available at www.sciencedirect.com

ScienceDirect

www.elsevier.com/locate/brainres

BRAIN RESEARCH

Research Report

A morphological and electrophysiological study on the postnatal development of oligodendrocyte precursor cells in the rat brain

Peng-hui Chen, Wen-qin Cai, Li-yan Wang, Qi-yue Deng*

Department of Neurobiology, Third Military Medical University, Chongqing 40038, China

ARTICLE INFO

Article history:
Accepted 10 September 2008
Available online 19 September 2008

Keywords:
Oligodendrocyte precursor cell
NG2
Astrocyte
Hippocampus
Cerebrum

ABSTRACT

A widespread population of cells in CNS is identified by specific expression of the NG2 chondroitin sulphate proteoglycan and named as oligodendrocyte precursor cell (OPC). OPCs may possess stem cell-like characteristics, including multipotentiality *in vitro* and *in vivo*. It was proposed that OPCs in the CNS parenchyma comprise a unique population of glia, distinct from oligodendrocytes and astrocytes. This study confirmed that NG2 immunoreactive OPCs were continuously distributed in cerebral cortex and hippocampus during different postnatal developmental stages. These cells rapidly increased in number over the postnatal 7 days and migrate extensively to populate with abundant processes both in developing cortex and hippocampus. The morphology of OPCs exhibited extremely complex changes with the distribution of long distance primary process gradually increased from neonatal to adult CNS. Immunohistochemical studies showed that OPCs exhibited the morphological properties that can be distinguished from astrocytes. The electrophysiological properties showed that OPCs expressed a small amount of inward Na^+ currents which was distinguished from Na^+ currents in neurons owing to their lower Na-to-K conductance ratio and higher command voltage step depolarized maximum Na^+ current amplitude. These observations suggest that OPCs can be identified as the third type of macroglia because of their distribution in the CNS, the morphological development in process diversity and the electrophysiological difference from astrocyte.

1. Introduction

The cellular composition of the vertebrate central nervous system (CNS) is traditionally thought of as consisting of neurons, astrocytes, oligodendrocytes, and microglia. However, a widespread population of cells in CNS is now being considered as another glial cell type [Nishiyama et al., 1999; Levine et al., 2001; Nishiyama, 2007] unique from oligodendrocytes, astrocytes and microglia [Butt et al., 2005]. They are thought to belong to the oligodendrocyte lineage, but do not express proteins characteristic of mature myelinating oligodendrocytes [Nishiyama et al., 2002; Fiedorowicz et al., 2008], and are identified by their specific expression of the NG2 chondroitin sulphate proteoglycan and named as oligodendrocyte precursor cell (OPC). NG2 immunoreactive OPCs are capable of cell division in the resting CNS [Dawson et al., 2003; Ohya et al.,

* *Corresponding author.*
E-mail address: wenqin@mail.tmmu.com.cn (W. Cai).

Abbreviations: CNS, central nervous system; GFAP, Glial fibrillary acidic protein; LSCM, laser scanning confocal microscope; OPC, oligodendrocyte precursor cell; P, postnatal day; PBS, phosphate-buffered saline; SP, streptavidin-peroxidase

0006-8993/$ – see front matter © 2008 Elsevier B.V. All rights reserved.
doi:10.1016/j.brainres.2008.09.029

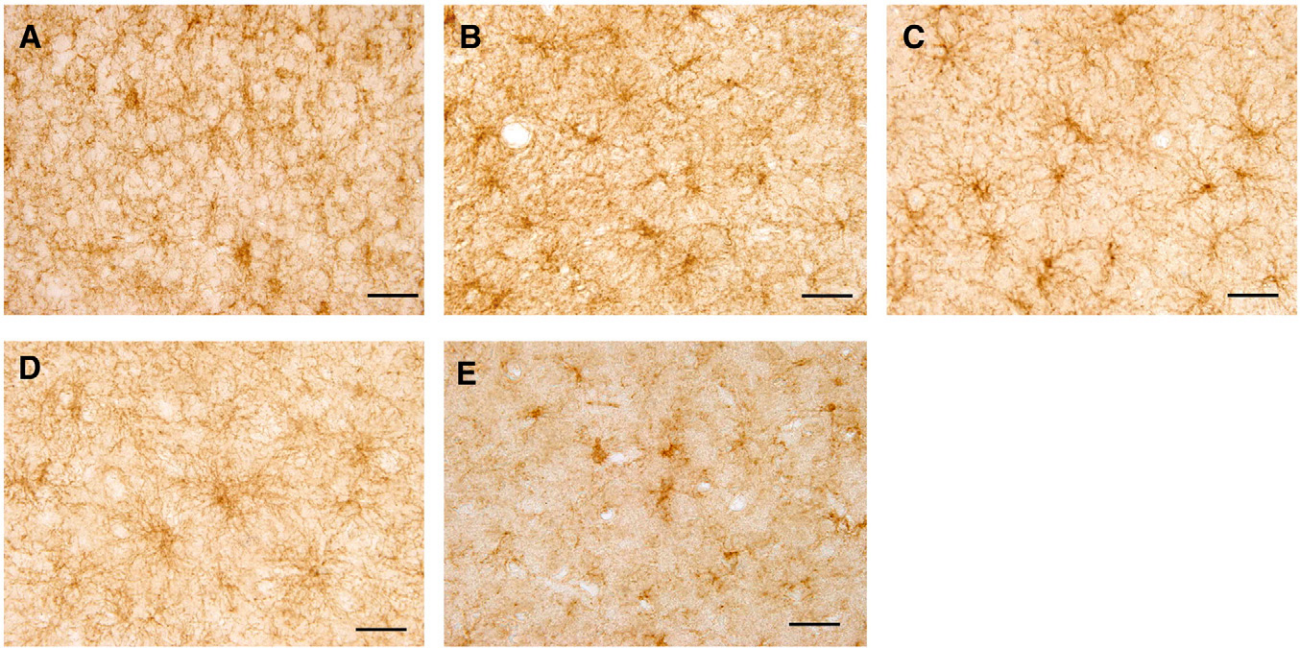

Fig. 1 – Morphological development of OPCs in cerebral cortex by anti-NG2 immunohistochemistry. (A) OPC at P0 exhibited "simple" morphology, with small round soma from which extended numerous short, fine, multibranching processes that passed in all directions. (B) OPCs at P7 showed a relative 'complex' morphology, with more fine primary dendrites, longer processes than those at P0. (C) OPC at P21 had an altered morphology of the increase in the number of fine processes, which gave the cells a more 'bushy' appearance. (D) OPCs at P50 (adult) exhibited a stellate morphology, a large cell body with plenty of processes. (E) OPCs at P450 (aged rats) appeared small and round cell bodies with thin branched processes. Scale bar, 50 μm.

2007] and can revert to a simpler morphology following injury [Reynolds et al., 2002; Alonso, 2005]. In addition to differentiating into myelinating oligodendrocytes, OPC has been proved to be able to generate neurons and astrocytes in culture, suggesting that $NG2^+$ OPCs in the CNS may possess stem cell-like characteristics, including multipotentiality *in vitro* and *in vivo* [Kondo and Raff, 2000; Diers-Fenger et al., 2001; Belachew et al., 2003; Baracskay et al., 2007].

Defining the role of NG2 cells *in vivo* has remained elusive despite their phenotypic characterization as part of the oligodendrocyte lineage. $NG2^+$ progenitors migrate throughout the developing CNS at a stage before axons have fully matured and before myelination begins [Nishiyama et al., 2007]. Although the majority of $NG2^+$ cells generated during CNS development do give rise to oligodendrocytes, a significant proportion of $NG2^+$ cells do not differentiate into myelin-forming oligodendrocytes but remain in the mature CNS with an immature phenotype [Rhodes et al., 2006]. Their distribution is relatively independent of the density of myelinated fibers and the persistence of these cells in both white and grey matter throughout life suggests that OPCs are not merely preserved as a pool of precursors from which to generate mature glia and neuron [Thallmair et al., 2006; Zhu et al., 2008].

Several lines of evidence suggest that $NG2^+$ cells in the adult CNS represent a population of reactive glial cells [Allen et al., 2005]. Their reactions to damage include dramatic changes in cell morphology, increases in NG2 immunoreactivity, and in some cases cell proliferation. These responses have been observed following stab wounds [Alonso, 2005], viral infection [Redwine and Armstrong, 1998], kainate excitotoxicity [Ong and Levine, 1999], and inflammation as seen in experimental autoimmune encephalomyelitis [Nishiyama et al., 1997]. In comparison with other glial cell types, NG2 positive cells are the first to react to the damage [Back et al., 2001; Sizonenko et al., 2008] and show the highest proliferation rate in response to demyelination [Watanabe et al., 2002].

Despite their abundance as a progenitor population and potential importance in maintenance and repair of neurological function, the developmental ontogeny of OPCs remains controversial. The persistence of numerous OPCs in the mature CNS has raised questions about their identities, relation to other type of CNS cells, and functions besides their progenitor role. OPCs express receptors for neurotransmitters, indicating that their behavior is likely to be influenced by neuronal activity [Bergles et al., 2000; Jabs et al., 2005; Ge et al., 2006]. Moreover, OPC can form direct synapses with glutamatergic and GABAergic neurons in the mouse cerebellum [Bergles et al., 2000; Butt et al., 2002; Bouslama-Oueghlani et al., 2005]. It was proposed that NG2 positive cells in the CNS parenchyma comprise a unique population of glia, distinct from oligodendrocytes and astrocytes [Kimelberg, 2004; Grass et al., 2004; Nishiyama et al., 2005]. However, there was insufficient developmental evidence supporting that proposal yet. The present study is undertaken to determine the morphological and electrophysiological features of these cells during postnatal developmental processes. The morphological and electrophysiological differences between OPC and astrocyte are also examined by laser scanning confocal microscope (LSCM) and whole-cell patch clamp recording on