Preface

Assembling *Glutamate and Addiction* was a two-and-a-half year labor of love. As editors, we all had the same goal in mind and pursued this with a fierce dedication. We felt that it was now time for a volume clarifying for the first time the relationship between glutamatergic systems and addiction. The past decade has seen a steady and escalating progression of scientific advances that have implicated a pivotal role of glutamatergic systems in cocaine, opiate, and alcohol dependence—both the etiology of these disorders and their treatment. As editors, we met as a group several times a year to discuss the progress and the ever emerging direction of the book. As senior editor, I am personally indebted to the superb job of the coeditors attracting the very best scientists in this field to contribute their important papers to this book.

To Philip H. Sheridan, MD of the Food and Drug Administration (FDA), for his marvelous ability to attract internationally known scientists to contribute to the first section of the book on the basic physiology and pharmacology of glutamate. The five stellar chapters in this section include ones by Borges and Dingledine; Witkin, Kaminski and Rogawski; Choi and Snider; Sanchez and Jensen; and Kaul and Lipton. A special thank you to Michael A. Rogawski, MD, PhD, Epilepsy Research Section, NINDS, NIH for being an early and avid supporter of this effort and bringing to our attention valuable contributors to this book. It is our hope that these five introductory chapters will provide a level playing field for all readers of this book to upgrade their basic understanding of glutamate before proceeding to the other research chapters focused on the relationship between glutamate and various addictive disorders.

To Jerry Frankenheim, PhD of the National Institute on Drug Abuse, the National Institutes of Health (NIH) for his wonderful role, as senior editor of Section II, in illustrating the role of glutamatergic systems in stimulant drugs of abuse including cocaine, amphetamine, and methamphetamine. Dr. Frankenheim displayed considerable care in editing this section. In addition, I am personally indebted to Dr. Frankenheim for his seamless job in serving as Acting Senior Editor of this volume for a two-month period when I was unavailable for this task. Section II is a truly remarkable part of the book in its thoroughness in covering virtually every aspect of the role of glutamate in stimulant drugs of abuse, with outstanding chapters by Pert, Post, and Weiss; Karler, Thai, and Calder; Wolf; Baker, Cornish, and Kalivas; Wang, Mao, and Lau; Pulvirenti; Vezina and Suto; Cadet; Burrows and Yamamoto; Itzhak, Martin, and Ali; Matsumoto and Pouw; Bisaga and Fischman; and Epping-Jordan. As we state in our dedication of this book, this effort also coincided with the tragic death of one of our beloved colleagues in the addiction field, Marian Fischman, PhD of Columbia University School of Medicine. Dr. Fischman was a vibrant human being, and one of the most vital forces in the research field of addiction medicine. A personal thank you to Adam Bisaga, MD who took over the task of writing and editing this chapter with Dr. Fischman in an extremely gracious and responsible fashion in the face of tragic circumstances.

We are extremely grateful to the authors who contributed to the valued third section of the book on glutamate and opiate drugs of abuse including heroin. The world-renown scientists in this section included Mao; Trujillo; Popik; and Rasmussen. An overview of this important topic is provided by Jianren Mao, MD, PhD of Harvard University School

of Medicine. It is of interest to note that the researchers in this section were some of the first to provide evidence of a relationship between glutamate and various aspects of the addiction process.

In the final section, the relationship between glutamate and alcohol abuse and alcoholism is explored. Our superb editors of Section IV are Forrest F. Weight, MD and Raye Litten, PhD, both of the National Institute of Alcohol Abuse and Alcoholism (NIAAA). Personally, I am particularly grateful for the continuous role provided by Dr. Litten, who managed to come to virtually every editorial meeting across building lines and to quickly get his section collated into a deliverable form to our publisher, Humana Press.

I would like to thank Craig Adams and Elyse O'Grady of Humana Press for their superb editorial and publishing skills and their tireless efforts in cheering this effort on to its completion. Craig and Elyse supported this effort from the beginning and until its completion, with a compassion and expertise that I will forever admire.

Finally, I would like to thank my institute, the National Institute on Drug Abuse, NIH, for being supremely generous in allowing me the time to pursue this effort for the last two and a half years. Particular thanks goes to Alan Leshner, Ph.D., former Director, NIDA, Glen R. Hanson, PhD, DDS, current and Acting Director, NIAA, Frank Vocci, PhD, Director, Division of Treatment Research and Development (DTR&D), NIDA and Ahmed Elkashef, MD, Chief, Clinical Medical Branch (CMB), DTR&D, NIDA for permitting this effort to occur. We also thank the institute directors of NIAAA, Enoch Gordis, MD (former director) and the present top official of the FDA Bernard A. Schwertz, DVM, PhD, Acting Principal Deputy Commissioner and the past commissioner of the FDA, Jane E. Henney, MD for enabling the participation of individuals from their respective institutions.

I am personally touched by the numerous cards, letters and flowers that I received from family, friends, professional colleagues, and folks from Humana while in the hospital.

Our interest in glutamatergic systems and drug abuse disorders stems back to at least 1991, when the first preclinical evidence was presented for a role of this system in the development of opiate tolerance and withdrawal (cf. 1, 2). Indeed, a few years earlier, research in the late 1980s suggested a role of glutamate in stimulant drug addiction (3). From there, we as a group launched several efforts to try to synthesize the knowledge base that was quickly accumulating in this exciting area. Thanks to the efforts of the National Institutes of Health (NIH) and the Food and Drug Administration (FDA), approaches to understanding the biological and behavioral basis of drug addiction and developing new modalities for the treatment of drug addiction are now attaining some level of consistency across the world. A highlight in this trend for unification in theory and practice, is illustrated by the conceptual writings of Alan I. Leshner, PhD former Director, National Institute on Drug Abuse, who has tirelessly pioneered to increase the research and scientific basis for understanding drug addiction as a disorder of the brain (e.g., 4, 5). A similar emphasis on drug abuse as a brain disorder is noted in the very basic preclinical research of Stephen E. Hyman, MD, former Director, National Institute on Mental Health (e.g., 6, 7). Similarly, in a monthly letter developed by the National Institute on Alcohol Abuse and Treatment (NIAAA), Enoch Gordis, MD, former Director, NIAAA has describe numerous scientific advances detailing the role of various biochemical systems in alcohol dependence and the role of medication treatment in alcohol dependence (cf., 8, 9). An esteemed partner in this endeavor is Jane Henney, MD, former Commissioner, FDA whose institute is responsible for making certain that the medications that are developed for this indication are both efficacious and safe. We very

much value the superb contributions of the authors in Section IV on glutamate and alcohol, who include Peoples; Crew, Rudolph, and Chandler; Becker and Redmond; Krystal, Petrakis, D'Souza, Mason, and Trevisan; Zieglgänsberger, Rammes, Spanagel, Danysz, and Parsons; Pasternak and Kolesnikov; and Potgieter. We all work together with these institutes and with the creative and brilliant scientists who undertake both the preclinical and clinical research to develop a rigorous science of drug addiction. It is our hope that this research will result in innovative treatments for drug abuse and addiction, and for understanding the basis of these disorders in the central nervous system.

The job of characterizing the role of glutamatergic systems in addiction disorders is now off to a solid beginning. With the recent advance and approval of glutamatergic antagonists for the indication of alcohol abuse and addiction in a variety of European countries, we have already started to witness some clinical payoff for the superbly innovative and thorough research of both preclinical and clinical sciences. We hope that this effort will launch a new decade starting in the year 2001, that will see yet even further advances in the glutamatergic field**,** both in the etiology and treatment of addiction disorders.

Barbara H. Herman, ***PhD***

References

1. Herman, B.H., Vocci, F., Bridge, P. The effects of NMDA receptor antagonists and nitric oxide synthase inhibitors on opioid tolerance and withdrawal. *Neuropsychopharmacology* **13**: 269–292, 1995.

2. Herman, B.H. and O'Brien, C.P. Clinical medications development for opiate addiction: focus on nonopioids and opioid antagonists for the amelioration of opiate withdrawal symptoms and relapse prevention. *Seminars in Neuroscience* **9**: 158–172, 1997.

3. Karler, R., et al., Blockade of "reverse tolerance" to cocaine and amphetamine by MK-801. *Life Sci* **45**: 599–606, 1989.

4. Leshner A.I., Koob G.F. Drugs of abuse and the brain. *Proc Assoc Am Physicians* **111**:99–108, 1999.

5. Leshner A.I. Addiction is a brain disease, and it matters. *Science* **278**: 45–47, 1997.

6. Hyman S.E., Hyman S.E., Malenka R.C. Addiction and the brain: the neurobiology of compulsion and its persistence. *Nat Rev Neurosci*.**2**: 695–703, 2001.

7. Berke J.D., Hyman S.E. Addiction, dopamine, and the molecular mechanisms of memory. *Neuron* **25**: 515–532, 2000.

8. Gordis E. Advances in research on alcoholism and what they promise for future treatment and prevention. *Med Health R I* **82**:121, 1999.

9. Gordis E. The neurobiology of alcohol abuse and alcoholism: building knowledge, creating hope. *Drug Alcohol Depend.* **51**:9–11, 1998.

4

Maturational Regulation of Glutamate Receptors and Their Role in Neuroplasticity

Russell M. Sanchez, PhD and Frances E. Jensen, MD

1. INTRODUCTION

Glutamate receptors mediate most excitatory synaptic transmission in the brain. Additionally, they mediate many forms of synaptic plasticity such as those thought to comprise the physiological basis of learning and memory. In the developing brain, glutamate receptor activation is required for appropriate synaptogenesis and activity-driven refinement of functional synaptic networks *(1,2)*. Thus, in early brain development, glutamate receptors additionally mediate highly age-specific forms of neuroplasticity that may not continue into maturity. Notably, activity-driven and maturational changes in the physiological roles glutamate receptors are paralleled by dynamic regulation of their molecular composition and functional properties. In this chapter, we review the glutamate receptor subtypes and discuss the possible relationships between their dynamic regulation during development and their ability to mediate various forms of synaptic plasticity.

2. GLUTAMATE RECEPTOR SUBTYPES

Glutamate is an ubiquitous excitatory neurotransmitter in the brain, and there are several subtypes of glutamate receptor (for reviews, see refs *3*–*5*). Glutamate receptors are broadly divided into the ionotropic glutamate receptors, which from glutamate-gated transmembrane ion channels, and the metabotropic glutamate receptors, which, when activated by glutamate, trigger intracellular signaling pathways via receptor-coupled second messengers. The ionotropic glutamate receptors are comprised of three subtypes whose names derive from selective agonist that bind each with highest affinity: the α-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid (AMPA), *N*-methyl-D-aspartate (NMDA), and kainate (KA) receptors *(3)*. The properties of each and their roles in neuroplasticity will be discussed separately.

2.1. NMDA Receptors

NMDA receptors are well known to be critically involved in many forms of activity- driven synaptic plasticity, and these have been extensively reviewed (see, for example, ref. *6*). Three general features of NMDA receptors give them a unique role in the activity-dependent regulation of synaptic function. First, NMDA receptors form ion channels that are highly permeable to Ca^{2+} (in addition to Na^{+} and K^{+}), and the influx of Ca^{2+} through NMDA receptors may trigger Ca^{2+}-dependent signaling pathways that regulate synaptic function and synaptogenesis *(7,8)* Second, NMDA receptors are highly voltage-dependent because their channels are blocked by Mg^{2+} at membrane potentials at or more negative to the resting potential, and Mg^{2+} is extruded from the channels only

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upon depolarization *(9)*. Thus, NMDA receptors require concurrent membrane depolarization (through the activation of non-NMDA ionotropic glutamate receptors) and glutamate binding to conduct appreciable current. This voltage dependence gives NMDA receptors the capacity to activate mechanisms of neuroplasticity in response only to specific patterns of synaptic input. The third key feature of NMDA receptors is that their channel kinetics are much slower than those of non-NMDA ionotropic glutamate receptors, and, therefore, their activation can result in relatively long-lasting membrane depolarization. This prolonged depolarization can further relieve the Mg^{2+} block of NMDA receptor channels, recurrently increasing membrane depolarization and activating high voltage- activated Ca^{2+} channels to additionally increase intracellular Ca^{2+}. In addition to their contributions to physiological forms of neuroplasticity, the Ca^{2+} permeability and kinetic properties of NMDA receptors give them a critical role in pathophysiological processes such as ictal seizure discharges and hypoxic/ischemic neuronal injury *(10–12)*.

The precise properties of native NMDA receptors are determined in large part by the particular combination of independently genetically encoded molecular subunits that comprise each receptor *(3)*. NMDA receptors are heteromerically assembled from subunits dubbed NRI, and NR2A, B, C, and D (for reviews, see refs *(13and 14)*. Evidence from recombinant expression studies indicates that only receptors composed of both NRI and NR2 subunits exhibit the functional properties of native NMDA receptors and, further, that certain properties (such as Mg^{2+} sensitivity or channel kinetics) differ subtly depending on the particular NR2 subunits expressed *(15,16)*. Notably, NRI subunits are expressed throughout the brain, whereas each of the NR2 subunits displays regionally and developmentally specific expression patterns *(17)*. Thus, regional and maturational differences in NMDA receptor properties appear to derive in large part from differences in the particular NR2 subunits expressed.

Differences in the key properties of NMDA receptors that result from different subunit combinations can have profound consequences for neuroplasticity and disease. The properties of NMDA receptors generally are such that their activity is enhanced during early postnatal development, and this is a period in which neuroplasticity is more robust and the brain is highly susceptible to epileptogenesis and excitotoxicity. For example, NMDA receptor-mediated synaptic currents appear generally to be more slowly decaying in the early postnatal brain compared to the adult *(18,19)*. In the forebrain, the ratio of NR2B to NR2A expression is much greater during early brain development compared to adulthood. Recombinant NMDA receptors that contain predominantly NR2B tend to exhibit slower decay times than those containing NR2A *(17,20,21)*, and native NMDA receptors in neurons that express NR2A exhibit more rapid decay kinetics compared to those in neurons that do not express detectable NR2A transcripts *(22)*. Therefore, activation of NMDA receptors in immature forebrain neurons would be expected to induce a much longer-lasting depolarization and possibly increase the capacity for glutamate-mediated neuroplasticity compared to their adult counterparts. Consistent with this notion, the capacity for NMDA receptor-mediated synaptic plasticity has been observed to be enhanced in the immature brain and decreases with maturation *(19,23)*. Additionally, transgenic mice overexpressing NR2B were observed to maintain enhanced NMDA receptor- dependent synaptic plasticity (and learning) into adulthood compared to wild-type mice *(24)*. These data indicate that NMDA receptor-mediated mechanisms of synaptic plasticity can be strongly influenced by relatively subtle differences in the properties of the NMDA receptor channels.

Notably, the NR2D subunit also is expressed at higher levels in subcortical structures in the immature brain compared to the adult brain *(17,25)*. Dimeric receptors composed of NRI and NR2D exhibit decreased channel block by Mg^{2+} and slower decay kinetics compared to receptors composed of other subunit combinations *(25,26)*. Thus, the transient developmental upregulation of NR2D could enhance NMDA receptor-mediated plasticity in the immature brain by decreasing the Mg^{2+}-block of NMDA receptor channels at resting membrane potentials and allowing significant membrane depolarization and Ca^{2+} influx in response to any pattern of afferent activation.

More generally, these data taken together indicate that NMDA receptor composition and function is geared toward increased synaptic plasticity in the developing brain, where synaptogenesis and activity-dependent refinement of synaptic networks is ongoing and a high level of plasticity is necessary. This scenario also renders the immature brain more susceptible to NMDA receptor-mediated injury compared to the adult brain *(27)*.

2.2. AMPA Receptors

In contrast to NMDA receptors, AMPA receptors mediate fast excitatory neuronal signaling, as they exhibit rapid activation and desensitization, operate linearly near the resting membrane potential, and mostly form ion channels that are virtually impermeable to Ca^{2+} for review, see ref. *(3)*. For these reasons, AMPA receptors historically were viewed as simply transmitting bits of information between neurons, and their role in neuroplasticity was thought to be only in the ability for Ca^{2+}-activated mechanisms to adjust the gain of the signals that AMPA receptors transmit. This role for AMPA receptors certainly has been firmly established, as studies over the last several years have revealed numerous posttranslational mechanisms by which AMPA receptor function is regulated by synaptic activity and Ca^{2+}-dependent pathways *(28)*.

It is now clear, however, that a subset of AMPA receptors in the brain and spinal cord exhibit relatively high permeability to Ca^{2+} and can directly activate Ca^{2+}-dependent mechanisms of neuroplasticity similarly to NMDA receptors. AMPA receptors are thought to be pentamers assembled from any combination of the molecular subunits GluR1, 2,3, and 4 (alternatively, GluRA–D) *(3)*. Notably, each AMPA receptor gene encodes a subunit that will form homomeric channels that are permeable to Ca^{2+} and other divalent cations. However, only the GluR2 mRNA undergoes posttranscriptional editing that results in the replacement of a neutral glutamine (Q) by a charged arginine (R) at a key site within the putative pore-forming region of the AMPA receptor channel to express a Ca^{2+}-impermeable channel *(29)*. Recombinant AMPA receptors that lack a GluR2 subunit exhibit significantly greater permeability to Ca^{2+} and other divalent cations compared to those that contain GluR2 *(30–33)*.

In embryonic rat brain, the proportion of Q/R edited to unedited GluR2 increases with age, with virtually 100% of GluR2 being edited in the postnatal brain *(34)* Thus, among native AMPA receptors, only those that lack a GluR2 subunit will exhibit appreciable divalent permeability. In the adult rat brain, the vast majority of AMPA receptors expressed in the forebrain contain GluR2. However, a number of molecular and electrophysiological studies in the last several years indicated that a subset of neurons in the postnatal nervous system express AMPA receptors that may lack GluR2 and exhibit relatively high permeability to Ca^{2+} and that these may have a role similar to that of NMDA receptors in activating Ca^{2+} dependent pathways of neuroplasticity. In spinal cord, for example, strong activation of Ca^{2+}-permeable AMPA receptors (with NMDA receptors pharmacologically blocked) was observed to potentiate AMPA receptor-mediated excitatory postsynaptic currents (EPSCs) in a subpopulation of dorsal root ganglion neurons *(35)*. In the hippocampus, Ca^{2+} influx through AMPA receptors was shown to be necessary for the induction of long-term depression (LTD) observed in type II interneurons in area CA3 *(36)*.

Notably,the ratio of expression of GluR2 subunits to that of other AMPA receptor subunits is significantly lower in the immature hippocampus compared to the adult *(37,38)*. and a larger number of principal neurons express divalent-permeable AMPA receptors in the neonatal hippocampus compared to the adult *(38)*. Thus, similar to NMDA receptors, maturational regulation of this key feature of AMPA receptors may confer upon them a specialized role in Ca^{2+}-dependent neuroplasticity during early brain development. The expression of these receptors in principal forebrain neurons selectively during early postnatal development suggests their possible role in normal development and age-dependent plasticity, as well as in the enhanced susceptibility of the immature brain to AMPA receptor-mediated epileptogenesis and excitotoxic injury *(38–40)*.

2.3. Kainate Receptors

Historically, kainate receptors were viewed as similar to AMPA receptors, largely because of their overlapping pharmacological sensitivities, fast kinetics, and lack of voltage dependence, but they are molecularly and functionally distinct *(41)*. kainate receptors are heteromerically assembled from the molecular subunits GluR5, 6, and 7, and KAI and KA2. The GluR5 and GluR6 subunits also undergo posttranscriptional editing at the codon for the Q/R site that results in significantly decreased Ca^{2+} permeability *(29)*. Similarly to AMPA receptors, the proportion of Q/R edited subunits increases with maturation *(42)*, and in spinal cord neurons, this has been shown to be correlated with a developmental decrease in Ca^{2+} permeability *(43)*, These observations suggest specialized roles of divalent-permeable kainate receptors in neuroplastic processes during early brain development.

The lack of adequately specific agonists and antagonists historically made it difficult to distinguish kainate receptor-mediated responses from those of AMPA receptors in native brain preparations to examine their potential roles in neuroplasticity. Recently, using an antagonist that specifically blocks GluR5-containing kainate receptors, Collingridge and colleagues were able to determine that a form of hippocampal long-term potentiation (LTP) known to be independent of NMDA receptor activation can be induced by the activation of postsynaptic kainate receptors in CA3 pyramidal neurons *(44)*. Kainate receptor-mediated postsynaptic currents in these neurons in hippocampal slices are extremely small compared to AMPA receptor-mediated or NMDA receptor-mediated EPSCs and require temporal summation following trains of repeated stimulation (with AMPA and NMDA receptors pharmacologically blocked) to be clearly distinguished from noise by conventional voltage-clamp recording methods *(45)*. Thus, at first glance, it would appear unlikely that such small events could result in a rise in intracellular Ca^{2+} that is sufficient to activate signaling pathways that fail to be activated following the much larger events that result from NMDA or AMPA receptor activation in the same cells. However, it is conceivable that kainate receptors could be specifically coupled to second messengers that do not interact with other glutamate receptors or are sequestered from Ca^{2+} entering through AMPA or NMDA receptors. Notably, the antagonist used by Collingridge's group was shown among recombinant homomeric kainate receptors to be highly selective for GluR5-containing receptors, yet *in situ* hybridization studies suggest that CA3 pyramidal neurons do not express mRNA for this subunit in abundance *(46,47)*. It certainly is possible that native kainate receptors respond differently than the homomeric receptors used to determine drug selectivity *(44)*, but, clearly, the precise role for kainate receptors in this form of neuroplasticity remains somewhat controversial.

In addition, similar to NMDA and AMPA receptors, kainate receptors appear to undergo developmental regulation in their expression and function in such manner as to promote neuronal excitability in early brain development. Kidd and Isaac have shown that low-affinity kainate receptors are activated at thalamocortical synapses in the early postnatal period and that their contribution to postsynaptic responses decreases with maturation. Notably, the immature brain is much more sensitive to the epileptogenic effects of kainate compared to the adult *(48,49)*. However, as kainate also is a potent AMPA receptor agonist, it is not yet clear if the maturational state of kainate receptor function is a critical mediator of the developmental changes in kainate sensitivity.

3. METABOTROPIC GLUTAMATE RECEPTORS

Whereas the ionotropic glutamate receptors are capable of secondarily directly or indirectly activating mechanisms of synaptic plasticity, the metabotropic receptors are directly coupled to second-messenger pathways that mediate forms of short-term plasticity. There are at least eight cloned metabotropic glutamate receptors (termed mGluR1–mGluR8) *(4)*. These have been classified into three groups based on sequence homology, coupling to second-messenger systems, and pharmacological sensitivities. Group I receptors are coupled to phosphoinositide (PI) hydrolysis that leads to Ca^{2+} mobilization from intracellular stores, whereas groups II and III receptors are negatively coupled to adenylyl cyclase (AC) activity.

Although the consequences of metabotropic glutamate receptor activation vary depending on receptor type, neuronal type, or brain region, some general principles regarding the outcomes of their activation have emerged *(5).* Postsynaptic group I metabotropic receptor activation, in general, causes and increase in the intrinsic excitability of principal neurons (particularly in hippocampal CA1 and CA3 subfields), mainly via down-modulation of voltage-gated potassium channels *(50),* groups II and III receptor activation tends to depress excitatory synaptic transmission by inhibiting glutamate release *(51).*

Evidence suggests that group I postsynaptic metabotropic glutamate receptors are mostly involved in the regulation of synaptic plasticity (for reviews, see refs. *52* and *53*). For example, mice lacking mGluR5 show reduced hippocampal CA1 LTP (although CA3 LTP was normal) *(54).* However, presynaptic metabotropic glutamate receptors also may play roles in synaptic plasticity, as Laezza et al. showed that LTD in CA3 interneurons only resulted from a synergistic effect that required both the activation of presynaptic metabotropic receptors and Ca^{2+} entry through postsynaptic AMPA receptors *(36).* Thus, either presynaptic or postsynaptic metabotropic glutamate receptors may contribute to glutamate-mediated synaptic plasticity under different conditions.

Interestingly, in the developing brain, as with the ionotropic glutamate receptors, metabotropic glutamate receptor function undergoes maturational regulation in such a manner as to promote neuronal excitability in early postnatal development. Agonist-stimulated PI turnover has been shown to be relatively robust in slices of immature rat brain, increasing from age P1 to P7–P10 before gradually decreasing to adult levels at around P24 *(55).* This contrasts with the activity of metabotropic receptors negatively coupled to AC, as cyclic AMP accumulation induced by the AC activator forskolin was shown to be inhibited by the nonspecific metabotropic glutamate receptor agonist 1*S*,3*R*(ACPD) in adult but not in neonatal (P1–P15) rat hippocampus *(56,57).* Notably, metabotropic glutamate receptors negatively coupled to AC are expressed in early postnatal development, but nonspecific metabotropic receptor activation in the neonatal hippocampus increases basal cyclic AMP levels *(58)* and, thus, would be expected to promote neuronal excitability in early postnatal development. Consistent with notion, the nonspecific mGluR agonist 1*S*,3*R*-1-aminocyclopentane-1,3,dicarboxylic acid [(1*S*,3*R*)ACPD], which activates both AC- and PI-coupled metabotropic receptors, was shown to elicit dose- dependent limbic seizures in neonatal (postnatal d 7) rats *(59).* This proconvulsant effect was similar to that observed for the specific group I mGluR agonist *(R,S)*-3,5-dihydroxyphenylglycine (3,5-DHPG) *(60),* suggesting that it was mediated by AC-coupled metabotropic receptors. Furthermore, specific group II agonists appear to be anticonvulsant,as intraventricular infusion of the group II *agonist* (2*S*,1′*R*,2′*R*,3′*R*)-2-(2,3-dicarboxycyclopropyl)glycine (DCG-IV) decreased the incidence of continuous limbic motor seizures induced by intraventricular kainate *(61),* and microinjection of *(S)*-4-carboxy-3-hydroxyphenylglycine]*(S)*-4C3HPG], a group I antagonist and group II agonist, into the inferior colliculus inhibited audiogenic seizures in genetically epilepsy-prone rats *(62).* However, the proconvulsant actions of nonspecific metabotropic glutamate receptor agonists may not be entirely age-selective, as microinjections of (1*S*,3*R*)ACPD in adult rat hippocampus *(63)* also elicited limbic seizures. Compared to the ionotropic glutamate receptors, the developmental pattern of metabotropic glutamate receptor function is not as clearly linked to the developmental patterns of mGluR gene expression and glutamate receptor-mediated pathogenesis.

4. SUMMARY

It has been long established that much neuroplasticity in the brain is mediated by the actions of glutamate at glutamate receptors. Additionally,a vast literature exists on the role of glutamate receptors in neurological disease states, and the selective vulnerability of the immature brain. Until recently, experimental evidence suggested that most glutamate-mediated plasticity and pathology relied on the activation of the NMDA subtype of glutamate receptor, mainly due to its high permeability to Ca^{2+}. However, it is now clear that all of the glutamate receptors have the capacity to mediate neuroplastic and neuropathological processes and that these processes may be enhanced in the developing brain.

REFERENCES

1. Fox, K., Schlaggar, B. L., Glazewski, S., and O'Leary, D. D. M. (1996) Glutamate receptor blockade at cortical synapses disrupts development of thalamocortical and columnar organization in somatosensory cortex. *Proc. Natl. Acad. Sci. USA* **93**, 5584–5589.
2. Constantine-Paton, M. (1994) Effects of NMDA receptor antagonists on the developing brain. *Psychopharmacol. Bull.* **30**, 561–565.
3. Dingledine, R., Borges, K., Bowie, D., and Traynelis, S. F. (1999) The glutamate receptor ion channels. *Pharmacol. Rev.* **51(1)**, 7–61.
4. Conn, P. J. and Pin, J.-P. (1997) Pharmacology and functions of metabotropic glutamate receptors. *Annu. Rev. Pharmacol. Toxicol.* **37**, 205–237.
5. Wong, R. K., Bianchi, R., Taylor, G. W., and Merlin, L. R. (1999). Role of metabotropic glutamate receptors in epilepsy. *Adv. Neurol.* **79,** 685–698.
6. Malenka, R. C. and Nicoll, R. A. (1993) NMDA-receptor-dependent synaptic plasticity: multiple forms and mechanisms. *Trends Neurosci.* **16(12),** 521–527.
7. Aamodt, S. M. and Constantine-Paton, M. (1999) The role of neural activity in synaptic development and its implications for adult brain function. *Adv. Neurol.* **79,** 133–144.
8. Vallano, M. L. (1998) Developmental aspects of NMDA receptor function. *Crit. Rev. Neurobiol.* **12(3),** 177–204.
9. Ascher, P. and Nowak, L. (1988) The role of divalent cations in the *N*-methyl-D-aspartate responses of mouse central neurons in culture. *J. Physiol.* **399,** 247–266.
10. Chapman, A. G. (1998) Glutamate receptors in epilepsy. *Prog. Brain Res.* **116,** 371–383.
11. Michaelis, E. K. (1998) Molecular biology of glutamate receptors in the central nervous system and their role in excitotoxicity, oxidative stress, and aging. *Prog. Neurobiol.* **54(4),** 369–415.
12. Meldrum, B. S. (1994) The role of glutamate in epilepsy and other CNS disorders. *Neurology* **44(Suppl. 8),** S14–S23.
13. Hollman, M. and Heinemann, S. (1994) Cloned glutamate receptors. *Annu. Rev. Neurosci.* **17,** 31–108.
14. Nakanishi, S. (1992) Molecular diversity of glutamate receptors and implications for brain function. *Science* **258,** 597–603.
15. Monyer, H., Sprengel, R., Schoepfer, R., Herb, A., Higuchi, M., Lomeli, H., et al. (1992) Heteromeric NMDA receptors: molecular and functional distinction of subtypes. *Science* **256,** 1217–1221.
16. Ishii, T., Moriyoshi, K., Sugihara, H., Sakurada, K., Kadotani, H., Yokoi, M., et al. (1992) Molecular characterization of the family of the *N*-methyl-D-aspartate receptor subunits. *J. Biol. Chem.* **268,** 2836–2843.
17. Monyer, H., Burnashev, N., Laurie, D. J., Sakmann, B., and Seeburg, P. H. (1994) Developmental and regional expression in the rat brain and functional properties of four NMDA receptors. *Neuron* **12,** 529–540.
18. Hestrin, S. (1992) Developmental regulation of NMDA receptor-mediated synaptic currents at a central synapse. *Nature* **357,** 686–689.
19. Crair, M. C. and Malenka, R. C. (1995) A critical period for long-term potentiation at thalamocortical synapses. *Nature* **375,** 325–328.
20. Zhong, J., Carrozza, D. P., Williams, K., Pritchett, D. B., and Molinoff, P. B. (1995) Expression of mRNAs encoding subunits of the NMDA receptor in developing rat brain.*J. Neurochem.* **64,** 531–539.
21. Burnashev, N., Jonas, P., Helm, P. J., Wisden, W., Monyer, H., Seeburg, P. H., et al. (1992) Calcium-permeable AMPA-kainate receptors in fusiform cerebellar glial cells. *Science* **256,** 1566–1570.
22. Flint, A. C., Maisch, U. S., Weishaupt, J. H., Kriegstein, A. R., and Monyer, H. (1997). NR2A subunit expression shortens NMDA receptor synaptic currents in developing neocortex. *J. Neurosci.* **17(7),** 2469–2476.
23. Fox, K. (1995) The critical period for long-term in primary sensory cortex. *Neuron* **15,** 485–488.
24. Tang, Y. P., Shimizu, E. Dube, G. R., Rampon, C. Kerchner, G. A., Zhuo, M., et al. (1999) Genetic enhancement of learning and memory in mice. *Nature* **401,** 25–27.
25. Kohr, G., Eckhardt, S., Luddens, H., Monyer, H., and Seeberg, P. H. (1994) NMDA receptor channels: subunit-specific potentiation by reducing agents. *Neuron* **12,** 1031–1040.
26. Wenzel, A., Villa, M., and Benke, D. (1996) Development and regional expression of NMDA receptor subtypes containing the NR2D subunit in rat brain. *J. Neurochem.* **66,** 1240–1248.
27. Ikonomidou, C., Mosinger, J. L., Shahid Salles, K., Labryere, J., and Onley, J. W. (1989) Sensitivity of the developing rat brain to hypobaric/ischemic damage parallels sensitivity to *N*-methylaspartate toxicity. *J. Neurosci.* **9(8),** 2809–2818.
28. Ziff, E. B. (1999) Recent excitement in the ionotropic glutamate receptor field. *Ann. NY Acad. Sci.* **868,** 465–473.
29. Sommer, B., Kohler, M., Sprengel, R., and Seeburg, P. H. (1991) RNA editing in brain controls a determinant of ion flow in glutamate-gated channels. *Cell* **67,** 11–19.
30. Burnashev, N., Monyer, H., Seeburg, P. H., and Sakmann, B. (1992) Divalent ion permeability of AMPA receptor channels is dominated by the edited form of a single subunit. *Neuron* **8,** 189–198.
31. Jonas, P., Racca, C., Sakmann, B., Seeburg, P. H., and Monyer, H. (1994) Differences in Ca^{2+} permeability of AMPA-type glutamate receptor channels in neocortical neurons caused by differential expression of the GluR-B subunit. *Neuron* **12,** 1281–1289.

32. McBain, C. J. and Dingledine, R. (1993) Heterogeneity of synaptic glutamate receptors on CA3 stratum radiatum interneurones of rat hippocampus. *J. Physiol.* **462,** 373–392.
33. Washburn, M. S., Numberger, M., Zhang, S., and Dingledine, R. (1997) Differential dependence on GluR2 expression of the three characteristic features of AMPA receptors. *J. Neurosci.* **17,** 9393–9406.
34. Pellegrini-Giampietro, D. E., Gorter, J. A., Bennett, M. V. L., and Zukin, R. S. (1997) The GluR2 (GluR-B) hypothesis: Ca(2+)-permeable AMPA receptors in neurological disorders. *Trends Neurosci.* **20,** 464–470.
35. Gu, J. G., Albuquerque, C. J., Lee, C. J., and MacDermott, A. B. (1996) Synaptic strengthening through activation of Ca^{2+}-permeable AMPA receptors. *Nature* **381,** 793–796.
36. Laezza, F., Doherty, J. J., and Dingledine, R. (1999) Long-term depression in hippocampal interneurons: joint requirement for pre- and postsynaptic events. *Science* **285,** 1411–1414.
37. Pellegrini-Giampietro, D. E., Bennett, M. V. L., and Zukin, R. S. (1991) Differential expression of three glutamate receptor genes in developing rat brain: an in situ hybridization study. *Proc. Natl. Acad. Sci. USA* **88,** 4157–4161.
38. Sanchez, R. M., Koh, S., Rio, C., Wang, C., Lamperti, E. D., Sharma, D., et al. (2001) Decreased GluR2 expression and enhanced epileptogenesis in immature rat hippocampus following perinatal hypoxia-induced seizures. *J. Neurosci.*, **21,** 8145–8163.
39. McDonald, J. W., Trescher, W. H., and Johnston, M. V. (1992) Susceptibility of brain to AMPA induced excitotoxicity transiently peaks during early postnatal development. *Brain Res.* **583(1–2),** 54–70.
40. McDonald, J. W., Trescher, W. H., and Johnston, M. V. (1990) The selective ionotropic-type quisqualate receptor agonist AMPA is a potent neurotoxin in immature rat brain. *Brain Res.* **526(1),** 165–168.
41. Lerma, J., Paternain, A. V., Rodriguez-Moreno, A., and Lopez-Garcia, J. C. (2001) Molecular physiology of kainate receptors. *Physiol. Rev.* **81(3),** 971–998.
42. Bernard, A., Ferhat, L., Dessi, F., Charton, G., Represa, A., Ben-Ari, Y., et al. (1999) Q/R editing of the rat GluR5 and GluR6 kainate receptors in vivo and in vitro: evidence for independent developmental, pathological and cellular regulation. *Eur. J. Neurosci.* **11**, 604–616.
43. Lee, C. J., Kong, H., Manzini, M. C., Albuquerque, C., Chao, M. V., and MacDermott, A. B. (2001) Kainate receptors expressed by a subpopulation of developing nociceptors rapidly switch from high to low Ca^{2+} permeability. *J. Neurosci.* **21(13)**, 4572–4581.
44. Bortolotto, Z. A., Clarke, V. R., Delany, C. M., Parry, M. C., Smolders, I., Vignes, M., et al. (1999) Kainate receptors are involved in synaptic plasticity. *Nature* **402**, 297–301.
45. Vignes, M. and Collingridge, G. L. (1997) The synaptic activation of kainate receptors. *Nature* **388**, 179–182.
46. Bahn, S., Volk, B., and Wisden, W. (1994) Kainate receptor gene expression in the developing rat brain. *J. Neurosci.* **14**, 5525–5547.
47. Paternain, A. V., Herrera, M. T., Nieto, M. A., and Lerma, J. (2000) GluR5 and GluR6 kainate receptor subunits coexist in hippocampal neurons and coassemble to form functional receptors. *J. Neurosci.* **20(1)**, 196–205.
48. Albala, B. J., Moshe, S. L., and Okada, R. (1984) Kainic-acid-induced seizures: a developmental study. *Brain Res.* **315(1)**, 139–148.
49. Stafstrom, C. E., Thompson, J. L., and Holmes, G. L. (1992) Kainic acid seizures in the developing brain: status epilepticus and spontaneous recurrent seizures. *Brain Res. Dev. Brain Res.* **65**, 227–236.
50. Gerber, U. and Gahwiler, B. H. (1994) Modulation of ionic currents by metabotropic glutamate receptors, in *The Metabotropic Glutamate Receptors* (Conn, P. J. and Patel, J., eds.), Humana, Totowa, NJ, pp. 125–146.
51. Glaum, S. R. and Miller, R. J. (1994) Acute regulation of synaptic transmission by metabotropic glutamate receptors, in *The Metabotropic Glutamate Receptors* (Conn, P. J. and Patel, J. eds.) Humana, Totowa, NJ, pp. 147–172.
52. De Blasi, A. D., Conn, P. J., Pin, J.-P., and Nicoletti, F. (2001). Molecular determinants of metabotropic glutamate receptors signaling. *Trends Pharmacol. Sci.* **22(3)**, 114–120.
53. Pin, J. -P. and Duvoisin, R. (1995) The metabotropic receptors: structure and function. *Neuropharmacology* **34**, 1–26.
54. Lu, Y.-M., Jia, Z., Janus, C., Henderson, J. T., Gerlai, R., Wojtowicz, J. M., et al. (1997) Mice lacking metabotropic glutamate receptor 5 show impaired learning and reduced CA1 long-term potentiation (LTP) but normal CA3 LTP. *J. Neurosci.* **17(13)**, 5196–5205.
55. Nicoletti, E., Aronica, E., Battaglia, G., Bruno, V., Casabona, G., Catania, M. V., et al. (1994) Plasticity of metabotropic glutamate receptors in physiological and pathological conditions, in *The Metabotropic Glutamate Receptors* (Conn, P. J., J Patel, J., eds.), Humana, Totowa, NJ, pp. 243–269.
56. Casabona, G., Genazzani, A. A., Di Stefano, M., Sortino, M. A., and Nicoletti, F. (1992) Developmental changes in the modulation of cyclic AMP formation by the metabotropic glutamate receptor agonist 1*S*,3*R*-aminocyclopentane-1,3-dicarboxylic acid in brain slices. *J. Neurochem.* **59**, 1161–1163.
57. Schoepp, D. D. and Johnson, B. G. (1993) Metabotropic glutamate receptor modulation of cAMP accumulation in the neonatal rat hippocampus. *Neuropharmacology* **32**, 1359–1365.
58. Schoepp, D. D., Johnson, B. G., and Monn, J. A. (1996) (1*S*,3*R*)-1-aminocyclopentane-1,3-dicarboxylic acid-induced increases in cyclic AMP formation in neonatal rat hippocampus are mediated by a synergistic interaction between phosphoinositide- and inhibitory cyclic AMP-coupled mGluRs. *J. Neurochem.* **66**, 1981–1985.

59. McDonald, J. W., Fix, A. S., Tizzano, J. P., and Schoepp, D. D. (1993) Seizures and brain injury in neonatal rats induced by 1*S*,3*R*-ACPD, a metabotropic glutamate receptor agonist. *J. Neurosci.* **13**, 4445–4455.
60. Camon, L., Vives, P., de Vera, N., and Martinez, E. (1998) Seizures and neuronal damage induced in the rat by activation of group I metabotropic glutamate receptors with their selective agonist 3,4-dihydroxyphenylglycine. *J. Neurosci. Res.* **51**, 339–348.
61. Miyamoto, M., Ishida, M., and Shinozaki, H. (1997) Anticonvulsive and neuroprotective actions of a potent agonist (DCG-IV) for group II metabotropic glutamate receptors against intraventricular kainate in the rat. *Neuroscience* **77**, 131–140.
62. Tang, E., Yop, P. K., Chapman, A. G., Jane, D. E., and Meldrum, B. S. (1997) Prolonged anticonvulsant action of glutamate metabotropic receptor agonists in inferior colliculus of genetically epilepsy-prone rats. *Eur. J. Pharmacol.* **327**, 109–115.
63. Sacaan, A. I. and Schoepp, D. D. (1992) Activation of hippocampal metabotropic excitatory amino acid receptors leads to seizures and neuronal damage. *Neurosci. Lett.* **139**, 77–82.