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The role of free radicals in cerebral hypoxia and ischemia☆

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Abstract

This review focuses on the effects that ischemia and hypoxia have on the cerebral cortex and the cerebellum during different periods of life. The acute interruption or reduction of cerebral blood flow, that can be induced by several factors and clinical pathologies, reduces available oxygen to the nervous system and this causes either focal or global brain damage, with characteristic biochemical and molecular alterations that can result in permanent or transitory neurological sequelae or even death. Under these circumstances, an increase in the activity of different isoforms of nitric oxide synthase occurs and nitric oxide is produced. This excess of nitric oxide reacts with cellular proteins yielding nitrotyrosine, thus contributing to cerebral damage. This phenomenon has been studied at different stages of perinatal and postnatal development, including aging animals. Both the duration and the intensity of the ischemic injury were evaluated. In all cases there is overproduction of nitric oxide in ischemia, which may represent an effort to reestablish normal blood flow. Unfortunately, in many cases this response becomes excessive and it triggers a cascade of free-radical reactions, leading to modifications of cerebral plasticity and overt injury.

Keyword: Free radicals

Contents

Abbreviations: CBF, cerebral blood flow; XDH, xanthine dehydrogenase; NO, nitric oxide; PMN, polymorphonuclear; NOS, nitric oxide synthase; LPS, lipopolysaccharide; EDRF, endothelium-derived relaxing factor; cNOS, constitutive NOS; iNOS, inducible NOS; nNOS, neuronal NOS; BH_4, tethrahydrobiopterin; SOD, superoxide dismutase; CNS, central nervous system; CP, cortical plate; MZ, marginal zone; SVZ, subventricular zone; ADC, apparent diffusion coefficient; NMDA, *N*-methyl-D-aspartate; CO, carbon monoxide; L-NAME, N^G-nitro-L-arginine methyl ester; L-NA, N^G-nitro-L-arginine; 7-NI, 7-nitroindazile; AIF, apoptosis-inducing factor; MPT, mitochondrial permeability transition; CsA, cyclosporine A.

☆ This article is part of a series of reviews on Free Radicals and Stroke. The full list of papers may be found on the home page of the journal.

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Introduction

Stroke is the third leading cause of death and a major cause of long-term disability in industrialized countries. More than 80% of all strokes are a consequence of a permanent or prolonged occlusion, caused by either thrombosis or embolism, of one of the main or secondary cerebral arteries [1]. In all cases there is a dramatic reduction of oxygen that may provoke ischemia in the whole brain (global ischemia) or in defined cerebral territories (focal ischemia), depending on the cerebral artery occluded. Ischemia produces severe focal or global damage of the brain tissue accompanied by biochemical and molecular alterations and neurological sequelae [2]. As a consequence of the arterial occlusion, if not reversed within a short period of time, ischemia usually results in tissue infarction by anoxemia within the perfusion territory of the affected vessel, depletion of tissue energy, and eventual death of all cells involved [3–5].

Oxygen delivery depends on two variables, as described by the Fick equation: volume flow rate of blood and arterial oxygen content. A reduction in either one of these two parameters gravely affects vital brain function.

The aetiology of acute reduction in brain oxygen after interruption of cerebral blood flow (CBF) is multifactorial. Patients with a history of cardiac arrest, shock, carotid occlusion, and some genetic factors are more likely to suffer common ischemic stroke [6].

Several situations may lead to overall cerebral hypoxia. These include the progressive restriction of oxygen in asphyxia, the lack of the carrier protein hemoglobin in anemia, the irreversible binding of carbon monoxide to hemoglobin, severe hypotension, and exposure to hypobaric conditions.

In both circumstances, ischemia/hypoxia, the situation can be permanent or transitory, producing a greater or lesser accumulation of toxic substances, with concomitant deterioration and damage of neural tissue. The resolution of the hypoxic insult requires restoration of the blood flow (reperfusion) and recovery of the normal oxygen levels (reoxygenation).

Cerebral ischemia results in a time-dependent cascade of molecular events, including (i) rapid depletion of intracellular adenosine triphosphate (ATP) stores, (ii) anaerobic glycolysis, lactic acidosis, and membrane depolarization, (iii) glutamate excitotoxicity, (iv) entry of calcium, sodium, and water into the cells, resulting in cell swelling, (iv) activation of calcium-stimulated enzymes, mitochondrial dysfunction, free radical production, (v) activation of the immune system, (vi) overexpression of particular genes, and (vii) increased neuronal death [7–9].

Ischemia compromises membrane functions as well as cellular enzymes such as xanthine dehydrogenase (XDH). This enzyme oxidizes hypoxanthine in an NADPH-dependent manner, producing xanthine and uric acid and catalyzing the reduction of molecular oxygen to form both superoxide anion radicals (O_2^-) and hydrogen peroxide (H_2O_2) [10].

The infarction volume in the brain is an important determinant of the long-term consequences of ischemic stroke, leading to extensive neuronal damage in the different parts of the brain and subsequent neurological impairment.

Delays in recirculation, during the reperfusion period, increase the damage of the ischemia-injured arterial wall, the cerebral inflammatory response, and the damage to the brain tissue [11–13].

Cerebral ischemia is accompanied by a marked inflammatory reaction, that is initiated by higher expression of cytokines, adhesion molecules, and other inflammatory mediators, including prostanoids and nitric oxide [14].

Although the mechanism of neuronal cell death has not exactly been demonstrated in ischemia, several mechanisms including oxidative stress and glutamate toxicity have been reported to be involved in cellular death induced by hypoxia/ischemia [15, 16].

Recent experiments have demonstrated that nitric oxide (NO) and proinflammatory cytokines released by microglial cells, which act as resident macrophage-like cells in the brain, are partly responsible for neuronal cell death [15]. This indicates that cellular death caused by ischemia results not only from direct neuron necrosis induced by a poor supply of oxygen and glucose, but also by the cytotoxic agents released by the microglia [17–19].

There is increasing evidence indicating that cerebral ischemia followed by reperfusión elicits an acute inflammatory response, which implicates infiltration of polymorphonuclear (PMNs) leukocytes into the ischemic tissue. The role of this infiltration in the development of focal cerebral ischemia-reperfusion-induced damage has been extensively studied using various anti-PMN strategies [20–22]. The mechanisms whereby PMNs could contribute to ischemic damage may include their participation in oxidative stress, nitric oxide synthase (NOS) activity, and synthesis of NO [22–25]. In agreement with this hypothesis, the production of NO has been shown to increase in the brain during ischemia [26–30].

The nitrergic system

Much attention has been directed in the last two decades to unravel the role of the free radical, NO, whose overproduction by NOS has been postulated to occur in a number of clinical disorders including acute cerebral ischemia [31–34] and chronic neurodegenerative diseases, such as schizophrenia, Alzheimer's and Parkinson's diseases, and aging dementia.