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The Concept of Neuroglia

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Abstract

Neuroglia represent a diverse population of non-neuronal cells in the nervous systems, be that peripheral, central, enteric or autonomic nervous system. Arguably, these cells represent about half of the volume of the human brain. This volumetric ratio, and by extension glia to neurone ratio, not only widely differ depending on the size of the animal species brain and its positioning on the phylogenetic tree, but also vary between the regions of an individual brain. Neuroglia derived from a dual origin (ectoderm and mesodermal) and in an assorted morphology, yet their functional traits can be mainly classified into being keepers of homeostasis (water, ions, neurotransmitters, metabolites, fuels, etc.) and defenders (e.g., against microbial organisms, etc.) of the nervous system. As these capabilities go awry, neuroglia ultimately define their fundamental role in most, if not, all neuropathologies. This concept presented in this chapter serves as a general introduction into the world of neuroglia and subsequent topics covered by this book.

Keywords

Neuroglia; Origin; Morphology; Function; Homeostasis; Defence; Physiology; Pathophysiology

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1.1 The Birth of the Concept of Homoeostatic Neuroglia

The complexity of the human brain is remarkable: a population of more than 200 billion (i.e. 2×10^{11}) neural cells (neurones and neuroglia) is packed within a limited volume (average human brain occupies 1200–1400 cm^3). These neural cells form complex networks, connected through 15–20 trillions of chemical and electrical synapses that provide for computing power of this organ. The logistical support underlying this highly complex organ is provided by a specific class of cells known as neuroglia.

The concept of connective tissue of the nervous system emerged in the nineteenth century [16, 47]; this concept was initially formalised by Rudolf Virchow who introduced the term neuroglia in the 1850s [100, 101]. According to Virchow the neuroglia was '…connective substance that forms in the brain, in the spinal cord and in the higher sensory nerves a type of putty (neuroglia), in which the nervous elements are embedded…' [100]. Prominent neuroanatomists of the second half of the nineteenth century characterised the cellular nature of glia in great detail, and described many types of glial cells [16]. At the same time numerous theories have considered the functional role of neuroglia in the brain homeostasis, nutritional support, regulation of blood flow, sleep and conscience, as well as in neuropathology [6, 29, 30, 72, 79]. The first major type of glia, the astroglia, has been defined in 1895, when Michal von Lenhossék suggested to name a sub-population of parenchymal glia astrocytes, star-like cells (from Greek αστρον κυτος). At the same time the parenchymal glia was also sub-classified into protoplasmic (grey matter) and fibrous (white matter) cells [6]. The myelinating cells of the central nervous system (CNS) were first drawn by the Scottish pathologist William Ford Robertson [74, 75], and subsequently Pío del Río Hortega named them oligodendrocytes and recognised their myelinating function [24]. It was also Pío Del Río Hortega who identified and named microglia as the defensive cellular elements of the CNS, by demonstrating that these cells undergo remarkable metamorphosis in pathology and suggesting their role as 'garbage collectors' [21–23]. Finally, in the 1980s the fourth type of neuroglia, the NG2 glia (also known as oligodendrocyte progenitor cells or polydendrocytes), was discovered by William Stallcup and colleagues, after they developed an antibody to a chondroitin sulphate proteoglycan, dubbed NG2 [88]. Based on their developmental origin (neuroepithelial or mesodermal), neuroglia of the CNS have been classified as macroglia (astrocytes, oligodendrocytes, NG2) and microglia, respectively.

1.2 The Definition of Neuroglia

The definition of neuroglia is based on the unifying fundamental function of these cells, which, regardless of their origin, is homeostasis of the nervous system. This function is fundamental for both physiological context, when glial cells perform their routine housekeeping duties, as well as for pathological context, when glial cells can undergo reactive remodelling in order to preserve, repair and restore brain homeostasis. Failure in this function results in the development of the neurological disease and damage to the nervous tissue. Therefore, neuroglia can be defined as homeostatic and defensive cells of the nervous system, represented by highly heterogeneous cellular populations of different origin, structure and function [94].

In this sense neuroglia are the ultimate supportive cells of the nervous system, keeping it in a functional state. This reflects upon evolution of the nervous system, which resulted in the division of labour: the information processing and electrical excitability became confined to the neuronal networks, whereas homeostatic support and defence became the sole prerogative of the neuroglia [95]. This homeostatic support occurs at all levels of organisation of the nervous system: at molecular level (control over homoeostasis of ions, neurotransmitters, protons, reactive oxygen species, metabolites, etc.), at cellular level (astrocytes involvement in neurogenesis), at network level (both astroglia and microglia regulate synaptogenesis, synaptic maturation and extinction), connectome level (which is maintained by myelinating oligodendroglia and Schwann cells), organ level (astrocytes control blood-brain barrier and glymphatic flow and regulate functional hyperaemia) and systemic level (glial cells emerge as central chemoceptors and contribute to systemic control over ventilation, ion homeostasis and energy metabolism); for comprehensive coverage of neuroglial homeostatic capabilities see [1, 2, 7, 19, 31, 34, 37, 41, 45, 46, 49, 50, 59, 68, 70, 93, 96, 97, 103, 105].

This ultimate homeostatic capability of neuroglia underlies their fundamental role in neuropathology, the latter being broadly defined as a homeostatic failure of the nervous system. Environmental stress and/or pathological insults trigger glial homeostatic response (when glial cells attempt to keep homeostatic equilibrium or steady state) and glial reactivity, which represents an evolutionary conserved programme of glial cells remodelling aimed at mounting defence of the nervous tissue. Neuroglial reactivity is manifested in reactive astrogliosis, microglial activation and Wallerian degeneration (for oligodendrocytes). At the same time glial asthenia or atrophy, which is observed in numerous neurological conditions, facilitates evolution of the disease because of compromised homeostatic and defensive capabilities. Although the fundamental role of neuroglia in neuropathology has been predicted by prominent neurologists of the nineteenth and the beginning of the twentieth centuries (such as Rudolf Virchow, Carl Frommann, Alois Alzheimer, Nicolas Achucarro and Franz Nissl), the pathophysiological role of glia begun to be universally recognised only in the recent decade; for references and concepts see [11, 12, 53, 66, 67, 76, 81, 84–86, 99]. The concept of astrotauopathology, recently introduced by Kovacz [51], supports the notion that the neuroglia emerges in the limelight when considering the evolution of neurological diseases.

1.3 Glial Numbers

The numerical preponderance of glial cells in the brains and spinal cords of different species and glial to neurone ratio (GNR) have been a matter of the most common fallacy over recent decades. The notion of glial cells outnumbering neurones in the human brain by a factor of 10 or even 50 [10, 18, 44] represented an undisputed general knowledge that has been repeatedly proclaimed in glial literature (for critical analysis see for example [39, 98, 102]). The story of exceedingly high number of glial cells in the human brain goes back to Franz Nissl [58]; this idea became rather popular and reached the climax in writings of Robert Galambos who considered that neuroglia represent the primary seat of intelligence, consciousness, emotions and are overall responsible for our ‘humanity’. ‘Glia is … conceived as genetically charged to organize and program neuron activity so that the best

interests of the organism will be served; the essential product of glia action is visualized to be what we call innate and acquired behavioural responses. In this scheme, neurons in large part merely execute the instructions glia give them' [28]. The notion was further promoted by the finding that the Einstein's brain had a rather higher glia to neurone ratio in his associated cortex than that found in the control human population [25], leading to speculations that this could be the reason for his remarkable intellectual abilities (https://www.theguardian.com/science/2007/feb/21/neuroscience.highereducation) (https://www.npr.org/templates/story/story.php?storyId=126229305). The public myth of glia has extended into that of an untapped part of the brain that we may not use, perhaps gloriously captured in Starbuck's The Way I see it? (http://www.stevekmccoy.com/blog/2005/08/starbucks_the_w) #236 quote 'Scientists tell us we only use 5% of our brains. But if they only used 5% of their brains to reach that conclusion, then why should we believe them?' Of course, based on any functional imaging, this myth has been debunked and the authors would like to assure the readers that we had used the vast majority if not all of our brains to write this chapter.

None of these concepts had experimental confirmation. Exceptionally high glia to neurone ratio of the human brain was not related to actual cell counts; to the contrary most of stereological investigations produced the GNR values in the neocortex somewhere around 1.5 (see Table 1 in [42, 102]), with the number of neuronal counts in the range of 20–30 billion and glial cells in the range of 27–38 billion. In the cerebellum, which contains the largest number of brain neurones (around 70 billion) the number of glial cells is much smaller, with GNR not exceeding 0.1 [3, 4]. These stereological data obviously made the total GNR estimate of 10:1 unrealistic. Further advances in defining the glial numbers are associated with the application of isotopic fractionation technique, which counts nuclei of neurones and non-neuronal cells in the homogenates of the nervous tissue [8, 40, 54]. This technique demonstrated that the total numbers of neuronal and non-neuronal cells in the human brain are more or less on par, both being in the range of ~80–100 billion. After subtracting the population of endothelial cells which may account for about 20% of all non-neuronal cells, the true number of glial can be estimated at ~60 billion and total GNR for the whole brain is therefore less than 1:1. The density of glia is quite different in various brain areas. For example, the GNR varies between 1.2 in the grey matter of the occipital cortex and 3.6 in the grey matter of frontal cortical regions [73, 82], it is technically an infinity in the white matter that does not contain neuronal cell bodies, and hence inclusion of white matter counts increases the total GNR in the cortex to ~3. As already alluded above the GNR in cerebellum is very low probably not exceeding 0.1. Much higher GNR values were reported for the striatum (3.7:1), for the superior colliculus (10:1), for the ventral pallidum (12.2:1), for the lateral vestibular nucleus (30–50:1), while for the globus pallidus a very high GNR of 160:1 has been calculated from stereological counts [5, 8, 64, 71, 80, 91, 102]. Similarly, the GNR for the spinal cord was determined at 5:1 in cynomolgus monkey and almost 7:1 in humans [9].

Evolution of the nervous system paralleled with an increase in GNR, which however was not entirely linear and was not directly related to the intelligence. The nervous system of invertebrates has, as a rule, relatively smaller numbers of glial cells, with a GNR between 0.01:1 and 0.2:1 (50 glial cells derived from neuronal/epithelial progenitors and six glial

cells that are mesodermally derived per 302 neurones in *Caenorhabditis elegans* [63, 89]; 10 glial cells per 400–700 neurones in every ganglion of the leech [20]; ~9000 glial cells per 90,000 neurones in the CNS of Drosophila [26, 52]). At the same time, the buccal ganglion of the great ramshorn snail *Planorbis corneus* contains 298 for example, in the cortex, the GNR is about 0.3–0.4 in rodents, ~1.1 in cat, ~1.2 in horse, 0.5–1.0 in rhesus monkey, 2.2 in Göttingen minipig, ~1.5 in humans and as high as 4–8 in elephants and the fin whale [15, 27, 38, 43, 55, 65, 92]. The largest absolute number of glial cells has been counted in the neocortex of whales [27, 56]; stereological cell counts in the neocortex of the long-finned pilot whale (*Globicephala melas*) brain determined there are approximately 37.2 billion neurones and 127 billion glial cells and this gives a GNR of 3.4 [56]. The largest GNR was found in the neocortex of the common Minke whale (*Balaenoptera acutorostrata*), which contained ~12.8 billion neurones and 98 billion of glia giving therefore a GNR of ~7.6 [27].

1.4 Classification and Main Functions

Neuroglia (Fig. 1.1, see also [94]) are classified into glia of the peripheral nervous system (PNS) and of the CNS. The glial cells of the PNS originate (similarly to peripheral neurones) from the neural crest and are classified into:

1. Schwann cells [48] associated with sensory, motor, sympathetic and parasympathetic axons; Schwann cells are further subdivided into (i) myelinating Schwann cells that myelinate peripheral axons; (ii) non-myelinating Schwann cells that surround multiple non-myelinating axons and (iii) perisynaptic Schwann cells, which enwrap peripheral synapses and maintain homoeostasis in the perisynaptic milieu.
2. Satellite glial cells [35, 36], which are surrounding neurones in sensory, sympathetic and parasympathetic ganglia. These satellite glial cells control local homeostasis and are capable of reactive remodelling in pathology.
3. Olfactory ensheathing cells [77], which are a part of the olfactory system. These cells extend very fine processes that enclose large numbers of unmyelinated olfactory axons
4. Enteric glia [32, 33], represented by homeostatic glial cells of the enteric nervous system.

Neuroglia of the CNS are subdivided into macroglia (cells of ectodermal, neuroepithelial origin) and microglia (cells of mesodermal, myeloid origin). The macroglia is further classified into:

1. Astroglia or astrocytes. Astrocytes are heterogeneous population of primary homeostatic glia residing throughout the brain and the spinal cord, in both grey and white matter. Astroglia include [94, 96]:
    i. protoplasmic astrocytes of grey matter;
    ii. fibrous astrocytes of white matter

iii. surface-associated astrocytes associated with the cortical surface in the posterior prefrontal and amygdaloid cortex;

iv. Velate astrocytes, which are localised in the parts of the brain that are densely packed with small neurones, for example in the olfactory bulb or in the granular layer of the cerebellar cortex;

v. Radial glia, which are the pluripotent neural cells precursors that generally disappear at birth in mammals

vi. Radial astrocytes, which include Bergmann glia in the cerebellum, Müller glia of the retina, radial glia-like neural stem cells of the neurogenic niches and tanycytes of the hypothalamus, hypophysis and the raphe part of the spinal cord;

vii. Pituicytes, which are the glial cells of the neurohypophysis;

viii. Gomori astrocytes rich in iron and positive for Gomori's chrome alum hematoxylin staining identified in the hypothalamus and in the hippocampus;

ix. Perivascular and marginal astrocytes, which are placed near the pia mater, where they form endfeet with blood vessels. These astrocytes do not establish contacts with neurones and their main function is in establishing the pial and perivascular glia limitans barriers.

x. Ependymocytes, choroid plexus cells and retinal pigment epithelial cells. These cells line up the ventricles and the subretinal space; the choroid plexus cells produce the cerebrospinal fluid. Ependymocytes possess small movable processes (microvilli and kinocilia), which by rhythmic movements produce a stream of cerebrospinal fluid.

In addition, the brain of higher primates (including humans) contains several types of specialised astrocytes [17, 98], which include:

xi. Interlaminar astrocytes;

xii. Polarised astrocytes;

xiii. Varicose projection astrocytes.

Function of these hominoid astroglia remain unknown.

Parenchymal astrocytes of the human brain are substantially larger and more complex compared with astroglial cells of rodents, and have distinct gene expression pattern [60–62, 87, 104]. Human protoplasmic astrocytes have about 10 times more primary processes and a more complex secondary process arborisation, with an average volume about 16.5 times larger than that of the corresponding astrocytes in a rat brain [61]. The larger human protoplasmic astrocytes also have extended outreach onto neuronal structures, on average contacting and encompassing up to 2 million synapses residing within astrocytic territorial domains, significantly more than the integrating capacity of rodent protoplasmic astrocytes,

which covers ~20,000–120,000 synaptic contacts [13, 61]. Similarly, human fibrous astrocytes have a 2.14-fold larger domain compared to that in rodents [61].

2. Oligodendroglia or oligodendrocytes, the myelinating cells of the CNS are subdivided into 4 classes [94]:
    i. *Type I oligodendrocytes* are most numerous in the cortex and grey matter; they have small rounded somata and fine branching processes that myelinate 30 or more small diameter axons;
    ii. *Type II oligodendrocytes* are similar to type I, but have parallel arrays of intermediate length internodes (100–250 μm), and are most common in white matter, such as the corpus callosum, optic nerve, cerebellum and spinal cord;
    iii. *Type III oligodendrocytes* have larger (than type I and II) irregular cell bodies, with one or more thick primary processes that myelinate a small number of large diameter axons with long internodes (250–500 μm). These cells are localised in the cerebral and cerebellar peduncles, the medulla oblongata, and the spinal cord funiculi;
    iv. *Type IV oligodendrocytes*, are somewhat similar to Schwann cells, being directly associated with a large diameter axon to form a single long internodal myelin sheath (as long as 1000 μm), and are restricted to tracts containing the largest diameter axons near the entrance/exit of nerve roots into the CNS.
3. NG-2 glia also known as oligodendrocyte progenitor cells or OPCs, or synantocytes, or polydendrocytes [14, 57]. The NG2 glia can have homeostatic role and contribute to adulthood myelination, albeit their functions are yet to be better characterised.

Microglia originate from the foetal macrophages that migrate into the neural tube very early in the embryonic development; arguably, microglia represent the first parenchymal glia to populate neural tissue in development. Microglial cells carry numerous physiological functions, including shaping neuronal synaptic connectivity, removing of redundant or apoptotic neurones in the development and regulating synaptic transmission [45, 46, 90]. Microglia form the main defence system of the CNS through evolutionary conserved programme of activation (microgliosis) which can produce numerous neuroprotective and neurotoxic phenotypes [78, 83].

In terms of numbers, the most numerous glia are oligodendrocytes and NG2 cells combined (40–60%), with astrocytes accounting for 20–40% and microglia for ~10% of neuroglia population, although there is, of course, a considerable variability between the brain regions, developmental stage and species.

1.5 Envoi and Outlook

One of the two goals of this chapter is to serve as a general introduction into the world of Neuroglia. The other goal is to pique an interest of the reader into subsequent chapters in

this book. As we tersely reviewed Neuroglia we establish the origin of these cells, their classification and their general functions in homeostasis and defence of the brain. In the following chapters, we explore the role of these cells in the progression of neuropathologies, especially neurodegenerative disorders. For a long time, the neurone-centric view dominated neuropathological thinking, and only recently the role of glia has been reassessed and the perception is mounting of specific importance of neuroglia that to a very large extent defines the progression and outcome of most (if not all) neurological diseases.

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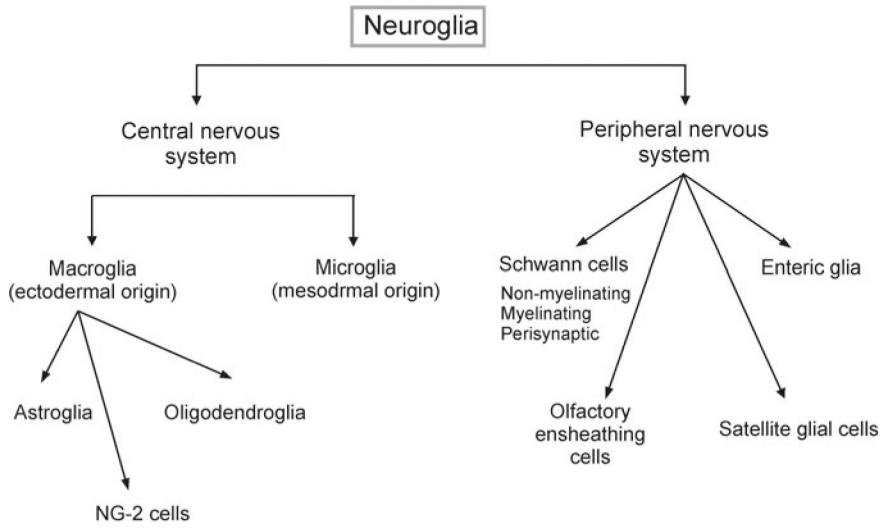

Fig. 1.1.
Classification of neuroglia