IL CERVELLO COME FUNZIONA 2:

LA GLIA
“Neuroglia” first coined by Rudolph Virchow (1850’s):
A connective substance that formed a sort of cement in the brain, spinal cord and higher sensory nerves

Glial cells → Santiago Ramòn y Cajal (1891)

Current designation: “Glia”, from the Greek word meaning “glue”
Group of cell types distinguishable from neurons that, together, form the predominant cell population of the nervous system

- Oligodendrocytes and Schwann cells → layers of myelin around axons in SNC and SNP
- Microglia → immunocompetent and specialized brain macrophages
- NG2-glia → glial cells that receive direct input from neurons
- Astrocytes → extend processes surrounding neurons and blood vessels, major glial component in the CNS, up to the 20-50% of the brain volume
O1 galactocerebroside processes and myelin-like sheaths

Fyn cell body and proximal processes
Oligodendrocytes originate from progenitors expressing NG2 chondroitin sulfate proteoglycan and PDGF-α receptor

Mature oligodendocytes form the myelin internodes in the CNS critical for fast saltatory conduction of action potential

Neurotransmitters released from neurons act on oligodendrocytes receptors (Glu, ATP, adenosine)

**Oligodendrocyte injury:**

**Oxidative damage:** very susceptible for their high metabolic rate and ATP requirement, high production of hydrogen peroxide in peroxisomes and large intracellular stores of iron

**Excitotoxicity:** highly susceptible, express EAAT1 e 2 and are predominant cells for Glu clearance in human white matter

ATP depletion → Glu release from oligodendrocytes and axons

ATP released from axons, astrocytes, or microglia in response to injury elicit oligodendrocyte toxicity

Glu and ATP toxicity depends on excessive Ca\(^{2+}\) influx which accumulate within mitochondria and leads to ROS, caspases activation, NOS

NMDA located on distal processes render the myelin sheaths vulnerable to excitotoxic insult

**Role of microglia:** microglia release Glu, proinflammatory cytokines which impair glu transporters function in astrocytes and oligodendrocytes and peroxynitrite
**Clinical correlations:**

### Multiple sclerosis:

Oligodendrocytes apoptosis and selective loss of myelin-associated glycoprotein

Glu toxicity due to release from microglia, excessive Glu production, reduced Glu degradation, reduced expression of Glu transporters in oligodendrocytes, upregulation of $\text{Ca}^{2+}$ permeable AMPA-R in MS lesions, high levels of iron in oligodendrocytes and MS lesions

upregulation nNOS $\rightarrow$ peroxynitrite

### Periventricular leukomalacia:

Diffuse injury of cerebral white matter initiated by cerebral ischemia and intrauterine or neonatal infection

The main target of injury is premyelinating oligodendrocytes, microglia has a critical role
NG2 cells
NG2 cells → “Polidendrocytes”, special glial cells that express NG2 proteoglycan on the cell surface, receive direct synaptic input from axons.

Some NG2+ cells are oligodendrocyte precursors, others are a distinct type of glia.

A subset of NG2 cells fire action potentials in response to Glu by AMPA-R.

NG2+ glia extend processes to nodes of Ranvier.

Physiologic role unclear → action potential to initiate differentiation into oligodendrocytes or to instruct nearby oligodendrocytes and axons to improve nodal axonal conduction.

Excitable NG2 are more vulnerable to toxicity than non-excitable.

Loss of excitable NG2+ glia leads to excitotoxic damage and impaired remyelination (NG2+ cells regenerate oligodendrocytes).
Astrocytes

GFAP Glial fibrillar acidic protein → intermediate filaments

Hoechst Nuclei
ASTROCYTES, FROM BRAIN GLUE TO COMMUNICATION ELEMENTS

Initially considered “brain glue” → inert scaffold necessary for neurons

Later → support cells

Till 20 yrs ago → astrocytes regarded as electrically silent elements

1990s → astrocytes express functional transmembrane proteins and share almost the same set of channels and receptors

1994 → increasing \(i[\text{Ca}^{2+}]\) in cultured astrocytes induces glu release

Astrocytes are direct communication partners of neurons and dynamically interact with synapses through the uptake and release of neurotransmitters and receptor mediated \(\text{Ca}^{2+}\) signalling

Different types of astrocytes within a given brain region and their properties vary in different subregions
Astrocytes are generated from radial glia residing in the embryonic VS (prevalently gray matter astrocytes) and SVZ progenitors (gray and white matter astrocytes and oligodendrocytes). Glial-restricted precursors give rise to astrocytic plasma membrane forming lamellae and fibers infiltrating among the neural processes.

Fibrous astrocytes (white matter) have long thin unbranched processes enveloping nodes of Ranvier.

Protoplasmic astrocytes (gray matter) have branching processes enveloping synapses and covering blood vessels.

The degree of synaptic ensheathment by astrocytic processes varies (57% in the hippocampus, 94% in the climbing fibers of the cerebellum). One hippocampal astrocyte can contact 100000 synapses.
PHYSIOLOGIC FUNCTIONS OF ASTROCYTES

1) Synaptic homeostasis

2) Tripartite Synapse-Neuronal Signaling

3) Regulation of neuronal glutamate receptors subunit expression

4) Blood brain barrier and control of blood flow

5) Neuronal energy homeostasis and neuroprotective properties of astrocytes

6) Neuronal replacement
1) Synaptic homeostasis

Removal of:

- Glu
- GABA
- K+ and H+

from the synapsis and dissipation through gap junctions coupling
**2) Tripartite Synapse-neuronal signaling**

**Gliotransmission**

Neuron dependent or spontaneous release of neuroactive molecules (Glu, D-serine, ATP, adenosine, GABA, TNFalpha, Prostaglandins)

- **Ca2+ dependent**
  - Vescicle and lysosome exocitosis

- **Ca2+ independent**
  - Reversal of Glu transporters
  - Hemichannels
  - Pore-forming P2X7 receptors
  - Swelling induced activation of volume regulated anion channels

**Release of**

- **Glu**↑ the frequency of both inhibitory (Kainate or mGLURs II or III) and excitatory (mGluRs I or NMDA) postsynaptic currents

- **ATP**↓ the glu release from presynaptic neurons

Intracellular calcium waves propagating through gap junctions modulate neuronal signaling
3) Regulation on neuronal GluR subunit expression

In vitro → astrocytes regulate the level of GluR2 subunit of AMPA receptors on neighboring MN regulating Ca2+ permeability of AMPA receptors and the susceptibility of neurons to Glu toxicity
4) Blood brain barrier and control of blood flow

Astrocyte end feet surround cerebral microvessels of the BBB and regulate:

- Tight junction formation
- Localization of transporters
- Production of anti-oxidant enzymes

Astrocyte secrete angiogenic factors (angiopoietin 1) and neurotrophins (GDNF and TGFbeta) while endothelial cells secrete LIF (leukemia inducing factor) which induce astrocyte differentiation.

Astrocyte end feet that contact vessels have AQP4, Glut-1 and Kir4.1 involved in ion, glucose and water homeostasis to be used in active material exchange.

BBB is dynamic and can be remodelled by inflammatory cytokines, angiogenic factors, glutamatergic toxicity, hypoxia and oxidative stress.
5) Neuronal energy homeostasis and neuroprotective properties

Astrocytes protect neurons when glucose lacks by releasing lactate and they have higher concentrations of anti-oxidant molecules (vit.E, ascorbate, GSH)

Astrocytes secrete GSH and increase the activity of glutamate cysteine ligase

Upregulate antioxidant genes after ROS and RNS challenge
6) Neuronal replacement

Neural stem cells are tightly regulated by their microenvironment (astrocytes, endothelial cells and secreted proteins)

SVZ astrocytes after neuronal ablation repopulate the entire SVZ including the neuronal precursors, in the presence of locally produced factors.
ASTROGLIOSIS

Changes in astrocytes in response to CNS injury and disease, varying with the nature and severity of the insult along a gradated continuum of progressive alterations in molecular expression, progressive cellular hypertrophy and proliferation and scar formation.

(a) Healthy tissue  (b) Moderate astrogliosis  (c) Severe astrogliosis

GFAP  Glial Scar  Inflam.
**ASTROGLIOSIS**

**Reactive astrocytes and acute CNS injury**

Reactive astrocytes $\rightarrow$ hypertrophy and upregulation of GFAP, iNOS and NFkB, re-expression of molecules associated with neonatal astrocytes

Astrogliosis is a protective response to acute injury.

Lack of reactive astrogliosis $\rightarrow$ $\uparrow$ neuronal and oligodendrocyte death, $\uparrow$ inflammatory infiltration, $\downarrow$ recovery of the BBB and $\uparrow$ functional deficits.

Astrogliosis is stimulated by cytokines released from microglia and damaged neurons

The type of the reactive astrocyte depends on the age of the organism and the nature of the insult $\rightarrow$ neonatal astrocytes after damage permit axonal growth; adult astrocytes inhibit axonal growth

Astrogliosis may be protective against acute trauma or injury but may be harmful in age related conditions such as neurodegenerative disorders.
ASTROGLIOSIS

**Chronic astrogliosis**

Prolonged activation of astrocytes leads to chronic disorders

Astrocytes release neurotrophins that in the adult can cause neuronal apoptosis

AD and MND → expression of neuronal C1q

Clearance of reactive astrocytes: return to a quiescent state or undergo programmed cell death → failure may result in toxicity

**Astrocytic inclusions**

Healthy elderly, more common in neurodegenerative disorders

Peroxidase positive inclusions → peri-ventricular area, hippocampus, striatum, periacqueductal grey matter

Tau-inclusions → subpial, subependymal and perivascular regions
REACTIVE ASTROCYTES: DELETERIOUS OR PROTECTIVE?

GFAP and Vimentin double ko mice →
improvement in axonal regeneration in the hippocampus after transection of the enthorinal cortex axons,
astrocytes more permissive for stem cell migration and neurite extension after retinal transplantation
3 times larger infact size after MCAO

Specific ablation of proliferative astrocytes →
Limited glial scar and greater neurite outgrowth in the forebrain stab wound model but increased severity of lesions, BBB repair impaired, higher immune cell infiltration, enhanced neuronal death

STAT3 or SOCS3 ko →
STAT3 ko reduced migration of reactive astrocytes towards injury, increased demielination and neuronal disruption and worsened clinical outcomes
SOCS3 ko improved all these outcomes
Astrocytes as neuronal killers

**Gliomas**
Expansion through the release of excess Glu with ensuing NMDA-dep neuronal excitotoxicity
Lack of Glu uptake leads to higher extracellular accumulation and neurotoxicity
AMPA-Rs lacking GluR2 subunit → $Ca^{2+}$ permeability which enhances proliferation and migration

**AIDS-related neuropathology**
Microglia infected by HIV triggers neurotoxic cascades that disregulate astrocytes
The viral coat protein gp120 activates CXCR4 amplifying Glu release from astrocytes and induces excitotoxic neuronal apoptosis

**Alzheimer's disease**
Healthy astrocytes degrade Aβ
In AD a defect in this digestive function and the abnormal expression of β-secretase switches the role of astrocytes to promoters of Aβ accumulation.
ApoE dependent signalling is crucial for astrocytes digestive function
**Amyotrophic lateral sclerosis**

fALS → point mutations in Cu/Zn SOD (SOD1)

**Mut SOD1 Astrocytes:**

activated and damaged,

focal loss of EAAT2 (also in sALS)

↓ Glu uptake,

↑ spill-over of synaptic Glu, activation of mGluR signalling in astrocytes with further Glu release, increased generation of NO that stimulates pro-apoptotic pathway in MN
The hSOD1-G93A transgenic mice

hSOD1-G93A mouse

Lumbar spinal cord

Pre-symptomatic

End-stage

Chiu et al., 1995
Presence of abnormal spheroid-shaped astrocytes in the lumbar spinal cord of hSOD1G93A mice

SGPCs appear selectively in the microenvironment of motor neurons before the symptomatic phase of ALS
Current evidence in favour of a role for PPARs in ALS


Pio improved motor performance and weight loss, attenuated MN loss, increased survival delaying the onset, reduced microglial activation and gliosis in the spinal cord, reduced iNOS, NFkB, COX2 expression
PPARg subcellular localization in hSOD1G93A astrocytes
PPARγ subcellular localization in hSOD1G93A motor neurons