HOMEOSTATIC EFFECTS OF ADENOSINE ON POTENTIALLY NEUROTOXIC GLIAL CELL ACTIVATION

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1. INTRODUCTION

The outcome of neurological impairment after stroke or traumatic brain injury depends largely on the amount of secondary nerve cell damage occurring as the result of a complex and not completely understood pathological process. This process includes a loss of the homeostatic control of the extracellular glutamate and intracellular Ca²⁺ by a number of interconnected vicious circles leading to neuronal Ca²⁺ overload and Ca²⁺ dependent structural damage. More recently, a pathological activation of glial cells has attracted increasing interest as possible causal factor for the generation of secondary nerve cell damage (see 1 for a review).

After stroke, progredient neuronal death is expanding into the so called penumbra, the primarily vital zone surrounding an ischemic infarct. Here, a disturbance of the extracellular ion homeostasis is discussed to impair physiologically required astrocyte functions, such as the uptake of neurally released K⁺ and glutamate from the extracellular space. This would favor nerve cell depolarization, seizuring and excitotoxic damage. An ischemia induced astrocyte reaction, as indicated by cell mitosis, hypertrophy and an increased formation of glial fibrillary acidic proteins (GFAP), has been observed in the gerbil hippocampus already at 2 days after transient forebrain ischemia, i.e. before apparent nerve cell damage (2). The “activation” of microglial cells, indicated by cell proliferation

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and the expression of immune surface receptors, occurred even earlier, i.e. within the first hours after transient brain ischemia (3, 4). By the release of interleukin 1 (5), reactive microglial cells are presumably involved in triggering the astrocyte reaction. The acquisition of immune competent properties enables microglial cells to contribute to a meaningful response of the cellular defense system under pathological conditions. However, if the microglia activation runs out of control or if it is hyperergic, additional and progradient nerve cell damage may be elicited by the newly adopted immunological weapons. Among those, free oxygen radicals are produced in particularly high amount by microglial cells and microglia-derived macrophages (6). In conjunction with NO, oxygen radicals form peroxynitrites, i.e. ONOO\(^-\), which may exert heavy damage the cytoskeleton and cell membranes. There is evidence that reactive oxygen intermediates are also involved in the formation of the aggregating and presumed neurotoxic \(\beta\)-amyloid by oxidating the C-terminal residues of the precursor protein (7). Such a pathological processing of \(\beta\)-APP in microglial cells may not only be relevant for the pathogenesis of neurodegenerative diseases but also for nerve cell death in response to brain ischemia which upregulates microglial \(\beta\)-APP formation (8). A common pathogenic significance of activated microglial cells for both types of diseases is supported by the finding that a positive immunostaining with antibodies, contained in cerebrospinal fluids of Alzheimer patients, was exhibited by microglial cells which had been activated by transient forebrain ischemia (9). A release of potentially neurotoxic cytokines presumably adds to microglia-related brain damage. In this context is interesting that the toxic \(\beta\)-amyloid fragment seems to stimulate the release of the cytokine TNF-\(\alpha\) from cultivated microglial cells. This favored the release of NO and caused, in conjunction with other factors, neuronal cell death in vitro (10). TNF-\(\alpha\) may further add to the impairment of neuronal function by inducing apoptosis in oligodendrocytes building the myelin sheaths of axons (11).

These findings suggest that pathologically activated microglial cells could maintain a vicious circle, i.e.: a microglial production of oxygen radicals favors the pathological processing of \(\beta\)-APP which, in turn, stimulates microglial activation with an increased formation of TNF-\(\alpha\), NO and the particularly aggressive ONOO\(^-\), formed in the presence of oxygen radicals. It follows that an interruption of this microglia-related vicious circle and the maintenance of physiologically required mature astrocyte functions may provide a new neuroprotective strategy for the treatment of diseases in which the activation of glial cells plays a pathogenic role. Prerequisite for the development of such a pharmacological strategy would be a detailed knowledge about the so far only vaguely understood molecular signaling that controls the potentially neurotoxic properties and the differentiation stage of activated glial cells. Alternatively, the sophisticated know how of an endogenous cell regulator may be used and pharmacologically reinforced. Such an ancient endogenous cell modulator, which has learned during evolution how to influence the complex molecular signaling in glial cells, is the nucleoside adenosine (12).

2. METHODOLOGICAL ASPECTS

Regulatory effects of adenosine on activated glial cell properties were studied in astrocyte and microglial cell cultures obtained from embryonic or new born rat brains. These cultured immature glial cells resemble pathologically activated glial cells in that they are less differentiated and show a high proliferation rate. Cultured rat astrocytes retained in vitro the appearance of so called type 1 astrocytes, cubic shaped cells, and they lack elongated cell processes which are taken as criteria for differentiated astrocytes. They also had not expressed