Age increases reactive oxygen species production in macrophages and potentiates oxidative damage after spinal cord injury

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There is an increasing incidence of spinal cord injury (SCI) in aged individuals. Previously, we demonstrated that aged mice exhibit worse functional deficits associated with differential macrophage activation following SCI. Reactive oxygen species (ROS)-mediated oxidative damage following CNS injury contributes to the secondary injury. It has been suggested that NADPH oxidase (NOX) plays an essential role in microglia/macrophage activation and subsequent inflammatory responses. We hypothesize that age increases oxidative damage in the injured spinal cord via increased NOX activity. In order to understand the mechanisms behind age-related differences in recovery, we compared oxidative stress associated with macrophage activation in 4- month-old (4 MO) and 14 MO mice after contusion SCI. By tracking oxidized dihydroethidine (ox- DHE), a superoxide marker, we identified that overall superoxide generation is significantly higher in 14 MO mice than 4 MO controls at 3 days post injury (dpi). Notably, the expression of NOX2, which we primarily detected in ROS-producing macrophages, is significantly increased in 14 MO mice, suggesting that macrophages and NADPH oxidase are the major cellular and subcellular sources of oxidative stress and may potentiate secondary injury in older animals. There is an increased activation of neurotoxic M1 macrophages (CD16/32-positive) in 14 MO SCI mice, while no difference in the level of protective M2 macrophage activation (Arginase-1-positive, ARG-1) was detected. Interestingly, we detected a larger percentage of ROS-producing ARG-1-positive macrophages in the injured spinal cords of 14 vs. 4 MO mice. These data indicate that age plays an important role in macrophage polarization in a way that normally protective M2 macrophages may potentiate secondary injury through ROS generation after SCI. Understanding the differences in inflammatory response and oxidative stress after SCI is important to determine how age at time of injury might affect endogenous repair processes, pathology, and clinical therapies.

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