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Chapter I

Excitotoxicity – Introduction

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The papers in the following chapter are all focused on a general theme of excitotoxicity. The work presented in each of the contributions, however, is representative of different trends in this field of research, and thus provides a broad-based, albeit necessarily incomplete, perspective on the field. The papers all describe basic research on the cellular basis of excitotoxicity, but the application of the results ranges from very fundamental explorations of new phenomena to studies with real potential for the development of new therapies in the short-term. Thus, our hope is that both basic neuroscientists and clinicians will find something of interest in this chapter.

The paper presented by Dr. A. M. Marini describes interesting data on the ability of NMDA and TrkB receptors to control the activation of Akt, a downstream activator of the PI-3 kinase pathway. Dr. Marini and her group have shown previously that appropiate concentrations of NMDA protect vulnerable neurons against glutamate excitotoxicity via a BDNF autocrine loop. Thus, NMDA increases BDNF levels in cerebellar granule cell culture medium within 2 min whereas TrkB receptor activation occurred within 10 min. The new data described herein, indicate that maximal Akt activation requires both TrkB and NMDA receptor stimulation. Based on studies of BDNF-mediated Akt activation, it appears that Akt is first activated via the TrkB receptor. Since TrkB receptor activation is sustained by the activity-dependent release of BDNF via NMDA, Akt activation can be maintained and sustained for hours. These exciting new findings of a cooperative effect of two receptors leading to a prolonged intracellular change, goes beyond the theme of excitotoxicity and extends to the field of neuronal development and regeneration.

The paper presented by Dr. R. Trullas deals with new evidence indicating that necrotic, but not apoptotic neuronal cell death, is preceded by choline release. The release of choline results from inhibition of phosphatidylcholine synthesis evoked by glutamate receptor overactivation. The evidence obtained by the Neurobiology Unit in Barcelona suggests that this glutamate receptor-mediated inhibition of phosphatidylcholine synthesis is a key early event, downstream of calcium overload, of the excitotoxic process. The results presented could be particularly important for the establishment of new and specific treatments for patients experiencing excitotoxic cell loss following either a traumatic event such as a stroke or a prolonged neurodegenerative process involving excitatory neurotransmission.

The possible link between excitotoxicity and the induction of cell cycle-related factors has been a matter of extensive investigation during the last 5–10 years and it is now supported by both in vivo and in vitro data. The paper presented by Dr. Memo and his group reports further evidence suggesting that a repertoire of events typical of proliferating cells is activated in degenerating neurons. In particular, Dr. Memo's group describes how exposure of primary cultures of cortical neurons to neurotoxic concentrations of NMDA induces a significant, long-lasting increase in p53 levels. The transcriptional activity of the over-expressed p53 is demonstrated by the concomitant enhancement of Gadd45, which is one of the p53 target genes. Although the mechanism(s) by which ionotropic glutamate receptor stimulation induces activation of

cell cycle related factors and apoptosis remains unclear, the authors hypothesize that over-stimulation of NMDA glutamate receptor types may induce DNA damage via the generation of oxygen free radicals (ROS). Activation of p53 and p53 target genes is thus a part of the cellular response to a genotoxic input and may participate in sensing DNA damage and initiating DNA repair processes. This work is clearly of potential relevance to future therapeutic approches to limit excitotoxicity.

The paper by Dr. Fernández-Sánchez and collaborators describes some novel pharmacological effects of a commercially available analgesic compound, nefopam. Despite being a cyclic analogue of orphenadrine, a known NMDA receptor antagonist, nefopam fails to protect against excitotoxicity resulting from direct exposure of cultured cerebellar neurons to glutamate, and does not displace MK-801 binding from rat hippocampal membranes. Rather, nefopam protects against both NMDA receptor-dependent as well as NMDA receptor-independent veratridine-induced toxicity. The authors go on to provide data indicating that these effects are not due to nefopam acting as a GABAa receptor agonist, and their work suggests that nefopam may represent a new class of agents that could be useful in the treatment of diseases involving an activity-dependent excessive release of glutamate.

The paper presented by Dr. Jakobsen and collaborators presented an elegant experimental system for studying excitotoxicity. The paper describes the results of a study in which they used 2–3 weeks old hippocampal slice cultures derived from 7 days old rats, to study the pharmacology of excitotoxicity produced by domoic acid, a naturally-occurring excitatory amino acid that is known to produce hippocampal damage in both humans and experimental animals. Based on densitometric measurements of the cellular uptake of propidium iodide in the different regions of the cultured hippocampal slices, they report that CA1 was the most susceptible hippocampal subregion (ie. displaying the lowest EC_{50} values), but that dentate granule cells and CA3 pyramidal cells were also susceptible. By using a variety of glutamate receptor specific antagonists they conclude that domoic acid neurotoxicity, at doses close to EC_{50} values, is primarily mediated by AMPA receptors in the hippocampal subregion CA3, whereas NMDA receptors are also involved in the degeneration of dentate granule cells and CA1 pyramidal cells.

Finally, the paper presented by Dr. Tasker describes an international collaboration in which laboratories in Spain, New Zealand and Canada combined their expertise to systematically evaluate the excitotoxic properties of three new commercial sources of kainic acid. Kainic acid is a potent excitotoxin that is widely used in basic science studies of non-NMDA receptor function and in clinically-relevant models of temporal lobe epilepsy. However, for the last several years the traditional source of this compound has been unavailable. Recently two new biological and one synthetic form of kainic acid have been introduced to the marketplace. The paper describes the use of cerebellar granule neurons in culture, hippocampal slice electrophysiology, and whole animal behavioural toxicity testing to compare these products. The authors conclude that all three sources represent a viable alternative to traditional kainic acid, but that the three are probably not identical; and they caution that results obtained with each should be evaluated accordingly.

As organizers of the symposium on Excitotoxicity, we are pleased to present these exciting, current and highly relevant papers, and to thank the many participants who made the symposium a success.