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Review Article

THE RELATIONSHIP BETWEEN EXCITOTOXICITY AND OXIDATIVE STRESS IN THE CENTRAL NERVOUS SYSTEM

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Abstract—Excitotoxicity and oxidative stress are two phenomena that have been repeatedly described as being implicated in a wide range of disorders of the nervous system. Such disorders include several common idiopathic neurological diseases, traumatic brain injury, and the consequences of exposure to certain neurotoxic agents. While there is evidence that metabolic derangements can lead to these adverse processes, and that these processes may synergize in their damaging effects, the degree of interdependence, and the causal relation between them is not clear. The intent of this review is to delineate potential mechanisms which may unit hyperexcitation to the excessive generation of reactive oxygen species. The degree of linkage between these events appears rather strong. It is likely that excitoxicity frequently leads to a pro-oxidant condition but that high rates of generation of reactive oxygen species are not invariably accompanied by a hyperexcited neuronal state Both excitoxic and 'oxidotoxic' states result from the failure of normal compensatory antiexcitatory and antioxidant mechanisms to maintain cellular homeostasis.

Keywords—Brain, Nervous system, Excitotoxicity, Reactive oxygen, Cerebral metabolism, Neurological disease, Free radicals

INTRODUCTION

Excitotoxicity was first described by Olney almost two decades ago,[1] when it was reported that death of neurons occurred as a result of excessive exposure to the EAAs glutamate and aspartate. This list of EAA has grown over the years to include kainic acid, ibotenic acid, quinolinic acid, cysteine sulfinic acid, β-*N*-oxalyl-L-*a*, β-diaminopropionic acid, homocysteic acid, and folic acid, all of which are considered to be analogs of glutamate and aspartate. The excitotoxicity hypothesis postulates a series of events leading to neuronal death, and is comprised of three assumptions:[1] (a) neuronal cell depolarization leading to the release of EAA; (b) a common EAA receptor mediates both excitation and toxicity; and (c) propagation of a chain of events leads to neuronal cell dysfunction and death. Since the advent of this hypothesis, numerous investigators have attempted to substantiate this theory of excitotoxicity. Both basic research and clinical efforts have led to the general proposal that excessive neuronal activity effected by abnormal extracellular levels of EAA plays a role in epilepsy, transient cerebral ischemia followed by restoration of the vascular oxygen supply, Alzheimer's disease, Parkinson's disease, and Huntington's disease.

The potential role that ROSs play in the central nervous system (CNS) has been thoroughly reviewed[2] and general properties of ROS, sites of ROS formation, methods of detection, and factors underlying excessive ROS formation in neural tissue have been discussed.[3,4] These works described the involvement of ROS and their relevance to abnormal conditions of the CNS, such as hyperoxia, hemorrhage, trauma, and aging. The literature is also abundant with refer-

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ences suggesting ROS involvement for each of the neurodegenerative disorders mentioned above that are proposed to be associated with excitotoxicity. Since excitotoxicity and ROS are both pathogenic events linked to a wide range of neurological disease states, the goal of this review is to examine the potential relationship between neuronal depolarization and ROS generation, and to gain an alternate perspective on the mechanisms underlying this relation.

EXCITOTOXICITY

As mentioned previously, Olney's excitotoxicity hypothesis was based on three assumptions, one as a depolarizing triggering mechanism, a second involving a receptor locus, and the last involving a chain of metabolic events leading to neuronal dysfunction. Numerous reports in the literature detail the foundation of this hypothesis.[5,6] This review will briefly summarize recent progress in this area of research.

There is ample support today to suggest that glutamate and other EAAs stimulate multiple subtypes of receptors. The most studied receptor site is that specific for NMDA.[7] The first two assumptions of the hypothesis, glutamate release effected by depolarization, and activation of the NMDA receptor, appear to be well established. Investigations on the subsequent chain of metabolic events that occurs after NMDA receptor activation is a complex and less clearly understood cascade. These include the following sequence. Stimulated NMDA receptors open calcium channels leading towards an increase in intracellular calcium. Concurrently, sodium influx increases through ion channels that are primarily mediated by the α-amino-3-hydroxy-5-methyl-4-isoxalolpropionic acid (AMPA)/kainate receptor. This cationic influx is followed by chloride ions and water which eventually lead to edematous cell swelling and dysfunction. Activation of another glutamate receptor subtype called the metabotropic receptor, signals increases in diacylglycerol and inositol 1, 4, 5-triphosphate. The process from depolarization to activation of the metabotropic receptors is collectively referred to as induction.[8] The stages of amplification and expression follow in a cascade of events. Excessive utilization of metabolic processes without timely replenishment of cellular energy reserves can lead to depletion of adenosine triphosphate (ATP) and disruption of anabolic processes. Such a deficit can result in partial collapse of ion gradients. The consequent increase in cytosolic levels of calcium and depletion of potassium can exacerbate initiating events by elevation of neuronal firing rates. In addition, lowered levels of substrates may retard mitochondrial oxidative phosphorylation and also impair the efficiency of electron transfer.

Recently a refinement of Olney's excitotoxic hypothesis has been proposed[9] in which it suggested that the central dogma of the hypothesis is termed "strong excitotoxicity." Albin and Greenamyre[9] propose that EAA receptor function may be chronically altered, and thus neuronal cell metabolism or membrane potential may be impaired. The culmination of these alterations ("weak excitotoxicity"), may leave neurons excessively susceptible to the direct excitatory effects of EAA.

Although a final common mechanism has yet to be delineated in the neurodegenerative disorders previously mentioned, a proposed descriptor in such disorders is the "excitotoxic index." This index was first used to describe the imbalance between excitatory and inhibitory processes at the neuronal level, where brain concentrations of glutamate and glycine, and the inhibitory amino acid gamma-aminobutyrate (GABA) were measured in rats subjected to transient global ischemia.[10] This index is intended to provide an overall assessment regarding the state of excitotoxicity and is thus a measure of neural imbalance. Under basal conditions the index would display homeostasis between excitatory and inhibitory processes. As excitatory events increase, a mild shift in the index occurs, but under most conditions, compensatory mechanisms in the brain respond and return the index to its resting state. Under conditions of excessive excitatory stress, an even greater shift occurs that may lead to neuronal dysfunction and tissue damage. Such a failure of equilibrium, which is represented by the excitotoxic index, may reflect many of the major dynamics involved in mechanisms of neurodegenerative disorders.

OXIDATIVE STRESS

The fact that oxygen is ubiquitous in aerobic organisms has lead to the concept of the oxygen paradox; namely the fact that this molecule which is essential for aerobic life, is also a precursor to the formation of harmful ROS.[11] The term "oxidative stress" was first coined by Sies[12] to account for the imbalance between ROS and the antioxidant opposing forces. ROS may be oxygen-centered radicals possessing unpaired electrons, such as superoxide anion and hydroxyl radical, or covalent molecules such as hydrogen peroxide.[13,14] These and other ROS have been implicated in a wide range of physiological, toxicological, and pathological phenomena.

Sources of ROS include mitochondria, where a significant fraction of molecular oxygen consumed is incompletely reduced and appears as ROS.[15] It has been suggested that this proportion may be increased when normal function of electron transport systems is com-

promised. However, this question, which is an active subject of current research, remains unresolved and conflicting data exist.[16] Lowered pH due to high levels of glycolytic activity may not only accelerate the process of liberating protein-bound iron in organisms, but it may also lead to an impairment of oxidative ATP generation, and to the appearance of the pro-oxidant protonated superoxide.[17] Iron liberation can ultimately enhance the degradation of important iron-binding proteins such as ferritin and transferrin. A small increase in levels of free iron within cells can dramatically accelerate rates of ROS production.[18]

Xanthine oxidase is a prime generator of superoxide anion and may be a significant exacerbating factor in several pathological states. Intracellular glutathione stores can be compromised by agents forming tight complexes with sulfhydryl groups, or by inhibition of the related glutathione reductase or peroxidase. Glutathione depletion can result in neurological deficit.[19] Extracellular formation of superoxide anion by phagocytes has long been recognized as a bacteriocidal mechanism. Similar oxidative activity has been observed in cerebral microglia.[14] Astroglial activation is a common event following neural trauma and reactive astrocytes are active in clearance of cell debris and ultimately, in the formation of glial scar tissue.

All organisms generate a basal level of ROS, where the pro-oxidant status is counterbalanced by non-enzymatic antioxidants and antioxidant enzymes. This balance between pro-oxidant and antioxidant components can be termed the "oxidative index," analogous in concept to the excitotoxic index previously described. In situations that place the brain under heightened oxidative conditions (increase in the oxidative index), such as genetic vitamin E deficiencies[20] or following exposure to a variety of neurotoxic agents,[21] the oxidative index of the brain shifts to a higher pro-oxidant status. When this oxidative stress occurs as a transient event, compensatory mechanisms in the brain may return the nervous system to normal function. Most organisms appear to respond to a flux in the redox potential by re-establishing homeostatis. The inability of the brain to respond to prolonged or severe shifts in the oxidative index will lead to ROS-induced alterations at the molecular and cellular levels and disrupt or halt normal cell function. It should be borne in mind that many phenomena not related to excitotoxicity, such as inflammation and radiation induced injury, may also involve generation of ROS.

THE RELATION BETWEEN EXCITATORY AND OXIDANT EVENTS

Several common neurological diseases are suspected to involve a combination of interacting excitatory and oxidative processes.[22] In addition, several neurotoxic agents may trigger both of these events simultaneously.[23] This, however, does not illuminate the precise relation between, and degree of interdependence of, these phenomena.

Elevated levels of calcium can enhance free radical-induced damage to cerebral morphological fractions,[24] and antioxidants can protect against cytotoxicity induced by high intracellular levels of calcium in the CNS.[25] Furthermore, reduction of cytosolic levels of ionic calcium may reduce damage to DNA effected by hydrogen peroxide,[26] and calcium channel blockers can prevent cyanide-induced lipid peroxidation and cell death.[27]

Glutamate toxicity in a neuronal cell line has been attributed to inhibition of cystine transport and consequent oxidative stress.[28] Activation of the NMDA receptor site has been implicated in the post-ischemic elevation of lipid peroxidation in the hippocampus.[29] There is a report of exacerbation of NMDA toxicity in glutathione-deficient cortical cultures.[30] Increased levels of cytosolic free calcium may result from either breakdown of the steep concentration gradient of calcium across the plasma membrane, or by liberation of the large amounts of calcium bound intracellularly within mitochondria or endoplasmic reticulum. Calcium stimulation of phospholipase A_2 and thence the arachidonic acid cascade, can lead to the release of arachidonic acid. The enzymic conversion of this compound to prostaglandins, leukotrienes, and thromboxanes by cyclooxygenases and lipoxygenases, which directly utilize molecular oxygen, leads to considerable ROS generation.[13,31,32] This may be a means by which excitatory events promote excess generation of ROS.[33] Phospholipase A_2-generated oxidative activity has also been shown to inhibit functioning of the GABA receptor gated channel, the major inhibitory receptor complex of the cell.[34] Calcium can also activate the respiratory burst of polymorphonuclear leukocytes leading to production of, and extracellular appearance of, superoxide anion. Such leukocytes are known to accumulate in post-ischemic cerebral tissues.[35] Finally, calcium activates the proteolytic conversion of xanthine dehydrogenase to xanthine oxidase.[36]

NMDA agonists are especially potent in the stimulation of nitric oxide synthetase,[37] and nitric oxide can interact with superoxide to form the intensely oxidant nitroperoxyl radical. There is evidence suggesting that the free radical nitric oxide mediates the neurotoxicity of glutamate.[38]

Kainate-induced damage to cerebellar neurons may also be mediated at least in part, by induction of superoxide.[39] In this latter study, allopurinol, a specific inhibitor of the superoxide-forming enzyme, xan-

thine oxidase, and hydroxyl radical scavengers such as mannitol, were protective against kainic acid-initiated tissue injury. A possible mechanism underlying this may be through the induction of nerve growth factor.[40] This factor has been found to induce several antioxidant enzymes such as catalase and glutathione peroxidase.[41] Such induction appears to involve genetic derepression, and a reciprocal mechanism also appears to exist whereby ROS can induce several growth factors.[42] However, kainate has also been found to promote ROS generation, not only in vivo,[43] but also directly in an isolated subcellular system.[44]

The relation between calcium and free radicals seems complex in that *extracellular* calcium may be an important defence against lipid peroxidation.[45,46] In this context, it may be relevant that the influx of calcium into the cell seems less critical that the liberation of calcium from intracellular storage sites.[47]

Excitatory events may stimulate ROS, but there is evidence for a reciprocal relation between these phenomena. Thus, ROS can lead to extracellular release of excitatory amino acids,[48] or can release mitochondrial calcium into the cytosol.[49,50] Peroxidative damage can also impair inhibitory processes as judged by inhibition of GABA-stimulated chloride uptake.[34] Glutamine synthetase appears to be especially sensitive to ROS-induced damage, and this may increase intracellular glutamate concentrations.[51] A bidirectional cooperation between excess neuronal activity and excessive ROS formation may therefore be relatively common.[52] Regardless of whether or not they can quantitatively intensify each other, ROS and increased levels of calcium within the cell can act synergistically, for example, by bringing about deleterious cross-linking reactions between proteins.[53]

There are, however, conflicting data suggesting that pro-oxidant conditions reduce the ionic gating of the NMDA receptor site,[54] and that lipid peroxides act pharmacologically as depressants.[55] Some of these apparent contradictions may be due to the key role of iron in ROS production. The presence of ionic iron is essential in the promotion of post-ischemic lipid peroxidation,[56] and iron liberation from sequestration sites within proteins is closely related to tissue pH.[17]

OXIDATIVE AND EXCITOTOXIC INTERRELATIONS IN NEUROLOGICAL DISEASES

Seizure activity

It is widely accepted that seizures alter many chemical and biophysical processes in the CNS, and there are several reports indicating that ROS may play a role in seizure phenomena. The generation of superoxide anion in brain was reported to increase during seizure activity in newborn pigs.[57] Accumulation of arachidonate metabolites[58] and free fatty acids[59] have been reported in drug-induced seizures. These data suggest that cyclooxygenase metabolism of arachidonate leading to ROS generation is an integral component of seizure activity.

Increased superoxide dismutase activity has been reported in brains isolated from kindled rats, and the administration of exogenous superoxide dismutase affords some protection against these seizures.[60,61] Alterations in antioxidant enzyme activities follow chemical-induced seizures.[62] We have found that a brief (1 sec) electroconvulsive shock is sufficient to cause elevated rates of ROS formation in rat cerebral cortex (unpublished data).

The attenuation of convulsive activity by phenytoin or corticosteroids reduces cerebral levels of lipid peroxidation.[63] It should also be recognized that in some seizures a prolonged hypertension may occur, which may contribute ROS generation as a result of accelerated arachidonate metabolism via prostaglandin H synthase.[3,4]

A reciprocal relation between seizures and ROS appears to exist, since free radicals induced in the brain by hyperbaric oxygen can precede, and may provoke, convulsions.[64]

Huntington's disease

The genetically inherited disorder known as Huntington's disease (HD) results from the spontaneous degeneration of neurons primarily localized in the striatum. While the etiology of HD remains unresolved, it is generally accepted that the corticostriatal afferents that innervate striatal neurons employ glutamate as an excitatory transmitter. Excessive glutamate activation of excitatory receptors could be paramount to the pathophysiologic etiology of HD. Evidence for the role that excitotoxic stress plays in HD has been progressively increasing for three decades. Glutamate was shown to destroy striatal neurons following direct intracerebral application.[65] Coyle and coworkers reported that lesion of striatal neurons with kainic acid leads to neuronal degeneration and behavioral deficits resembling Huntington's Chorea.[66]

More recently, decreases in glutamate receptors from the caudate and putamen isolated from HD patients were reported.[67] These findings probably reflected the loss of cells in the same brain regions that typically occur in HD. Bioenergetic defects, such as mitochondrial metabolic dysfunction, have also been reported in HD brain.[68–70] Most recently it has been shown that direct injection of the mitochondrial energy impairing agent amino-oxyacetate into the stria-

tum produced HD type lesions.[71] Energy impairment may be a result of excess demand arising from excitatory and cerebral oxidative stress in the HD brain.

Reference to a direct association between oxidative stress and HD has not been reported. Perry and Hansen[72] found no statistically significant differences between total glutathione content in the caudate nucleus isolated from patients with HD compared to tissue from neurologic or non-neurologic diseased patients. However, accurate assessment of brain glutathione content in humans is complicated by the artifactual oxidation of glutathione in isolated tissues.[73] Vitamin E may be a better marker for measurement of oxidative stress in brain, and Adams and coworkers have shown increases in brain regional vitamin E content in patients with Alzheimer's and Parkinson's disease.[73] Overall, a relation between excitatory and oxidative stress remain circumstantial possibilities in pathogenesis of HD.

Aging and Alzheimer's disease

A combination of oxidative and excitatory stress has been proposed to underlie some of the behavioral deficits associated with aging.[74] Both normal cerebral aging and Alzheimer's disease (AD) involve a degree of cell loss. The "free radical theory of aging," proposes that the accumulation of free radical-induced damage is a main contributor to the aging process. Some investigators[75] have asked how random events such as ROS-induced alterations could account for a final common aging phenomenon. Our theory is that the brain goes through periodic oxidative stress where the oxidative index shifts to pro-oxidant conditions.[4] The progressive accumulation of damage from these intermittent, stressful conditions over a long period can contribute to the aging process, but by no means is the single cause to aging. It is likely that cumulative oxidative stress incurred over many years plays a major role in aging. This philosophy is directly in line with the "weak excitoxicity" hypothesis proposed by Albin and Greenamyre.[9] Their suggestion is that bioenergetic disturbances produce this weak but prolonged hyperexcitation, and that this may render an organism increasingly susceptible to episodes of severe excitotoxicity.

For several years, theories on the etiology of AD have drawn an association between aging and AD. However, it is clear that the pathology of AD is complex and involves multiple abnormal fibrous protein deposits. AD is typically characterized by the presence of neuritic plaques and neurofibrillary tangles. Beta-amyloid, the abnormal protein that is widely dispersed in AD brain, has been shown not only to enhance glutamate toxicity in culture,[76] but also to exert a degree of neurotoxicity when injected directly into rat brain.[77] Additionally, and in parallel to Huntington's disease, AD is associated with bioenergetic deficits,[78,69,79,68] and this may lead to induction of ROS.[80]

Superoxide dismutase levels are elevated in AD.[81] Since this is an inducible enzyme, a rise in levels implies a compensatory response to excess levels of ROS. In parallel, potentially adaptive changes have been reported in levels of alpha-tocopherol and glutathione in brains from AD patients.[73] Aluminosilicate deposits are known to be present within senile plaque cores, and this material has recently been shown to induce ROS generation.[82] The presence of ferritin in the senile plaque, has also been proposed as evidence for a role for free radicals in amyloid formation.[83] There is less evidence for a relation between AD and chronic low level hyperexcitatory events. However a correlation of extended dietary ingestion of a glutaminergic agonist, β-methylamino-L-alanine, with a delayed occurrence of a neurological complex (Amyotrophic Lateral Sclerosis-Parkinsonism-Dementia) has been claimed.[84] In addition, application of excitotoxins to the brain can both selectively destroy cholinergic neurons, and effect late-occurring neuropathological changes resembling neuritic plaques and neurofibrillary tangles.[85]

Parkinson's disease

A role for oxidative stress in the processes underlying MPTP neurotoxicity has been proposed. This compound, a contaminant of an illicitly manufactured meperidine analogue, has been the subject of much interest since the neurological damage that it can cause closely resembles Parkinson's disease. MPTP is a very specific dopaminergic neurotoxin. Dopaminergic circuitry is especially vulnerable to neurotoxic damage, and this is at least in part due to the readiness with which dopamine is auto-oxidized in the presence of trace amounts of metals with multivalence potential. In addition, dopamine can be enzymically oxidized by monoamine oxidases to 3,4 dihydroxyphenyl acetaldehyde and H_2O_2. Glutamate antagonists such as MK-801 have been found to attenuate the behavioral deficits associated with dopaminergic neurotoxicants, and thus chronic excitotoxicity has been proposed to constitute a component of Parkinson's disease.[86]

There is considerable support for the "mitochondrial theory" of MPTP toxicity, which postulates that 1-methyl-4-phenylpyridinium (MPP^+), the ultimate oxidation product of MPTP, blocks the reoxidation of NADH dehydrogenase by coenzyme Q_{10} and eventu-

ally leads to ATP depletion in a rotenone-like fashion.[87] However, there are also several studies that suggest oxygen radicals may play a role in the MPTP-induced neuronal damage.[88,89] These concepts may be reconciled by the finding that the metabolic inhibitors rotenone and antimycin can increase the generation rate of oxygen radicals in crude rat synaptosomes and mitochondria.[21,90]

Emerging information concerning MPTP has led to several new ideas concerning Parkinson's Disease. These concepts include both the possibility of an environmental agent being contributory to the pathogenesis of this disease,[91] and of the potential for antioxidant therapy of this disorder.[92] Parkinsonism has been associated with abnormally high levels of superoxide dismutase within the substantia nigra,[93] implying an induced response to oxidative stress. Abnormally low levels of ferritin are associated with Parkinson's Disease.[94,95] This suggests that iron may be present in a low-molecular weight form in Parkinson's disease. While the level of ferritin in Parkinson's disease patients remains controversial, recent studies, using both x-ray microanalysis and laser microprobe mass analysis, have found a significant excess accumulation of iron in neuromelanin in Parkinson's disease.[96] This diminution of iron-sequestering capacity could also enhance ROS generation.[97] Evidence for a role of iron and oxidant stress in the Parkinsonian brain has been the subject of a recent symposium.[98]

The findings presented support the weak excitotoxicity hypothesis, and further buttress the proposal of a dual involvement of oxidative and excitoxic stress in the pathogenesis of this disorder.

Stroke and ischemia

Stroke leads to cerebral ischemia that can be of varying duration as the capillary supply may be restored. In addition to hemorrhage caused by the extravasating type of stroke, all transient interruptions of vascularity (including traumatic injury to the brain or spinal cord) have the potential to lead to severe post-ischemic damage. The presence of free hemoglobin in nerve tissue can exacerbate potential for ROS-effected damage, and that this is due to the appearance of free iron salts is suggested by protection afforded by desferrioxamine.[22,99] Such events have been proposed to account for posttraumatic epileptogenesis.[100]

Free radical generation during cerebral ischemia may underlie delayed neuronal death.[101,102] Ischemic injury and edema within the CNS have frequently been found to involve excessive oxidative activity as judged by lipid peroxidation, induction of superoxide dismutase, and the protection afforded by antioxidant chemicals, such as alpha-tocopherol, iron chelators, or 21-aminosteroids.[103–111] The potential for therapy of spinal cord injury by use of novel pharmacological antioxidants is already very promising.[112] In many cases, attenuation of pro-oxidant activity alone, appears to be protective against neural insult. Oxidative damage may occur independently of the excitotoxicity, which is often an indirect secondary event with the potential for exacerbating the initial trauma.

The pathological changes consequent to restoration of the normal blood supply may be initially related to excessively high levels of cytosolic calcium,[17] and this appears to be the basis for subsequent elevations of ROS.[28,39,113] The high metabolic rates associated with reperfusion injury can lead to excessive ROS production.[114] At the onset of ischemia there is an accumulation of hypoxanthine due to breakdown of adenine nucleotides. Upon reperfusion, a combination of oxidative and proteolytic events converts xanthine dehydrogenase to the direct oxygen-acceptor, xanthine oxidase. The final combination of elevated enzyme and substrate leads to superoxide production and consequent oxidative stress.[36] It is significant that focal cerebral ischemic injury is attenuated in transgenic mice constitutively overexpressing superoxide dismutase.[115]

The complex relation between ROS-induced events and anoxia is illustrated by the finding that morphological damage to the penumbra region around an ischemic region is generally more severe than at the core.[107] The presence of excess oxygen free radicals in post-ischemic tissue has been directly demonstrated.[104] Even a brief period of ischemia can lead to ROS generation and thence to delayed neuronal death.[101] Hypoxia has been directly shown to stimulate ROS production by astroglia.[116]

It has been proposed that during ischemia, ROS and EAA may cooperate in effecting neuronal damage.[117–119] Transient ischemia elevates cerebral levels of both EAA and rates of hydroxyl radical formation.[120] Whether one of these events gives rise to the second is not well established. Excitatory events may stimulate ROS, but there is also evidence that ROS can lead to release of EAA and thus a bidirectional relationship is an attractive hypothesis.[48]

The most likely primary course of events following ischemia may be as follows. The resultant low oxygen supply (ischemia) can lead to reduced energy supply. This anabolic deficit may then result in diminution of the ionic gradients across the plasma membrane. In addition, the capacity of the energy-requiring high affinity reuptake systems is diminished, and extracellular levels of glutamate rise. Thus, a hyperexcitable

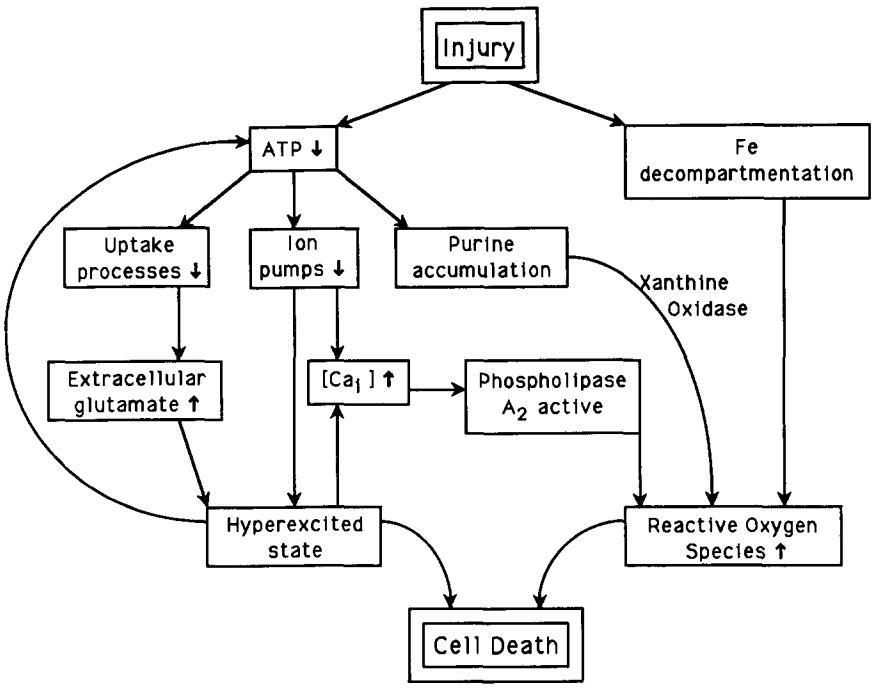

Fig. 1. Interactions between excitatory and oxidative events.

state ensues, resulting from both a reduced axonal membrane potential, and increased calcium-stimulated neurotransmitter release. Subsequent reperfusion leads to an abrupt return of glucose and oxygen to neurons, which disrupts mitochondrial function. Such uncoupling then increases rates of generation of ROS. Elevated intracellular calcium also exacerbates levels of ROS by way of phospholipase activation. This gives rise to a cycle leading to increasingly severe neuronal damage. This may explain why protection is afforded against ischemic states by both ROS scavengers and calcium channel antagonists.

CONCLUSION

Under normal circumstances excess ROS production is prevented by a host of antioxidant agents and detoxifying enzymes. Extracellular EAA levels are regulated by potent high affinity transmitter re-uptake mechanisms found in both neurons and glia. Failure of cell energy generation processes to keep up with demand can lead to both the hyperexcited state and to the appearance of excessive amounts of ROS. Disease almost invariably involves increased cellular entropy, and so it is reasonable to expect that oxidative stress and excitotoxicity can be contributory factors to many superficially unrelated neurological diseases. In addition to the definitive characteristics of a given disease, a less specific feature involving anabolic failure is likely to be present, and this may account for phenomena under discussion being common to several neurodegenerative diseases.

Major surges in intracellular levels of free ionic calcium occur normally, while basal rates of ROS production are not known to flux as dramatically. In pathological states, abnormally high resting calcium levels are thus perhaps more likely to initially precede extended changes in ROS. Some of the potential interactions between excitatory and ROS-enhancing events are illustrated in Figure 1.

An unequivocal causal relation between these two broad classes of events remains to be uncovered. While the general outlines of the co-occurrence and partial interdependence of these events is clear, the precise cause–effect relationships taking place in the living brain are unknown. Many correlations have been made in isolated systems but the quantitative role played by these events in the intact animal awaits elucidation. However, the potential therapeutic validation of antioxidants and calcium channel blockers, need not await final clarification of the interrelation of these processes that are initiated by failure of intracellular generation of energy and by collapse of ion gradients.

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ABBREVIATIONS

AD—Alzheimer's disease
EAA—excitatory amino acids
HD—Huntington's disease
NMDA—*N*-methyl-D-aspartic acid
MPTP—1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine
ROS—reactive oxygen species