GLUTAMATE RECEPTORS; FROM BASIC PHARMACOLOGY TO CLINICAL APPLICATIONS

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ABSTRACT

The existence of glutamate receptors as important neurotransmitter receptors was not fully accepted until the 1980s. It is now generally agreed that glutamate is the major fast excitatory neurotransmitter in brain, and plays a major role in brain development, affecting neuronal migration, neuronal differentiation, axon genesis, and neuronal survival through its action on the glutamate receptors. There are two classes of glutamate receptors; the ionotropic glutamate receptors (iGluRs), and the metabotropic glutamate receptors (mGluRs). Glutamate receptors are important mediators of a wide range of neuronal functions from primary sensory perception to cognition and also play a role in a number of neurodegenerative diseases. Understanding the role of glutamate in these neurological diseases may highlight treatment potentials of different agonists and antagonists to glutamatergic transmission. In this review, the basic biology of glutamate receptors will be explored. The second part of this article presents a review of the literature for the role of glutamate agonists and antagonists in neurological disease.

1. INTRODUCTION

Since the late 1950s, glutamate, which was previously considered to be solely a metabolic precursor, has been known to excite neurons (Curtis et al., 1959). However, as most central neurons are excited by glutamate, this was originally interpreted as a nonspecific effect. The existence of glutamate receptors as important neurotransmitter receptors was not fully accepted until the 1980s. The realization that glutamate activates a number of pharmacologically distinct subtypes (Ascher and Nowak, 1988; Collingridge and Lester, 1989) and plays an important role in long-term potentiation (Bliss and Collingridge, 1993) led to the recognition that most excitatory neurotransmission in the central nervous system is mediated by glutamate receptors. It is now generally agreed that glutamate is the major fast excitatory neurotransmitter in brain. Glutamate plays a major role in brain development, affecting neuronal migration, neuronal differentiation, axon genesis, and neuronal survival (Coyle et al., 2002; Hassel and Dingledine, 2006). Glutamate receptors are also important mediators of a wide range of neuronal functions from primary sensory perception to cognition (Collingridge and Bliss, 1995; Collingridge and Singer, 1990; Danyasz et al., 1995; Dingledine et al., 1999). It is also known that glutamate receptors play a role in a number of neurodegenerative diseases and epilepsy (Gillessen et al., 2002). In addition, glutamate receptors seem to have functional roles outside of the nervous system (as in insulin secretion, bone resorption, cardiac pacemaking, and tactile sensation) (Ault and Hildebrand, 1993; Carlton et al., 1995; Chenu et al., 1995; Chenu et al., 1998; Gill et al., 1998; Inagaki et al., 1995; Jorgensen et al., 1995; Patton et al., 1998; Weaver et al., 1996).

1.1. Glutamate Synthesis and Release

L-glutamate can be synthesized in nerve terminals from a-ketoglutarate (formed from gluconeogenesis and the tricarboxylic acid cycle) by the enzyme glutamate dehydrogenase, and from glutamine by the glutaminase enzyme (Hashimoto et al., 2005;...
Hashimoto and Hattori, 2007). Upon depolarization glutamate is released into the cleft, where the glutamate either (i) is bound to pre- and post-synaptic receptors, (ii) is actively taken back up via a glutamate transporter and repackaged, (iii) diffuses away from the cleft, or (iv) is internalised by glial glutamate transporters (Anderson and Swanson, 2000; Attwell, 2000). In the glial cells, glutamate is converted to glutamine by the glial enzyme glutamine synthetase. Glutamine is transported from glia into nerve terminals by specific transporters, followed by re-conversion to glutamate via glutaminase and repackaged in synaptic vesicles, completing the so called ‘glutamine cycle’ that is proposed to ensure the efficient replenishment of neurotransmitter glutamate (Chaudhry et al., 2002).

There are two classes of glutamate receptors; First the ionotropic glutamate receptors (iGluRs): which are ligand gated ion channels receptors consist of four subunits. The assembled subunits may or may not be homologous, these different combinations of subunits result in channels with different characteristics (Dingledine et al., 1999). These receptors mediate fast synaptic transmission between neurons in mammalian brain.

The second class is the metabotropic glutamate receptors (mGluRs): which are G-protein coupled receptors that have transmembrane segments similar to other G-protein coupled receptors, that have extracellular Amino terminal domain (ATD) process some structural homology to that in iGluRs (O’Hara et al., 1993).

2. Glutamate Receptors

2.1. Ionotropic Glutamate Receptors (iGluRs)

There are three major types of ionotropic glutamate receptors (iGluRs), which are named after the agonists that were originally identified to activate them selectively, and are, thus, called N-methyl-D-aspartate (NMDA), α-amino-3-hydroxy-5-methyl-4-isozolepropionic acid (AMPA) and 2-carboxy-3-carboxymethyl-4-isopropenylpyrrolidine (kainate) receptors (Collingridge and Lester, 1989). Native receptors of all of these families are likely tetrameric assemblies comprising more than one type of subunit (Laube et al., 1998), and the subunits that comprise these are specific for each of the three families (Dingledine and Conn, 2000). The subunit composition determines the biophysical properties of the receptor and to a variable extent its pharmacology.

Ionotropic glutamate receptor subunits possess an extracellular amino terminal domain, followed by a first transmembrane domain and then a pore forming membrane-residing domain that does not cross the membrane but forms a reentrant loop entering from and exiting to the cytoplasm. The second and third transmembrane domains are linked by a large extracellular loop and the third transmembrane domain is followed by an intracellular carboxy terminus (Dingledine et al., 1999; Mayer and Armstrong, 2004). The crystal structure of the iGluR ligand binding domains, which comprise polypeptides in both the amino terminus (S1 domain) and the extracellular loop between transmembrane domains 3 and 4 (S2 domain), has confirmed this topology model for the ionotropic glutamate receptor family (Armstrong et al., 1998; Mayer and Armstrong, 2004).

The ionotropic receptors themselves are ligand gated ion channels. Once the ligand is bounded to the receptor, charged ions such as Na\textsuperscript{+} and Ca\textsuperscript{2+} pass through a channel in the centre of the receptor complex. This flow of ions results in a depolarization of the plasma membrane and the generation of an electrical current that is propagated down the processes (dendrites and axons) of the neuron to the next in line (Mayer and Westbrook, 1987). These ionotropic glutamate receptors are an extremely diverse group of receptors. This diversity is generated both before and after gene transcription. Individual receptors are multimeric assemblies of subunits, transcribed from separate genes. However, following gene transcription, the resultant pre-mRNA may be modified. For instance, different regions of this mRNA molecule can be spliced together, giving rise to multiple mRNAs that are
translated into different proteins. This is known as 'splice variation' and is very common among neuroreceptors (Sommer et al., 1990).

A further modification leading to diversification is RNA editing, in which selected nucleotides in the mRNA sequence transcribed from the gene sequence are enzymatically modified, changing the amino-acid coded for (Melcher et al., 1996).

2.1.1. NMDA Receptors
NMDA receptors are essential for normal physiological processes in the central nervous system. As these receptors are implicated in neuronal survival and maturation (Simon et al. 1984; Balazs et al., 1989), neuronal migration (Komuro and Rakic, 1993), induction of long-term potentiation (LTP) (Bliss and Collingridge, 1993), formation of sensory maps (Cline et al. 1987; Simon et al. 1992), and neurodegeneration (Meldrum and Garthwaite, 1990; Lipton and Rosenberg, 1994; Cull-Candy et al., 2001). Excessive activation of NMDARs can lead to neuronal damage in many acute (hypoxic ischemic injury) and chronic neurodegenerative diseases (Alzheimer, Parkinson, Huntington).

The NMDA receptor family is composed of seven subunits, GluN1, GluN2A–D and GluN3A and B, which are all products of separate genes. Functional NMDA receptors appear to be comprised of GluN1 in combination with one or more GluN2 subunits or GluN1 in combination both GluN2 and GluN3 subunits (Watanabe et al., 1992, 1993; Monyer et al., 1992; Zhong et al., 1995; Wenzel et al., 1997). NMDA receptor is unique amongst ligand-gated ion channels in its requirement for two obligatory co(agonists (Kleckner and Dingledine, 1988). As not only the binding of glutamate is required to activate the channel, but a micromolar concentrations of glycine must also be present (Johnson and Ascher, 1987, Kleckner and Dingledine, 1988). The binding of glycine and glutamate is on different binding sites localized on the GluN1 (Kuryatov et al., 1994; Wafford et al., 1995; Kew et al. 2000) and GluN2 (Laube et al., 1997; Anson et al., 1998) subunits, respectively. Electrophysiological studies have demonstrated that NMDA receptor activation requires occupation of two independent glycine sites and two independent glutamate sites (Benveniste and Mayer, 1991; Clements and Westbrook, 1991). Thus, the minimal requirement for a functional NMDA receptor is likely to be a tetramer composed of two GluN1 and two GluN2 subunits. In agreement, recent studies have provided convincing evidence that, like the other iGluRs, NMDA receptors are of tetrameric structure and are likely composed of pairs, or dimers, of dimers (e.g. an GluN1 dimer in combination with an GluN2X dimer) (Schorge and Colquhoun, 2003; Mayer and Armstrong, 2004). In receptors containing GluN1, GluN2 and GluN3 subunits, it seems likely that an GluN3 subunit substitutes for one of the GluN2 subunits. Thus, GluN1 subunits are combined with different GluN2 subunits and GluN3 to generate a large number of different NMDA receptors with differing pharmacological and biological properties.

NMDA receptors have also separate binding sites for magnesium ions (Mg$^{2+}$), zinc ions (Zn$^{2+}$), as well as a polyamine recognition site (Lodge, 1997). Mg$^{2+}$ ions provide a voltage-dependent block of NMDA-gated channels. Where, at resting membrane potentials, NMDA receptors are inactive. This is due to a voltage-dependent block of the channel pore by magnesium ions, preventing ion flows through it (Nowak et al., 1984). Sustained activation of AMPA receptors by, for instance, a train of impulses arriving from a pre-synaptic terminal, depolarizes the post-synaptic cell, releasing the channel inhibition and thus allowing NMDA receptor activation.

On the other hand, the zinc and polyamine sites are not needed for activation of NMDA receptors, but affect the efficacy of the channel. Zinc blocks the channel in a voltage-independent manner (Westbrook and Mayer, 1987). The polyamine site, is a regulatory site for molecules that modulate the functioning of the NMDA receptor, binds compounds such as spermine or spermidine, either potentiating (Ranson and Stec, 1988; Williams et al., 1994) or inhibiting (Williams et al., 1994) the activity of the receptor, depending on the combination of subunits forming each NMDA receptor (Williams et al., 1994).

The unique property of being both voltage-dependent and ligand gated gives the NMDAR the ability to act as a coincidence detector for presynaptic activity (glutamate release) and post-
synaptic activity (adequate depolarization of the post-synaptic membrane). Glutamate binding onto an NMDA receptor opens non-selective cation channels, resulting in a conductance increase. The high conductance channel associated with these receptors is more permeable to Ca\(^{2+}\) than Na\(^+\) ions (Mayer and Westbrook, 1987). Thus NMDA receptor activation leads to a calcium influx into the post-synaptic cells, a signal that is instrumental in the activation of a number of signaling cascades that can subsequently affect synaptic plasticity and gene transcription (Bading et al., 1993). Induction of NMDAR-dependent forms of synaptic plasticity, such as LTP, is thought to underlie many types of learning and memory formation (Nicoll, 2003).

However, overactivation of the receptor relieves the Mg\(^{2+}\) block and causes an excessive amount of Ca\(^{2+}\) influx into the nerve cell, which then triggers a variety of processes that can lead to necrosis, apoptosis, or dendritic/synaptic damage. These detrimental processes include Ca\(^{2+}\) /overload of mitochondria, resulting in oxygen free radical formation, activation of caspases, and release of apoptosis inducing factor; Ca\(^{2+}\)-dependent activation of neuronal NOS, leading to increase NO production and the formation of toxic peroxynitrite (ONOO–) and stimulation of mitogen-activated protein kinase p38 (MAPK p38), which activates transcription factors that can go into the nucleus to influence neuronal injury and apoptosis (Dawson et al., 1991; Bonfoco et al., 1995; Okamoto et al., 2002; Hara et al., 2005).

2.1.2. AMPA Receptors
AMPA receptors are the receptors responsible for most rapid excitatory transmission within the vertebrate CNS and their modulation is the ultimate mechanism that underlies much of the plasticity of excitatory transmission that is expressed in the brain. However, the affinity of AMPA receptors for L-glutamate, the endogenous ligand for these receptors, is much lower than the affinity at NMDA receptors (Dingledine et al., 1999). The associated channels are rapidly activated and inactivated, and appear to be present on all neurons within the CNS (Tzschentke, 2002).

There are four known subunits GluA1 to GluA4 which are widely, and differentially distributed throughout the CNS (Hollmann and Heinemann, 1994). The types of subunits forming these receptors determine their biophysical properties and pharmacological sensitivity. Two alternative splice variants for all GluA1 to GluA4 subunits designated as ‘flip’ and ‘flop’ have been shown to differ in their expression throughout the brain and during development, and to impart different pharmacological properties (Monyer et al., 1990; Sommer et al., 1990).

Native AMPA receptor channels are impermeable to calcium, a function controlled by the GluA2 subunit. The calcium permeability of the GluA2 subunit is determined by the post-transcriptional editing of the GluA2 mRNA. Where, in the primary transcript of GluA2, a codon for glutamine (Q) in the transmembrane II (TMII) region is edited to one for arginine (R) by adenosine deaminase that converts an adenosine to an inosine (Seeburg and Hartner, 2003). This is the so called Q/R editing site, where GluA2(Q) is calcium permeable whilst GluA2(R) is not. Almost all the GluA2 protein expressed in the CNS is in the GluA2(R) form, giving rise to calcium impermeable AMPA receptors. This, along with the interactions with other intracellular proteins, makes GluA2 perhaps the most important AMPA receptor subunit (Dingledine et al., 1999; Seeburg and Hartner, 2003).

2.1.3. Kainate Receptors
Kainate receptors constitute a separate group from the NMDA and AMPA receptors, although they share many of the same structural characteristics. Kainate receptors are composed of two related subunit families, GluR5–7 and KA-1 and KA-2. Once again, native receptors are likely tetrameric combinations, possibly of both homomeric and heteromeric combinations (Bettler et al., 1990; Egebjerg et al., 1991). KA-1 and KA-2 subunits do not form functional homomeric receptors but do form high-affinity kainate binding sites and can, thus, bind agonist ligands. KA-1 and KA-2 combine in heteromeric assemblies with members of the GluR5–7 subfamily to form functional receptors resembling native kainate receptors (Bleakman et al., 2002). Unlike the Kan subunits, the GluR5, 6 and
7 subunits can form functional homomeric receptors as well as combine with KA-1 and KA-2 to form heteromeric receptors with distinct pharmacological properties (Alt et al., 2004).

KA receptors are permeable to Na\(^+\) and K\(^+\) ions and, like NMDA and AMPA receptors, contribute to excitatory postsynaptic currents. The role of KA receptors in synaptic plasticity is less well-defined, however, KA receptors have been found to be localized presynaptically where they can modulate neurotransmitter release (Huettner, 2003).

### 2.2. Metabotropic Glutamate Receptors (mGluRs)

It was first suggested that metabotropic glutamate receptors (mGluRs) might exist in 1985, after it was noted that glutamate could stimulate phospholipase C through the activation of a receptor that did not belong to any of the ionotropic glutamate receptor families (Temple et al., 2001). The suspicion that mGluRs existed was confirmed in 1987, and in 1991 the first mGluR was cloned (Temple et al., 2001). They are found in pre- and postsynaptic neurons in synapses of the hippocampus, cerebellum (Hinoi et al., 2001), and the cerebral cortex, as well as other parts of the brain and peripheral tissues (Chu and Hablitz, 2000). These mGluRs perform a variety of functions in the central and peripheral nervous systems: for example, they are involved in learning, memory, anxiety, and the perception of pain (Ohashi et al., 2002). Unlike ionotropic receptors, mGluR are not directly linked to ion channels, but may affect them by activating biochemical cascades. As these mGluRs are members of guanine nucleotide-binding protein -coupled receptors (GPCRs) (Bonsi et al., 2005).

Many GPCRs can be activated as individual, monomeric receptors, however, most also exist as homodimers. This is particularly true of Class C GPCRs like the mGluR ,GABAB, Ca\(^{2+}\)sensing pheromone and taste receptors (Kuang et al., 2005; Pin et al., 2003). Receptor dimerization at the ATD is crucial for receptor activation, since binding of L-glutamate to one or two of the ATDs of the dimer increase the probability of the ATDs to close and cause a conformational change of the ATDs relative to each other (Pin et al., 2003). Most likely, this conformational change brings the lower parts of the two ATDs of the dimer closer together resulting in a similar relative movement of the two seven transmembrane domains (7TMD) to an active receptor conformation capable of activating G-proteins (Kunishima et al., 2000). Recently, it was shown that binding of agonist to one ATD partially activated the receptor, while binding of agonist to both ATDs was required for full receptor activation (Kniazeff et al., 2004; MRC, 2009; Brock et al., 2007). This binding of an agonist as glutamate to ATD of a mGluR causes G proteins bound to the intracellular region to be phosphorylated, affecting multiple biochemical pathways and ion channels in the cell (Platt, 2007). Because of this, mGluRs can both increase or decrease the excitability of the post synaptic cell, thereby causing a wide range of physiological response. In addition to producing excitatory and inhibitory postsynaptic potentials, mGluRs serve to modulate the function of other receptors (such as NMDA receptors), and changing the synapse's excitability (Chu and Hablitz, 2000; Endoh, 2004; Bonsi et al., 2005, Platt, 2005).

There are eight metabotropic glutamate receptors (mGlu1 to mGlu8) that are placed into three groups; I, II, and III on the basis of sequence homology, agonist pharmacology, and coupling to intracellular transduction mechanisms (Chu and Hablitz, 2000; Hinoi et al., 2001; Endoh, 2004; Bonsi et al., 2005).

#### 2.2.1. Group I

The mGluRs in group I, including mGlu1 (splice variants: mGlu1a, -b, -c, -d, -e) and mGlu5 (splice variants: mGlu5a and -b) receptors, are stimulated most strongly by the excitatory amino acid analog L-glutamyl acid (Chu and Hablitz, 2000; Bates et al., 2002). These receptors are coupled to postsynaptic inositol phosphate metabolism, as stimulation of these receptors causes an associated phospholipase C molecule to hydrolyze phosphoinositide phospholipids in the cell's plasma membrane (Chu and Hablitz, 2000; Endoh, 2004; Bonsi et al., 2005). This leads to formation of inositol 1,4,5-trisphosphate (IP3) and diacylglycerol (DAG) as second messengers. Due to its hydrophilic character IP3 can travel to the endoplasmic reticulum where it induces the opening of calcium channels.
The lipophilic DAG remains in the membrane acting as a cofactor for the activation of protein kinase C (PKC), increasing by these ways the cytosolic calcium concentrations. These receptors are usually found on postsynaptic membranes (Endoh, 2004) and are rarely found presynaptically. They are also associated with Na\(^+\) channels and K\(^+\) channels (Chu and Hablitz, 2000). Their action can be excitatory, increasing conductance, causing more glutamate to be released from the presynaptic cell (Chu and Hablitz, 2000). They can also inhibit glutamate release and can modulate voltage-dependent calcium channels (Endoh, 2004). mGluR5, are positively coupled to NMDA receptor function via PKC (Brakeman et al., 1997; Tu et al., 1999; Xiao et al., 2000; Szumlinski et al., 2006).

2.2.2. Group II

The receptors in group II, including mGluRs 2 and 3 (Flor et al., 1995a; Emile et al., 1996), are negatively coupled to adenylyl cyclase thereby reducing intracellular cyclic adenosine monophosphate (cAMP) formation in many expression systems, but it also may be coupled to other transduction mechanisms under physiological conditions (Chu and Hablitz, 2000; Hinoi et al., 2001; Bonsi et al., 2005; MRC, 2009). They are found on both pre- and postsynaptic membranes, and are involved in presynaptic inhibition (Endoh, 2004). However, they do not appear to affect postsynaptic membrane potential by themselves. Receptors in groups II and III reduce the activity of postsynaptic potentials, both excitatory and inhibitory, in the cortex (Chu and Hablitz, 2000).

2.2.3. Group-III

This group of mGlu receptors include the subtypes mGlu4 (splice variants: mGlu4a and -b), mGlu6, mGlu7 (splice variants mGlu7a and -b), and mGlu8 (splices variants: mGlu8a and -b). Group III mGluR are all coupled to Gi proteins in transfected cells (i.e. they are negatively coupled to adenylyllycycase where activation inhibits cAMP formation) (Okamoto et al., 1994; Flor et al., 1995b; 1997; Duvoisin et al., 1995; Corti et al., 1997). mGlu4, mGlu7 and mGlu8 receptors are predominantly localized to axon terminals where they are in a position to control the synaptic availability of glutamate, as well as of GABA, dopamine, and serotonin (Cartmell and Schoepp, 2000; Ferraguti and Shigemoto, 2006). As opposed to other group III mGlu receptors, mGlu7a and b receptors are restricted to the center of the presynaptic active zone of axon terminals (i.e., the site of synaptic vesicle fusion) (Ferraguti and Shigemoto, 2006). As such, mGlu7 receptors are well positioned to play the role of the main glutamatergic autoreceptor responsible for the regulation of glutamate release under normal physiological conditions (Schoepp, 2001).

3. Glutamate Receptors Pharmacology

3.1. Inotropic Glutamate Receptors Pharmacology

3.1.1. NMDA Receptor Agonists

i. Agonist at the glutamate site

Early structure–activity studies established that an ideal structure for activating NMDARs is represented by (S)-glutamate. NMDA is several-fold weaker as an agonist than (S)-glutamate. However, NMDA has a low affinity for the plasma membrane transporters and thus can appear more potent than glutamate in some physiological assays. NMDA shows little selectivity between the NMDA receptor subtypes, however, it displays no activity at other glutamate receptors (Watkins, 1981).

By incorporating ring systems into the glutamate structure, rigid glutamate analogues that are potent NMDAR agonists have been developed. They mimic the active, partially folded, conformation of (S)-glutamate and include homoquinolinate (Brown et al., 1998), (2S,1′R,2′S) 2-(carboxycyclopropyl) glycine (L-CCG-IV) (Kawai et al., 1992), (1R,3R) 1-aminocyclopentane-1,3-dicarboxylic acid (ACPD) (Sunter et al., 1991), and 1-aminocyclobutane-1,3-dicarboxylic acid (ACBD) (Jane et al., 1994). Other selective, reasonably full agonists at NMDA receptors include L-aspartate, quinolinate and homocysteate, whilst cis-2,3-piperidinedicarboxylic acid is a fairly low efficacy partial agonist (Priestley and Kemp, 1994).

ii. Agonist and partial agonist at the glycine co-agonist site
Initially it was thought that endogenous levels of extracellular glycine were enough to saturate the glycine binding site; however, later studies suggest that this is not the case and it may be possible to develop positive modulators of NMDAR function via interaction with the glycine binding site (Danysz and Parsons, 1998). Amino acids such as \((R)\)-alanine and \((R)\)-serine display high affinities for the glycine site and behave as full agonists (Leeson and Iversen, 1994). A number of compounds were identified as having agonist, or partial agonist, activity at the glycine co-agonist site on the NR1 subunit. Conformationally constrained analogues of glycine such as ACPC, a cyclopropylanalogue (Watson and Lanthorn, 1990), and ACBC, a cyclobutane analogue (Hood et al., 1989), are partial agonists with different degrees of efficacy. At lower doses, they show antischizophrenic properties in animal models but this effect is reversed at higher doses when they act like antagonists (Javitt, 2002). Additionally, some vinyl substituted glycine derivatives, such as S-hydroxyethylvinyl glycine, act as reasonably potent glycine site agonists. Glycine-site partial agonists, in order of decreasing levels of intrinsic activity, include L-alanine, D-cycloserine, R(+)-3-amino-1-hydroxypyrrolid-2-one \([(+)\text{-HA-966}]\) and R(+)-cis-\(\beta\)-methyl-3-amino-1-hydroxypyrrolid-2-one \((L-687,414)\) (Kemp and Leeson, 1993).

3.1.2. NMDA Receptor Antagonists
NMDAR antagonists can be categorized pharmacologically into three major groups according to site of action on the receptor-channel complex (Wong and Kemp, 1991). Competitive NMDAR antagonists acting on either (i) the glutamate (agonist) recognition site, or (ii) the glycine (co-agonist) site; uncompetitive NMDAR antagonists (NMDAR open channel blockers; that block the channel pore following activation by the agonists); and allosteric inhibitors act at modulatory sites, such as the high-affinity \(\text{Zn}^{2+}\) site, and the polyamine site.

3.1.2.1. Competitive Antagonists
i. Antagonists of the glutamate recognition site
A large number of competitive antagonists of the glutamate recognition site of the NMDA receptor have been synthesized. The vast majority are conformationally constrained, \(\alpha\)-amino carboxylic acids with an appropriately placed \(\omega\)-phosphonic acid group (Jane et al., 1994). Examples of high-affinity glutamate site antagonists include \((R)-2\)-amino-5 phosphonopentanoate (DAP5), \((\pm)-\text{cis-4-phosphonomethyl-2-piperidine carboxylic acid (CGS 19755), D,L-(E)-2-amino-4-propyl-5-phosphono-3-} \text{pentenoic acid (CGP 39653), (2-amino-4,5-(1,2-cyclohexyl))-7-phosphonooctanooic acid (NPC 12626), (\pm)-6-phosphonomethyl-decahydroisoquinoline-3-carboxylic acid (LY 274614), (S)-alpha-amino-5-phosphonomethyl[1,1′-biphenyl]-3-propanoic acid (SDZ EAB-515) and (S)-alpha-amino-5-phosphonomethyl[1,1′:4′,1″-terphenyl]-3-propanoic acid (SDZ 215-439). Although these compounds have in vivo activity, they penetrate the blood–brain barrier (BBB) relatively poorly due to their highly charged nature and equilibrate slowly in the brain.

In general, these compounds show only modest NMDA, receptor subtype selectivity, although there are some notable exceptions. For example, some 5-phosphonomethylquinolinediones have been shown to have marked preference for the human NR1/NR2A, rather than NR1/NR2B, subunit containing NMDA receptors. The best of these are \((1RS,1′S)-(PEAQX (Auberson et al., 2002) and its active diastereomer, NVP-AAM077 (Chaperon et al., 2003). Conantokin G (Con G) is a 17-amino-acid peptide antagonist of NMDA receptors isolated from the venom of the marine cone snail, Conusgeographus, which also acts at the glutamate recognition site but shows considerable selectivity for NR2B subtype containing receptors (Donevan and McCabe, 2000). As expected of a large peptide, conantokin G is not active following systemic administration.

ii. Antagonists of glycine recognition site
The first full antagonist found to bind to the glycine site was kynurenic acid, after that a large number of glycine recognition site antagonists have been identified. Many of these were derived from medicinal chemistry efforts based around kynurenic acid (Bristow et al., 1996). However, compounds of this class tend to bind very tightly to plasma proteins and this compromises their BBB penetration and in
vivo activity (Rowley et al., 1997). Nevertheless, numerous high-affinity derivatives (e.g. quinolines, quinolones and indoles) possessing systemic activity (Leeson and Iversen, 1994), exemplified by the following: 7-chloro-4-hydroxy-3-(3-phenoxy)phenyl-2(H) quinoline (L-701-324) (Bristow et al., 1996); (E)-3-[(phenylcarbamoyl)ethenyl]-4,6-dichloroindole-2-carboxylic acid (Gavestinel/GV150 526A) (Di Fabio et al., 1997); 5-nitro-6,7-dichloro-1,4-dihydro-2,3-quinazalininedione (licostinel/Acea 1021) and derivatives (Zhou et al., 2003); 7-chloro-4-hydroxy-2-(4-methoxy-2-methylphenyl)-1,2,5,10-tetrahydropyridazino[4,5-b]quinoline-1,10-dione (ZD9379) (Takano et al., 1997); (E)-3-(2-phenyl-2-carboxyethenyl)-4,6-dichloro-1H-indole-2-carboxylic acid (MDL 105,519) (Baron et al., 1997); (S)-7-chloro-3-[2-((1R)-1-carboxyethoxy)-4-aminomethylphenyl] aminocarbonylmethyl-1,3,4-tetrahydrobenz[e,d]indole-2-carboxylic acid hydrochloride (SM-31900) (Ohtani et al., 2002).

These compounds act as full antagonists at the glycine site, i.e. they completely inhibit NMDA responses at sufficiently high concentrations.

3.1.2.2. Uncompetitive Antagonists
To be clinically acceptable antagonist of NMDAR, the drug must block excessive activation of the NMDAR while leaving normal function relatively intact in order to avoid side-effects. Drugs that simply compete with glutamate or glycine at the agonist binding sites block normal function and therefore do not meet this requirement, and have thus failed in clinical trials to date because of side-effects (drowsiness, hallucinations, and even coma; Hickenbottom and Grotta, 1998; Lutsep and Clark, 1999; Palmer, 2001). This directed the clinical research more to uncompetitive antagonists. Blockers of the ion channel pore of the NMDA receptor have been a fruitful source of potent, selective and drug like antagonists. A number of compounds that block NMDAR channels by a use-dependent (channels must be opened via binding of glycine and glutamate to their respective binding sites such that they can access to, then dissociation from the binding site) and voltage-dependent mechanism have been identified (Huettner and Bean, 1988). These compounds include the dissociative anaesthetics, phencyclidine and ketamine (Aniset al., 1983). Phencyclidine,1-(1-phenylcyclohexyl) piperidine commonly initialized as (PCP), and also known as angel dust, was formerly used as an anesthetic agent. Ketamine is NMDAR antagonists that have been used in clinical practice for many years but for non-neurological indications (MacDonald et al., 1991). Ketamine is commonly used as a dissociative anesthetic. Other NMDAR antagonist is dextromethorphan which is a commonly used cough suppressant and its metabolite, dextrorphan, antagonizes the NMDAR by binding to a site within the channel pore (Franklin and Murray, 1992; LePage and Ishmael, 2005).

Initial identification of ketamine and phencyclidine as selective NMDA receptor antagonists led to the development of selective high affinity NMDAR channel blockers such as dizocilpine (MK-801) (Gill et al., 1987). The kinetic action of channel blocking and unblocking exhibited by MK-801 depends on the NR2 subunit composition of the NMDAR complex. Slower channel blocking kinetics were observed for NR2C-containing receptors compared to those containing NR2A or NR2B (Monaghan and Larson, 1997). This is consistent with the shorter open times of NR2C-containing receptors.

Subsequently, numerous compounds have been identified as selective blockers of the NMDA receptor ion channel e.g. N-1-naphthyl-N′-(3-ethylphenyl)-N′-methylguanidine (aptiganel/CNS1102) (Reddy et al., 1994) These compounds act as open channel blockers and can become trapped inside the closed channel, which, coupled with their high affinity and slow reversibility, results in them having very steep dose–response curves and little or no separation between their therapeutic effect and unacceptable side effects (Kemp and Kew, 1998).

Amantadine is NMDAR antagonist that was the first member of a class of organic molecules called aminoadamantanes to be introduced into clinical use. It was first marketed in the 1960s for prophyaxis of respiratory infections due to influenza A virus but was serendipitously discovered to have beneficial...
effects on extrapyramidal symptoms in a patient with Parkinson’s disease taking amantadine for influenza prophylaxis (Schwab et al., 1969). Initially, amantadine was assumed to have its antiparkinsonian effects through direct dopaminomimetic activity based on indirect in-vivo evidence (Danysz et al., 1997). Later studies have shown that the dominant mechanism of action for amantadine is its NMDAR antagonistic properties, acting as an open-channel blocker (Kornhuber et al., 1994).

Memantine, 1,3-dimethyl-5-aminoadamantane, is NMDAR antagonists that has been approved for human use (Farlow, 2004). It exhibits fast on-and-off kinetics, allowing it to rapidly bind to and, quickly dissociate from the receptor and reduced tendencies to produce adverse reactions such as psychotomimetic effects (Parsons et al., 1999). In addition, memantine has pronounced voltage-dependency and, therefore, will dissociate from the NMDAR channel upon strong postsynaptic depolarization, as occurs during normal physiological activation, but will remain blocking the channel during moderate long-lasting depolarization, as during chronic excitotoxic conditions (Parsons et al., 1999). Therefore, memantine’s favorable clinical profile might also result from preservation of normal synaptic activity while inhibiting excitotoxicity. Memantine also affects the cholinergic neurotransmitter system; in particular, it inhibits α7 nicotinic acetylcholine receptors (nAChRs) (Aracava et al., 2005). This effect might also contribute to its favorable clinical profile as there is some evidence that α7 nAChR inhibition results in attenuation of pathological processes associated with Alzheimer’s disease (Wang et al., 2003). Memantine is now in clinical use for treatment of cognitive deficits in moderate to severe Alzheimer’s disease.

Riluzole is another NMDAR antagonist that was originally synthesized by researchers in France, and early laboratory studies in the 1980s suggested it had anticonvulsant properties (Mizoule et al., 1985). However, the discovery that riluzole can disrupt glutamate neurotransmission to prevent NMDAR-mediated neuronal death in experimental models (Malgouris et al., 1994) promoted its further development. The entire mechanism of its neuroprotective action has not yet been fully delineated but is due, at least in part, to multiple effects on the NMDAR-glutamate system. First, riluzole inhibits Na⁺ channels on glutamate-containing neurons and thereby selectively reduces presynaptic release of glutamate (Prakriya and Mennerick, 2000). Second, riluzole might block NMDAR activation, preventing Ca²⁺ entry via the channel. Riluzole either acts directly on the NMDAR, although a binding site for riluzole on the receptor has not been identified (Debono et al., 1993), or indirectly, possibly via a G-protein dependent signalling pathway (Hubert et al., 1994). Third, riluzole facilitates glutamate reuptake by increasing the activity of glutamate transporters expressed on neurons and glia (Fumagalli et al., 2008).

Finally, two new NMDAR-targeting drugs, neramexane and dimebon, are in clinical trials. Neramexane belongs to a recently described group of NMDAR open-channel blockers known as the amino-alkyl-cyclohexanes (Danysz et al., 2002). The drug has similar kinetics and voltage-dependency to memantine, and comparable clinical tolerability. Although there are no clinical trials of neramexane registered, this drug is being developed for treatment of Alzheimer’s disease (Rammes and Schierloh, 2006). Dimebon, a drug predominantly developed in Russia, is being assessed in clinical trials in patients with Alzheimer’s (Doody et al., 2008) and Huntington’s diseases. A small preliminary clinical trial done in Moscow suggested some improvement in cognitive function and reduction in neuropsychiatric symptoms with dimebon in patients with mild to moderate Alzheimer’s disease (Bachurin et al., 2001). Dimebon was initially classified as an antihistamine but its mechanism of action seems to be more complex (Lermontova et al., 2001; Bachurin et al., 2003). Studies suggest dimebon blocks NMDARs but likely at a site distinct from memantine (Grigorev et al., 2003).
3.1.2.3. **NMDA Receptor Negative Allosteric Modulators**

One of the potentially most important recent developments in NMDA receptor pharmacology has been the identification of highly subtype selective antagonists which act allosterically through an interaction with the extracellular ATD of the NR2 subunits (Perin-Dureau et al., 2002; Malherbe et al., 2003). Most of these compounds are selective for NR2B subunit containing receptors and, so far, only zinc has been identified as being NR2A subunit selective through an interaction with this domain. Zinc displays a voltage-dependent inhibition of NMDAR responses in heteromeric NR1/NR2A and NR1/NR2B receptors. At lower concentrations, it shows a voltage-independent inhibition of NR1/NR2A receptors (Chen et al., 1997). Since zinc is co-released with glutamate from pre-synaptic terminals, zinc modulation of NMDARs may be physiologically relevant (Aniksztejn et al., 1987). A large number of pharmacological agents bind and inhibit NMDAR activity specifically at NR2B containing receptors. The prototype is ifenprodil, a phenylethanolamine that binds at a site distinct from the glutamate- and glycine-binding sites (Legendre and Westbrook, 1991). Ifenprodil exhibits greater than a 100-fold selectivity for NR2B over NR2A containing receptors (Williams, 1993), and very low affinity at NR2C- and NR2D-containing receptors (Williams, 1995).

In addition to their activity at NMDA receptors, ifenprodil and its analogue eliprodil are also antagonists of α1-adrenergic receptors, serotonin receptors and calcium channels, suggesting that the therapeutic effectiveness of these compounds might be compromised by accompanying cardiovascular interactions (Gogas, 2006). There have been several ‘second generation’ ifenprodil analogues developed, which readily penetrate the brain and are active systemically including traxoprodil (Chenard et al., 1995), and (R-(R*, S*)-α-(4-hydroxyphenyl)-β-methyl-4-(phenylmethyl)-1-piperidine propanol (Ro 25-6981) (Fischer et al., 1997). Both compounds exhibit greater selectivity for NR2B over other receptor subtypes and ion channels, suggesting a reduced probability of cardiovascular side effects (MCCAuely et al., 2004). They have been useful for defining the actions of NR2B-containing receptors in the brain. Troxoprodil possesses high selectivity and has been through phase III clinical trials for the treatment of traumatic brain injury. However, the clinical utility of these compounds for chronic indications has been compromised by their poor oral bioavailability and pharmacokinetic profiles, mainly due to high first pass liver metabolism and high clearance rates (Gogas, 2006).

3.1.3. **NMDA Receptor Positive Allosteric Modulators**

The polyamines, spermine and spermidine, were the first potentiators of NMDA receptor function to be described (Ransom and Stec, 1988). They appear to enhance NMDA receptor function through two mechanisms, one glycine-dependent, mediated by enhancing receptor affinity for glycine, and one glycine-independent characterized by increases in the maximal amplitudes of NMDAR responses, which persists in the presence of saturating concentrations of glycine, and is only present at NR2B subunit containing receptors (Williams, 1997). In the absence of glutamate and glycine, polyamines have no effect on NMDAR activity. However, they increase glycine affinity (Sacaan and Johnson, 1989; McGurk et al., 1990), and thus increase NMDAR responses at subsaturating glycine concentrations by increasing glycine association. Under saturating glycine conditions, polyamines still potentiate NMDAR responses (glycine-independent potentiation).

Moreover, at negative potentials, polyamines reduce channel conductance by partial channel block. Consistent with early studies (Ransom and Stec, 1988), these polyamine effects are noncompetitive with glutamate, glycine, and channel blockers, suggesting distinct binding sites for polyamines (McBain and Mayer, 1994). Spermine, spermidine and other compounds active at the polyamine site are all highly positively charged and, thus, it appears difficult to target this site with systemically active drugs. Magnesium mimics all of the potentiating effects of the polyamines (Kew and Kemp, 1998) and may be an endogenous ligand for this site (Paoletti et al., 1995).
3.1.4. AMPA Receptor Agonists

The agonists glutamate, AMPA, and kainate, are widely employed for activating AMPARs, and they display modest selectivity among homomeric receptors (Keinanen et al., 1990; Kew and Kemp, 2005). Moreover, a large number of AMPA receptor agonists have been described and many of them, like AMPA itself, have been derived from classic structure activity studies using ibotenic acid, quisqualic acid and willardiine as leads (Stensbol et al., 2002). One of the interesting aspects of AMPA receptor agonists is that they can vary dramatically in the amount of receptor desensitization that they induce. Thus, whilst glutamate and AMPA act as full agonists and induce rapidly desensitizing responses, kainate acts as a partial agonist and induces AMPA receptor responses that show little desensitization. A number of 5-substituted willardiines act as partial agonists with varying levels of intrinsic activity at AMPA receptors whilst the potent AMPA analogue, (R,S)-(2-(amino(3-(3-carboxy(5-(methyl(4-isoxazolyl)propionic acid (ACPA) induces much less steady-state desensitization than AMPA itself (Stensbol et al., 2002; Mayer and Armstrong, 2004).

3.1.5. AMPA Receptor Competitive Antagonist

The first somewhat selective and useful AMPA receptor antagonists were 6-cyano-7-nitroquinoxaline-2,3-dione (CNQX) and 6,7-dinitroquinoxaline-2,3-dione (DNQX) (Drejer and Honore, 1988). However, these compounds also had activity at the glycine binding site on NMDA receptors and, thus, lacked sufficient selectivity to be really useful agents (Birch et al., 1988). Nevertheless, they served as the starting point for various more selective competitive AMPA receptor antagonists, such as 2,3-dihydroxy-6-nitro-7-sulfamoyl-benzo(F) quinoxaline (NBQX) (Sheardown et al., 1990); 1,4,7,8,9,10-hexahydro-9-methyl-6-nitropyrido[3,4-f]-quinoxaline-2,3-dione (PNQX) (Oomori et al., 1994), 2,3-dioxo-7-(1H-imidazol-1-yl)-6-nitro-1,2,3,4-tetrahydro-1-quinoxalinyl]aceticacid (YM90K) (Kohara et al., 1998); and [1,2,3,4-tetrahydro-7-morpholinyl-2,3-dioxo-6-(trifluoromethyl) quinoxalin-1-yl]methylphosphonate (ZK200775) (Turskiet al., 1998).

Most of these quinoxaline derivatives have limited water solubility and, as a consequence, representatives have failed clinical trials due to nephrotoxicity (Turskiet al., 1998). However, ZK200775 and YM872 are water soluble, AMPA receptor selective and active in in-vivo models of stroke (Turski et al., 1998; Takahashi et al. 2002). Both these compounds entered clinical trials for the treatment of acute ischemic stroke: the trial with ZK200775 was stopped prematurely due to safety concerns (Elting et al., 2002) and the trial with YM872 was completed but no further information is available.

3.1.6. Non-Competitive AMPA Receptor Antagonists

Several chemical series of non-competitive AMPA receptor antagonists that are also known as negative allosteric modulators have been reported. These ligands are comprised of several distinct chemical classes, including the 2,3-benzodiazepines, quinazolinones, aryolphthalazines, and tetrahydrosioquinolines. The latter two series are structural derivatives of 2,3-benzodiazepines (Pelletier et al., 1996; Gitto et al., 2003). Following the identification of 1-(4-aminophenyl)-4-methyl-7,8-methylenedioxy-5H-2,3-benzodiazepine (GYKI 52466) as a selective, non-competitive AMPA receptor antagonist (Solyom and Tarnawa, 2002), a family of 2,3-benzodiazepine compounds was elaborated. The 2,3-benzodiazepines include(R)-1-(4-aminophenyl)-4-methyl-7,8-methylenedioxy-4,5-dihydro-3methylcarbomyl-2,3-benzodiazepine (GYKI 53784/LY303070) and (R)-7-acetyl-5-(4-aminophenyl)-8,9-dihydro-8-methyl-7H-1,3-dioxolo(4,5-h)(2,3)benzodiazepine (GYKI53773/LY300164) (Tarnawa et al., 1993; Ruel et al., 2002). These compounds exhibit relatively little selectivity between AMPA receptor subtypes but are selective for AMPA over kainate and NMDA receptors (Ruel et al., 2002). The 2,3-benzodiazepines are systemically bioavailable (Szabados et al., 2001) and indeed GYKI 53773/LY300164 has entered clinical trials for indications including epilepsy (Langan et al., 2003).

A series of quinazolinone derivatives has also been identified as selective non-competitive AMPA
receptor antagonists, typified by 3-(2-Chloro-phenyl)-2-[2-(6-diethylaminomethyl-pyridin-2-yl)-vinyl]-6-fluoro-3H quinazolin-4-one (CP-465,022) and 2-[2-[3-(2-Chloro-phenyl)-6-fluoro-4-oxo-3,4-dihydro-quinazolin-2-yl]-vinyl]-nicotinonitrile (CP-526,427) (Welch et al. 2001; Lazzaro et al. 2002). The binding site of CP-526,427 is distinct from the glutamate binding site but appears to overlap with that of the 2,3-benzodiazepines (Menniti et al. 2000). Like the 2,3-benzodiazepines, CP-465,022 is systemically bioavailable (Menniti et al. 2003) and is selective for AMPA over kainite and NMDA receptors whilst exhibiting little selectivity between AMPA receptor subtypes (Lazzaro et al. 2002).

Initial studies evaluating the effects of naturally occurring polyamine toxins across multiple AMPAR subtypes demonstrated that these compounds generally are selective for a subset of AMPARs—namely, channels lacking the GluA2 subunit (i.e., GluA1, GluA3, GluA4). For example, the natural product Joro spider toxin (JSTX-3) blocks nearly all of the current from GluA1, GluA3 and GluA4 homomers, while having no effect on the heteromeric receptors tested (i.e., GluA1/A2 and GluA2/A3) (Blaschke et al., 1993). Similarly, argiotoxin-636 (ArgTx-636) inhibits GluA1, GluA3, and GluA4 homomers with nearly equal potency, and is more than 10-fold less potent at GluA1/A2 heteromers (Herlitze et al., 1993).

3.1.7. AMPA Receptor Positive Allosteric Modulators
A number of AMPA receptor positive allosteric modulators have been identified. These compounds potentiate AMPAR-mediated currents by attenuating receptor desensitization and/or deactivation, but do not activate the receptor when applied alone. Such compounds have been shown to facilitate AMPA receptor-mediated synaptic activity both in-vitro and in-vivo and have exhibited activity in a variety of behavioral assays, most notably in models of cognition and have been pursued for their potential as cognition enhancing or ‘nootropic’ drugs. Indeed representatives have entered clinical trials which have yielded encouraging results (Danysz, 2002; O’Neill et al., 2004a).

Several distinct chemical classes of these compounds have been reported, including the benzamides (e.g., aniracetam, and CX-614), the benzothiadiazines (e.g., cyclothiazide), and a broad collection of arylsulfonamides (e.g., LY-404187 and PEPA) (Kew and Kemp, 2005; Morrow et al., 2006). The observation that cognitive enhancing ‘nootropic agents’ such as 1-(4-methoxybenzoyl)-2-pyrrolidinone (aniracetam) facilitated AMPA receptor-mediated activity (Ito et al., 1990) led to the elucidation of a structurally related series of benzamide compounds, also known as ‘AMPAkines=ampakines’, including 1-(quinazolin-6-ylcarbonyl)piperidine (CX516), 1-(4-benzothiadiazine1,1-dioxide (CX546) and 2H,3H,6aH-pyrrolidino[2″,1″(3′2′]1,3-oxazine[6′5′,4]benzo[e]1,4-dioxan-1-one (CX614) (Danysz, 2002; Lynch, 2004; O’Neill et al., 2004a).

A second series of modulators were identified following the observation that the benzothiadiazides, 7-Chloro-3-methyl-2H-1,2,4-benzothiadiazine 1,1-dioxide (diazoxide) and 6-Chloro-3,4-dihydro-3-(5-norbornen-2-yl)-2H-1,2,4-benzothiadiazidine sulfonamide-1,1-dioxide (cyclothiazide) also enhanced AMPA receptor-mediated currents (Yamada and Tang, 1993). Subsequent studies have resulted in the identification of the related 7-chloro-3-methyl-3,4-dihydro-2H-1,2,4-benzothiadiazine 1,1-dioxide (IDRA-21), and ((4-[2-(phenylsulphonylamino)ethylthio]-2,6-difluorophenoxyacetamide (PEPA) (Sekiguchi et al., 1997; O’Neill et al., 2004a).

More recently, a series of biarylpropysulphonamides has been reported including (R,S)-N-2(4(3thienyl)phenyl)propyl-2-propane sulfonamide (LY392098), N-2-[4-(4-cyanophenyl)phenyl]propyl2-propanesulphonamide (LY404187), N-2-(4-(N-benzamido) phenyl) propyl-2-propanesulphonamide (LY395153) and (R)-4′-[1-fluoro-1-methyl-2-(propane-2-sulphonlamino)-ethyl]-biphenyl-4-carboxylic acid methylamide (LY503430) (Miuetal., 2001; Murray et al., 2003; O’Neill et al. 2004a).
The positive modulators exhibit distinct mechanistic profiles and selectivity for individual AMPA receptor subunits and their splice variants. For example, PEPA attenuates receptor desensitization without effect on deactivation, cyclothiazide inhibits receptor desensitization but has little effect on receptor deactivation, whereas CX614 both blocks desensitization and slows deactivation (Sekiguchi et al., 2002). LY404187 also suppresses receptor desensitization with a distinct time-dependence in the presence of agonist that may reflect receptor ‘desensitization’ (Quirk and Nisenbaum, 2002).

3.1.8. Kainate Receptor Agonists
Historically, the study of kainate receptor physiology has been hampered by the lack of selective ligands. In addition to glutamate, both recombinant and native kainate receptors are activated by agonists including kainate and domoate. However, both compounds also elicit non-desensitizing responses at AMPA receptors and exhibit only moderate selectivity for kainate receptors (Lerma et al., 2001). Several kainate receptor selective agonists have now been identified (Lerma et al., 2001; Bleakman et al., 2002). This includes 5-tert-butyl-4-isoxazole propionic acid (ATPA), (S)-5-iodowillardiine, (2S,4R) 4-methyl glutamic acid (SYM2081) and (2S,4R,6E)-2-amino-4-carboxy-7-(2-naphthyl)hept-6-enolic acid (LY339434). ATPA, a substituted analogue of AMPA and (S)-5-iodowillardiine are potent, selective GluR5 agonists that display low affinity at AMPAR and GluR6 or GluR7-containing receptors (Swanson et al., 1998; Alt et al., 2004). AMPA, ATPA and (S)-5-iodowillardiine can also activate GluR6/KA2 heteromeric receptors, despite functioning only as partial agonists (Swanson et al., 1998; Alt et al., 2004).

The gamma substituted glutamate analogues SYM2081 and LY339434 are also more selective for kainate than for AMPA receptors (Small et al., 1998; Alt et al., 2004). However, SYM2081 inhibits currents through kainate receptors by a process of fast agonist-induced desensitization (Zhou et al., 1997) and thus essentially performs as an antagonist of these receptors. The naturally occurring marine toxins, dysiherbaine and its natural analogue neodysiherbaine, also act as potent kainate-receptor agonists (Swanson et al., 2002; Sanders et al., 2005).

3.1.9. Kainate-Receptor Antagonists
A small number of compounds have been described as selective antagonists of kainate receptors, they mainly target GluR5. Compounds of the quinoxalinedione family, including CNQX and NBQX, act as competitive antagonists at native and recombinant kainate receptors; whereas CNQX exhibits little selectivity for kainate over AMPA receptors, NBQX is far more potent at AMPA receptors (Mayer et al., 2006). Some pyrrolilquinazolinedione derivatives have been found to have a higher affinity for kainate than for AMPA receptors. This is particularly true for LU97175, which displays an extremely high selectivity for GluR5 and GluR6 and, especially, for GluR7 (Loscher et al., 1999).

Compounds of a new series of 6-substituted decahydroisoquinolines display more selectivity towards kainate receptors and act as potent antagonists at certain subunits. This is the case for LY382884, a compound derived from the non-competitive AMPA receptor antagonist LY293558, which displays selectivity towards the GluR5 subunit (Alt et al., 2004), and for LY377770 (O’Neill et al., 1998). The willardiine derivative UBP296 has been reported as the most potent and selective antagonist at GluR5-containing kainate receptors, with activity residing in the S enantiomer, UBP302 (More et al., 2004).

More recently, another series of willardiine derivatives has been synthesized and tested for antagonist activity. The N3-2-carboxybenzyl substituted analogue (UBP310) has been found to be a potent and selective antagonist of GluR5-containing kainite receptors when tested on native rat and human recombinant AMPA and kainate-receptor subtypes (Dolman et al., 2005). Interestingly, MSVIII-19, a synthetic analogue of the natural agonist dysiherbaine, also acts as a potent antagonist of homomeric GluR5 (Sanders et al., 2005). In addition, kainate-receptor function can also be antagonized by lanthanides, particularly lanthanum and gadolinium (Huettner et al., 1998), with a
significantly higher potency than for AMPA receptors.

3.1.10. Allosteric Modulators

Plant lectins, such as concanavalin A, succinylconcanavalin A, soybean agglutinin and wheat germ agglutinin, have been proposed to block the process of desensitization of kainate receptors, being more effective on homomeric assemblies (Wong and Mayer, 1993; Yue et al., 1995). On GluR6 homomeric receptors, concanavalin A exerts its action via non-specific binding to any of the N-glycosylation sites, even those artificially introduced (Everts et al., 1999), and have been suggested to “lock” the receptor in the activatable state, inhibiting the conformational changes required to shift the receptor to the desensitized state (Wong and Mayer, 1993; Yue et al., 1995). An alternative mechanism has been proposed. Neurons also express endogenous lectins but whether they are involved in regulating kainite receptor function is not yet known.

3.2. Metabotropic Glutamate Receptors

3.2.1. Group I mGluR Agonist

The most potent group I mGluR agonist identified to date is quisqualic acid, which exhibits potent activity at rat recombinant mGluR1 and 5 and is selective over the group II and III receptors (Bräuner-Osborne and Krosggaard-Larsen, 1998). However, its utility is severely limited by its agonist activity at AMPA receptors (Watkins et al., 1990). The first selective group I mGluR agonist to be identified was (S)-3,5-dihydroxyphenylglycine [(S)-DHPG] (Schoepp et al., 1994) which exhibits low micromolar potency at mGluR1 and 5 and is selective vs the group II and III mGluRs (Schoepp et al., 1999). No agonists selective for mGluR1 over mGluR5 have been reported to date. However, (R,S)-2- chloro-5-hydroxyphenylglycine (CHPG) was identified as a low-potency agonist at rat recombinant mGluR5 and is selective vs mGluR1, whilst its activity at the group II and III mGluRs has not been reported (Doherty et al., 1997).

3.2.2. Group I mGluR Antagonist

A number of phenylglycine derivatives have been identified as group I mGluR antagonists, some of which exhibit selectivity for mGluR1 vs mGluR5 (Schoepp et al., 1999). Many of the early compounds also exhibited activity at mGluR2, with the best example, (S)-4-carboxyphenylglycine [(S)-4CPG] exhibiting approximately tenfold selectivity for mGluR1 vs mGluR2 (Thomsen et al., 1994). Subsequent optimization of this template resulted in the identification of more selective ligands with low micromolar activity including (R,S)-1- aminoindan-1-5-dicarboxylic acid (AIDA) (Pellicciani et al., 1995; Moroni et al., 1997) and (S)-2-methyl-4-carboxyphenylglycine (LY367385) (Clark et al., 1997), both of which are selective for mGluR1 vs mGluR5 (Kingston et al., 2002). Generally, the pharmacokinetic profile of both competitive agonist and antagonists has restricted their utility as in vivo tools with CNS delivery only possible via intracerebroventricular administration.

3.2.3. Group I mGluR Negative Allosteric Modulators

High-affinity, non-competitive modulators have been identified which are selective between mGluR1 and mGluR5. The first non-competitive mGluR ligand to be identified was 7-hydroxyiminocyclopropan[b]chromen-1a-carboxylic acid ethyl ester (CPCCOEt) (Annoura et al., 1996). CPCCOEt inhibited mGluR1 activity, and without effect on glutamate binding and is selective for mGluR1 vs mGluR5 and the group II and III mGluRs (Hermans et al., 1998; Litchig et al., 1999). Subsequently, a number of higher affinity, selective mGluR1 negative modulators have been identified including [(3aS,6aS)(6a(Naphtalen(2(ylmethyl(5(methyliden(hexahydro(cyclopental [c] furan-1-on) (BAY36-7620) (Carroll et al., 2001); 1-(3,4-dihydro-2H-pyran[2,3-b]quinolin-7-yl)-2-phenyl-1-ethanone (R214127) (Lavreysen et al., 2003); and 1-ethyl-2-methyl-6-oxo-4-1,2,4,5-tetrahydro-benzo [d] azepin-3-yl)-1,6-dihydro-pyrimidine-5-carbonitrile (EMTBPC) (Malherbe et al., 2003).

The first non-competitive mGluR5 antagonists to be described were 6-methyl-2-(phenylazo)-3-pyridinol (SIB-1757) and (E)-2-methyl-6-(2-phenylethenyl)pyridine (SIB- 1893) (Varney et al., 1999). Both compounds are selective for mGluR5 vs mGluR1 and the family II and III mGluRs Structural
optimization around SIB-1757 and SIB-1893 led to the identification of 2-methyl-6-(phenylethynyl)-pyridine (MPEP), a significantly more potent ligand (Gasparini et al., 1999) which exhibits inverse agonism (Pagano et al., 2000). Further elaboration has led to the discovery of 3-[2-methyl-1,3-thiazol-4-yl) ethynyl] pyridine (MTEP), which exhibits improved selectivity and CNS bioavailability (Cosford et al., 2003).

3.2.4. Group I mGluR Positive Allosteric Modulators

Two chemical series have been reported as positive allosteric modulators for mGluR1: 2-phenyl-1-benzensulfonyl-pyrrolidine derivatives, exemplified by (S)-2-(4-fluoro-phenyl)-1-(toluene-4-sulfonyl)pyrrolidine (Ro 67-7476); diphenylacetyl- and(9H-xanthene-9-carbonyl)-carbamic acid esters, exemplified by diphenylacetyl-carbamic acid ethyl ester (Ro 01-6128) and (9H-xanthene-9-carbonyl)-carbamic acid butyl ester (Ro 67-4853) (Knoflach et al., 2001). In both rat recombinant and native receptor assay systems, these compounds did not exhibit intrinsic agonist activity but markedly facilitated agonist-induced responses, increasing potency and maximum efficacy. Notably, both Ro 67-7476 and Ro 01-6128 also increased the binding affinity of the agonist ligand [3H]quisqualate, without a significant effect on the number of binding sites. All these compounds were mGluR1 selective vs group II and III mGluRs with Ro 67-7476 and Ro 01-6128 also selective vs mGluR5, whilst positive modulator activity at mGluR5 was noted with Ro 67-4853 at high concentrations (Wichmann et al., 2002).

The first mGluR5 positive allosteric modulators to be described derive from two structural classes, the first represented by 3,3′-difluorobenzaldazine (DFB) (O’Brien et al., 2003) and the second by N-{4-chloro-2-[(1,3-dioxo-1,3-dihydro-2H-isoinol-2-yl)methyl] phenyl}-2-hydroxybenzamide (CPPHA) (O’Brien et al., 2004). Both DFB and CPPHA have no intrinsic agonist activity but potentiate agonist responses at both human and rat recombinant as well as rat native mGluR5, increasing agonist potency with no apparent increase in maximum response and without affecting the binding affinity of [3H]quisqualate (O’Brien et al., 2003, 2004). Interestingly, both DFB and CPPHA appear to increase the intrinsic efficacy of partial agonists at the orthosteric binding site (O’Brien et al., 2003, 2004). Both compounds were devoid of activity at mGluR1 and the group II and III mGluRs but exhibited weak antagonist activity at mGluR4 and mGluR8.

3.2.5 Group II mGluR Agonist

A number of potent group II mGluR agonists have been identified with selectivity vs the group I; however, they do not discriminate between mGluR2 and 3. These include 2′3′-dicarboxycyclopropylglycine (DCG-IV) (Brabet et al., 1998) and a series of highly potent compounds including (1S,2S,5R,6S)-2-amino-6-fluorobicyclo[3.1.0]hexane-2,6-di-carboxylic acid (LY354740) (Monn et al., 1997), (−)-2-oxa-4-aminobicyclo[3.1.0]hexane-4,6-dicarboxylic acid (LY379268) (Monn et al., 1999) and (1R,2S,5S,6S)-2-amino-6-fluro-4 oxobicyclo[3.1.0]hexane 2,6-dicarboxylic acid (MGS0028) (Nakazato et al., 2000). Notably, LY354740, LY379268 and MGS0028 exhibit nanomolar potency at mGluR2 and -3 and are systemically active (Cartmell et al., 1999; Nakazato et al., 2000). The only agonist reported to date which discriminates between mGluR2 and mGluR3 is the endogenous neuropeptide N-acetyl aspartylglutamate (NAAG), which is a relatively low potency mGluR3 agonist with moderate selectivity over mGluR2 and the other mGluR family members (Schweitzer et al., 2000).

3.2.6 Group II mGluR Antagonist

2S-2-amino-2-(1S,2S-2-carboxycycloprop-1-yl)-3-(xanth-9-yl) propanoic acid (LY341495) is the most potent group II mGluR antagonist identified, which exhibits nanomolar affinity at both mGluR2 and -3 (Kingston et al., 1998). LY341495 is systemically bioavailable and has been used to block the activity of group II agonists in vivo (Cartmell et al., 1999). Several other selective competitive ligands have been identified (Schoepp et al., 1999) including (2S,4S)-2-amino-4(2,2-diphenylethyl) pentanedioic acid which exhibits low micromolar activity at mGluR2 and -3 and greater than tenfold selectivity.
vs the group I and III mGluRs and ionotropic glutamate receptors (Escribano et al., 1998).

### 3.2.7 Group II mGluR Negative Allosteric Modulators

Three 8-arylethynyl-1,3-dihydrobenzo[b][1,4]diazepin-2-one derivatives, 3-(4-oxo-7-phenylethynyl-4,5-dihydro-3H-benzo[b][1,4]diazepin-2-yl)benzonitrile (Ro 67-6221), 8-(4-fluoro-phenylethynyl)-7-hydroxy-4-(3-imidazol-1-yl-phenyl)-1,3-dihydro-benzo[b][1,4]diazepin-2-one (Ro 71-8216) and 8-(4-fluoro-phenylethynyl)-7-hydroxy-4-(3-[1,2,3]triazol-1-yl-phenyl)-1,3-dihydro-benzo[b][1,4]diazepin-2-one (Ro 71-8218) were described that exhibited high affinity, non-competitive antagonism at rat mGluR2 with selectivity vs group I and III mGluRs. Preliminary observations also revealed the antagonist activity of Ro 67-6221 and Ro 71-8218 at mGluR3. Interestingly, whilst these ligands exhibited a noncompetitive pharmacological profile, they partially displaced binding of the orthosteric agonist [3H](LY354740 at mGluR2, suggesting an allosteric interaction between the two binding sites (Kew and Kemp, 2005).

### 3.2.8 Group II mGluR Positive Allosteric Modulators

A series of pyridylmethyl sulfonamide mGluR2 positive allosteric modulators typified by N-(4-(2-methoxyphenoxy)phenyl)-N-(2,2,2-trifluoroethylsulfonyl)prid-3-ylmethylamine (LY487379) has been identified via functional screening (Johnson et al., 2003). LY487379 was without intrinsic activity but potentiated submaximal glutamate responses, increasing agonist potency and maximum efficacy, and mGluR2 was selective over mGluR3, the group I, and III mGluRs (Johnson et al., 2003; Schaffhauser et al., 2003). LY487389 also increased the binding affinity of the orthosteric agonist ligand, [3H]DCG-IV, but not the antagonist, [3H]LY341495, at mGluR2, this was suggested to be due to an increase in receptor/G-protein coupling in the presence of the positive modulator (Schaffhauser et al., 2003). To date, mGluR2-selective positive allosteric modulators appear to exhibit an in vivo profile similar to that of the mixed mGluR2/3 agonists, suggesting that agonist efficacy is predominantly mediated via mGluR2. Recently, a novel class of mGluR2-selective positive modulators typified by 1-(2-hydroxy-3-propyl-4-{4-[4-(2H-tetrazol-5-yl)phenoxy]-phenyl} ethanone was reported (Pinkerton et al., 2004). This compound attenuated ketamine-induced noradrenaline release in rat ventral hippocampus and also inhibited ketamine-induced hyperactivity. However, due to limited brain penetration, its utility was restricted to delivery via intracerebroventricular administration.

### 3.2.9 Group III mGluR Agonist

The discovery of selective ligands for group III mGluRs has trailed behind that of the group I and II receptors. (S)-4-Phosphono-2-aminobutyric acid (S-AP4) exhibits low micromolar agonist activity at mGluR4, -6 and 8 and is selective vs the group I and II mGluRs and vs mGluR7 at which it exhibits high micromolar potency (Schoepf et al., 1999). In fact, it is notable that mGluR7 is differentiated from other group III mGluRs by its relatively low sensitivity to both glutamate and synthetic agonists (Ahmadian et al., 1997). A number of group III mGluR subtype selective ligands have been identified. (S)-2-Amino-4-(3-hydroxy-5-methylisoxazol-4-yl) butyric acid (S-homo-AMPA) is a selective agonist at mGluR6 (Ahmadian et al., 1997). (R,S)-4-Phosphonophenylglycine (PPG) is a potent group III mGluR agonist which exhibits greater than tenfold selectivity for mGluR8 vs mGluR4, 6 and 7 (Gasparini et al., 1999). (S)-3,4-Dicarboxyphenylglycine (DCPG) is a potent and selective mGluR8 agonist (Thomas et al., 2001).

### 3.2.10 Group III mGluR Antagonist

A number of competitive group III mGluR antagonists have been identified as (R,S)-α-cyclopropyl-4-phosphonophenylglycine (CPPG) and (R,S)-α-methylserine-O-phosphate (MSOP) (Schoepf et al., 1999). However, pharmacological characterization of these compounds against the mGluR family members is incomplete. In addition, the group II selective antagonist, LY341495, also exhibits activity at the group III mGluRs (Kingston et al., 1998). LY341495 also exhibits low micromolar antagonist activity at the group I mGluRs.
3.2.11. Group III mGluR Allosteric Modulators

No non-competitive group III mGluR antagonists (negative allosteric modulators) have been reported to date. The mGluR5 positive allosteric modulators, DFB and CPPHA, both exhibit weak antagonist activity at both mGluR4 and mGluR8 (O’Brien et al., 2003, 2004). (−)-N-Phenyl-7-(hydroximino)cyclopropa[b]chromen-1a-carboxamide ((−)-PHCCC) is a close structural analogue of the mGluR1 antagonist, CPCCOEt, and is itself a weak antagonist at mGluR1 (Annoura et al., 1996), but acts with equivalent potency as a positive allosteric modulator of mGluR4 (Marino et al., 2003). (−)-PHCCC was devoid of intrinsic agonist activity but increased agonist potency and efficacy at mGluR4. The mGluR5 antagonists, SIB-1893 and MPEP, also exhibit mGluR4 positive allosteric modulator activity (Mathiesen et al., 2003). Similar to (−)-PHCCC, both compounds were devoid of intrinsic agonist activity but increased agonist potency and efficacy.

4. Clinical Application of Glutamate Receptors’ Agonists and Antagonists

4.1. Major Depressive Disorder

There is growing evidence that the glutamatergic system plays an important role in the neurobiology and treatment of major depressive disorder MDD (Sen and Sanacora, 2008). As there are a number of papers reporting alterations in glutamate levels of blood and cerebrospinal fluid (CSF) in patients with MDD (Mauri et al., 1998; Mitani et al., 2006). There is also a positive correlation between plasma glutamate levels and severity of depressive symptoms in patients with MDD (Mitani et al., 2006). Glutamate is also known to regulate neurogenesis, synaptogenesis, and neuron survival in the developing and adult mammalian brain (Mattson, 2008). Stress during childhood and adolescence has been implicated in the extent of depression at adulthood. Stressful events lead to the regression of synapses with the loss of synaptic spines and in some cases whole dendrites of pyramidal neurons in the prefrontal cortex, a process that leads to the malfunctioning of neural networks in the neocortex (Bennett, 2008). The possible mechanism of such regression involves the loss of synaptic spines associated with excessive activation of glutamate receptors by increased glutamate levels, under conditions of oxidative and metabolic stress (Mattson, 2008; Bennett, 2008).

The non-competitive NMDA receptor antagonist ketamine has been demonstrated to have antidepressant effects in animal models of depression. Where, Chaturvedi et al. (1999) has reported the antidepressant effects of dizocilpine and ketamine on shock-induced behavioral changes in mice. Furthermore, ketamine showed antidepressant effects in a forced-swim test (Yilmaz et al., 2002). Very recently, Zarate et al. (2006a) demonstrated the robust and rapid antidepressant effects of a single dose of ketamine (0.5 mg/kg, i.v. infusion for 40 min) in treatment-resistant major depression. It is assumed that directly targeting the NMDA receptor complex may bring about rapid and relatively sustained antidepressant effects (Zarate et al., 2006a). However, the clinical applicability of the non-competitive open-channel NMDA receptor antagonists such as PCP and ketamine is limited by their propensity to cause psychomimetic effects (Javitt and Zukin, 1991; Krystal et al., 2005).

Memantine is a derivative of amantadine, a low affinity, non-competitive, open-channel NMDA receptor blocker. However, unlike ketamine, memantine failed to improve depression (Zarate et al., 2006b). The precise reasons for this discrepancy are unclear. One possibility is that high-affinity, strong open-channel blockers with slow off-rates, such as ketamine, exert antidepressant effects whereas low-affinity, weak open channel blockers with fast off-rates, such as memantine, are ineffective (Tsai, 2007). However, Kollmar et al. (2008) reported the successful treatment of a patient with MDD using a two-step therapeutic regimen consisting of two infusions of ketamine followed by oral administration of memantine. Still, the antidepressant effects of memantine remain unproven.

The NMDA receptors containing the NR2B subunit are localized primarily in the forebrain including hippocampus, a region implicated in the pathophysiology of MDD (Campbell and Macqueen, 2004). Considering the acute psychomimetic side effects of ketamine, the selective antagonists of the NR2B subtype of NMDA
receptors appear to pose little risk. The NR2B antagonist Ro 25-6981 had antidepressant-like properties in the forced-swim test (Maeng et al., 2008). Moreover, a recent double-blind, randomized, placebo-controlled study demonstrated that the selective NR2B antagonist traxoprodil had significant antidepressant effects in patients with treatment-resistant MDD (Preskorn et al., 2008). Therefore, the selective NR2B antagonists may be a fruitful target for the development of a new antidepressant with more robust effects and a faster onset compared with those currently available.

Recent findings suggest the emerging role of AMPA receptors in the treatment and etiology of mood disorders (Mathew et al., 2008; Alt et al., 2006). Where, it was reported that ketamine might exert its rapid antidepressant-like effects by enhancing AMPA receptors relative to NMDA receptors throughout critical neuronal circuits (Maeng and Zarate, 2007; Maeng et al., 2008). This results raise the possibility that the combination of AMPA receptor potentiators with even lower doses of NMDA receptor antagonists might be useful in the treatment of major depression. Positive modulators of AMPA receptors do not activate AMPA receptors themselves but slow the rate of receptors desensitization and/or deactivation in the presence of antagonist. Several positive modulators (piracetam, aniracetam, cyclothiazide, CX516, CX614, LY392098, LY404187, LY451395, LY503430) of AMPA receptors have shown antidepressant-like effects in animal models of depression (Alt et al., 2006; O’Neill and Witkin, 2007).

Lavreysen et al. (2004) demonstrated that JNJ16259685; 3-4-Dihydro-2H-pyra[n[2,3-b]quinolin-7-yl-(cis-4-methoxy-cyclohexyl)-methanone is a highly potent, selective, and systemically active mGluR1 antagonist produced a significant reduction of offensive behaviors (e.g., threat and attack) without affecting immobility. This suggest that mGlu1 receptors are involved in aggression regulation (Navarro et al., 2008). EMQCMCM, (JNJ16567083): 3-ethyl-2-methyl-quinolin-6-yl-(4-methoxycyclohexyl)-methanenemethanesulfonate), is another potent and highly subtype-selective mGluR1 antagonist that has antidepressant-like effects in the rat forced-swim and the mouse tail-suspension tests (Belozertseva et al., 2007). Several studies have demonstrated that the selective mGluR5 antagonists, MPEP, and the more selective and metabolically stable analog MTEP, have antidepressant-like effects in animal models of depression. Administration of MPEP or MTEP decreased immobility in the tail-suspension test in mice (Belozertseva et al., 2007; Palucha et al., 2005).

Group II mGlu receptors (mGluR2/3) exhibit moderate to high expression in brain regions (e.g., hippocampus, prefrontal cortex, and amygdala) that are associated with MDD. The mGluR2/3 antagonists, as LY341495 and MGS0039, exhibit antidepressant-like effects in behavioral animal models of depression (Sanacora et al., 2008; Pilc et al., 2008). Administration of MGS0039 for 14 days increased cell proliferation in the adult mouse hippocampus (Yoshimizu and Chaki, 2004), which is implicated in the pathophysiology of MDD as well as in the underlying mechanisms of antidepressants (Sahay and Hen, 2007). However, till now no clinical studies have been conducted to explore the antidepressant efficacy of mGluR2/3 antagonists in patients with MDD (Sanacora et al., 2008; Pilc et al., 2008).

On the other hand, selective group III mGlu receptor agonist ACPT-1 showed antidepressant-like effects in a forced-swim test (Palucha et al., 2004). The effects of ACPT-1 were antagonized by the group III mGluR antagonist CPPG (Kłak et al., 2007). Furthermore, administration of mGlu8 receptor agonist (R,S)-4-phosphonophenylglycine ((R,S)-PPG) showed dose dependent antidepressant-like effects in the forced-swim test in rats (Palucha et al., 2004). Moreover, Palucha et al. (2007) reported that the first selective and bio-available mGluR7 agonist N,N’-dibenzylhydril-ethane-1,2-diaminedihydrochloride (AMN082) showed antidepressant-like effects in the forced-swim test and in the tail-suspension test. These results
suggested that group III mGlu receptor agonists might show antidepressant-like effects.

4.2. Schizophrenia

Schizophrenia is characterized by positive (hallucinations and delusions), negative (anhedonia and poverty of speech), cognitive, mood and motor symptoms (Tandon et al., 2009). In addition to the well-established “dopamine hypothesis of schizophrenia”, abnormalities in glutamate transmissions have been suggested to be involved in the pathophysiology of schizophrenia. The glutamate hypothesis stemmed from the following findings: 1) significantly lower levels of glutamate are found in the cerebrospinal fluid (CSF) and postmortem brain tissue of schizophrenic patients (Kim et al., 1980; Tsai et al., 1995); 2) CSF glutamate levels are inversely correlated with the severity of positive symptoms in unmedicated patients (Faustman et al., 1999); and 3) phencyclidine (PCP) and ketamine, two non-competitive NMDA receptor antagonists, produced transient psychosis and disrupted affect and cognitive impairment in healthy volunteers, similar to the symptoms of schizophrenia (Javitt and Zukin, 1991; Lahti et al., 2001), and also led to the profound exacerbation of preexisting symptoms in schizophrenic patients (Lahti et al., 2001) and indicated the NMDAR hypofunction hypothesis in schizophrenia. This NMDAR hypofunction leads to excessive release of glutamate, leading to overstimulation of glutamatergic neurotransmission at AMPA/kainate receptors (Moghaddam et al., 1997). Thus, glutamatergic dysfunction, particularly the hypofunction of NMDA receptors may have an important role in the pathophysiology of schizophrenia.

Based on this NMDA receptor hypofunction hypothesis, compounds that enhance NMDA receptor function could alleviate symptoms of this disorder. In this case, the NMDAR D-serine/glycine site has been proposed as a potential therapeutic target, as increasing its activation offers a safer alternative to elevations in glutamate levels that can promote neurotoxicity (Coyle, 2006). To date, clinical trials have been conducted with the partial agonist D-cycloserine, the full agonists glycine and D-serine, and the glycine transporter-1 inhibitor sarcosine. D-serine and sarcosine, both of which increase NMDA receptor activities, have reportedly improved cognitive impairment in rodent models and in schizophrenic patients (Karasawa et al., 2008; Coyle and Tsai, 2004). Though administration of direct agonists, such as D-serine and glycine, are of therapeutic value, such treatments require large doses and exhibit difficulties in penetrating the blood–brain-barrier (Labrie and Roder, 2010). Consequently, agents targeting proteins involved in D-serine metabolism may be a more effective strategy. Inhibition of D-amino acid oxidase (DAO), the D-serine catabolic enzyme, activity in the brain is of particular interest as it would circumvent any nephrotoxicity associated with the catabolism of high levels of systemic D-serine (Maekawa et al., 2005b). Development of DAO antagonists has recently begun (Adage et al., 2008; Hashimoto et al., 2009). Intravenous injections of the DAO inhibitor AS057278 were found to readily cross the blood–brain-barrier and enhance D-serine contents in the rat brain (Adage et al., 2008). Chronic administration of AS057278 in mice was shown to normalize PCP-induced deficits in behavioral tasks relevant to the cognitive and positive symptoms of schizophrenia (Adage et al., 2008). However, elevations in D-serine following DAO inhibition are considerably lower than with exogenous D-serine treatments (Ferraris et al., 2008). Indeed, coadministration of D-serine and a DAO antagonist were shown to produce greater ameliorative effects in animals treated with an NMDAR antagonist than either compound applied alone (Hashimoto et al., 2009). Thus, DAO inhibition in combination with D-serine administration may be a valuable therapeutic approach for the treatment of schizophrenia.

Although the capacity of D-cycloserine to improve negative symptoms of schizophrenia has been replicated in some placebo-controlled studies (Heresco-Levy et al., 2002; Goff et al., 1999b), evidence supporting the effectiveness of D-cycloserine in treatment of schizophrenia is weak according to several other clinical studies. (Tuominen et al., 2005, 2006; Buchanan et al., 2007).
Several lines of evidence suggest that mGlu1 receptor positive allosteric modulators may have potential for the treatment of positive and cognitive symptoms of schizophrenia. This is because; first, compounds that activate mGlu1 receptors are documented to facilitate NMDA and AMPA receptor responses. As mGluR1 are expressed in regions that play a crucial role in schizophrenia: the cortex, hippocampus and basal ganglia (Simonyi et al., 2005), and it facilitate NMDA receptor-induced currents in cortical neurons (Lindemeyer et al., 2006), the CA3 region of the hippocampus (Benquet et al., 2002), and in the thalamus (Salt and Binns, 2000), as revealed in electrophysiological recordings of synaptic plasticity (Anwyl, 2009). Second, mGluR1 levels are altered in brains of schizophrenic patients. Where, mGlu1 receptor expression is increased in the prefrontal cortex of schizophrenic patients, which has been interpreted as a compensatory change to the NMDA receptor hypofunction (Gupta et al., 2005). And third, mGlu1 receptors can modulate dopamine release. Although few studies have investigated whether mGlu1 receptors can modulate dopamine release, but glutamate spillover from the corticostriatal synapse could not only activates glutamate receptors on medium spiny neurons, but may also activate presynaptic mGlu1 receptors on neighboring dopaminergic substantia nigra afferents, to inhibit electrically-evoked dopamine release (Zhang and Suzler, 2003). These may all add to the rationale in support of mGlu1 receptor positive allosteric modulators as potential therapy for the treatment of schizophrenia. However, for our knowledge, there is no mGlu1 receptor selective agonist published, and the effect of the mGlu1 positive allosteric modulators in animal models of schizophrenia has not been reported (Sheffler and Conn, 2008).

In contrast, several reports have been describing the effects of mGlu1 receptor antagonists in animal models that pick up antipsychotic-like activity. Different mGluR1 antagonist as ; FTIDC , 4-[1-(2-Fluoropyridine-3-yl)-5-methyl-1H-1,2,3-triazol-4-yl]-N-isopropyl-N-methyl-3,6 dihydropridine-1(2H)-carboxamide; and CFMTI, 2-cyclopropyl-5-[1-(2-fluoro-3-pyridinyl)-5- methyl-1H-1,2,3-triazol-4-yl]-2,3-dihydro-1H-isooindol-1-one (Satow et al., 2008, 2009) were tested for their antipsychotic effect in the most commonly used animal models for schizophrenia that involve the measurement of hyperactivity and increased stereotypic behaviors in response to amphetamine, MK-801, ketamine or phencyclidine. However, the data with mGluR1 antagonists were not very consistent.

Antagonists of mGluR5 can be also used for treatment of schizophrenia. Activation of presynaptic mGluR5 facilitates synaptic glutamate release whereas postsynaptic mGluR5 increase neuronal excitability by facilitating NMDA currents. Consequently, a reduction of the hyperglutamatergic PFC state could be achieved with drugs like acamprosate which attenuate the excitatory effect of mGluR5 on presynaptic glutamate release and by decreasing NMDA-dependent postsynaptic excitability. Acamprosate, interfere with mGluR5-dependent regulation of glutamate release (De Witte et al., 2005). Additionally, preclinical studies suggest that acamprosate normalizes glutamate release and NMDA receptors function without altering the normal glutamatergic neurotransmission (De Witte et al., 2005). On the other hand, mGluR5 regulates non-synaptic release of glutamate from glia and astrocytes via stimulation of the cystine–glutamate antiporter (Melendez et al., 2005). Accordingly, acamprosate would prevent activation of the this antiporter by antagonizing mGluR5, which in turn would reduce the non-synaptic extracellular glutamate.

In addition, clinical reports documented that activation of mGluR2/3 receptors provides symptomatic improvements in schizophrenic patients without major adverse effects (Patil et al., 2007). Recent results of a phase 2 clinical trial examining LY2140023, an mGlu2/3 receptor agonist, showed improvements in both positive and negative symptoms of schizophrenia. Importantly, patients takingLY2140023 did not show the typical side effects associated with medications targeting dopamine receptors, such as extrapyramidal syndrome or elevated serum prolactin levels; furthermore, unlike olanzapine, which has negative metabolic side effects such as weight gain, LY2140023 did not increase body weight rather, it
resulted in a loss of body weight (Patil et al., 2007). Therefore, mGlu2/3 receptor agonists might be superior to the antipsychotics currently used for the treatment of schizophrenia.

4.3. Epilepsy

Epileptic syndromes have very diverse primary causes, which may be genetic, developmental or acquired (McNamara, 1994). During an epileptic seizure, large populations of neurons in selected portions of the central nervous system abandon their normal activity and begin to fire in periodic synchronous discharges. This pathological synchronized activity is transmitted from one neuron to the next primarily through excitatory glutamatergic transmission (Doherty and Dingledine, 2002). Regardless of the primary cause, synaptically released glutamate acting on ionotropic and metabotropic receptors appears to play a major role in the initiation and spread of seizure activity (Chapman, 2000; Meldrum, 1994).

Epilepsy was one of the early targets for NMDA receptor antagonists (Czuczwar and Meldrum, 1982). Despite being effective in some animal models, those NMDA receptor antagonists that have been tested in man have not been proven effective. The first NMDAR channel blocker to be tested in epileptic patients was (+)MK-801 (dizocilpine) which showed no efficacy at doses producing appearance of side-effect (Leppik et al., 1988). On the one hand, side effects have seriously restricted the clinical trials of classical NMDA antagonists and, some doubts have been cast on the use of global seizure models induced by chemicals and sound as predictors of activity in partial seizures, the greatest population of epilepsy patients (Dansysz and Parsons, 1998). Kindled animals have been suggested as a better model (Lisscher and Schmidt, 1993). ADC1 (5-aminocarbonyl-lo,11-dihydro-5h-dibenzo[a,d] cyclohepten-5,10-imine) is a low affinity uncompetitive NMDA receptor antagonist showing anticonvulsive activity in various animal models and a favorable side effect profile, however it was discontinued in Phase I of clinical trials (Rogawski et al., 1995).

Glycine site antagonists could have some advantages over NMDA channel blockers such as lower potential for inducing psychotomimetic side effects. However, they have not yet been tested in clinical trials for epilepsy most likely due to numerous pharmacokinetic problems such as low solubility potentially resulting in kidney damage due to crystallization (e.g. ACEA 1021), low penetration to the CNS, short half-life, and last but not least by doubtful efficacy in kindled seizure models (Wlaz and L’oscher, 1998). Paradoxically, the best pre-clinical data for modulating the glycine site have come from D-cycloserine, a partial agonist (Lisscher et al., 1994).

Felbamate is one of NR2B selective antagonists that has been under development for epilepsy (Harty and Rogawski, 2000). It has favorable profile in animal models including kindling (Wlaz and L’oscher, 1997). However, felbamate had to be subsequently withdrawn due to the occurrence of aplastic anaemia cases connected with the drug use (Pennell et al., 1995). Conantokin G which also preferentially interacts with NR2B subunit is still under development for epilepsy, as it is in phase I clinical trials (Donevan and McCabe, 2000; White et al., 2000).

AMPA/kainate receptor antagonists had been or are still under development for treatment of epilepsy. However, competitive AMPA antagonists show several obstacles such as low solubility in neutral pH, short half-life and weak anticonvulsive effects. In fact most of these substances are either mixed antagonists of AMPA and other receptors such as kainate (NS-1209) or glycine (LU-73068) or act through noncompetitive, modulatory sites, e.g. talampanel. (Lisscher and Schmidt, 2006). On the other hand, the fundamental role of kainate in synaptic physiology, in particular in the balance between excitation and inhibition in many brain regions suggests that they may participate selectively in some forms of epilepsy in humans (Vincent and Mulle, 2009). In view of their role in the regulation of hippocampal networks, kainite receptors antagonists may represent a promising anticonvulsant. There is some growing evidence that GluR5 containing KARs represent a novel target for antiepileptic drugs development. Topiramate is a new antiepileptic drugs that has been beneficial for
the treatment of refractory epilepsy. Although there is clear evidence that topiramate affects the function of GluR5-containing receptors, its mode of action needs to be better understood. On the other hand, there is also the intriguing possibility that in fact GluR5 agonists, by increasing the inhibitory tone, might paradoxically be protective, under conditions that remain to be clarified (Khalilov et al., 2002).

It has been reported that the mGluR1 antagonist LY456236 and mGluR5 antagonist MPEP are effective as antiepileptic in mice (Barton et al., 2003; Shannon et al., 2005). Another mGluR1 antagonist, BAY36-7620 has been shown to inhibit PTZ seizures but failed to provide protection against kindled seizures (Chapman et al., 2000; Spooren et al., 2003). However, it should be pointed out that the use of either mGluR1 and mGluR5 antagonists may be connected with possible learning impairing effects (Steckler et al., 2005).

Several reports indicate potential anticonvulsant utility of metabotropic group II agonists. The orthosteric agonist LY354740 has been shown to attenuate clonic convulsions produced by PTZ or picrotoxin, but not by NMDA (Klodzinska et al., 2000). Moreover, the GluR2/3 agonists LY379268 and LY389795 inhibited sound-induced clonic seizures in mice (Moldrich et al., 2001). However, these agonists did not inhibit sound induced seizures in genetically epilepsy-prone rats, but were even proconvulsant following the sound stimulus (Moldrich et al., 2001). Little is known about the role of group III metabotropic receptors due to the very limited number of selective ligands. In principle, anticonvulsant effects could be expected from either agonists or positive modulators. ACPT-I (preferential mGluR4 agonist) inhibited sound convulsions in mice and rats (Chapman et al., 2001) which indicate that mGluR4 may be a candidate for anticonvulsant therapy.

4.4. Alzheimer’s disease
Alzheimer’s disease (AD) is an irreversible neurodegenerative disease causing dementia severe enough to hamper daily living. Both neuropathological and neurochemical studies of the AD brain have shown that degeneration of glutamate mediated pathways occurs early in the disease and in a pattern that corresponds with the distribution of plaques and tangles (Procter, 2000).

Pyramidal neuron loss is a feature of AD (Morrison and Hof, 1997), and glutamatergic pyramidal neurons account for many of the neurons lost in the cerebral cortex and hippocampus in AD (Morrison and Hof, 1997). Similarly, a large decrease in glutamate receptors has been observed in the hippocampus (Greenamyre et al., 1987; Procter et al., 1989) and cortex (Greenamyre et al., 1985) of the brains of AD patients presumably due in part to the accompanying neuronal loss described above. Subsequent reports of subunit-specific abnormalities in the expression and pharmacological properties of NMDA receptors (Hynd et al., 2004; Procter et al., 1989) and mGluR (Albasanz et al., 2005) suggests these changes do not simply reflect generalized cortical atrophy, but may indicate disease-specific processes. Abnormalities in AMPA receptor expression have also been observed in the AD (Wakabayashi et al., 1999), while KA receptors seem to be unaffected (Cowburn et al., 1989).

In addition to the consequences of glutamatergic cell loss there is evidence for dysfunction in remaining neurons. Where, the ability of glial cells to remove glutamate from the synaptic cleft was impaired in several brain regions (Procter et al., 1988). Similarly there was a reduction in the vesicular glutamate transporter in parietal but not temporal cortex (Kirvell et al., 2006) and the activity of this protein was lower in AD temporal cortex than in controls (Westphalen et al., 2003). As a consequence of these alterations, it has been proposed (Francis, 2003) that in AD there is inadequate removal of glutamate in the synaptic cleft between the pre and postsynaptic neuron, creating an excessive level of background “noise” at glutamate receptors within the synapse and adversely affecting the ability of the NMDA receptor to generate LTP (Danysz et al., 2000). This disruption of glutamatergic neurotransmission may contribute to cognitive impairment in AD (Francis, 2003). Furthermore, the pathologic glutamate acting at all classes of glutamate receptors can cause neuronal...
death or at least can be a contributory factor in AD. (Danysz et al., 2000).

Several approaches to correct glutamatergic dysfunction in AD have been attempted, including positive modulation of both AMPA and NMDA receptors. AMPAKines, which are considered to work by increasing the sensitivity of these receptors, were in clinical trial for mild cognitive impairment (Johnson and Simmon, 2002), but no drugs have yet come to the clinic. Modulation of the NMDA receptor has been also attempted via the glycine co-agonist site with clear indication in preclinical studies that the partial agonist D-cycloserine improved learning and memory (Myhrer and Paulsen, 1997). Clinical studies have suggested some benefit but full-scale trials have not been initiated (Schwartz et al., 1996).

Since NMDARs are the primary mediators of glutamate mediated excitotoxicity, one line of therapeutical research has focused on the development of NMDA antagonists for treatment of AD. One would normally consider that such an approach, blockade of a receptor that would normally be activated in learning and memory, would be counter-intuitive. However, the most surprising development is the success of the non-competitive NMDA antagonist memantine (1-aminoo-3,5-dimethyladamantane) in clinical trials in moderate and severe AD (Parsons et al., 2007). Memantine could decrease pathological activation of NMDA receptors without affecting physiological NMDA receptor activity (Scarpini et al., 2003). As this molecule acts like magnesium ions, able to prevent background activation of the NMDA receptor (“noise”), whilst allowing activation of this receptor for LTP formation (Francis, 2003; Parsons et al., 2007). Most importantly, clinical trials have shown that memantine treatment leads to substantial increases in cognitive function relative to placebo in AD patients with severe dementia (Winblad and Poritis, 1999; Kirby et al., 2006), and it is well tolerated (Winblad and Poritis, 1999). On the basis of successful clinical trials, the use of memantine in the modulation of glutamatergic function may therefore represent a novel approach for the treatment of AD.

4.5. Parkinson’s disease

Parkinson’s disease (PD) is a common neurodegenerative disorder characterized by severe motor impairments such as bradykinesia, tremor and rigidity. These motor symptoms arise as a consequence of a progressive loss of dopaminergic neurons in the substantia nigra, which project into the striatum regulating activity through the basal ganglia (Hassler, 1938).

Glutamatergic pathways to the striatum and basal ganglia output nuclei become overactive in Parkinson’s disease owing to the depletion of nigrostriatal dopamine. Glutaminergic neurons themselves do not appear to degenerate in Parkinson’s disease, however NMDA receptors have been implicated in mediating excitotoxicity in the basal ganglia, a process that has been linked to dopaminergic neuronal cell death in PD. There is some evidence that blocking glutamate input to the striatum with NMDA receptor antagonists reverses akinesia (Uyama et al., 1991), improves parkinsonian motor symptoms and improves dyskinesia (Blanchett et al., 1996).

Amantadine, is an NMDA receptor antagonist, that is accepted for treatment of Parkinson’s disease. Originally being found to be remarkably effective in treating the motor symptoms of the disorder in a patient taking it as a flu prevention agent. A number of reports followed which described the benefit of the drug in terms of symptomatic improvement, time of response to the medication, and the coprescribing of L-DOPA (Schwab and England, 1995; Factor et al., 1998). Where improvement in levodopa-induced dyskinesias were reported (Blanchet et al., 1998), but no studies specifically examined cognition. Side effects of the drug include ankle edema, insomnia, nausea, restlessness and in some cases psychosis. There have also been some reports of worsening of cognition and withdrawal delirium with the use of amantadine in PD (Cash et al., 1987).

Pharmacological studies using NR2B receptor antagonists in several animal models further support a role for this subtype in Parkinson’s disease (Nash et al., 2004). Ifenprodil showed efficacy in a
reserpine-treated rat model of Parkinson’s disease (Nash et al., 1999). However, a small clinical trial in which ifenprodil was used as an add-on therapy with L-DOPA in Parkinson’s disease patients failed to demonstrate significant therapeutic benefit (Montastruc et al., 1992). Traxoprodil has been shown to reduce rigidity and akinesia in both rodent and non-human primate Parkinson’s disease models (Steece-Collier et al., 2000). These preclinical data, taken together with results from clinical studies of NMDA antagonists, suggest that NR2B-selective antagonists hold therapeutic utility as antiparkinsonian agents.

Targeting mGlu receptors is another interesting possibility for treatment of PD. Recent evidence suggests that profound alterations in depotentiation, a process that probably utilizes mGlu receptors could underlie dyskinesias (Picconi et al., 2003). The mGlu4 orthosteric agonist LSP1-2111 has demonstrated efficacy in rodent models of PD, specifically reversal of haloperidol-induced catalepsy and 6- hydroxydopamine-induced motor deficits (Beurrier et al., 2009). This has been linked to the inhibition of striato-pallidal GABAergic transmission through mGlu4 activation (Cuomo et al., 2009). However, positive allosteric modulators have several advantages over orthosteric agonists. N-phenyl-7-(hydroxylimino) cyclopropa[c]- chromen-1a-carboxamide (PHCCC) was the first selective mGlu4 positive allosteric modulator with high selective potentiation on mGluR4, and with no direct agonist activity and no potentiator activity at any other mGluR receptor subtype (Marino et al., 2003). This compound was originally identified as an allosteric antagonist of mGluR1. PHCCC increases the potency of glutamate in recombinant systems as well as at several synapses including the striato-pallidal synapse (Marino et al., 2003; Valenti et al., 2005). Moreover, PHCCC decreases reserpine-induced akinesia (Marino et al., 2003). Clearly, there is much promise for the development of mGlu4 positive allosteric modulator as novel therapeutics for PD.

Another prime target in treating PD is the AMPA receptor, which mediates most fast excitatory synaptic transmission in the brain. AMPA receptor antagonists, such as GYKI-47261 and the non-competitive inhibitor talampanel, have entered into clinical trials as potential neuroprotectants in PD. Conversely, several strategies have been used to potentiate AMPA receptor function, with a view to providing cognitive enhancement and neurotrophic effects (O’Neill et al., 2004b).

4.6. CNS injury
Primary traumatic brain injury is a direct result of the initial insult, that is complete at the time of presentation, and cannot be altered. Secondary traumatic brain injury is an alteration of brain tissue, either anatomic or physiologic, which occurs subsequent to the primary injury (Leonard and Kirby, 2002). Secondary injury may include edema and elevations of intracranial pressure (Chen and Swanson, 2003). Traumatic brain injury has been shown to cause marked elevation in glutamate concentrations, which is responsible through excitotoxicity for neuronal injury or death, making secondary injury as potentially lethal as primary injury (Leonard and Kirby, 2002). Recently, growing evidence has suggested that apoptosis significantly contributes to central neuronal death following head trauma. NMDA receptor activation as well as AMPA receptor activation have been implicated in the initiation of apoptosis following trauma (Bullock et al., 1998; Wada et al., 1999). Sustained intracranial pressure elevations and poor patient outcome have been significantly associated with high levels of CNS glutamate in humans (Bullock et al., 1998). It was proposed that failure of the high affinity uptake system of glutamate is the cause for its high concentrations following trauma to the nervous tissue (Bullock et al., 1998). Essentially, transporter-mediated glutamate homeostasis fails dramatically in ischaemia: instead of removing extracellular glutamate to protect neurons, transporters release glutamate, triggering neuronal death (Rossi et al., 2000). Several studies using tissue cultures or intact animals provided evidence that the accumulation of synaptically released glutamate was responsible for anoxic cell death, and that the removal of glutamatergic excitatory input reduced ischaemic damage.

Blockade of NMDA receptors attenuates ischaemic damage in the brain (Simon et al., 1984).
Studies showing that high levels of glutamate are maintained for many hours during prolonged focal ischemia and that NMDA receptor antagonists are protective when administered either before or during focal ischaemic insult support this hypothesis (Parsons et al., 1998). For example, the non-competitive NMDA open channel blocker memantine was recently shown to be neuroprotective without adverse side-effects in animal models of CNS ischaemia (Rao et al., 2001). Additionally, NMDA receptor antagonist MK-801, was effective in ameliorating the effects of CNS trauma (Yum and Faden, 1990). Based on these very positive, if in hindsight misleading (Gladstone et al., 2002), preclinical data, where NMDA antagonists proved to be very effective neuroprotective drugs in vitro and in animal models of stroke, traumatic brain injury and spinal-cord injury, clinical trials of NMDA antagonists were initiated. However, sequentially the clinical trials were terminated based on efficacy and side effects (Kemp and McKernan, 2002; Muir, 2006). Failure may be due to deficient pharmacokinetics with the plasma levels achieved in studies consistently below those needed for maximal neuroprotection in animal models (Kemp and McKernan, 2002). Moreover, NMDA antagonists have a number of mechanism-based adverse CNS effects, including hallucinations, a centrally mediated increase in blood pressure and, at high doses, catatonia which have limited the doses used clinically (Kemp and McKernan, 2002). Together, it seems unlikely that nonselective channel blockers will ever pass the safety hurdles to receive approval for widespread use (Kemp and McKernan, 2002; Muir, 2006).

However, several NR2B receptor selective antagonists have been evaluated in animal models of ischaemic brain injury, and it was proven to be effective. Several studies using rat, cat and mouse middle cerebral artery occlusion models show that ifenprodil reduces infarct volume in a dose-related manner in the absence of hypothermia (Wang and Shuaib, 2005). However, ifenprodil and its analogue eliprodil are also antagonists of α1-adrenergic receptors, serotonin receptors and calcium channels, suggesting that the therapeutic effectiveness of these compounds might be compromised by accompanying cardiovascular interactions. Traxoprodil and Ro 25-6981 exhibit greater selectivity for NR2B over other receptor subtypes and ion channels, suggesting a reduced probability of cardiovascular side effects (Chenard and Menniti, 1999; Mutel et al., 1998). Clinical trial data for traxoprodil show that it is well tolerated, with none of the side effects typically seen with non-selective NMDA antagonists (Saltarelli et al., 2004). However, the efficacy results for traxoprodil in the acute ischemic cortical stroke trial were somewhat disappointing (Saltarelli et al., 2004).

Owing to the signal transduction coupling and synaptic localization of mGluR subtypes, antagonizing the postsynaptic excitatory group I mGluRs should have a protective effect whereas activation of the presynaptic inhibitory group II or III mGluRs should be required. Indeed, this concept has been demonstrated with several orthosteric group I mGluR antagonists. After spinal cord contusion injury, injections of the mGluR group I antagonist AIDA into the lesion site improved early locomotor recovery (Mills et al., 2002a). mGluR1 antagonists have also proved protective in traumatic brain injury models. AIDA administration after lateral fluid percussion in rats significantly improved neuroscores (Faden et al., 2001), as well as reducing overall neuronal cell death (Lyeth et al., 2001). Furthermore, the mGluR1 antagonist YM-202074 is neuroprotective after cerebral ischemia (Kohara et al., 2008).

Additionally, mGluR5 agonists have shown antiapoptotic properties in neuronal cultures (Vincent et al., 1999; Allen et al., 2000). Although a number of reports indicate that treatment with the specific mGluR5 agonist CHPG is neuroprotective in vitro and in vivo (Lea et al., 2002; Deng et al., 2004), treatment with the mGluR5 antagonist MPEP may also provide neuroprotection. The fact that the group I receptors mGluR1 and mGluR5 have similar signaling pathways and intracellular effects led to the theory that inhibition of mGluR5, similar to mGluR1, would be neuroprotective. Indeed, administration of MPEP significantly reduced neuronal death after glutamate or NMDA exposure (O’Leary et al., 2000). This apparent confusion...
regarding mGluR5 agonists and antagonists was resolved by work of Lea et al., 2005 who definitively showed that neuroprotective actions of the mGluR5 antagonists MPEP or MTEP do not reflect actions at the mGluR5 receptor. Instead, MPEP acts to directly inhibit NMDA receptor signaling, and application of the antagonists in cultures lacking the mGluR5 receptors (mGluR5 knockouts) yields the same neuroprotective effects as in cultures from wild-type animals.

Moreover, group II and III mGluRs provide neuroprotection against models of CNS damage, such as spinal cord injury, traumatic brain injury, or excitotoxic injections (Mills et al., 2002b). As activation of group II and III receptors reduces glutamate release and GABAergic transmission in neurons (Pinheiro and Mulle, 2008). Thus potentially reducing excitotoxic cell death. The group II agonist LY379268 reduced neuronal loss in the hippocampus after global ischemia and application of LY379268 up to 2 h after occlusion was neuroprotective in a rat model of focal ischemia (Bond et al., 1999).

When considering allosteric modulators, the positive mGluR4 modulator, PHCCC has also shown neuroprotection in vitro (Maj et al., 2003) but it should be kept in mind that this compound also antagonize group I mGluRs.

4.7. Pain

Chronic pain is now thought to involve plastic changes resulting from sustained electrical activity. An accumulating body of evidence supports the notion that modulation of glutamate receptors may have potential for therapeutic utility in several categories of persistent pain, including neuropathic pain resulting from injury and/or disease of central (e.g., spinal cord injury) or peripheral nerves (e.g., diabetic neuropathy) and inflammatory or joint-related pain (e.g., rheumatoid arthritis, osteoarthritis) (Dougherty and Willis, 1991).

Evidence in support of a therapeutic potential for AMPA receptor antagonists in chronic pain states comes from studies implicating a role for AMPA receptors in nociceptive signaling, as well as the actions of selective compounds in animal models of persistent pain. For example, the development, of spinal sensitization and secondary hyperalgesia in first degree burn models can be prevented by pretreatment with intrathecal AMPA/kainate receptor antagonists NBQX or CNQX (Pogatzki et al., 2000; Nozaki-Taguchi and Yaksh, 2002). Agents that selectively block calcium permeable AMPA/kainate receptors, such as philathotoxins or joro spider toxin, indicate also a role for this type of receptor in secondary mechanical allodynia resulting from thermal injury (Sorkin et al., 1999), and incisional pain (Pogatzki et al., 2003). The involvement of these may extend to neuropathic pain models such as the chronic constriction injury model, where intrathecal AMPA/kainite receptor antagonists appear to block nerve-induced thermal hyperalgesia and mechanical allodynia (Garry et al., 2003). All of these data suggest a potential role for this receptor in pathological pain signaling and the potential utility of AMPA receptor antagonists as therapeutic treatments for chronic pain conditions. A key challenge to this therapeutic approach will be dissociating the well-known side-effects of AMPA receptor antagonists, including ataxia and sedation, from their analgesic actions.

In addition to the animal data for the role of NMDA antagonist in pain some clinical observations with medicines that are known to block NMDA receptor activation are observed. Thus, low-dose ketamine reduced chronic pain associated with spinal cord injury (Eide et al., 1995) and could reduce pain in patients with peripheral nerve injury and with peripheral vascular disease (Felsby et al., 1996; Eisenberg and Pud, 1998). Amantadine was proven to significantly reduce pain in cancer patients and in those with trauma-induced neuropathic pain (Pud et al., 1998). On the other hand, NR2B subunit-selective compounds as, Ifenprodil, traxoprodil and Ro 25-6981 are effective in inflammatory and/or neuropathic pain models in animals at doses that are not accompanied by motor effects (Chizh and Headley, 2005). In addition, traxoprodil demonstrated efficacy in a randomized, double-blind, placebo-controlled crossover trial in patients with neuropathic pain (spinal cord injury) without producing side effects typically associated with non-selective NMDA antagonists (Sang et al., 2003). For that NR2B antagonists hold promise for therapeutic intervention in pain states, particularly given their
potential for efficacy with reduced liability of psychomotor effects.

In addition to NMDA receptor inhibitors, the prominence of functional kainate receptors and the GluR5 subunit in dorsal root ganglia suggests that this might also be a target for chronic pain. A selective GluR5 antagonist LY382884 that has poor affinity for AMPA receptors and for GluR6 showed analgesic activity in the rat formalin model (Palecek et al., 2004). Another decahydroisoquinoline, LY466195, has also been shown to be effective in these animal models of migraine (Weiss et al., 2006). Also antagonism of mGlu1 receptors may provide a useful therapeutic strategy for treatment of persistent inflammatory pain conditions. For example, local infusion of the selective group I mGlu receptor antagonists (AIDA or LY367385) into the spinal cord attenuates the development and maintenance of thermal hyperalgesia induced by kaolin/carrageenan injection into the knee joint (Zhang et al., 2002). In addition, intrathecal administration of selective mGlu5 receptor antagonists reduces mechanical hypersensitivity to subdermal hindpaw injection of capsaicin in rats (Soliman et al., 2005). Collectively, these data demonstrate that group I mGlu receptor antagonists can have analgesic effects in multiple models of inflammatory pain.

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