Mini-Symposium

Extrasynaptic $GABA_A$ Receptors: Form, Pharmacology, and Function

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GABA is the principal inhibitory neurotransmitter in the CNS and acts via $GABA_A$ and $GABA_B$ receptors. Recently, a novel form of $GABA_A$ receptor-mediated inhibition, termed "tonic" inhibition, has been described. Whereas synaptic $GABA_A$ receptors underlie classical "phasic" $GABA_A$ receptor-mediated inhibition (inhibitory postsynaptic currents), tonic $GABA_A$ receptor-mediated inhibition results from the activation of extrasynaptic receptors by low concentrations of ambient GABA. Extrasynaptic $GABA_A$ receptors are composed of receptor subunits that convey biophysical properties ideally suited to the generation of persistent inhibition and are pharmacologically and functionally distinct from their synaptic counterparts. This mini-symposium review highlights ongoing work examining the properties of recombinant and native extrasynaptic $GABA_A$ receptors and their preferential targeting by endogenous and clinically relevant agents. In addition, it emphasizes the important role of extrasynaptic $GABA_A$ receptors in GABAergic inhibition throughout the CNS and identifies them as a major player in both physiological and pathophysiological processes.

Introduction

It is only recently that two seemingly unrelated phenomena, the existence of a $GABA_A$ receptor ($GABA_AR$)-dependent "tone" in some neurons (Otis et al., 1991; Salin and Prince, 1996) and the presence of $GABA_A$Rs outside synaptic specializations (Somogyi et al., 1989; Soltesz et al., 1990), have been unified: GABA spillover from the synaptic cleft activates extrasynaptic or perisynaptic $GABA_A$Rs to generate a persistent or tonic inhibition (for review, see Semyanov et al., 2004; Farrant and Nusser, 2005; Glykys and Mody, 2007). Tonic inhibition is distinct from the transient activation of synaptic $GABA_A$Rs leading to classical inhibitory postsynaptic currents (phasic inhibition) and the slow, but still transient, response of the metabotropic $GABA_B$Rs. The initial finding in cerebellar granule cells (Brickley et al., 1996; Wall and Usowicz, 1997; Nusser et al., 1998; Brickley et al., 2001; Hamann et al., 2002) was followed by subsequent discoveries in, among others, the dentate gyrus and hippocampus (Bai et al., 2001; Nusser and Mody, 2002; Semyanov et al., 2003; Wei et al., 2003; Caraiscos et al., 2004a,b; Scimemi et al., 2005; Glykys et al., 2007), neocortex (Drasbek and Jensen, 2006; Yamada et al., 2007; Krook-Magnuson et al., 2008), thalamus (Belelli et al., 2005; Cope et al., 2005; Jia et al., 2005), striatum (Ade et al., 2008; Janssen et al., 2009), hypothalamus (Park et al., 2006, 2007), and spinal cord (Takahashi et al., 2006; Wang et al., 2008), and also in humans (Scimemi et al., 2006). The occurrence of tonic $GABA_A$ inhibition coincides with the expression of relatively rare receptor subunits, particularly the $\alpha 4$, $\alpha 6$, and δ subunits, and as a general rule-of-thumb, δ subunit-containing receptors are extrasynaptic, but not all extrasynaptic $GABA_A$Rs contain δ subunits. In comparison, the ubiquitous $\gamma 2$ subunit is a major component of synaptic $GABA_A$Rs and drives receptor clustering at the synapse (Essrich et al., 1998). The presence of the δ subunit in recombinant receptors conveys properties ideally suited to generating tonic inhibition, namely activation by low concentrations of GABA, such as may be found in the extracellular space and reduced desensitization (Saxena and Macdonald, 1994; Haas and Macdonald, 1999; Bianchi and Macdonald, 2002; Brown et al., 2002). The δ subunit can also govern receptor pharmacology, extrasynaptic $GABA_A$Rs typically being insensitive to benzodiazepine agonists (Nusser et al., 2002; Cope et al., 2005) but highly sensitive to the $GABA_AR$ "super agonist" 4,5,6,7-tetrahydroisoxazolo[4,5-c]pyridine-3-ol (THIP/gaboxadol) (Brown et al., 2002; Wohlfarth and Macdonald, 2002).

More recently, studies have begun to identify extrasynaptic $GABA_A$Rs as novel targets for a diverse array of endogenous and clinically relevant agents, including certain neuroactive steroids (Belelli et al., 2002; Wohlfarth et al., 2002; Stell et al., 2003; Cope et al., 2005) and the amino acid taurine (Jia et al., 2008a), as well as ethanol (Sundstrom-Poromaa et al., 2002; Wallner et al., 2003; Glykys et al., 2007; Jia et al., 2008b), several anesthetic and hyp-

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notic agents (Bai et al., 2001; Caraiscos et al., 2004b; Belelli et al., 2005; Cheng et al., 2006a; Takahashi et al., 2006; Jia et al., 2008c), analgesics (Krogsgaard-Larsen et al., 2004), and some anticonvulsant drugs (Cheng et al., 2006b). What is more, the functional role of tonic inhibition is beginning to be elucidated, such as the dynamic regulation of neuronal output, firing mode, and gain-control of neurotransmission (Hamann et al., 2002; Mitchell and Silver, 2003; Semyanov et al., 2003; Chadderton et al., 2004; Cope et al., 2005; Park et al., 2006; Bright et al., 2007; Rothman et al., 2009). Last, aberrant tonic inhibition has been implicated in multiple pathophysiological conditions, including fragile X mental retardation (Curia et al., 2009), γ-hydroxybutyric acid (GHB)-uria (Drasbek et al., 2008), stress (Maguire and Mody, 2007), disorders associated with the menstrual cycle and puberty (Maguire et al., 2005; Shen et al., 2007), and idiopathic generalized and temporal lobe epilepsies (Dibbens et al., 2004; Naylor et al., 2005; Scimemi et al., 2005; Feng et al., 2006; Zhang et al., 2007). Thus, extrasynaptic $GABA_A$Rs may be candidates for therapeutic treatment in a range of neurological disorders.

The mini-symposium described below, therefore, provided an overview of previous work on extrasynaptic $GABA_A$Rs and tonic inhibition and documented ongoing studies in this exciting field of research. Findings ranging from molecular studies to behavioral experiments were discussed. Individual presentations focused on the biophysical properties and structure-function relationships of putative extrasynaptic $GABA_A$Rs (Biophysical properties of recombinant δ subunit-containing $GABA_A$R channels), their selective targeting by endogenous and clinically important agents (Neuron-selective actions of neurosteroids at synaptic and extrasynaptic $GABA_A$Rs and Thalamic extrasynaptic $GABA_A$Rs are a target for ethanol and volatile anesthetics), and the contribution of tonic $GABA_A$ inhibition to both physiological and pathophysiological processes (Steroid hormones regulate extrasynaptic $GABA_A$Rs during the ovarian cycle, pregnancy, and associated disorders, Enhanced tonic $GABA_A$ inhibition in thalamic neurons is necessary and sufficient for absence seizures, and The contribution of tonic $GABA_A$ inhibition to physiological and pathological hippocampal excitability).

Biophysical properties of recombinant δ subunit-containing $GABA_A$R channels

Molecular biological and biochemical techniques have been instrumental in determining the basic properties of ligand-gated ion channels, and by inference, the functional properties of native receptors in the CNS. Recombinant receptors have, therefore, been used to distinguish the stoichiometry, electrophysiological, and pharmacological properties of $GABA_A$Rs. Macdonald and colleagues have examined the properties of recombinant $\alpha\beta\delta$ and $\alpha\beta\gamma$ receptors, i.e., putative extrasynaptic and synaptic receptors, respectively, and the possible roles of δ subunit variants in the pathophysiology of idiopathic generalized epilepsies.

$\alpha\beta\gamma$ receptors likely have a stoichiometry of $2\alpha:2\beta:1\gamma$ (Chang et al., 1996; Tretter et al., 1997), and while it is generally believed that the δ subunit can substitute for the γ subunit, the stoichiometry of $\alpha\beta\delta$ receptors remains uncertain (Kaur et al., 2009). The functional properties between the two receptor subtypes differ. Compared with $\alpha1\beta\gamma2$ receptors, $\alpha1\beta\delta$, $\alpha4\beta\delta$, and $\alpha6\beta\delta$ receptors have smaller macroscopic current amplitudes, increased outward rectification, slower desensitization, and absence of fast desensitization (Saxena and Macdonald, 1994; Fisher and Macdonald, 1997; Haas and Macdonald, 1999; Bianchi et al., 2002). Single-channel recordings indicate that $\alpha1\beta\delta$ receptors exhibit brief, isolated openings with two open states, whereas $\alpha4\beta\delta$ and $\alpha1\beta\gamma2$ receptors open to three states (Fisher and Macdonald, 1997; Feng et al., 2006). Furthermore, $\alpha1\beta\delta$ receptors have a lower GABA EC_{50} than $\alpha1\beta\gamma2$ receptors (Saxena and Macdonald, 1994; Fisher and Macdonald, 1997), and the presence of α4 or α6 subunit confers even higher GABA sensitivity (Saxena and Macdonald, 1996; Brown et al., 2002). The slower and less extensive desensitization, and high sensitivity to GABA, of $\alpha\beta\delta$ receptors, and α4 and α6 containing receptors in particular, makes these receptors ideal candidates to generate tonic $GABA_A$ inhibition.

Pharmacologically, $\alpha\beta\delta$ and $\alpha\beta\gamma$ receptors are distinct. Not only are $\alpha\beta\delta$ receptors benzodiazepine insensitive, they have increased sensitivity, compared with $\alpha\beta\gamma$ receptors, to allosteric modulators including zinc and lanthanum (Saxena et al., 1994, 1996), neurosteroids (see Neuron-selective actions of neurosteroids at synaptic and extrasynaptic $GABA_A$Rs), ethanol (see Thalamic extrasynaptic $GABA_A$Rs are a target for ethanol and volatile anesthetics), barbiturates (Saxena and Macdonald, 1996; Feng et al., 2004), certain anesthetics (see Thalamic extrasynaptic $GABA_A$Rs are a target for ethanol and volatile anesthetics), the nonbenzodiazepine anxiolytic tracazolate (Zheleznova et al., 2008), and protons (Feng and Macdonald, 2004). In addition, GABA exhibits high efficacy at $\alpha\beta\gamma$ receptors, whereas at $\alpha\beta\delta$ receptors it has a low efficacy, suggesting GABA is a partial agonist at $\alpha\beta\delta$ receptors (Bianchi and Macdonald, 2003). Changes in GABA efficacy of $\alpha\beta\delta$ receptors may be a general mechanism by which allosteric modulators bring about their actions.

A role for dysfunction of $\alpha\beta\delta$ receptors in the pathophysiology of idiopathic generalized epilepsies has been suggested. Two δ subunit variants (E177A and R220H) have been identified as susceptibility alleles for generalized epilepsy with febrile seizures plus and juvenile myoclonic epilepsy (Dibbens et al., 2004). In HEK293T cells, recombinant h$\alpha1\beta2\delta$(E177A) and h$\alpha1\beta2\delta$(R220H) receptors exhibited reduced receptor currents, although the GABA EC_{50} was no different from wild-type receptors (Dibbens et al., 2004). In recombinant h$\alpha4\beta2\delta$(E177A) and h$\alpha4\beta2\delta$(R220H) receptors, GABA EC_{50}s were also similar to wild-type receptors, but reduced macroscopic currents were caused by reduced single-channel currents attributable to shorter mean open durations and to loss of cell-surface receptor expression (Feng et al., 2006). Thus, disruption of $\alpha\beta\delta$ receptor function indicates a possible role for aberrant extrasynaptic $GABA_A$Rs in epileptogenesis (see Enhanced tonic $GABA_A$ inhibition in thalamic neurons is necessary and sufficient for absence seizures and The contribution of tonic $GABA_A$ inhibition to physiological and pathological hippocampal excitability).

Neuron-selective actions of neurosteroids at synaptic and extrasynaptic $GABA_A$Rs

Neurosteroids, typified by the progesterone metabolite allopregnanolone, potently modulate neuronal excitability through endocrine, paracrine, or autocrine actions at $GABA_A$Rs (Belelli and Lambert, 2005). Estimated brain and plasma levels of neurosteroids (10–300 nM) are dynamically regulated during certain (patho)physiological conditions, including development, later stages of pregnancy, and episodes of stress. Thus, neurosteroid modulation of $GABA_A$R function may play an important role in these conditions (see Steroid hormones regulate extrasynaptic $GABA_A$Rs during the ovarian cycle, pregnancy, and associated disorders). However, given the ubiquitous expression of $GABA_A$Rs, it might be predicted that neurosteroid actions would be widespread, causing a nonspecific enhancement of neuronal inhibition that would seem incompatible with a physiological

role. Belelli and colleagues have investigated neurosteroid actions and shown that they are neuron selective.

Neuronal selectivity may be the product of a range of molecular mechanisms, including subunit composition (Herd et al., 2007) so that different populations of receptors within a given neuron may exhibit different neurosteroid sensitivity. Recombinant $\alpha\beta\gamma$ receptors are sensitive to neurosteroids, but the identity of the α or β subunit isoform has little influence on receptor responses (Belelli et al., 2002). In contrast, in native neurons, synaptic $GABA_AR$ responses are highly heterogeneous (Cooper et al., 1999; Belelli and Herd, 2003; Harney et al., 2003). For instance, synaptic $GABA_A$Rs of thalamocortical (TC) neurons of the ventrobasal (VB) thalamus are sensitive to only high concentrations of allopregnanolone (Mitchell et al., 2007), whereas even low concentrations (10 and 100 nM) enhance the synaptic inhibition of CRF-releasing parvocellular neurons of the paraventricular nucleus of the hypothalamus and inhibit their output (Gunn et al., 2009). Compared with synaptic $GABA_A$Rs, δ subunit-containing extrasynaptic receptors have been proposed to be highly sensitive to low, physiologically relevant concentrations of neurosteroids, a suggestion supported both by experiments on recombinant receptors (Belelli et al., 2002; Wohlfarth et al., 2002) and the reduced behavioral sensitivity of δ subunit knock-out mice to endogenous and synthetic neuroactive steroids (Mihalek et al., 1999). Furthermore, some native δ subunit-containing receptors are indeed sensitive to low concentrations of allopregnanolone, for instance cerebellar granule cells (Stell et al., 2003). However, extrasynaptic $GABA_A$Rs in TC neurons are relatively insensitive to even high concentrations of allopregnanolone (Brown et al., 2009) or 5α-THDOC (5α-tetrahydrodeoxycorticosterone) (Porcello et al., 2003). Moreover, the modest 5α-THDOC-dependent effects seen in TC neurons are still present in δ subunit knock-out mice (Porcello et al., 2003). Additional mechanisms have been shown to contribute to the neuronal-selectivity of neurosteroid actions, including the phosphorylation state of native $GABA_A$Rs (Harney et al., 2003; Koksma et al., 2003) and local steroid metabolism (Belelli and Herd, 2003), although their precise roles remain to be elucidated.

Thus, both synaptic and extrasynaptic $GABA_A$Rs represent targets for the actions of neurosteroids. The imminent generation of transgenic mice harboring neurosteroid-insensitive receptor isoforms will greatly aid the exploration of the relative contribution of distinct synaptic and extrasynaptic $GABA_A$RS to the putative (patho)physiological roles of neurosteroids.

Thalamic extrasynaptic $GABA_A$Rs are a target for ethanol and volatile anesthetics

It has been well documented that TC neurons of the VB thalamus exhibit tonic $GABA_A$ inhibition (Belelli et al., 2005; Cope et al., 2005; Jia et al., 2005). Coimmunoprecipitation studies show that antibodies to the δ subunit precipitate the α4 subunit (Jia et al., 2005), and it has been estimated that as much as 30% of the total TC neuron $GABA_AR$ population may contain the α4 subunit (Sur et al., 1999). Furthermore, α4 and δ subunits colocalize with each other and are found predominantly at extrasynaptic sites (Jia et al., 2005), a feature confirmed by the loss of tonic inhibition in TC neurons of α4 subunit knock-out mice (Chandra et al., 2006). Because the thalamus plays a crucial role in sleep regulation (Steriade, 2000), and GABA levels have been shown to fluctuate over the sleep–wake cycle (Kékesi et al., 1997), agents that target extrasynaptic $GABA_A$Rs in the thalamus may play a significant role in governing sedation, hypnosis, and consciousness.

Harrison and colleagues showed that this is the case for two clinically important agents, ethanol and the volatile anesthetic isoflurane. In control mice, low concentrations of ethanol, such as may cause intoxication, elicited a sustained current in TC neurons of the VB, that was associated with a decrease in neuronal excitability and firing rate (Jia et al., 2008b). The steady current was completely abolished by the $GABA_AR$ antagonist gabazine, and ethanol had no effect on TC neurons from α4 subunit knock-out mice. In a similar vein, volatile anesthetics are used clinically to produce analgesia, amnesia, unconsciousness, blunted autonomic responsiveness, and immobility (Campagna et al., 2003), and, at lower doses, sedation (Dwyer et al., 1992). Harrison and colleagues demonstrated that even low concentrations of isoflurane, such as may be sufficient to cause sedation, elicited a sustained current in TC neurons of the VB, associated with a conductance increase (Jia et al., 2008c). The reversal potential of the isoflurane-evoked current was close to the Cl^- reversal potential, was blocked by gabazine, and, as for ethanol, there was no effect of isoflurane in α4 subunit knock-out mice, even at doses that produce unconsciousness (Jia et al., 2008c). Thus, in the thalamus, as elsewhere (Sundstrom-Poromaa et al., 2002; Wallner et al., 2003; Caraiscos et al., 2004b; Glykys et al., 2007), extrasynaptic $GABA_A$Rs appear to be a preferential target for both ethanol and certain volatile anesthetics.

Although the effects of ethanol and volatile anesthetics may be mediated by multiple mechanisms in the brain, the work by Harrison and colleagues, and in agreement with other studies, implicates extrasynaptic $GABA_A$Rs as a major player in their principal modes of action. Furthermore, the pivotal role played by the thalamus in controlling sleep–wake states suggest that modulation of extrasynaptic $GABA_A$Rs in TC neurons may contribute to global states of arousal and be a candidate target for the treatment of thalamocortical related disorders such as sleep disturbances (Belelli et al. 2005; Herd et al., 2009) and epilepsy (see Enhanced tonic $GABA_A$ inhibition in thalamic neurons is necessary and sufficient for absence seizures). For instance, disruption of normal sleep patterns by alcohol has been well documented and can be both economically important (Stoller, 1994) as well as a factor in alcoholic relapse.

Steroid hormones regulate extrasynaptic $GABA_A$Rs during the ovarian cycle, pregnancy, and associated disorders

Altered levels of neurosteroids in the CNS are associated with numerous psychiatric and neurological disorders, including premenstrual syndrome (PMS), premenstrual dysphoric disorder (PMDD), catamenial epilepsy, and postpartum depression. It has long been assumed that the pathophysiology of these disorders was attributable to an adverse reaction to changing steroid hormone levels. Recent evidence, however, indicates that exogenous administration of some neurosteroids alters the expression of certain $GABA_AR$ subunits, for example the α1, α4, and γ2 subunits (Smith et al., 1998; Follesa et al., 2000, 2002; Smith, 2002), whereas γ2 subunit expression is inversely correlated to neurosteroid levels (Follesa et al., 2004). In addition, certain neurosteroids are known to potentiate the effects of GABA at selective $GABA_A$Rs, particularly those containing the δ subunit (see Neuron-selective actions of neurosteroids at synaptic and extrasynaptic $GABA_A$Rs). Thus, the targeting of specific $GABA_AR$ populations by neurosteroids may underlie some of their actions. Jamie Maguire and colleagues have examined the contribution of extrasynaptic $GABA_A$Rs to the ovarian cycle and throughout pregnancy and the role of dysfunctional neurosteroid regulation of these receptors in associated pathophysiological disorders.

In the hippocampus, changes in $GABA_AR$ subunit expression occur over the estrous cycle, in particular a reciprocal increase in δ and decrease in γ2 subunit expression at periods of the cycle associated with elevated levels of the steroid hormone progesterone (Maguire et al., 2005; Maguire and Mody, 2007). In dentate gyrus granule cells, elevated progesterone and δ subunit levels correlates with an increase in tonic inhibition and a decrease in both anxiety levels and seizure susceptibility (Maguire et al., 2005; Maguire and Mody, 2007). Postpartum is also a particularly vulnerable period for mood disorders. In the hippocampus, $GABA_AR$ subunit expression changes during pregnancy, in particular a reduction in the expression of δ and γ2 subunits 18 d after conception, which rebounds to virgin levels by 48 h postpartum. The loss of δ and γ2 subunits is accompanied by a decrease in both tonic and phasic inhibition in the dentate gyrus (Maguire and Mody, 2008) and may represent a homeostatic compensatory mechanism to maintain excitability levels during pregnancy. In δ subunit knock-out mice, failure to regulate $GABA_ARs$ during pregnancy and postpartum is reflected in an abnormal phenotype including depression-like behavior, failure to build a nest, and increased pup mortality attributable to neglect and/or increased cannibalism (Maguire and Mody, 2008). Thus, not only do extrasynaptic $GABA_ARs$ represent a target for certain steroid hormones but the subsequent regulation of neuronal excitability may likely play an important role in the normal function of these hormones during the ovarian cycle, pregnancy, and postpartum. However, dysfunction in $GABA_AR$–steroid hormone interactions may underlie multiple associated neuropsychiatric disorders including PMS, PMDD, and postpartum depression.

Enhanced tonic $GABA_A$ inhibition in thalamic neurons is necessary and sufficient for absence seizures

Absence seizures are a feature of many idiopathic generalized epilepsies and are characterized by bilaterally synchronous spike-and-wave discharges (SWDs) generated in reciprocal cortico-thalamo–cortical networks (Crunelli and Leresche, 2002). Despite not being directly involved in seizure initiation, the thalamus is required for both the full electrographic and behavioral expression of seizures (Polack et al., 2007), and $GABA_ARs$ are clearly important. For instance, selective application of GABAmimetics to sensory thalamic nuclei can exacerbate or initiate seizures (Danober et al., 1998). However, only modest alterations in phasic $GABA_A$ inhibition have been documented in TC neurons (Bessaïh et al., 2006), and the role of tonic inhibition is unknown, despite systemic THIP administration having previously been described as a model of absence seizures (Fariello and Golden, 1987). Cope and colleagues investigated tonic inhibition in TC neurons of a prototypical sensory thalamic nucleus, the VB, from a polygenic model of SWDs, the genetic absence epilepsy rats from Strasbourg (GAERS), and showed that it was larger compared with nonepileptic controls. Importantly, this increase was seen before the onset of seizures and may therefore contribute to seizure genesis. Furthermore, tonic inhibition was also increased in other genetic models, the monogenic mutant mice stargazer (*stg*) and lethargic (*lh*), and after the application of the SWD-inducing agents GHB and THIP. In GAERS, *stg* and *lh* animals, enhanced tonic inhibition is caused not by overexpression of extrasynaptic $GABA_ARs$ but by compromised GABA uptake by the GABA transporter GAT-1, leading to an increase in ambient GABA concentration. The critical importance of thalamic GAT-1 in seizure genesis was highlighted by the presence of seizures in GAT-1 knock-out mice and their induction in normal Wistar rats after the intrathalamic microinjection of a selective GAT-1 blocker.

Cope and colleagues then examined whether enhanced tonic inhibition in thalamic neurons was important in seizure genesis, or just an interesting phenomenon. GHB failed to induce SWDs in δ subunit knock-out mice that show dramatically reduced, albeit not abolished, tonic inhibition compared with wild-type littermates (Herd et al., 2009). By comparison, GHB-induced seizures were readily apparent in the littermates. In addition, spontaneous seizures in GAERS were susceptible to the intrathalamic microinfusion of an antisense oligodeoxynucleotide (ODN) to the δ subunit (Maguire et al., 2005), whereas there was no affect after microinfusion of a missense ODN. Importantly, the antisense, but not the missense, ODN reduces tonic inhibition. Last, intrathalamic microinfusion of THIP in normal Wistar rats induces both SWDs and the behavioral correlates of seizures, i.e., the full electrographic and behavioral repertoire of absence seizures was replicated. Collectively, these findings demonstrate that enhanced tonic inhibition in TC neurons is common to multiple and diverse models of absence seizures and is both necessary and sufficient for the full, i.e., electrographic and behavioral, expression of absence seizures. Furthermore, extrasynaptic $GABA_ARs$ and GABA transporters in the thalamus may represent a novel therapeutic target for the treatment of absence epilepsy.

The contribution of tonic $GABA_A$ inhibition to physiological and pathological hippocampal excitability

Activation of extrasynaptic $GABA_ARs$ in the hippocampus can have a profound effect on neuronal excitability (Semyanov et al., 2003). Although neuronal gain (the slope of the relationship between excitatory input and firing rate) can be altered by subthreshold synaptic "noise" (Wolfart et al., 2007; Rothman et al., 2009), it has been suggested that tonic inhibition may also play a role in gain control (Mitchell and Silver, 2003; Rothman et al., 2009). In CA1 hippocampal neurons, Walker and colleagues have demonstrated that tonic $GABA_A$ inhibition exhibits a strong outward rectification (Pavlov et al., 2008) and thus has a greater modulatory effect on excitatory inputs at or close to threshold, compared with a much reduced effect on subthreshold noise. Furthermore, using a dynamic clamp system, tonic inhibition predominantly affects the offset (left–right position) of the relationship between input and firing while having only a minimal effect on gain. Thus, at least in the hippocampus, extrasynaptic $GABA_A$ receptors modulate network excitability without altering the sensitivity of neurons to changing inputs.

Because extracellular GABA can vary during different physiological and pathological conditions, tonic inhibition may be expected to change according to brain state. Temporal lobe epilepsy frequently results from a brain insult leading to the emergence of spontaneous seizures after alterations in cellular and network properties during the subsequent latent period. This can be mimicked in animal models by chemically or electrically inducing status epilepticus as the initiating insult. In poststatus epilepticus models, there is a loss of δ and α5 $GABA_AR$ subunits (Schwarzer et al., 1997; Peng et al., 2004) that usually mediate tonic inhibition under control conditions (Caraiscos et al., 2004a; Scimemi et al., 2005). This has lead to the hypothesis that epileptogenesis is accompanied by a loss of, or reduction in, tonic inhibition. However, this is not the case, and in temporal lobe epilepsy models either during induced status epilepticus or after seizure onset, tonic inhibition can be either unaltered (Zhang et al., 2007) or

indeed increased (Naylor et al., 2005; Scimemi et al., 2005). The lack of effect of the loss of subunits that normally generate tonic inhibition appears to be attributable to the upregulation of other receptor subunits and/or the translocation of receptors that are typically found at synaptic specializations into the extrasynaptic membrane (Peng et al., 2004; Scimemi et al., 2005; Zhang et al., 2007). Furthermore, Walker and colleagues showed that the preservation of tonic inhibition also occurs in a posttraumatic epilepsy model (Kharatishvili et al., 2006), where a reduction in phasic $GABA_A$ inhibition is not accompanied by a loss of tonic inhibition. These findings are in agreement with human epileptic tissue where tonic inhibition is preserved (Scimemi et al., 2006) and support a common paradigm in which tonic inhibition is maintained or enhanced during temporal lobe epilepsy, perhaps as a homeostatic mechanism to counter the concomitant loss of phasic inhibition.

Concluding remarks

In conclusion, the field of $GABA_AR$ research is excitingly poised to make significant advances in our understanding of the distinct contributions of both synaptic and extrasynaptic $GABA_ARs$ to physiological and pathophysiological CNS function. The advent of global $GABA_AR$ subunit-specific knock-out and knock-in mice has greatly aided the identification of synaptic and extrasynaptic $GABA_AR$ isoforms in discrete neuronal populations. However, notwithstanding the fact that synaptic and extrasynaptic $GABA_ARs$ can be distinguished by classical $GABA_AR$ antagonists in some neurons (Park et al., 2006), the specific roles of different subtypes of $GABA_ARs$ will only be determined by the development of neuron-specific and/or conditional transgenic mice (Gavériaux-Ruff and Kieffer, 2007; Wulff et al., 2007) or receptor subtype-specific antagonists and inverse agonists.

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