assembling lot of problems and inconveniencies. Various modifications are happening as the cells of our body are aging. They are pursuing lot of changes and some of them are even losing the ability to recover. Therefore, our body, including the brain, is more susceptible to any damage.

Yearly, there is about 4,6 million new cases of dementia (Kalaria et al., 2008), one of the most common being Alzheimer's disease. Part of the reasons why this number is so high, is that the main cause of this illness, named after its discoverer Alois Alzheimer who first described it in 1910, is still to be discovered. For years, the main direction of the research was discussing neurons and their role in the disease as the neurodegeneration is well observed in patients with Alzheimer's disease, causing most of the symptoms. It was just lately that the attention of scientists was turned to glial cells. There are many types of glial cells in the brain and they have numbers of different functions, some of which are not yet clarified. Nevertheless, today we know that glial cells play an important role in the development and progression of Alzheimer's disease. The discovery of the new type of glial cells, the $NG2^{+}$ glia also called synantocytes, brought some fresh air in the research that is now dwelling on their character of glial but also neuronal precursors considering them to be a new hope for the understanding of the disease likewise the basic brain functions.

In my work, I summarize the basic facts about both the Alzheimer's disease and glial cells, and outline some of the functions or malfunctions of glial cells in this disease. In three parts, I am introducing the Alzheimer's disease, glial cells and their role in the illness, respectively.

Alzheimer's disease

Dementia is a broad term enclosing group of disorders which cause impaired memory and cognition for a period of time of at least 6 months. It differs from mental retardation by changing thinking abilities fundamentally ("Dementia," n.d.). Alzheimer's disease (AD) belongs to a group of dementia disorders. In fact, this devastating neurodegenerative disease is one of the most common types, affecting mostly older people (Maccioni, Muñoz, & Barbeito, 2001).

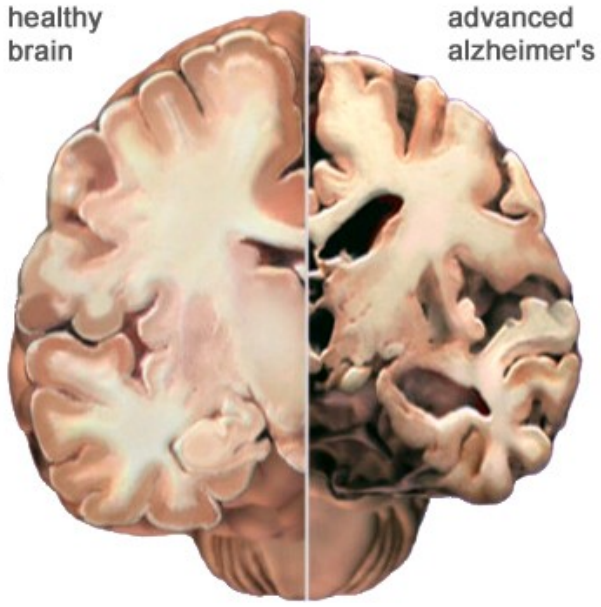

Figure1 – Comparison of a healthy brain and brain of a patient with advanced AD. ("Brain Tour," n.d.)

Main characteristic signs of AD are progressive memory loss, cognitive decline and widespread loss of neurons and their synapses in the cerebral cortex, entorhinal area, hippocampus, ventral striatum and basal forebrain (Nagele et al., 2004). We distinguish two types of AD, familial and sporadic. Phenotypes of these two forms are almost identical. The only difference is an earlier age of onset of familial AD. This type is inherited in the autosomal dominant way. Until these days, we are unable to say to what degree the genetic factors are responsible for sporadic AD (Selkoe, 2001). Many studies suggest that allele ε4 of polymorphic apolipoprotein E (ApoE) gene plays a key role in sporadic late-onset AD (Altamura et al., 2007; Sangmee et al., 2006). However, results of the studies of ApoE polymorphism and AD still vary and more experiments have to be carried out (Cosentino et al., 2008).

The disease has 4 stages: pre-dementia, early (or mild), moderate and advanced (or severe) stage.

- During pre-dementia stage, starting already several years before clinical diagnosis, there is an observable cognitive impairment (Bäckman, Jones, Berger, Laukka, & Small, 2004); however, afflicted persons do not have any serious problems with the "Activities of Daily Living" (ADLs) (Förstl & Kurz, 1999).
- In the early stage, there are some difficulties with ADLs caused especially by memory impairment interfering with various cognitive domains. Because of shrinking vocabulary, decreasing word fluency, and less precise expressive language, communication may be getting worse (Förstl & Kurz, 1999).
- Decline in logical reasoning, planning, and organizing is significant at moderate stage. Communication problems are even more obvious. Patients are losing basic skills necessary for the activities of daily living without close supervision and longer sequences of action cannot be performed. Using household appliances, dressing, and eating are becoming increasingly problematic until these abilities are completely lost. Spatial disorientation is getting more severe. Patients no longer realize their condition (Förstl & Kurz, 1999).
- At the late stage of illness, cognition is seriously impaired. Almost all cognitive functions are lost, even early biographical memories can be affected. Communication skills are reduced to minimum, patients lose the ability to use simple phrases or even single words and express even the simplest of their needs (Förstl & Kurz, 1999).

Life expectancy of a patient with AD is lowered by one third. Mortality is affected by many risk factors, the major being a long persistence of symptoms, the severity of illness, old age, male sex, and physical disease (Förstl & Kurz, 1999).

Pathophysiology

When performing the autopsy of the brain of a person afflicted by AD, we cannot observe any grossly apparent changes compared to any other brain of elderly individual with normal cognitive functions. The definitive diagnosis can be made only upon the histological examination. In this analysis, neurofibrillary tangles and neuritic plaques have to be present to determine AD, as these are both generally recognized as cardinal microscopic lesions associated with this disease (Perl, 2010).

The hippocampus, entorhinal cortex, and amygdale are the main regions where one can find the anomalous structures of neurofibrillary tangles (NFTs). They are composed of clusters of paired helical filaments (PHFs). PHFs are a dense filamentous network formed by self-aggregation of hyperphosphorylated forms of the tau protein. Tau is a multifunctional microtubule-associated protein. It participates in stabilization of microtubules against dynamic instability, and bridging of these polymers with other cytoskeletal filaments. In normal brain, whether this protein is phosphorylated or not, tau plays a major role in modulating the stability of the cytoskeleton and thus axonal morphology. The action of diverse protein kinases (or alternatively the lack of action of various phosphatases) promoting hyperphosphorylations of tau in AD results in structural and conformational changes in this protein, leading to incorrect binding with tubulin, thus affecting the microtubule assembly (Dickson, 1997).

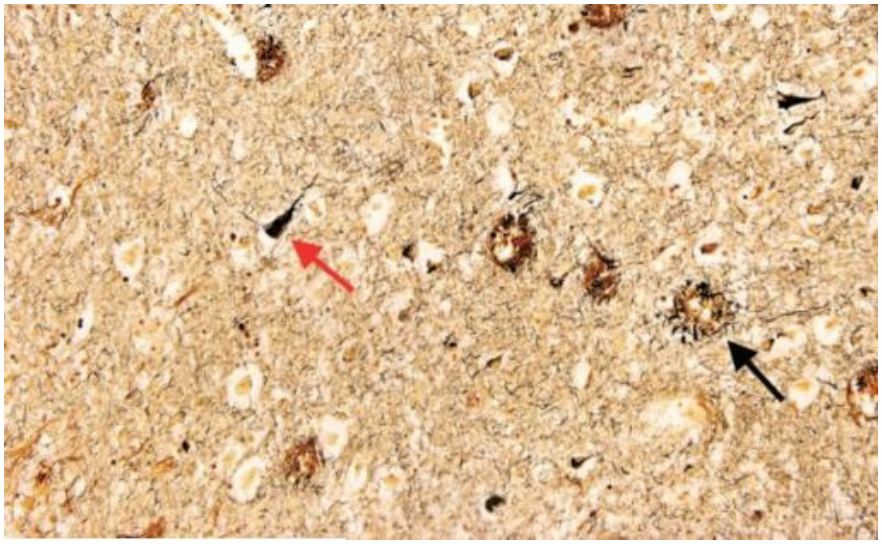

Figure 2 – NFTs (red arrow) in temporal cortex of a patient with AD; visualised by modified Bielschowski staining at 40x magnification. (Perl, 2010)

Diffuse, fibrillar or dense-cored neuritic plaques are formed by extracellular deposition of ß amyloid (Aß) protein (Perl, 2010). In their close surrounding, there are degenerating neuronal processes and clusters of microglia and astrocytes (Dickson, 1997). These plaques can be found in the brains of healthy elderly subjects; however, their number is substantially increased in the brains of people with AD (Maccioni et al., 2001), especially in specific brain regions such as entorhinal cortex (Cummings, Pike, Shankle, & Cotman, 1996).

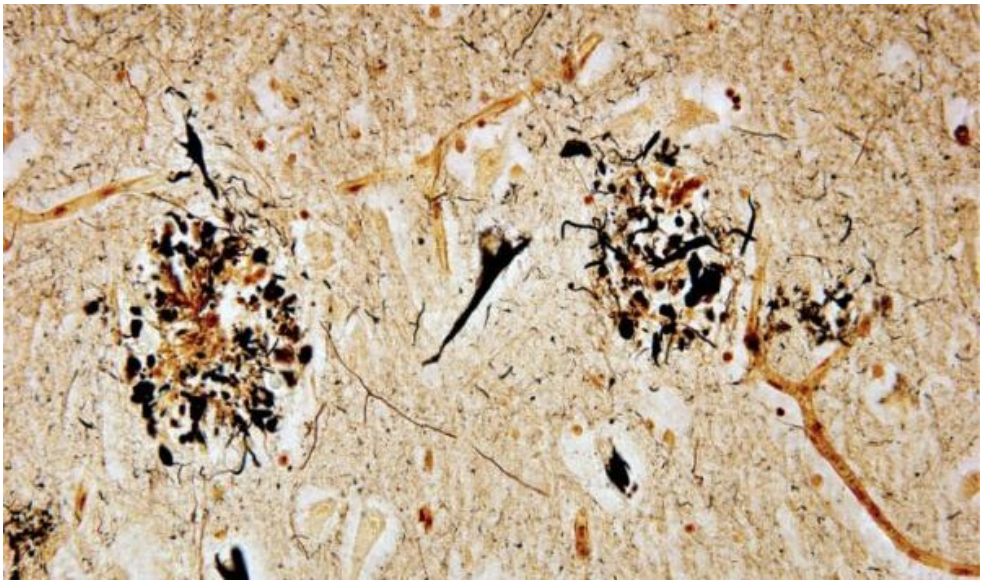

Figure 3 – Neuritic plaques in the temporal cortex of a patient with AD. A NFT can be seen between the plaques. The sample was stained using modified Bielschowski method; original magnification is 400×. (Perl, 2010)

Even though Aß deposition seems to be one of the promoting factors of extracellular and intracellular changes that are responsible for neuronal death in AD, it is not the only cause of neurodegeneration. There are other processes triggering molecular events leading to neuronal dysfunctions, e.g. oxidative stress (Maccioni et al., 2001) or neuroinflammation (Heneka, Rodríguez, & Verkhratsky, 2010).

Diagnosis

To diagnose AD, histopathological methods are employed and special criteria have to be fulfilled. First, a dementia syndrome must be confirmed and just then the criteria based on AD phenotype can be applied. These are memory disorders and impairment in at least one supplementary cognitive domain, both interfering with social life and ADLs. Other brain or systematic disorders that could lead to the deficits in memory and cognition have to be excluded (Dubois et al., 2007).

Magnetic resonance imaging (MRI) is used to observe structural changes in the brain of a patient. These changes are observable mainly in medial temporal lobe (Dubois et al., 2007).

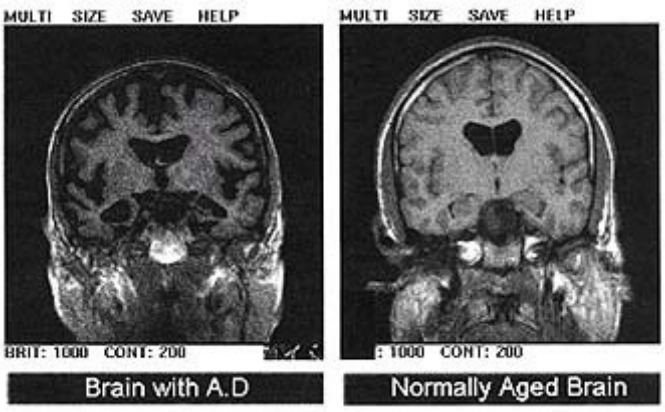

Figure 4 – MRI images of the normally aged brain and of the brain of a patient affected by AD. (Akre, 2008)

To analyze the pathophysiological metabolic changes, positron emission tomography (PET) is used with the main signs of AD being the hypometabolism or hypoperfusion in temporoparietal areas, accompanied by changes in cerebrospinal fluid biomarkers (Dubois et al., 2007).

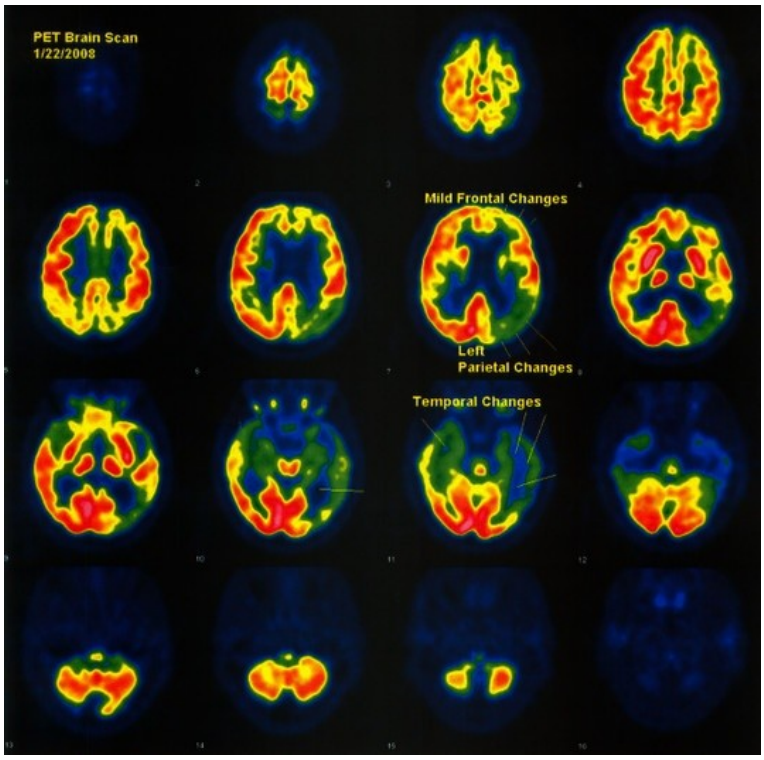

Figure 5 – PET scan of the brain of a person with AD. (Tobinick & Gross, 2008)

A definitive diagnosis can be made only upon meeting both the AD diagnostic criteria and histopathological confirmation. The identification of the disease in its earliest stage is still impossible, therefore more studies refining the definition of AD are needed (Dubois et al., 2007).

Treatment

There is no cure for Alzheimer's disease; however, there are medications slowing down its course available and psychosocial interventions. Caregiving is also referred to as a treatment.

- There are 4 drugs to treat main symptoms of AD. Three of them are cholinesterase inhibitors ("Drug treatments," n.d.), which block the function of acetylcholinesterase and thus prevent the hydrolysis of acetylcholine which makes it more available at the cholinergic synapses. These synapses are very important in the processes of learning, memory and attention (Geula & Mesulam, 1995). However, these drugs can be used only for a limited period of time during early to mid-stage of AD and they cannot stabilize all of the symptoms ("Drug treatments," n.d.). The fourth drug in use is a non-competitive antagonist of the N-methyl D-aspartate receptor for the chemical transmitter glutamate, which is neurotoxic in high concentrations. Hence, excessive activation of glutamate receptors is damaging (Danbolt, 2001). A number of other treatments have shown some promising results; however, they are not yet authorized for routine use ("Drug treatments," n.d.).
- Specific psychosocial treatments for dementia can be divided into four broad groups: behaviour oriented, emotion oriented, cognition oriented, and stimulation oriented. Mostly, these methods are not supported by controlled trials, however there are some favourable research findings and they have acquired clinical acceptance.(APA Work Group on Alzheimer's Disease and other Dementias, 2007)
- Caregiving is a very important part of the treatment. Caregiver is often someone from the family, but usually, as the disease advances, person with dementia ends up in a special establishment with professional help. It is important that the caregiver is taking care not only of the patient but also of himself. Very important is to understand the diagnosis, especially the prognosis and the clinical course of the disease. It was proved that the patients are less likely to get aggressive near the end of life when the caregiver is fully familiar with all the facts (Mitchell et al., 2009).

Prevention

There have been several studies about prevention of AD. However, there are not many clear results as such studies have many limitations (Scarmeas, Stern, Tang, Luchsinger, & Mayeux, 2006). The risk of AD may be reduced by physical activity that might decrease

vascular risk factors such as hypertension or hypercholesterolemia, promoting brain plasticity and/or neurotrophic factors, and reducing amyloid burden. Non-physical activity lowering AD risk could be some kind of cognitive activity building so called “cognitive reserve”. It can improve brain capability to manage on-going neuropathogenesis related to older age (Szekely, Breitner, & Zandi, 2007). Diet might influence the risk of developing AD as well, for example the Mediterranean diet is known to lower the possibility of getting this disease (Scarmeas et al., 2006).

Research directions

Currently, the most reliable research tools to study the new AD treatments are the mouse models. However, among the existing ones, none possesses all AD characteristics. Many AD models were generated in the past years as AD has various phenotypic features, which manifest differently in individual models. In nearly all of the models, cognitive deficits and amyloid plaques can be observed; however, it is not the case with NFTs, which were described only in mice with inserted gene for human tau. The main problem represents mimicking the neuronal loss that can be seen in only very few models (Hall & Roberson, 2012). Important data about most common transgenic mouse models are presented in Table 1.

One of the first models was a transgenic mouse overexpressing human amyloid precursor protein (hAPP) with the Indiana mutation called PDAPP (Games et al., 1995) and other lines containing hAPP transgene with different mutations were generated and widely used. Due to overexpression of hAPP, amyloid pathology typical for AD causing memory deficits is observable. However, these mouse lines do not exhibit neuronal loss (Hall & Roberson, 2012). Amyloid precursor protein gene does not produce only Aß, but also other biologically active elements. To eliminate their influence, lines overexpressing only Aß have been generated (P. A. Lewis et al., 2001).

Other AD mouse models are based on mutated presenilin gene (Hall & Roberson, 2012). Presenilin is a component of a specific secretase cleaving APP to its final products (Strooper et al., 1998). Mice carrying mutations in presenilin gene were crossed with hAPP transgenic mice and expressed extensive depositions of Aß and behavioural disorders (Savonenko et al., 2005). More common is the use of hAPP/PS1 mice carrying one single transgene created by co-injection of the presenilin and hAPP transgenes (Hall & Roberson, 2012).

Table 1 – Summary of the main transgenic mouse models of AD and their neuropathological features (Schaeffer, Figueiro, & Gattaz, 2011)

Mouse model	Gene (mutation)	Intraneuronal Aβ	Parenchymal Aβ plaques	Hyperphosphorylated Tau	Neurofibrillary tangles	Neuronal loss	Synaptic loss	CAA	Primary reference
PDAPP	APP (V717F)	-	Yes	Yes	No	No	Yes	-	Games et al. 1995
Tg2576	APP (K670N/M671L)	Yes	Yes	-	-	No	No	-	Hsiao et al. 1996
TgCRND8	APP (K670N/M671L, V717F)	-	Yes	-	No	No	-	-	Chishti et al. 2001
APP/PS1	APP (K670N/M671L), PS1 (M146L)	-	Yes	-	-	-	-	-	Holcomb et al. 1998
APP23	APP (K670N/M671L)	-	Yes	Yes	No	Little	Yes	Yes	Sturchler-Pierrat et al. 1997
Tg-SwDI	APP (E693Q, D694N)	-	Yes	-	-	-	-	Yes	Davis et al. 2004
APPDutch	APP (E693Q)	-	Little	-	-	-	-	Yes	Herzig et al. 2004
APPDutch/PS1	APP (E693Q), PS1 (G384A)	-	Yes	-	-	-	-	Little	Herzig et al. 2004
hAPP-Arc	APP (E693G, K670N/M671L, V717F)	-	Yes	-	-	-	-	Little	Cheng et al. 2004
Tg-ArcSwe	APP (E693G, K670N/M671L)	Yes	Yes	-	-	-	-	Yes	Lord et al. 2006 Knobloch et al. 2007
APPArc	APP (E693G)	-	Yes	-	-	-	-	Yes	Rönnbäck et al. 2011
TAPP	APP (K670N/M671L), Tau (P301L)	-	Yes	-	Yes	-	-	-	Lewis et al. 2001
3xTg-AD	APP (K670N/M671L), Tau (P301L), PS1 (M146V)	Yes	Yes	Yes	Yes	-	No	-	Oddo et al. 2003
APP_{SL}/PS1	APP (K670N/M671L, V717I), PS1 (M146L)	Yes	Yes	-	-	Yes	Yes	-	Wirths et al. 2002
APP/PS1KI	APP (K670N/M671L, V717I), PS1 (M233T/L235P)	Yes	Yes	-	-	Yes	Yes	-	Casas et al. 2004
5xFAD	APP (K670N/M671L, I716V, V717I), PS1 (M146L/L286V)	Yes	Yes	-	-	Yes	Yes	-	Oakley et al. 2006

CAA = cerebral amyloid angiopathy; Dash (-) = not reported.

To produce tau pathology next to plaque depositions, the hAPP/tau double transgenic mouse was generated by crossing the hAPP transgenic mice and mice expressing human mutated tau (J. Lewis et al., 2001).

A big milestone in AD research was generation of a triple-transgenic mouse model (3×Tg-AD) expressing presenilin, hAPP and tau transgenes. 3×Tg-AD mice gradually develop NFTs and amyloid plaques. Synaptic dysfunctions including long-term potentiation are observed even before accumulation of these histological signs. The deficiency in long-term synaptic plasticity is associated with the aggregation of intraneuronal Aß (Oddo et al., 2003).

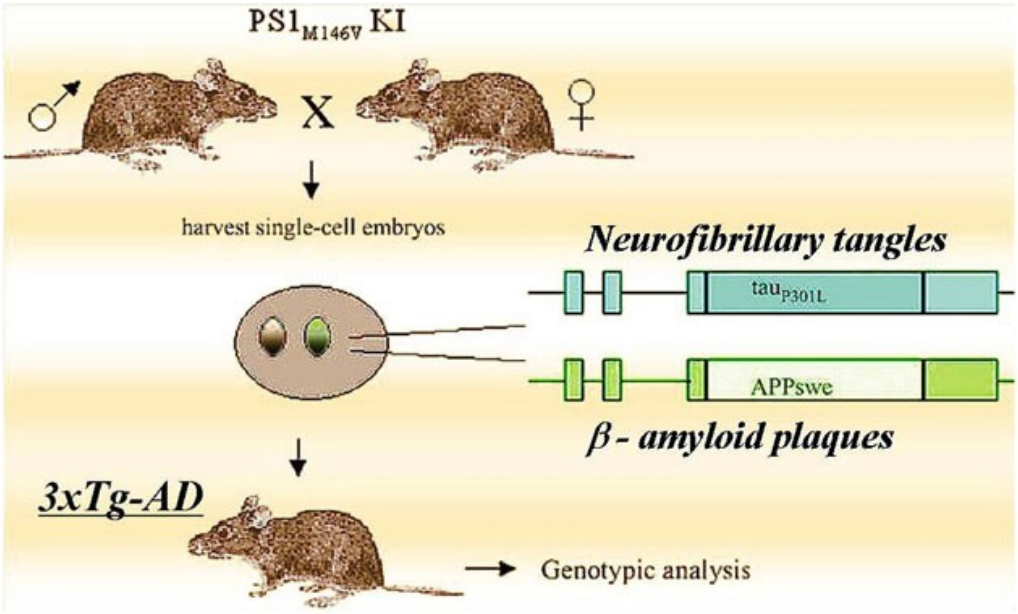

Figure 6 –Development of 3 × Tg-AD mice. First, single-cell embryos need to be harvested from mutant homozygous PS1M146Vknockin mice. Two independent transgene constructs encoding human APPSwe and tauP301L (4R/0N) (both under the control of the mouse Thy1.2 regulatory elements) are injected into them using the pronuclear microinjection technique. Then, the embryos are reimplanted into foster mothers. Finally, the offspring are genotypically analyzed to distinguish the 3xTg-AD mice. (Oddo et al., 2003)

Hundreds of clinical trials of various immunotherapy approaches are being in progress worldwide and the research is oriented mainly towards treatment of what is for now considered the main AD pathologies, and that is Aß deposits and NFTs (Delrieu, Ousset, Caillaud, & Vellas, 2012).

Numerous studies are specialized on vaccines against Aß peptide in AD transgenic mice that brought good results and hopes for an effective treatment of AD. The method is based on Aß immunization in mice. It improves cognitive dysfunctions by promoting clearance of Aß plaques via specific anti-Aß antibodies. As Aß immunotherapy diminishes

both extracellular Aß plaques and intracellular Aß accumulation which is linked to NFTs generation, it also significantly decreases tau pathology in early stages (Solomon, 2009). Despite these promising results, there have already been several studies doubting the efficacy of Aß vaccination (Nemirovsky, Shapiro, Baron, Kompaniets, & Monsonego, 2012). Various mechanisms are responsible for the removal of Aß depositions, and further understanding of these mechanisms is necessary for the development of an optimal therapeutic approach.

Some studies are orientated towards tau aggregation inhibitors however, these represent only an additional support in treatment of AD (Bulic, Pickhardt, Mandelkow, & Mandelkow, 2010).

Glial cells

Glial cells are important components of central and also peripheral nervous system, without them, the nervous system wouldn't be functional. Besides providing physical support to neurons, their role is also to respond to injury, regulate the ionic and chemical composition of the extracellular milieu, participate in the blood-brain barrier and blood-retinal barrier, form the myelin insulation of nervous pathways, guide neuronal migration during development, and exchange metabolites with neurons. On the glial cell membrane, there are high-affinity systems for ion/neurotransmitter uptake such as voltage-dependent K^+, Cl^- ion channels, neurotransmitter-gated ion channels and transporters. These cells are even able to release neurotransmitters ("Neuroglia" 1999). In contrast to neurons, they are not able to generate action potentials; however, they are excitable non-electrically (Volterra & Meldolesi, 2005).

Unlike neurons, glial cells keep their ability to divide. Some of the glial cells have conserved the stem cell properties and are engaged not only in gliogenesis but also in neurogenesis. These are mainly astrocytes of the germinal centers of adult brain. Not only they function as the precursor cells, astrocytes may also regulate the rate of proliferation of hippocampal stem cells, and encourage the generation of new neuronal cells (Rodríguez & Verkhratsky, 2011).

Since AD is the disease affecting central nervous system (CNS), following chapters will focus on some of the main glial cell types residing in the CNS, namely microglia, astrocytes, oligodendrocytes and recently discovered synantocytes.

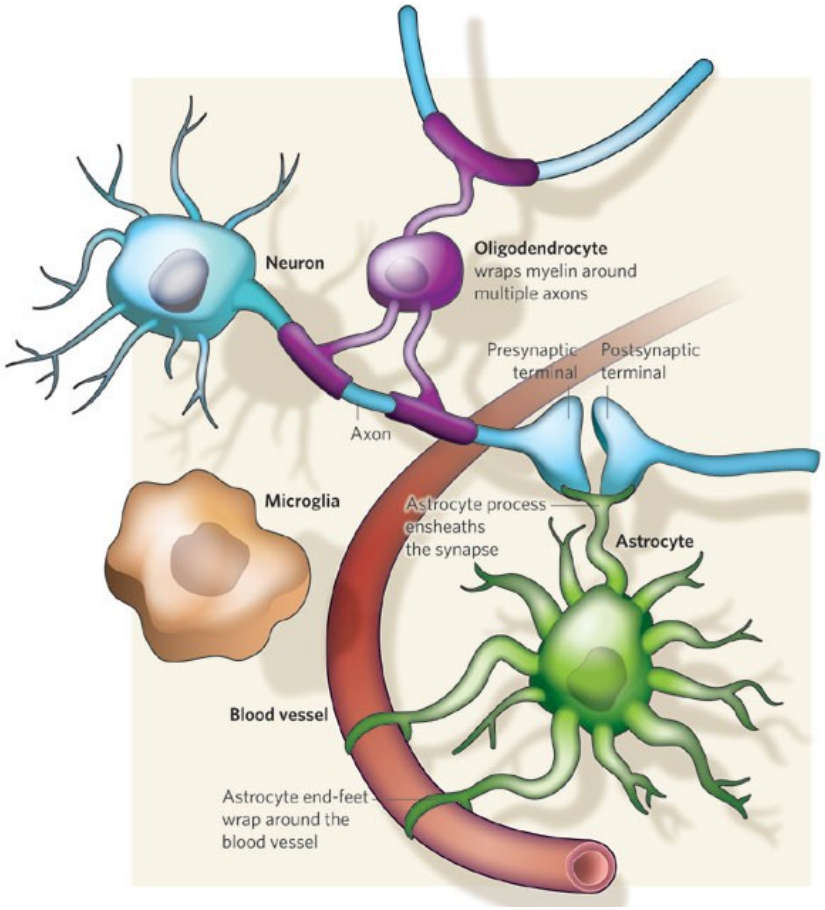

Figure 7–Schematic description of neuron-glia interactions in CNS. Oligodendrocytes provide myelination for neurons, microglia work as effectors of immune system and astrocytes contact blood vessels and synapses. (Allen & Barres, 2009)

Microglia

Unlike the majority of cells in the CNS that are derived from ectoderm, microglial cells are of mesodermal origin. Shortly after birth, they migrate into the CNS where they disperse and transform into resting microglia with particular morphological structure. That is a small cell soma with little perinuclear cytoplasm, and a portion of fine, branched processes that are covered in slender protrusions (Rodríguez, Witton, Olabarria, Noristani, & Verkhratsky, 2010). They represent the main effectors of immune system surveillance in CNS, working as resident macrophages. They undergo many changes of states according to current health situation of the organism, from surveillant mode to effector microglia (Ransohoff & Perry, 2009).

In the normal physiological conditions, microglial processes are constantly scanning the microenvironment within their anatomical domains. Each of these non-overlapping domains is occupied by a single microglial cell and they never protrude with neighboring microglia (Heneka et al., 2010). Microglial activation can be induced by various stimuli, for

example serum constituents or alterations in neurotransmitter (e.g. glutamate) concentrations (Ransohoff & Perry, 2009). As almost every other cell of immune system, microglia in their activated state are able to proliferate, move and perform phagocytosis. In addition, they function also as secretory cells, releasing cytokines, chemokines, proteolytic enzymes, reactive oxygen species (ROS), complement proteins and other molecules, which may further influence other microglia and astrocytes to sustain ongoing inflammation. Being a part of the immune system, microglial cells possess membranes equipped with receptors and other molecules important for communication with other types of immune cells and interaction with relevant molecules (Lue, Walker, & Rogers, 2001).

Microglia play a key role in maintaining healthy CNS, but their prolonged activation can lead to neurotoxicity and must be therefore tightly regulated (Ransohoff & Perry, 2009).

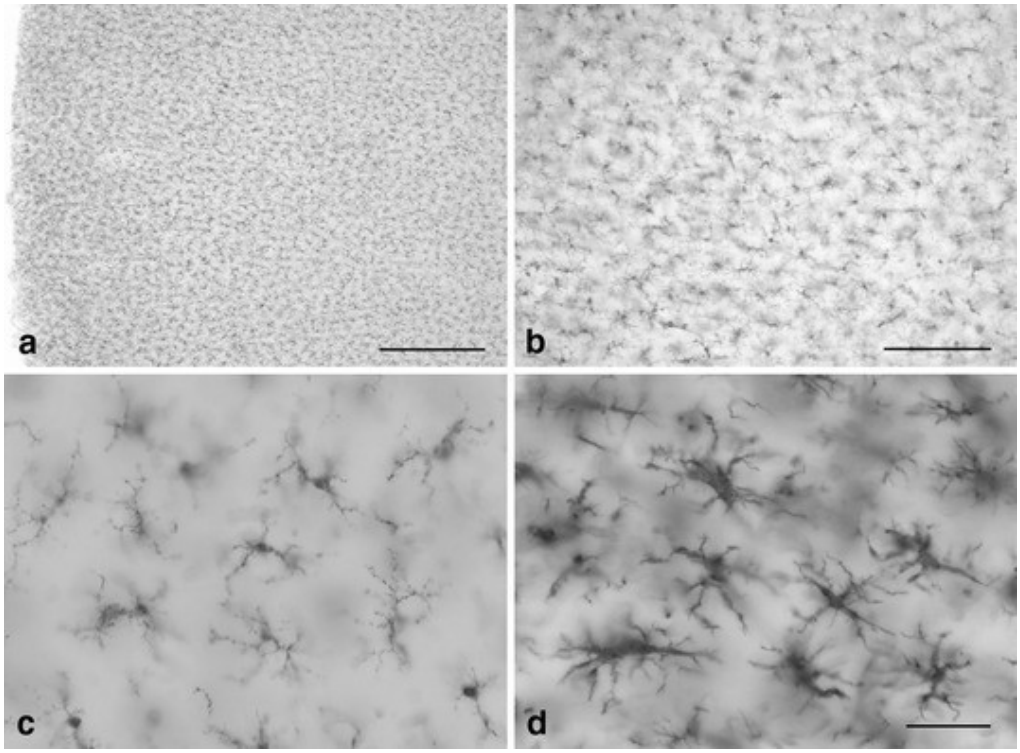

Figure 8–Microglia from archival human brain tissue stained using anti-iba1 immunohistochemistry. Using low magnification (scale bar a-500µm and b-100µm), we observe an abundance of resting microglia in the cortical grey matter of the entorhinal cortex in a non-pathological control subject. Note well-defined non-overlapping anatomical domains. Images c and d (scale bar 50µm) represent resting and activated microglia, respectively. The main difference is cytoplasmic hypertrophy in activated microglia. (Streit, Braak, Xue, & Bechmann, 2009)

Astrocytes

Astrocytes are referred to as "brain glue", as they have an important structural role. They serve as a scaffold for neuronal distribution and interactions. But they fulfill a variety of other functions, one of them being the maintenance of optimal environment for neuronal functioning. Similarly to microglia, each astrocyte controls a distinct domain that might enclose thousands of synapses and blood vessels in the astrocyte vicinity (Volterra & Meldolesi, 2005). Two major types of astroglia are distinguished in CNS based on their morphology; protoplasmic and fibrous astrocytes. Fibrous astrocytes, located mainly in white matter, have long unbranched processes and they have more regular contours than protoplasmic astrocytes with short and highly branched processes. The latter can be found mostly in gray matter (Miller & Raff, 1984). Concerning immunohistochemistry, antibodies that recognize glial fibrillary acidic protein (GFAP) are used to label thick main processes of fibrous astrocytes since GFAP shows more predominant expression in white matter. On the other hand, aldehyde dehydrogenase 1 family, member L1 (ALdh1L1) gene is expressed in every astrocytic cell, that is why polyclonal antibodies against ALdh1L1 protein is used as universal marker for astroglia (Cahoy et al., 2008).

Astrocytes function as the bridging units in CNS, due to their ability to integrate and respond to non-electrical excitatory external inputs by generation of Ca^{2+} waves and gliotransmitter release. Thus, they might interconnect neurons, vascular cells and distant glia (Volterra & Meldolesi, 2005). They also contribute to the control of the extracellular concentrations of ions, metabolites and neuroactive molecules. The regulation of extracellular K^{+} is particularly important as excessive amount of this ion interferes with neuronal excitability. Fluxes of ions necessarily provoke movement of water and astrocytes are crucial for maintaining water homeostasis of the CNS. In addition, astroglia regulate extracellular levels of the main excitatory neurotransmitter, glutamate, which is neurotoxic in high levels (Heneka et al., 2010). Glutamate released during synaptic activity is taken up by astrocytic glutamate transporters and further converted by glutamine synthase to glutamine, which is then transported back to the neuronal presynaptic terminal, where it is converted into glutamate and stored in synaptic vesicles (Shen, 2013). Astrocytes also play a key role in energetic metabolism of the CNS. They mediate the transport of glucose from blood to neurons and are able to stock it in the form of glycogen (Kreft, Bak, Waagepetersen, & Schousboe, 2012). In addition, they are able to produce lactate as an alternative source of energy for neurons (Bouzier-Sore, Merle, Magistretti, & Pellerin, 2002).

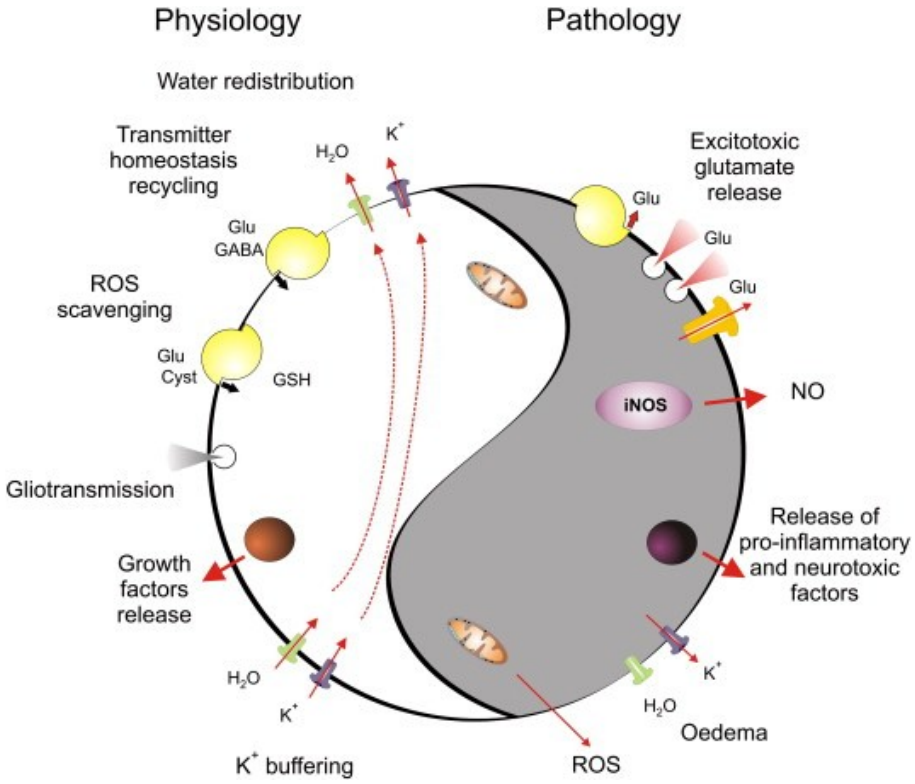

Figure 9 – Homeostatic cascades maintained by astrocytes in normal conditions and under metabolic stress. In the first case, astrocytes contribute to water homeostasis, K^+ spatial buffering, uptake of neurotransmitter glutamate or ROS scavenging. In case of CNS disorders, these processes can be disrupted, causing an additional brain/spinal cord damage. Failures in glutamate uptake lead to glutamate excitotoxicity; high levels of K^+ induce overexcitation of neurons; ineffective water transport promotes oedema and release of ROS supports inflammation. (Heneka et al., 2010)

Oligodendrocytes

The main function of oligodendrocytes (OLs) is to form myelin sheath around axons. During the CNS maturation, there are more OLs produced than is required to provide myelination. Neuronal axons are supplying OLs ensheathing them with necessary survival factors, while those OLs, which do not receive survival signals, undergo apoptosis. Myelination is an essential process and excessive OLs death might have devastating consequences (depending on the extent and brain region) because it may cause impaired neuronal signaling (McTigue & Tripathi, 2008).

Myelination is not the only function of OLs. They secrete soluble mediators that induce the clustering of the neuronal sodium channels, which is fundamental for saltatory conduction of nerve impulses. In addition, OLs release trophic factors for neuronal cell bodies as was observed in dopaminergic, cerebellar, and embryonic cortical neurons (McTigue & Tripathi, 2008).

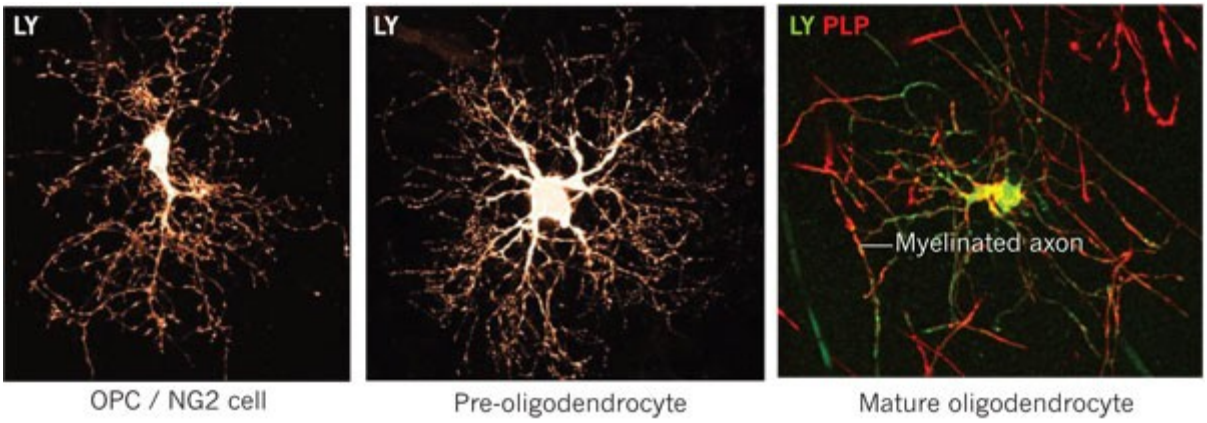

Figure10 –Development of OLs. (Nave, 2010)

Synantocytes

Synantocytes are also referred to as polydendrocytes or NG2-glia because they express the NG2 chondroitin sulphate proteoglycan. They are generally perceived as OL progenitor cells. Several recent studies using transgenic mice showed that these cells are able to generate OLs, astrocytes(Zhu, Bergles, & Nishiyama, 2008) and even neurons(Rivers et al., 2008).

Synantocytes have a centrally placed cell body and numerous primary processes which overlap slightly with processes of neighboring synantocytes, but are not interconnected via gap junctions. They are present in both white and grey matter. In the white matter, they are scattered, together with astrocytes, between the rows of oligodendrocytes. In the gray matter, they envelope neuronal cell bodies and their processes are in contact with numerous synapses. Synantocytes might also participate in the perineuronal net formation by producing various extracellular matrix molecules stabilizing synapses. They might also influence synapses by their putative ability to guide axonal growth (Butt, Hamilton, Hubbard, Pugh, & Ibrahim, 2005). They express sufficient number of voltage-gated Na^+ channels to generate action potentials (Heneka et al., 2010). Whether these channels are present on membranes of each synantocyte or just of some, e.g. in some specific regions, is not yet entirely established and several studies are giving contradictive results (De Biase, Nishiyama, & Bergles, 2010; Káradóttir, Hamilton, Bakiri, & Attwell, 2008). Also the function of the channels is not yet exactly known, besides generating action potential they might have numerous other functions (Frohlich, Nagy, Hovhannisyan, & Kukley, 2011).

Several studies have proved that synantocytes are capable of proliferation in the adult rat CNS and that they represent the major percentage of cycling cells in healthy adult rat brain (Dawson, Polito, Levine, & Reynolds, 2003; Horner et al., 2000). Proliferation of

synantocytes can be also observed during injury and inflammation. These activated cells might give rise to new astrocytes or oligodendrocytes (Levine, Reynolds, & Fawcett, 2001). The increase in their number is also very important during the process of remyelination (Levine & Reynolds, 1999). The functions of non-dividing synantocytes that do not give rise to OLs are unknown. It is believed that the functions of synantocytes and astrocytes are different since their communication pathways are overlapping, hence same functions would be highly redundant (Wigley & Butt, 2009).

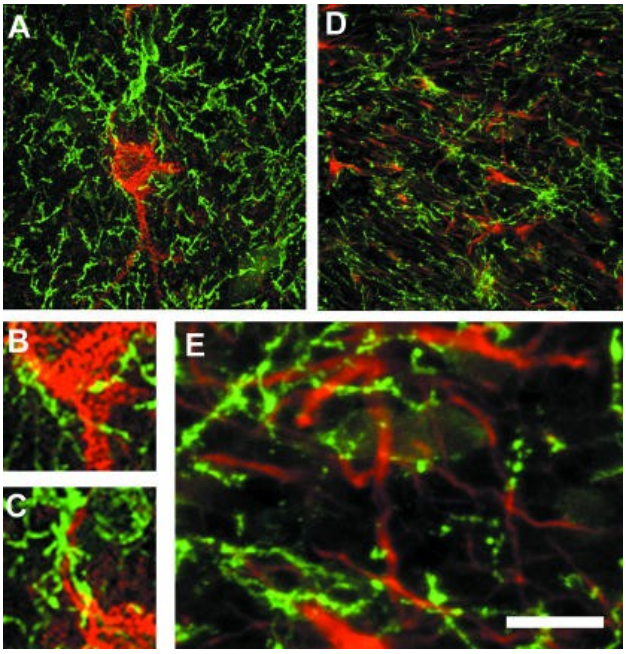

Figure 11 –Synantocytes in the cortex (A-C) and the optic nerve (D, E). The tissue was stained by double immunofluorescent labelling for NG2 proteoglycan (green) and neuronal marker calbindin (red, A-C) or astrocytic marker GFAP (red, D,E). These images show numerous contact points between synantocytes and neurons or astrocytes. Multiple synantocytes contact the soma (A), axon (B) and dendrites (C) of single neuron. D and E represent synantocytes and astrocytes dispersed in white matter. Astrocytes and NG2 glia can contact each other. Scale bar in A and D is 50 μm and in B, C, and E 12.5 μm. (Butt et al., 2005)

Glial cells in AD

Glial cells carry out a number of tasks that are crucial for normal brain function - they maintain brain homeostasis, provide energetic supply for neurons, protect them from various neurotoxic elements etc.; however in damaged/degenerating CNS their functions can be markedly altered. The loss/alteration of glial cell functions might ultimately lead to further damage of surrounding tissue (Heneka et al., 2010).

Microglia

The severity of the inflammatory response in AD corresponds to the amount of activated microglia. Microglial activation is induced upon stimulation of various receptors such as receptor for advanced glycation end products or the class A macrophage scavenger receptor, both for modified or degraded proteins including Aß (Lue et al., 2001).

Resting microglia and activated microglia have different functions, which changes throughout the disease. Microglial activation can be both beneficial and harmful. In the early stage of the disease, activated microglia are probably neuroprotective and delay the course of AD, especially due to their capability of phagocytosis which allows them to clear Aβ and degrade neuritic plaques. The actual mechanism of phagocytosis is influenced by the nature of Aβ, i.e. whether it is fibrillar or soluble (Solito & Sastre, 2012).

In vitro experiments have shown that cultured microglia are able to phagocytize Aß. However, they are not capable of effective phagocytosis of the excessive amount of Aβ accumulated during the progression of AD. This chronic struggle of microglia to digest enormous quantity of Aβ may promote the inflammatory processes in AD (Lue et al., 2001) and in later stages of the disease, microglial sustained production and release of proinflammatory cytokines leads to neuronal damage. In addition to their role in the immune response, inflammatory molecules, such as cytokines and chemokines, are able to alter neuronal functions. There is evidence that overactivated microglia can trigger uncontrolled inflammation, contributing to AD progression by promoting Aβ aggregation and inducing neuronal death (Solito & Sastre, 2012).

Furthermore, neuroinflammation is engaged in tau-mediated neurodegeneration. Tau-positive nerve cells in animal models of tauopathy such as the P301S tau transgenic mice (expressing P301S tau gene mutation) are surrounded by activated microglial cells. Tau phosphorylation is induced by specific kinases. These can be modified by several proinflammatory cytokines. For example the substrate specificity of kinases/phosphatases leading to tau phosphorylation may be altered and these changes would lead to hyperphosphorylation of Tau protein and creation of NFTs at pathological sites (Solito & Sastre, 2012).

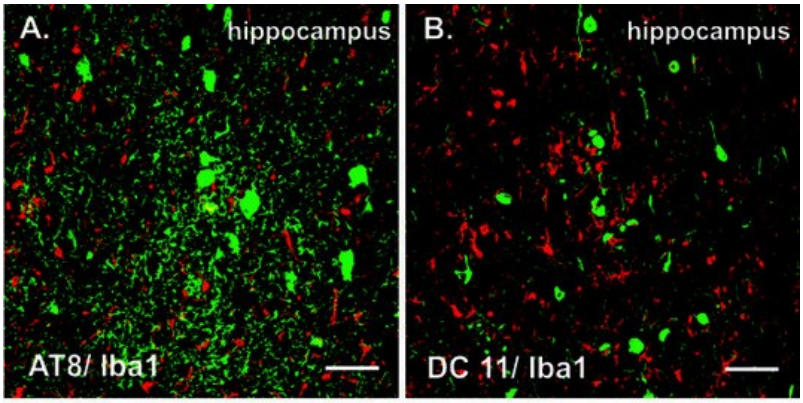

Figure 12 – NFTs surrounded by activated microglia in the hippocampus of an AD patient. Activated microglia are stained by Iba1 antibody (red) and NFTs by AT8 (tagging phospho-tau) and DC11 (recognising AD-specific misfolded tau) (both green). Scale bars: 100 μm. (Zilka et al., 2012)

Astrocytes

Astrocytic reactions to the AD are heterogeneous, depending on their location in respect to amyloid plaques. Peri-plaque astrocytes undergo reactive gliosis, while distant astrocytes become atrophic (Rodríguez, Olabarria, Chvatal, & Verkhratsky, 2009).

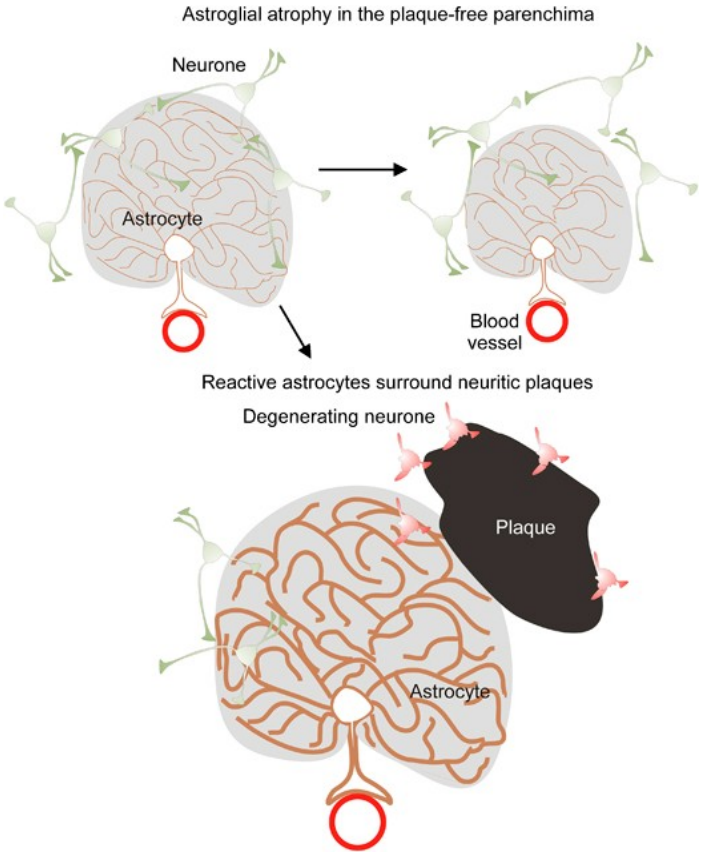

Figure 13 – Astroglia in AD – hypothesis. (Rodríguez et al., 2009)

One of the indicators of astrogliosis is an increased level of GFAP which is linked with accumulation of plaques, and to some extent with the amount of NFTs. It is inversely correlated with cognitive functions. In both human brain affected by AD and in AD mouse models, the presence of amyloid depositions is connected to the presence of reactive astrocytes strongly expressing GFAP. This astrocytic activation has devastating effects. It causes increase in neurotoxic substances such as glutamate and therefore an increase in neuronal loss. Furthermore, reactive astrocytes can form a glial scar, which inhibits CNS reparation. On the other hand, gliosis has also beneficial effects. Healthy functioning astrocytes play a key role in glutamate uptake, thus lowering its neurotoxicity. They also surround and isolate affected tissue, which helps to limit the inflammation (Kamphuis et al., 2012). Hence, reactive astrocytes are supposed to be beneficial and help with the clearance and degradation of amyloid deposits. However, later experiments have shown that only astrocytes from healthy brain are able to do so. Astrocytes derived from the brain of APP transgenic mouse weren't capable of Aß degradation. Moreover, conditions developed by AD may even turn astrocytes to Aß producers (Verkhratsky, Olabarria, Noristani, Yeh, & Rodriguez, 2010). Other astrocytic functions are affected by astrogliosis during AD as well and astrocytes can lose entirely all the beneficial effects. Processes contributing to homeostatic maintenance, such as glutamate uptake, or the transport of substances between blood vessels, astrocytes and neurons can be impaired as a result of changes in reactive astrocytes (Heneka et al., 2010). Extracellular Aß can modulate $[Ca^{2+}]_i$ oscillations in proximate astrocytes and provoke spontaneous Ca^{2+}signaling, which was shown to be neurotoxic. Activated astrocytes also contribute to neuroinflammation, since they release cytokines and other proinflammatory molecules and ROS (Verkhratsky et al., 2010).

As mentioned above, astrocytes distant to the amyloid deposits and plaques undergo atrophy. The reduction of primary, secondary and distal processes of astrocytes is observable before all the other signs of AD, already in 1 month old 3xTg-AD mice. Astrocytes remain atrophic until advanced age (12 months), and additionally, Aß plaques and NFTs develop. Astrocytes that lose their processes can no longer support synapses, what can lead to synaptic loss and form the basis for cognitive and memory impairments seen in the early stages of AD (Yeh, Vadhwana, Verkhratsky, & Rodríguez, 2011). Moreover, astrocytic atrophy may lead to homeostatic imbalance. Brain homeostasis is crucial for proper CNS functioning and its disturbances increase neuronal vulnerability and damage, e.g. by glutamate excitotoxicity (Rodríguez et al., 2009).

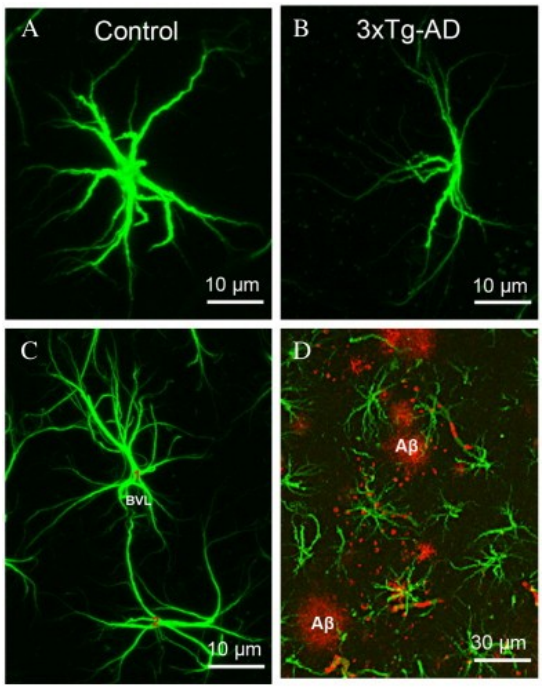

Figure 14 – Astrogliosis and astroglial atrophy during the development of AD pathology in the brains of 3xTg-AD mice. Confocal micrographs show normal astrocytes in control animals (A, C) in comparison with atrophic astrocyte from 3xTg-AD mouse (B) and activated astrocytes from 3xTg-AD mouse undergoing astrogliosis (D) induced by the Aβ accumulation (dual labelling was used, GFAP in astrocytes is shown in green, Aβ in red). The atrophic astrocyte (B) has not only reduced cell body size but also decreased number of processes.(Heneka et al., 2010)

Oligodendrocytes

Generally, it is believed that degeneration caused by AD occurs mainly in the grey matter. However, various studies proved that AD also affects the white matter in animal models of AD as well as human brain, where it is manifested as a loss of myelination and glial cells, especially OLs (Horiuchi et al., 2012). It may be due to the ability of Aβ to damage with particular severity membranes that are rich in cholesterol, such as oligodendrocytic membranes (Roth, Ramírez, Alarcón, & Von Bernhardi, 2005). Reduction or loss of OLs capability to form and maintain myelin sheaths would severely affect the conduction of signal in CNS, leading to general malfunction of the neural network and eventually, neurodegeneration (Horiuchi et al., 2012). Even if the damage of myelin isn't a direct consequence of Aß accumulation but is caused secondarily by e.g. lack of survival signals from degenerated axons, the loss of trophic support from the OLs increases the vulnerability of neurons, inflammation, and further cell damage (Roth et al., 2005).

Synantocytes

As already mentioned, synantocytes are not only glial, but also neuronal precursors (Rivers et al., 2008). Neurogenesis during AD might be beneficial and could even reverse the course of the disease. However, *in vitro* experiments show that increased levels of Aβ inhibit neurogenesis in both AD progenitor cells and healthy control progenitor cells treated with Aβ, the induction of new neuronal cells was depressed. This is happening mainly through mechanism of β-catenin signaling interference, which is diminishing expression of proneural genes (Xu, Zhao, & Li, 2011).

Wnt/β-catenin signaling cascade plays important role in neuronal induction (Lie et al., 2005). β-catenin together with other necessary elements forms a complex that is fundamental for regulation of transcription of target genes of Wnt/β-catenin signaling cascade, such as genes inducing neurogenesis (Xu et al., 2011).

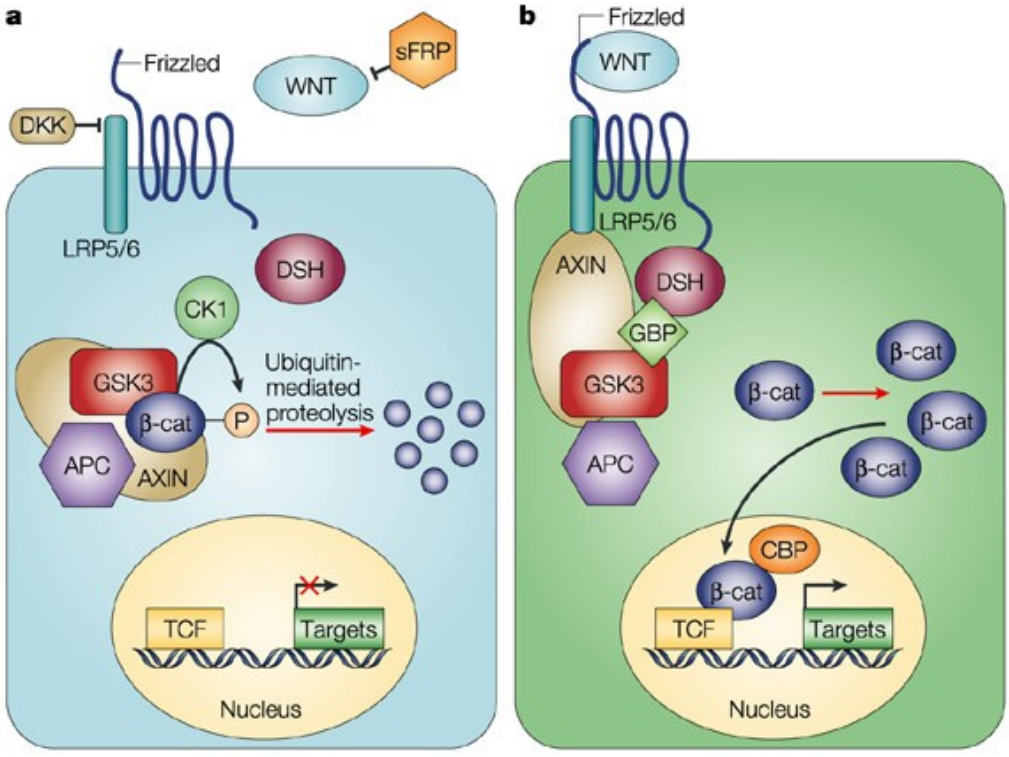

Figure 15 - Wnt/β-catenin signalling. In the first case (a), there is no Wnt bound to the membrane receptor of the Frizzled family, β-catenin is degraded and target genes are not transcribed. When Wnt binds to its receptor (b), β-catenin doesn't undergo degradation and enters the nucleus where it activates the transcription, together with other elements. APC, adenomatous polyposis coli; β-cat, β-catenin; CBP, CREB-binding protein; CK, casein kinase; DKK, Dickkopf; DSH, Dishevelled; GBP, GSK3-binding protein; GSK, glycogen synthase kinase; LRP, LDL-receptor-related protein; P, phosphorylation; sFRP, secreted Frizzled-related protein; TCF, T-cell factor.(Moon, Kohn, Ferrari, & Kaykas, 2004)

Accumulation of Aβ leads to increased activation of glycogen synthase kinase 3 (GSK-3), an enzyme that among other things phosphorylates β-catenin, causing is degradation (Moon et al., 2004). Even very small amount of Aβ can increase the level of GSK-3 and thus decrease the amount of β-catenin. This mechanism leads to downregulation of proneural gene transcription and neurogenesis abolition (He & Shen, 2009).

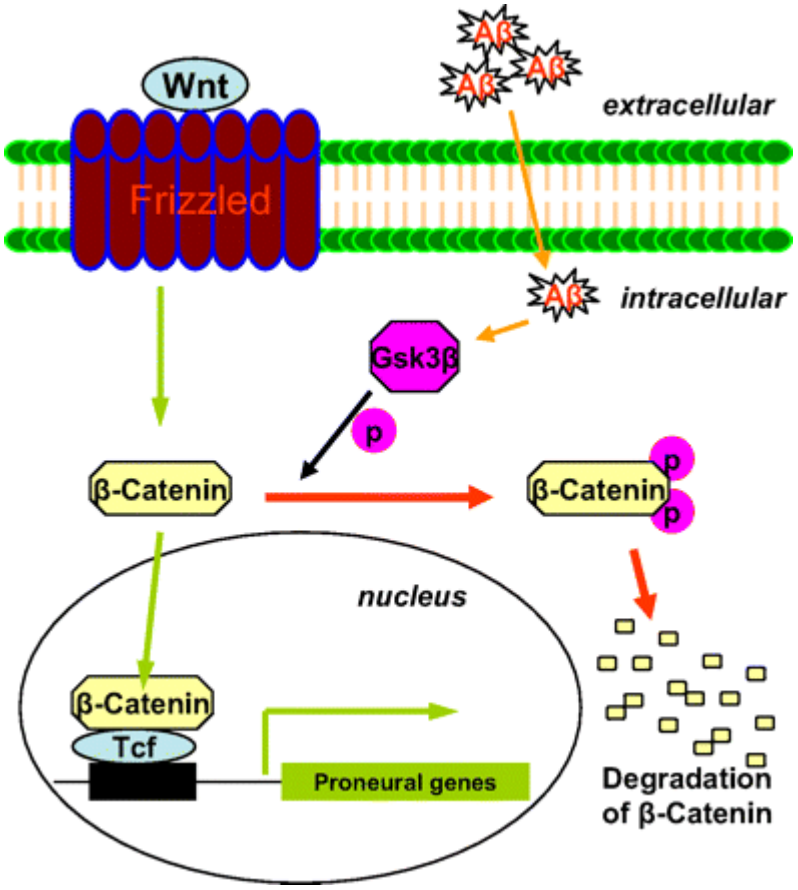

Figure 16 – Schematic representation of how Aβ accumulations lead to interruption of Wnt/β-catenin signalling. Aβ increases activation of GSK-3 that enhance phosphorylation and degradation of β-catenin. Reduced β-catenin signalling then results in down regulated expression of target proneural genes. (He & Shen, 2009)

In conclusion, although synantocytes are still present in the brains of patients with AD, they lose their capability for multipotential differentiation and are no more able to generate new neurons(Xu et al., 2011).

Conclusion

Alzheimer's disease is a devastating disorder caused by multiple factors many of which have to be further studied. In several stages, patients are gradually losing memory and basic abilities for activities of daily living due to neurodegeneration caused mainly by aggregation of β amyloid and tau protein. There is no cure, only few treatments that could slow down the progress of the disease for some period of time.

Based on recent findings, glial cells can be beneficial and slow down the course of Alzheimer's disease by maintaining proper brain homeostasis, immune response, myelination of the neurons and many other functions they ensure. However, overreaction or malfunctioning of glial cells can seriously worsen the damage of the affected brain. Uncontrolled inflammation can occur or they can also support the aggregation of β amyloid, ergo the formation of neuritic plaques.

Newly discovered synantocytes might play a key role in neurogenesis during Alzheimer's disease what could notably change the course of the disease and slower if not even stop the impact of neurodegeneration on memory and cognition. How exactly synantocytes function, is not yet fully understood. However, it is known that their capability of neurogenesis is stopped by the presence of aggregation of β amyloid through Wnt/β-catenin signaling. Therefore more studies are necessary so this process could be reversed and that might lead to finally defeating Alzheimer's disease, or at least markedly slowing down it progress.

Bibliography

Akre, J. (2008). MRI May Be Useful For Early Alzheimer's Diagnosis. *The Legal Examiner*, 1.

Allen, N. J., & Barres, B. A. (2009). Glia - more than just brain glue. *Nature*, 457, 675–677.

Altamura, C., Squitti, R., Pasqualetti, P., Tibuzzi, F., Silvestrini, M., Ventriglia, M. C., Cassetta, E., et al. (2007). What is the relationship among atherosclerosis markers , apolipoprotein E polymorphism and dementia ? *European Journal of Neurology*, 14, 679–682. doi:10.1111/j.1468-1331.2007.01714.x

APA Work Group on Alzheimer's Disease and other Dementias. (2007). American Psychiatric Association practice guideline for the treatment of patients with Alzheimer's disease and other dementias. Second edition. *The American Journal of Psychiatry*, 5–56.

Bouzier-Sore, A.-K., Merle, M., Magistretti, P. J., & Pellerin, L. (2002). Feeding active neurons: (re)emergence of a nursing role for astrocytes. *Journal of physiology, Paris*, 96(3-4), 273–82.

Brain Tour. (n.d.). Retrieved January 6, 2013, from http://www.alz.org/braintour/healthy_vs_alzheimers.asp

Bulic, B., Pickhardt, M., Mandelkow, E., & Mandelkow, E. (2010). Tau protein and tau aggregation inhibitors. *Neuropharmacology*, 59, 276–289. doi:10.1016/j.neuropharm.2010.01.016

Butt, A. M., Hamilton, N., Hubbard, P., Pugh, M., & Ibrahim, M. (2005). Synantocytes: the fifth element. *Journal of anatomy*, 207(6), 695–706. doi:10.1111/j.1469-7580.2005.00458.x

Bäckman, L., Jones, S., Berger, a-K., Laukka, E. J., & Small, B. J. (2004). Multiple cognitive deficits during the transition to Alzheimer's disease. *Journal of internal medicine*, 256(3), 195–204. doi:10.1111/j.1365-2796.2004.01386.x

Cahoy, J. D., Emery, B., Kaushal, A., Foo, L. C., Zamanian, J. L., Christopherson, K. S., Xing, Y., et al. (2008). A Transcriptome Database for Astrocytes , Neurons , and Oligodendrocytes : A New Resource for Understanding Brain Development and Function. *The Journal of Neuroscience*, 28(1), 264–278. doi:10.1523/JNEUROSCI.4178-07.2008

Cosentino, S., Scarmeas, N., Helzner, E., Glymour, M. M., Brandt, J., Albert, M., Blacker, D., et al. (2008). APOE ε4 allele predicts faster cognitive decline in mild Alzheimer disease. *Neurology*, 70, 1842–1849.

Cummings, B. J., Pike, C. J., Shankle, R., & Cotman, C. W. (1996). ß-Amyloid deposition and other measures of neuropathology predict cognitive status in Alzheimer's disease. *Neurobiology of aging*, 17(6), 921–933.

Danbolt, N. C. (2001). Glutamate uptake. *Progress in Neurobiology*, 65, 1–105.

Dawson, M. R. L., Polito, A., Levine, J. M., & Reynolds, R. (2003). NG2-expressing glial progenitor cells : an abundant and widespread population of cycling cells in the adult rat CNS. *Molecular and Cellular Neuroscience*, 24, 476–488. doi:10.1016/S1044-7431(03)00210-0

De Biase, L. M., Nishiyama, A., & Bergles, D. E. (2010). Excitability and Synaptic Communication within the Oligodendrocyte Lineage. *The Journal of Neuroscience*, 30(10), 3600–3611. doi:10.1523/JNEUROSCI.6000-09.2010.Excitability

Delrieu, J., Ousset, P. J., Caillaud, C., & Vellas, B. (2012). "Clinical trials in Alzheimer"s disease': immunotherapy approaches. *Journal of Neurochemistry*, 120, 186–193. doi:10.1111/j.1471-4159.2011.07458.x

Dementia. (n.d.). Retrieved from http://www.mdguidelines.com/dementia/definition

Dickson, D. W. (1997). The pathogenesis of senile plaques. *Journal of neuropathology and experimental ...*, 56, 321–339.

Drug treatments. (n.d.). Retrieved from http://www.alz.co.uk/caring/drug-treatments

Dubois, B., Feldman, H. H., Jacova, C., Dekosky, S. T., Barberger-Gateau, P., Cummings, J., Delacourte, A., et al. (2007). Research criteria for the diagnosis of Alzheimer's disease: revising the NINCDS-ADRDA criteria. *Lancet neurology*, 6(8), 734–46. doi:10.1016/S1474-4422(07)70178-3

Frohlich, N., Nagy, B., Hovhannisyan, A., & Kukley, M. (2011). Fate of neuron–glia synapses during proliferation and differentiation of NG2 cells. *Journal of Anatomy*, 219, 18–32. doi:10.1111/j.1469-7580.2011.01392.x

Förstl, H., & Kurz, a. (1999). Clinical features of Alzheimer's disease. *European Archives of Psychiatry and Clinical Neuroscience*, 249(6), 288–290. doi:10.1007/s004060050101

Games, D., Adams, D., Alessandrini, R., Barbour, R., Berthelette, P., Blackwell, C., Carr, T., et al. (1995). Alzheimer-type neuropathology in transgenic mice overexpressing V717F beta-amyloid precursor protein. *Nature*, 373, 523–527.

Geula, C., & Mesulam, M. (1995). Cholinesterases and the pathology of Alzheimer disease. *Alzheimer disease and associated ...*, 9, 23–28.

Hall, A. M., & Roberson, E. D. (2012). Mouse models of Alzheimer ' s disease. *Brain Research Bulletin*, 88, 3–12. doi:10.1016/j.brainresbull.2011.11.017

Haydon, P., & Carmignoto, G. (2006). Astrocyte control of synaptic transmission and neurovascular coupling. *Physiological reviews*, 1009–1031. doi:10.1152/physrev.00049.2005.

He, P., & Shen, Y. (2009). Interruption of ß-Catenin Signaling Reduces Neurogenesis in Alzheimer ' s Disease. *The Journal of Neuroscience*, 29(20), 6545–6557. doi:10.1523/JNEUROSCI.0421-09.2009

Heneka, M. T., Rodríguez, J. J., & Verkhratsky, A. (2010). Neuroglia in neurodegeneration. *Brain research reviews*, 63(1-2), 189–211. doi:10.1016/j.brainresrev.2009.11.004

Horiuchi, M., Maezawa, I., Itoh, A., Wakayama, K., Jin, L.-W., Itoh, T., & Decarli, C. (2012). Amyloid β1-42 oligomer inhibits myelin sheet formation in vitro. *Neurobiology of aging*, 33(3), 499–509. doi:10.1016/j.neurobiolaging.2010.05.007

Horner, P. J., Power, A. E., Kempermann, G., Kuhn, H. G., Palmer, T. D., Winkler, J., Thal, L. J., et al. (2000). Proliferation and Differentiation of Progenitor Cells Throughout the Intact Adult Rat Spinal Cord. *The Journal of Neuroscience*, 20(6), 2218–2228.

Kalaria, R., Maestre, G., Arizaga, R., Friedland, R. P., Galasko, D., Hall, K., Luchsinger, J. A., et al. (2008). Alzheimer's disease and vascular dementia in developing countries: prevalence, management, and risk factors. *Lancet neurology*, 7(9), 812–826. doi:10.1016/S1474-4422(08)70169-8.Alzheimer

Kamphuis, W., Mamber, C., Moeton, M., Kooijman, L., Sluijs, J. a, Jansen, A. H. P., Verveer, M., et al. (2012). GFAP isoforms in adult mouse brain with a focus on neurogenic astrocytes and reactive astrogliosis in mouse models of Alzheimer disease. *PloS one*, 7(8), e42823. doi:10.1371/journal.pone.0042823

Kreft, M., Bak, L. K., Waagepetersen, H. S., & Schousboe, A. (2012). Aspects of astrocyte energy metabolism, amino acid neurotransmitter homoeostasis and metabolic compartmentation. *ASN neuro*, 4(3), 187–199. doi:10.1042/AN20120007

Káradóttir, R., Hamilton, N. B., Bakiri, Y., & Attwell, D. (2008). Spiking and non-spiking classes of oligodendrocyte precursor glia in CNS white matter. *Nature Neuroscience*, 11(4), 450–456. doi:10.1038/nn2060.Spiking

Levine, J. M., & Reynolds, R. (1999). Activation and Proliferation of Endogenous Oligodendrocyte Precursor Cells during Ethidium Bromide-Induced Demyelination. *Experimental Neurology*, 160, 333–347.

Levine, J. M., Reynolds, R., & Fawcett, J. W. (2001). The oligodendrocyte precursor cell in health and disease. *Trends in neurosciences*, 24(1), 39–47.

Lewis, J., Dickson, D. W., Lin, W., Chisholm, L., Corral, A., Jones, G., Yen, S., et al. (2001). Enhanced Neurofibrillary Degeneration in Transgenic Mice Expressing Mutant Tau and APP. *Science*, 293, 1487–91. doi:10.1126/science.1058189

Lewis, P. A., Piper, S., Baker, M., Onstead, L., Murphy, M. P., Hardy, J., Wang, R., et al. (2001). Expression of BRI-amyloid ß peptide fusion proteins: a novel method for speci¢c high-level expression of amyloid ß peptides. *Biochimica et Biophysica Acta*, 1537, 58–62.

Lie, D., Colamarino, S. A., Song, H., Desire, L., Mira, H., Consiglio, A., Lein, E. S., et al. (2005). Wnt signalling regulates adult hippocampal neurogenesis. *Nature*, 437, 1370–1375. doi:10.1038/nature04108

Lue, L., Walker, D. G., & Rogers, J. (2001). Modeling microglial activation in Alzheimer ' s disease with human postmortem microglial cultures. *Neurobiology of aging*, 22, 945–956.

Maccioni, R. B., Muñoz, J. P., & Barbeito, L. (2001). The Molecular Bases of Alzheimer's Disease and Other Neurodegenerative Disorders. *Archives of Medical Research*, 32(5), 367–381. doi:10.1016/S0188-4409(01)00316-2

McTigue, D. M., & Tripathi, R. B. (2008). The life, death, and replacement of oligodendrocytes in the adult CNS. *Journal of neurochemistry*, 107(1), 1–19. doi:10.1111/j.1471-4159.2008.05570.x

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

Mitchell, S. L., Teno, J. M., Kiely, D. K., Shaffer, M. L., Jones, R. N., Prigerson, H. G., Volicer, L., et al. (2009). The Clinical Course of Advanced Dementia. *The New England Journal of Medicine*, 361(16), 1529–1538. doi:10.1056/NEJMoa0902234.The

Moon, R. T., Kohn, A. D., Ferrari, G. V. De, & Kaykas, A. (2004). Wnt and β-catenin signalling: diseases and therapies. *Nature Reviews Genetics*, 5, 689–699. doi:10.1038/nrg1427

Nagele, R. G., Wegiel, J., Venkataraman, V., Imaki, H., Wang, K.-C., & Wegiel, J. (2004). Contribution of glial cells to the development of amyloid plaques in Alzheimer's disease. *Neurobiology of aging*, 25(5), 663–74. doi:10.1016/j.neurobiolaging.2004.01.007

Nave, K. (2010). Myelination and support of axonal integrity by glia. *Nature*, 468, 244–252. doi:10.1038/nature09614

Nemirovsky, A., Shapiro, J., Baron, R., Kompaniets, A., & Monsonego, A. (2012). Active Aβ vaccination fails to enhance amyloid clearance in a mouse model of Alzheimer's disease with Aβ42-driven pathology. *Journal of neuroimmunology*, 247(1-2), 95–9. doi:10.1016/j.jneuroim.2012.03.017

Neuroglia. (1999). Retrieved from http://www.nlm.nih.gov/cgi/mesh/2011/MB_cgi?mode=&term=Glia

Oddo, S., Caccamo, A., Shepherd, J., Murphy, M. P., Kayed, R., Mattson, M. P., LaFerla, F. M., et al. (2003). Triple-transgenic model of Alzheimer's disease with plaques and tangles: intracellular AÎ^2 and synaptic dysfunction. *Neuron*, 39, 409–421.

Perl, D. P. (2010). Neuropathology of Alzheimer ' s Disease. *Mount Sinai Journal of Medicine*, 77(1), 32–42. doi:10.1002/msj.20157.Neuropathology

Ransohoff, R. M., & Perry, V. H. (2009). Microglial physiology: unique stimuli, specialized responses. *Annual review of immunology*, 27, 119–45. doi:10.1146/annurev.immunol.021908.132528

Rivers, L. E., Young, K. M., Rizzi, M., Jamen, F., Psachoulia, K., Wade, A., Kessaris, N., et al. (2008). PDGFRA / NG2 glia generate myelinating oligodendrocytes and piriform projection neurons in adult mice. *Nature Neuroscience*, 11(12), 1392–1402. doi:10.1038/nn.2220

Rodríguez, J., Olabarria, M., Chvatal, A., & Verkhratsky, A. (2009). Astroglia in dementia and Alzheimer's disease. *Cell death and differentiation*, 16(3), 378–85. doi:10.1038/cdd.2008.172

Rodríguez, J., & Verkhratsky, A. (2011). Neurogenesis in Alzheimer's disease. *Journal of anatomy*, 219(1), 78–89. doi:10.1111/j.1469-7580.2011.01343.x

Rodríguez, J., Witton, J., Olabarria, M., Noristani, H. N., & Verkhratsky, A. (2010). Increase in the density of resting microglia precedes neuritic plaque formation and microglial activation in a transgenic model of Alzheimer's disease. *Cell death & disease*, 1, e1. doi:10.1038/cddis.2009.2

Roth, A. D., Ramírez, G., Alarcón, R., & Von Bernhardi, R. (2005). Oligodendrocytes damage in Alzheimer's disease: Beta amyloid toxicity and inflammation. *Biological Research*, 38(4), 381–387. doi:10.4067/S0716-97602005000400011

Sangmee, A. J., Kyungsook, A., Ji-Hyun, K., Byung-Hak, K., Eunkyung, K., Inho, J., & Doh Kwan, K. (2006). ApoE-ε 4-dependent association of the choline acetyltransferase gene polymorphisms (2384G > A and 1882G > A) with Alzheimer ' s disease. *Clinica Chimica Acta*, 368, 179–182. doi:10.1016/j.cca.2005.12.037

Savonenko, A., Xu, G. M., Melnikova, T., Morton, J. L., Gonzales, V., Wong, M. P. F., Price, D. L., et al. (2005). Episodic-like memory deficits in the APPswe / PS1dE9 mouse model of Alzheimer ' s disease : Relationships to B-amyloid deposition and neurotransmitter abnormalities. *Neurobiology of Disease*, 18, 602–617. doi:10.1016/j.nbd.2004.10.022

Scarmeas, N., Stern, Y., Tang, M., Luchsinger, J. A., & Mayeux, R. (2006). Mediterranean Diet and Risk for Alzheimer's Disease. *Annals of Neurology*, 59(6), 912–921. doi:10.1002/ana.20854.Mediterranean

Schaeffer, E. L., Figueiro, M., & Gattaz, W. F. (2011). Insights into Alzheimer disease pathogenesis from studies in transgenic animal models. *Clinics*, 66, 45–54.

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

Shen, J. (2013). Modeling the glutamate – glutamine neurotransmitter cycle. *Frontiers in Neuroenergetics*, 5, 1–13. doi:10.3389/fnene.2013.00001

Solito, E., & Sastre, M. (2012). Microglia function in Alzheimer's disease. *Frontiers in pharmacology*, 3(February), 14. doi:10.3389/fphar.2012.00014

Solomon, B. (2009). Immunotherapeutic strategies for Alzheimer's disease treatment. *TheScientificWorldJournal*, 9, 909–19. doi:10.1100/tsw.2009.99

Streit, W. J., Braak, H., Xue, Q.-S., & Bechmann, I. (2009). Dystrophic (senescent) rather than activated microglial cells are associated with tau pathology and likely precede neurodegeneration in Alzheimer ' s disease. *Acta Neuropathologica*, 475–485. doi:10.1007/s00401-009-0556-6

Strooper, B. De, Saftig, P., Craessaerts, K., Vanderstichele, H., Guhde, G., Annaert, W., Figura, K. Von, et al. (1998). Deficiency of presenilin-1 inhibits the normal cleavage of amyloid precursor protein. *Nature*, 391(January), 387–390.

Szekely, C. a, Breitner, J. C. S., & Zandi, P. P. (2007). Prevention of Alzheimer's disease. *International review of psychiatry (Abingdon, England)*, 19(6), 693–706. doi:10.1080/09540260701797944

Tobinick, E. L., & Gross, H. (2008). Rapid improvement in verbal fluency and aphasia following perispinal etanercept in Alzheimer ' s disease. *BMC Neurology*, 1–9. doi:10.1186/1471-2377-8-27

Verkhratsky, A., Olabarria, M., Noristani, H. N., Yeh, C., & Rodriguez, J. J. (2010). Astrocytes in Alzheimer ' s Disease. *Neurotherapeutics*, 7(October), 399–412.

Volterra, A., & Meldolesi, J. (2005). Astrocytes, from brain glue to communication elements: the revolution continues. *Nature reviews. Neuroscience*, 6(8), 626–40. doi:10.1038/nrn1722

Wigley, R., & Butt, A. M. (2009). Integration of NG2-glia (synantocytes) into the neuroglial network. *Neuron glia biology*, 5(1-2), 21–8. doi:10.1017/S1740925X09990329

Xu, J.-P., Zhao, J., & Li, S. (2011). Roles of NG2 glial cells in diseases of the central nervous system. *Neuroscience bulletin*, 27(6), 413–21. doi:10.1007/s12264-011-1838-2

Yeh, C.-Y., Vadhwana, B., Verkhratsky, A., & Rodríguez, J. J. (2011). Early astrocytic atrophy in the entorhinal cortex of a triple transgenic animal model of Alzheimer's disease. *ASN neuro*, 3(5), 271–9. doi:10.1042/AN20110025

Zhu, X., Bergles, D. E., & Nishiyama, A. (2008). NG2 cells generate both oligodendrocytes and gray matter astrocytes. *Development*, 135, 145–157. doi:10.1242/dev.004895

Zilka, N., Kazmerova, Z., Jadhav, S., Neradil, P., Madari, A., Obetkova, D., Bugos, O., et al. (2012). Who fans the flames of Alzheimer ' s disease brains ? Misfolded tau on the crossroad of neurodegenerative and inflammatory pathways. *Journal of Neuroinflammation*, 9(1), 47. doi:10.1186/1742-2094-9-47