Neurotoxicity And Neurodegeneration: Implications For Neuropsychiatric Disorders

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The word neurotoxicology generally signifies damaging effects of chemicals, drugs or poisons on key structural elements of the central nervous system (CNS): cell bodies, axons and neuroglia. The chemically-induced damage to these elements would predictably produce functional deficits which may be manifested as impairments of sensorimotor functions and or behaviour. However, in recent years the term neurotoxicology has assumed a much broader definition which is forced by the observations that profound changes in the function of the CNS (*e.g.*, such as those associated with certain drugs of dependence) may result from very subtle or regionally localized damage to the neuronal structures, or these changes may occur in the absence of obvious structural damage. Additionally, effects of drugs or xenobiotics on the brain, especially during development, may be expressed some time after initial exposure to such chemicals. While the term neurotoxicology very often signifies that damaging chemicals are exogenous to the nervous system, it is now widely recognized that injury to the nervous system may be inflicted by abnormal activity of chemicals, such as L-glutamate and other excitatory amino acids, that are endogenous to the brain. Whatever the definitions or dimensions of this phenomenon, it is becoming increasingly clear that neurotoxicant exposure can produce debilitating symptoms of the neurological or psychiatric diseases. Indeed, it has been recognized for some time that neurodegenrative diseases such as Alzheimer's and Parkinson's disease may have a neurotoxicological basis. In the past years there has been appreciation of the fact that psychiatric diseases such as schizophrenia, which show no overt neuronal pathology, may also involve neurodegeneration. The following chapters in this section focus on these new dimensions of neurotoxicology; the findings reported here may be expected to increase appreciation of the role of diverse neurotoxicants in neurological and psychiatric disorders.

In Chapter 2, Weiss and Koob introduce the concept of drug addiction as a "functional neurotoxicity" phenomenon, implying that CNS exposure to certain drugs on a chronic basis profoundly alters the function of specific brain neurons without significantly damaging such neurons. Such modifications, impacting specifically on neurons implicated in the reward circuitry, are manifested as compulsive behaviours where seeking the drug becomes a paramount feature. Historically, rewarding behaviours, including those based on drugs, have been attributed to the activation of dopaminergic neurons that are localized to the ventral tegmental area and that project to the corticolimbic area via the mesolimbic pathway (Yorkel and Wise 1975; Phillips and Fibiger, 1987). However, on the basis of recent neurochemical and anatomical evidence, Weiss and Koob suggest that it is altered activity of structures in the " extended amygdala", incorporating specific basal forebrain regions including shell of the nucleus accumbens, that is involved in drug-induced disruptions of the brain reward mechanism. Surgical or pharmacological manipulations of structures in this system can

modulate reinforcing drug effects. The authors also suggest that activity of the same system also contributes to the negatively reinforcing effects of drug withdrawal, the neurochemical changes (*e.g.*, reduced dopamine release) being opposite to those occurring in reinforcement (*e.g.*, increased dopamine release). Additional contributions in this respect may come from the brain stress system (Koob and Le Moal), especially those involving corticotrophin releasing factor (CRF). Thus, persistent changes in both the reward and stress systems may direct the drug seeking responses. Finally, on the basis of evidence that animals with access to cocaine will continue to augment drug intake, Weiss and Koob theorize that chronic drug exposure can change the "set point" for the reward system. The identification of biochemical and molecular mechanisms underlying this constitute a major challenge to future investigations.

Two experimental studies in Chapters 4 and 5 address the notion that early CNS exposure to psychoactive drugs or xenobiotics has important implications for CNS function at maturity. In a highly comprehensive and integrated study, Harvey *et al.* (Chapter 4) demonstrate the biochemical and behavioural sequale of prenatal intravenous cocaine exposure in the rabbit. The choice of the animal model used in this study may at first seem rather unusual as most neurobehavioural studies focusing on prenatal drug effects employ rodent models, however, the authors put forward strong arguments in favour of this choice, an important one being its similarity to humans in respect to drug sensitivity (Denenberg *et al.*, 1982). Using this model, Harvey and colleagues report on startling changes in the neurochemical, behavioural and electrophysiological indices assessed following *in utero* cocaine exposure. Neurochemically, prenatal cocaine produces both presynaptic and post synaptic changes in dopaminergic function: impaired dopamine release (Wang *et al.*, 1995a) and a significant decoupling of the D_1 dopamine receptor from its second messenger in the neostraitum and mesolimbic cortex (Wang *et al.*, 1995b). The behavioural significance of reduced D_1 receptor function is reflected in the impairment of sensorimotor function and discrimination learning in an instrumental avoidance task. The electrophysiological analysis of cortical neuron function suggests that deficits in dopamine receptor function alter development of the cingulate cortex which in turn produces attentional and learning deficits in maturity. Dopaminergic neurons in the cingulate cortex are known to interact with GABA neurons and the investigators suggest that altered D_1 receptor function is the contributing factor in the increase in the number of cingulate GABA neurons resulting from exposure to prenatal cocaine. Thus, impairments of the D_1 receptor function by the drug appear to be a fundamental basis of the structural and behavioural changes that occur in the progeny after exposure of the developing brain to cocaine. It is highly significant that the neurochemical and behavioural changes that have been identified in the progeny exhibit high level of specificity and, importantly, they occur in the absence of altered physical characteristics.

The investigation by Ericksson *et al.* (Chapter 5) examines neonatal effects of nicotine, possibly the commonest addictive substance, and, polychlorinated biphenyls (PCBs) and polybrominated diphenylethers, two classes of environmental toxicants, on the neurochemical and behavioural development in the mouse model. Nicotine is recognized to modulate activity of the central dopaminergic and cholinergic neurons via distinct receptor sites - the high and low affinity sites (Westfall *et al.*, 1989; Norberg, 1993). The low affinity site corresponds to the site that binds the snake toxin, alpha-bungarotoxin. The investigators have found that low nicotine dose exposure during a specific neonatal period (days 10-14) *prevents* the development of low affinity nicotine binding sites that normally appear between post-natal days 5 and 17. Interestingly, this effect of nicotine persists in adult mice. Behaviourally, the progeny of nicotine-exposed mice show no changes in spontaneous locomotion, however, they show altered responses to the activation of

nicotinic receptors, an effect that could be related to the absence of the low-affinity sites in the brain. This possibility is suggested by an interesting observation that animals lacking such sites also show altered responses to nicotine administration. Remarkably, similar neonatal exposure of mice to the PCBs also impairs the development of low-affinity nicotine sites. The progeny of these mice exhibit poor performance in memory-related tasks, likely reflecting impaired pre-synaptic modulation of the brain cholinergic neurons that play a key role in cognitive functions. Thus, both studies clearly demonstrate that early exposure of the nervous system to psychotropic drugs or xenobiotics can produce enduring changes in central neurotransmission processes and that this can lead to significant changes in motor and/or cognitive function long after the exposure.

Whether drug or xenobiotic-based changes in neurotransmission have a role in the etiology of disorders involving motor, affective and cognitive impairments is a difficult but challenging issue. In Chapter 9, Farber and colleagues address this issue by examining the role of neurotoxicological process in schizophrenia, especially those that impinge on L-glutamate, a major excitatory neurotransmitter which, through its ionotropic and metabotropic receptors, plays a critical role in neural development, synaptic function and plasticity, and neuronal survival. The authors put forward the proposal that hypoactivity of glutamate receptor activity in the mature or developing brain can lead to symptoms associated with schizophrenia (Olney and Farber, 1995) It is now widely recognized that overactivity of the NMDA type of glutamate receptors produces neuronal loss and that this phenomenon of excitotoxicity may contribute to the brain cell death in certain neurodegenerative diseases. However, the authors propose the novel idea that a *hypoactivity* of the NMDA receptors, specifically governing the output of corticolimbic projections, induces neurotoxic responses and this may contribute to both the symptoms and natural course of schizophrenia. This proposal is rooted in the observation that agents producing blockade of NMDA receptors, including the hallucinogen phencyclidine, elicit a schizophrenia-like behavioural state (Javit and Zukin, 1991). Additionally, in the adult brain NMDA receptor blockade produces vacuolation in the cingulate cortex while in the developing brain this blockade produces apoptotic cell death in corticolimbic structures implicated in schizophrenia (Olney *et al.*, 1989; Ikonomidou *et al.*, 1999). Thus, competitive and non-competitive blockers of NMDA receptors not only mimic schizophrenia symptoms but they also induce neuronal pathology. The neuronal damage produced is attributed to weakening of NMDA receptor-mediated drive to neurons that normally inhibit activity of the basal forebrain projecting to corticolimbic structures The loss of this inhibition and consequent unrestrained activity of these projections, that are glutamatergic or cholinergic in nature, injures neurons in the corticolimbic structures (Olney and Farber, 1995). An attractive aspect of the NMDA hypoactivity model is that it can accommodate the classical dopaminergic hyperactivity model of schizophrenia. Since dopamine inhibits the release glutamate, Farber et. al argue that dopaminergic overactivity could produce hypofunction of the NMDA receptors by reducing L-glutamate release at these receptors. While the NMDA hypoactivity model raises several unresolved questions, among them the regional selectivity of neuronal changes and nature of genetic or non-genetic factors that might produce NMDA receptor hypofunction, it provides a novel concept that embraces two important dimensions of schizophrenia: neurotransmitter dysfunction and loss of brain neurons. Indeed, neurotoxicants may produce degeneration by interfering with glutamate release or with activity of NMDA receptors governing corticolimbic activity.

While the brain in schizophrenia clearly does not exhibit the devastating structural changes associated with Alzheimer's disease or the highly specific neuronal damage seen in Parkinson's disease, there is emerging evidence that suggests the occurrence of neurodegeneration in this disorder (Gur *et al.*, 1991; Woods *et al.*, 1996). The clinical study by Molina *et al.* (Chapter 8) addresses this issue through neuroimaging and neurophysiogical assessements in schizophrenic patients. They suggest that degenerative phenomenon in some patients might be expressed through resistance of negative symptoms to drug treatment. Thus, their brain imaging study explores potential links between the existence of neurodegeneration, as revealed by changes in cerebrospinal fluid volume, and resistance of negative disease symptoms to the neuroleptic, clozapine. On the basis of evidence provided the authors suggest that changes in the sulcal cerebrospinal fluid volume may serve to predict success with clozapine therapy. The observation adds to the evidence favouring schizophrenia as a neurodegenrrative disorder, a concept that opens new avenues in the pharmacotherapy of the disorder: prevention of schizophrenia by inhibition of neurodegeneration.

The basis of neurodegeneration in schizophrenia and indeed several disorders characterised by frank neuronal pathology remains unknown. However, elevated oxidative stress due to the formation of reactive oxygen species (ROS) is beginning to emerge as a highly important factor in the neuron loss associated with these disorders. Chapter 6 by Rupniak *et al.* describes the therapeutic potential of a novel class of agents that mimic activity of enzymes such as superoxide dismutase and catalase that are an important part of the natural anti-oxidant defence system. The authors put forward the theory that inflammatory processes constitute a major source of ROS production the Alzheimer's disease. They demonstrate that exposure of astroglial cells to amyloid, a protein implicated in the pathophysiology of AD, leads to production of nitric oxide which, on combining with the reative oxygen produces peroxynitrate, a free radical agent that damages mitochondria and destroys cells. They show experimentally that their novel anti-oxidant compounds impair the production of the free radical species, an effect that harbors potential for neuroprotection. Indeed, the authors go on to demonstrate that some of these agents protect against the corticolimbic damage which is produced by injections of kainic acid and which is mediated by ROS (Bruce *et al.*, 1996; Rong *et al.*, 1999). The significance of oxidative stress in schizophrenia is unclear but if studies in the future were too reveal a link between oxidative stress and neurodegeneration, the novel agents developed by the authors may find therapeutic utility in reducing neurodegeneration or preventing its onset.

The inflammatory cascades may also have a role in demylinating disorders of the central nervous system (Archelos *et al.*, 1999). The study by Molina-Holgado (Chapter 3) focuses on the role of inflammatory cytokines in multiple sclerosis, a chronic inflammatory demylinating disease in which the neuronal pathology appears to be immune-based. Although the causitive factors in this disease are far from clear, the authors develop the idea that certain proinflammatory cytokines (interleukin 1 ., tumor necrosis factor- and. -interferon) have the potential to induce neuronal damage while other cytokines such as IL-10 have the ability to counteract the action of these agents (Moore *et al.*, 1993). In primary cultures of oligodendocyte precursors, exposure to IFN-diminshed the number of these cells, an effect that was enhanced by lipopolysacharide (LPS). The authors demonstrate that the damaging effects of cytokines involve nitric oxide production via activity of the inducible form of nitric oxide synthase (iNOS). Interestingly exposure of the oligodendrocyte precursors to IL-10 reduces the effect of IFN-, possibly by inhibiting the induction of iNOS. However, other interlukins that inhibit the induction of iNOS do not

counter the action of IFN-. Thus, it is not clear if NO is the factor in production of the damaging effects of cytokines on oligodendrocytes. Nevertheless, the study raises the possibility of harnessing the damaging effects of inflammatory cytokines by promoting levels or activity of anti-inflammatory cytokines. Indeed, IFN-, a cytokine that has therapeutic potential in multiple sclerosis, augments levels of IL-10 in the serum and cerebrospinal fluid and thus may express its beneficial effect by this mechanism.

The paper by Izquierdo *et al.* (Chapter 7) focuses on the important problem of memory failure which is associated with age, anxiety, depression or dementias. The authors review previous studies and provide data from their own experimental studies which shed light on some important biochemical mechanism underlying memory consolidation and retrieval. There is now wide recognition that glutamate receptor-driven activity of protein kinases has a central role in memory formation (see Izquierdo and Medina, 1997). While previous studies have emphasized the role of protein kinases C and A, recent studies (see Atkins *et al.*, 1998) point to the critical role of the mitogen activated protein kinase (MAPK), which influences memory processes through phosphorylation of specific presynaptic proteins, glutamate receptors and transcription factors. The activity of MAPK in hippocampal areas increases in following learning trials in the Morris water maze while inhibition of the enzyme activity impairs spatial memory. Reflecting on the question of whether pharmacological manipulation can ameliorate memory dysfunction, the authors emphasize the need to prevent the loss or dysfunction of neurons that contribute to memory impairment in the first place. However, they concede the need for treatments aimed at the retrieval processes. Given the critical role of protein kinases in memory processes it is likely that these enzymes will be targeted to develop novel cognition enhancers.

Chapter 10 in this series, by Archer *et al.*, describes the results of experiments which examine the influence of central noradrenergeic denervation on the locomotor effects of clonidine. In the first part of the study, the authors have investigated acute effects of clonidine following treatment with DSP4, a toxin that destroys noradrenergic neurons that originate in the locus coeruleus (Jonsson *et al.*, 1981). In the second part of the study, they have explored the interaction of clonidine with apomorphine. Acute clonidine inhibits locomotor activity but treatment with DSP 4 attenuates this action and reverses the effects of clonidine. The authors show that apomorphine augments locomotor activity and clonidine adminstration increases this effect. The effect of clonidine in this respect is further increased by the DSP treatment. The effects of clonidine are antagonized by yohimbine, suggesting involvement of $_2$ receptors in the actions of this agonist. These and other observations cited by the authors support the notion that dopamine receptor function is modulated by noradrenergic acitivty, and this may be important in influencing dopaminergic drug action in parkinsonism.

The above studies, revealing the breadth and subtleties of neuronal toxicity, can be expected to increase our understanding of multiple and complex factors that play a role in the development of neuropsychiatric diseases. A number of these studies reported here show that the action of neurotoxicants with respect to the neural elements affected can be both selective and regional in nature. Drugs of abuse such as amphetamine, cocaine and opioids, while presented to the entire brain following injection or inhalation, selectively usurp functions of neuron circuitry that constitute parts of the brain reward mechanism. In respect to the neurotoxicity of psychoactive drugs, the morphological or neurochemical targets of toxicity may differ from those mediating the behavioural effects. The early brain exposure to nicotine produces developmental changes in the low-affinity but not the high-affinity brain nicotinic sites that are an important target of this alkaloid. Similarly, prenatal

cocaine exposure selectively decouples the D_1 dopamine receptor and the presynaptic dopamine release process. Remarkably, the prenatal exposure to this agent does not appear to affect the development of brain dopamine transporter, the inhibiton of which is thought to mediate behavioural effects of this stimulant drug. In a similar vein, the neuro-toxicological syndrome produced by glutamate receptor hypofunction exhibits a regional selectivity, the NMDA receptor blockade producing apoptotic lesions in the forebrain areas involved in cognitive and motor functions, but not in caudal areas regulating visceral or endocrine functions. The specific behavioural deficits resulting from the neurotoxicant exposure very likely are a reflection of the selective morphological or neurochemical damage in a specific brain area However, the nature of factors that dictate the selectivity or subtlety of the toxicant action on neurons remains a puzzle that will continue to provide an impetus for additional work in this area. The solution of this puzzle, following the advent of newer investigative tools and approaches, should clarify the role of early or late neurotoxicant exposure in disorders such as schizophrenia that have neurodevelopmental and neurodegenerative dimensions.

The degenerative phenomenon underlying several neurological and neurospsychiatirc disorders has been linked to neurotoxicants that are exogenous: psychoactive drugs, dietary factors and environmental chemicals. However, structural, neurochemical or functional deficits may also be produced by unregulated activity of chemicals that are endogenous to the brain.. The cytokine family includes a plethora of chemicals whose abnormal activity can have significant adverse effects on neuronal structure and function. Similarly, tryptophan catabolism by the kynurenine pathway produces a number of metabolites, including quinolinic acid, which can damge or protect neurons. Nitric oxide, a gaseous messenger that is intermediary in the expression of physiological and toxic responses due to NMDA receptor activity, carbon monoxide, glutamate and aspartate can also be viewed as endogenous candidate neurotoxicants whose activity may contribute to the neurodegenerative phenomenon. The role of a number of these factors in the human degenerative disorders is currently under active investigation in many laboratories, however, it should be emphasized that to-date none of the endogenous toxicants can be considered to have a causative role in these disorders.

The cellular and molecular mechanisms that contribute to neurotoxicant- induced brain damage are equally diverse and no single mechanism can be viewed as the basis of neurodegeneration leading to the development of neuropsychiatric disease. However, in recent years oxidative neuronal damage, due to free radicals resulting from mitochondrial dysfunction, has assumed high level of significance in the pathophysiology associated with neurdegenerative and immune-based disorders. As the role of this damage in these disorders becomes clearer, the case for limiting or preventing such damage by pharmacological approaches will become compelling. The therapeutic options in this area would include agents that have the potential to prevent energy failure by augmentation of mitochondrial function and/or to sequester free radicals inflicting damage on brain neurons. However, a highly original approach in this respect involves development of antioxidants that mimic activity of the enzymes (superoxide dismutase and catalase) constituting a defence mechanism against the free radical activity. While prevention of neuron loss clearly constitutes an ideal approach for therapeutic interventions, the augmentation or revival of activity of compromised neurons whose abnormal activity gives rise to the sensorimotor and cognitive deficits also remains a desirable therapeutic goal. Thus, development of agents that selectively influence activity of specific neurotransmitter systems and protein kinase cascades, such as the MAPK, that are involved in memory and learning, could make

it possible to reverse cognitive dysfunction in neurodegenerative disorders. The range of ideas and approaches that are represented here and that explore links between neurotoxicology and neurodegeneration will hopefully open new vistas in therapeutics.

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