Microglia Cells, The Brain Innate Immune System: Friend or Foe?

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Highlights

- Microglial cells are the innate immune system of the brain.
- Microglia continuously survey the brain environment maintaining homeostasis.
- Sex differences have been observed in the microglia of naïve adult animals.
- Space radiation induces chronic aberrant activation of microglia in a sex-specific manner
- Microglia depletion and complete repopulation prevents cognitive impairments after galactic cosmic radiation exposure in male mice.

Abstract Microglial cells are the resident immune cells of the Central Nervous System (CNS). Under physiological conditions, microglia constantly surveil their surrounding parenchyma and act as scavenger cells to maintain a healthy environment within the CNS. Following different insults to the CNS, microglia turn into a “reactive” state characterized by the production of inflammatory mediators that promote tissue repair to restore homeostasis. If inflammation is not in check, chronic microglia activation results in damage to the brain and leads to persistent cognitive impairments. Microglia display sex-specific features in adult mice; specifically, microglia from female mice have been found to be less reactive. Exposure to space radiation results in chronic activation of microglia in male but not in female mice. Interestingly, manipulating microglia after exposure to space radiation can prevent the development of cognitive deficits in adult male mice. These discoveries may provide clues in how to protect astronauts’ cognitive functions both during the missions and after return.

Keywords

Microglia; Space Radiation; Galactic Cosmic Ray; Cognitive functions

Abbreviations

CNS, Central Nervous System; CSF-1R, Colony-Stimulating Factor 1 Receptor

Microglia in the central nervous system: Microglia cells are the resident immune cells in the central nervous system (CNS). Humans and rodents' microglia show similar functions and partially overlapping gene expression patterns and regional distribution[1,2]; thus, rodent studies have been used to investigate how these cells develop, proliferate and react to insults. Microglia originate from myeloid lineage precursors in the yolk sac and migrate into the CNS during embryonic development[3]. Upon entry into the CNS, microglia remain isolated from the peripheral immune system[3,4] and maintain their homeostasis solely by self-renewal[5,6]. Unlike other types of mononuclear phagocytes in the CNS (meningeal macrophages, choroid plexus macrophages, epiplexus cells and perivascular macrophages) which have specific locations, marker profiles and functions, microglia are highly dynamic and mobile macrophages in the brain parenchyma that present also CNS glia features[7]. Microglia present different morphological[8] and transcriptomic[2] features during different phases of development. In the embryonic and early postnatal time window, microglia display a "generic macrophage"-like amoeboid morphology and expression profile that indicate activation[9] and are highly proliferative[10]. In the neonatal mouse brain, microglia play critical roles in the remodeling of neuronal networks by synaptic pruning, a process orchestrated by signals from complement cascades, IL33 and CD47, to wire the developing brain[11-14]. Homeostatic adult microglia exhibit a characteristic ramified morphology with long and dynamic processes that can cover areas up to 10x their soma[9]. Thanks to these ramified and dynamic extensions, these cells can continuously scan their environment and have interactions with neighboring cells[15]. Under physiological conditions, microglia are crucial in maintaining homeostasis and regulating different processes in the CNS. Some of the mechanisms by which microglia exert their function are engulfing metabolites and debris from apoptotic cells, secreting trophic factors to support neurons and modeling synapses[15-19].

Microglia and CNS pathology: One of the key features of microglia is the ability to quickly respond to pathological conditions (such as insult, infection, disease). Microglia's morphology, transcriptome, secretome, migration, and proliferation can quickly change in response to alterations in the environment. Once "danger signals" such as materials containing pathogen associated molecular patterns (PAMPs) and damage-associated molecular patterns (DAMPs) are detected, microglia migrate to the damage site and turn into an amoeboid-like "reactive" morphology to engulf those materials[20]. Alongside phagocytosis, microglia respond to CNS changes by secreting proinflammatory cytokines such as IL-1β, IL-6 and TNF-α and reactive oxygen species (ROS) that mediate wound healing and inflammatory responses[9,21]. Properly and tightly regulated microglia activation is essential for the brain to return to homeostasis after insults. Nevertheless, when these cells are overly activated for a long time, the production of proinflammatory cytokines and ROS can lead to neuronal damage[22] and cognitive dysfunctions[23-28].

Sex Dimorphism of Microglia: Microglia display sex differences during development and in the adult rodent brain[29]. Sex-specific distinctions have been observed in gene expression profile, brain distribution and density of microglia in physiological conditions[17,30-32]. Partial overlaps in the transcriptomic signature of murine sex-dimorphic microglia and human's brain[33] and fetal microglia[34] have been observed, but whether human microglia also show sex differences is not known and should be assessed. Importantly, microglia sex dimorphism has been also observed in rodents exposed to various challenges. Insults to the female mouse brain (such as stroke, Lipopolysaccharide stimulation and trauma) display a dampened inflammatory response when compared to their male counterparts[35-38]. Microglia gene expression analyses have shown sex-dependent differential expression patterns that corresponded with a neuroprotective state of female microglia, and microglia isolated from adults female brain maintain sex-specific features

when transplanted in the male brain[35]. In line with these results, we previously demonstrated that space radiation exposure induces a robust and chronic microglia response in males but not in female adult mice[24]. Furthermore, we found a profound sex dimorphism related to microglia cell function between female and male mice at chronic time points after radiation exposure. While irradiation resulted in cognitive and behavioral deficits in adult male mice, females were protected. The altered behavioral and cognitive responses observed only in male mice corresponded to microglia activation and synapse loss in the hippocampus, the brain region responsible for learning and memory[24]. Nevertheless, studies on sex-dimorphic activation of microglia after space radiation should be carefully interpreted considering the different radiation protocol/dosage/mouse strain and brain region analyzed.

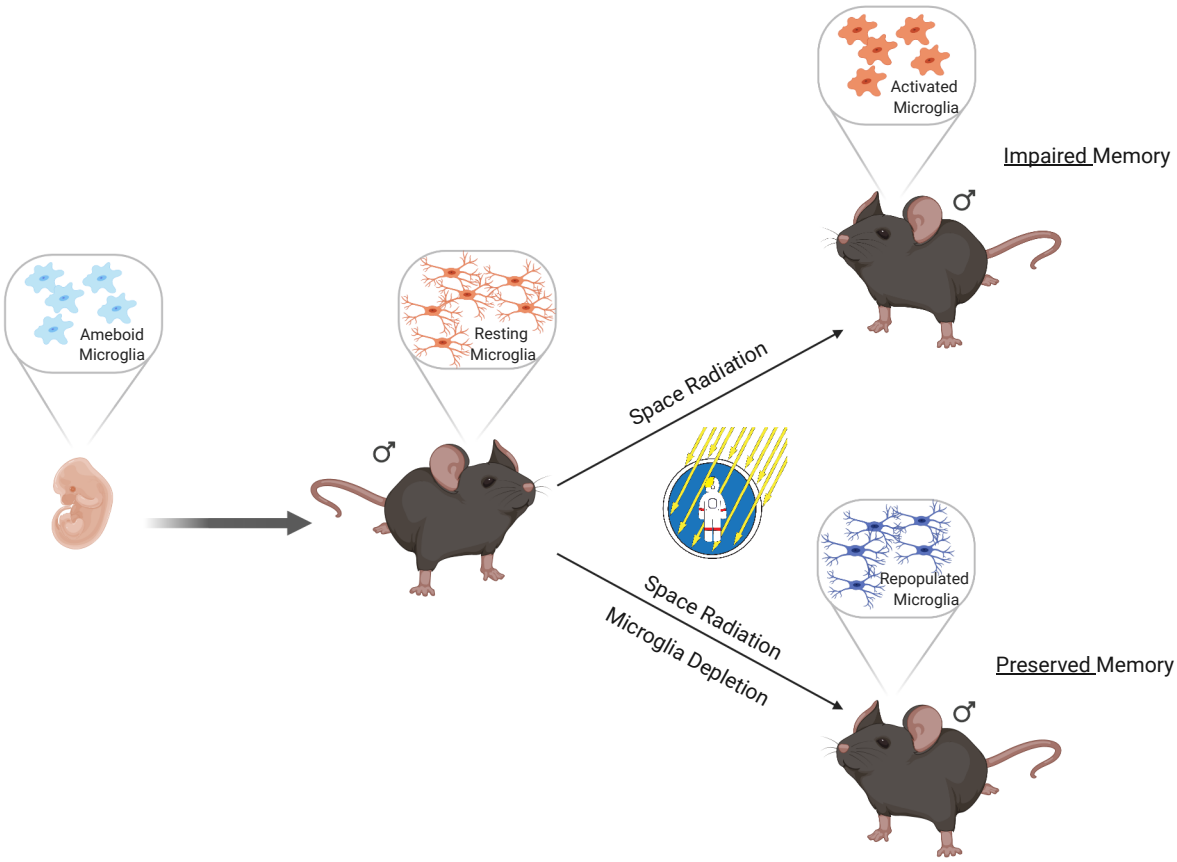
Therapeutic strategies to manipulate microglia: Given the duality of microglia function in response to pathological conditions, manipulation of these cells has been explored in hope of ameliorating or preventing the deleterious effects associated with chronic microglia over activation[23,25,26,39]. Microglia depletion is a novel potential therapeutic strategy for CNS diseases. Depletion can be effectively achieved through either pharmacological approaches or genetic manipulations[40]. The Colony-Stimulating Factor 1 Receptor (CSF-1R), a tyrosine kinase receptor expressed in leukocytes of the monocytic lineage -including microglia, monocytes and tissue macrophages- is the main receptor target for microglia manipulation. In the CNS, the CSF-1R signaling is essential for the migration, differentiation and survival of microglia[41]. Mice with intrinsic disruption of the CSF-1R signaling are born with brain deficits, further suggesting microglia's critical roles in CNS development[42]. Disruption of the CSF-1R signaling in the adult brain has not been reported to cause noticeable cognitive deficits or neurological dysfunctions[23,25,26,43-47]. PLX5622 is a CSF-1R inhibitor widely used to deplete microglia[23,25,44,45,48-51]. At the dose of 290 parts per million (ppm, supplemented in chow), PLX5622 can deplete about 50% of CNS microglia in

mice[25,45]; and 99% depletion of microglia can be achieved at the dose of 1200 ppm without associated cognitive deficits[23,44,48-51]. Another CSF-1R inhibitor, PLX3397, has been tested in clinical trials to treat glioblastoma[52-54]. However, further studies are needed to evaluate the suitability of CSF-1R inhibitor for astronauts. It is important to consider that CSF1R inhibitors, however, can potentially affect also other cells of the peripheral immune response when delivered systemically. Indeed, it has been demonstrated that 3 weeks of PLX5622 treatment leads to long-term changes in monocytes, macrophages, hematopoietic progenitor cells and stem cells, that do not recover after cessation of the inhibitor[55]. On the other hand, genetic manipulation methods allow microglia ablation with high cellular specificity and efficiency[56]. With this approach, the expression of a suicide gene – a gene encoding an enzyme that converts nontoxic prodrug into toxic metabolites, such as diphtheria toxin receptor (DTR) and thymidine kinase of the herpes simplex virus (HSVTK)[57] - is regulated by a microglia-specific promoter, and cell ablation is triggered by the administration of the corresponding toxin. The main advantage of these approaches is the reduced side-effects on other peripheral tissues and hence higher microglia specificity than pharmacological treatments with CSF-1R inhibitor[56]. Due to their self-renewal properties, another peculiar feature of microglia is that after depletion these cells can fully repopulate the brain within a few days[40]. In light of these reports, microglia depletion and repopulation has been tested as a therapeutic strategy for neurocognitive deficits in both therapeutic irradiation and space radiation models[23,25,45,46,49,58].

Microglia and space radiation-induced cognitive deficits: Chronic microglia activation along with behavioral and chronic deficits can be measured months after space radiation exposure in rodents[23,24,59-65]. Our group discovered that temporary microglia depletion with PLX5622 (1200 ppm) shortly after helium irradiation prevented the development of long-term cognitive deficits in male mice[23]. In this study we administered PLX5622 beginning one week after radiation exposure. The animals received PLX5622 for

15 days and then were returned to normal chow for the remainder of the study. Three months later we tested the ability of the animals to remember a previously encountered item. This tasks measures recognition memory and is dependent on the integrity of the hippocampus. Our results demonstrated that brief microglia depletion even after radiation exposure, followed by full repopulation, completely prevented the development of chronic recognition memory deficits in male mice. When compared to microglia from the irradiated mice, the repopulated microglia had a modified functional phenotype with reduced expression of lysosome membrane protein and complement receptors, all shown to be involved in microglia-synapses interaction[23]. The lower phagocytic activity observed in the repopulated microglia was paralleled by improved synaptic protein expression. Just recently, another group replicated these findings[66].

These data provide a mechanistic role for microglial cells in the development of cognitive deficits in male mice exposed to space irradiation. Furthermore, these reports have remarkable translatability as they demonstrate the potential for microglia depletion agents to be administered post radiation exposure to protect astronauts' cognitive functions during mission.



Microglia development and space radiation-induced memory deficits:
Space radiation exposure triggers chronic microglia activation and subsequent memory impairments. Microglia depletion and complete repopulation prevent memory deficits after space radiation.

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Declaration of Competing Interest

The authors declare no competing interest.

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