



Functional role of ambient GABA in refining neuronal circuits early in postnatal development

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Early in development, γ -aminobutyric acid (GABA), the primary inhibitory neurotransmitter in the mature brain, depolarizes and excites targeted neurons by an outwardly directed flux of chloride, resulting from the peculiar balance between the cation-chloride importer NKCC1 and the extruder KCC2. The low expression of KCC2 at birth leads to accumulation of chloride inside the cell and to the equilibrium potential for chloride positive respect to the resting membrane potential. GABA exerts its action *via* synaptic and extrasynaptic GABA_A receptors mediating phasic and tonic inhibition, respectively. Here, recent data on the contribution of “ambient” GABA to the refinement of neuronal circuits in the immature brain have been reviewed. In particular, we focus on the hippocampus, where, prior to the formation of conventional synapses, GABA released from growth cones and astrocytes in a calcium- and SNARE (soluble *N*-ethylmaleimide-sensitive-factor attachment protein receptor)-independent way, diffuses away to activate in a paracrine fashion extrasynaptic receptors localized on distal neurons. The transient increase in intracellular calcium following the depolarizing action of GABA leads to inhibition of DNA synthesis and cell proliferation. Tonic GABA exerts also a chemotropic action on cell migration. Later on, when synapses are formed, GABA spilled out from neighboring synapses, acting mainly on extrasynaptic $\alpha 5$, $\beta 2$, $\beta 3$, and γ containing GABA_A receptor subunits, provides the membrane depolarization necessary for principal cells to reach the window where intrinsic bursts are generated. These are instrumental in triggering calcium transients associated with network-driven giant depolarizing potentials which act as coincident detector signals to enhance synaptic efficacy at emerging GABAergic and glutamatergic synapses.

Keywords: development, hippocampus, tonic GABA_A conductance, network activity, extrasynaptic GABA_A receptor

γ -aminobutyric acid (GABA) signaling plays a crucial role for processing and storage of information in the brain. By releasing γ -aminobutyric acid (GABA) into distinct targeted sub-cellular compartments, GABAergic interneurons regulate cells excitability and dictate the temporal dynamics of principal cells firing giving rise to networks oscillations thought to support distinct brain states and high cognitive functions (Klausberger and Somogyi, 2008). The action of GABA relies on the temporally and spatially regulated expression of GABA_A receptors which mediate two distinct forms of inhibition: phasic and tonic (Semyanov et al., 2004; Cavalier et al., 2005; Farrant and Nusser, 2005). Phasic inhibition is mediated by local release of GABA from presynaptic vesicles (Cherubini, 2012). Once released, GABA binds to synaptic GABA_A receptors facing presynaptic release sites and trigger fast inhibitory postsynaptic potentials (IPSPs), regulating point-to-point communication between neurons. In this case, synaptic GABA_A receptors are exposed for a very short period of time to high concentrations of GABA. GABA diffuses throughout the neuropil before being taken up by selective plasma membrane transporters, which contribute to the clearance of the neurotransmitter and to shape synaptic currents (Cherubini and Conti, 2001). This transient inhibitory action is important for timing-based signaling, setting the temporal window for synaptic integration

(Pouille and Scanziani, 2001) and synchronization of neuronal networks (Cobb et al., 1995).

Tonic inhibition is mediated by “ambient” GABA originated from spillover of the neurotransmitter escaping the synaptic cleft (Kaneda et al., 1995; Brickley et al., 1996; Wall and Usowicz, 1997), from astrocytes *via* a non-vesicular calcium-independent process (Liu et al., 2000; Schousboe, 2003) or from the reversed transport (Attwell et al., 1993; Wu et al., 2001). In all these cases extrasynaptic GABA_A receptors are persistently exposed to sub-micromolar concentrations of GABA present in the extracellular space. This requires extrasynaptic GABA_A receptors with high affinity for GABA and relatively insensitive to desensitization. Selective plasma membrane transporters contribute to the clearance of GABA thus regulating its concentration in the extracellular space, in particular during massive release (Bragina et al., 2008). The resulting GABA-mediated tonic conductance is involved in regulating network excitability, cell firing and oscillatory behavior. In addition, the persistent increase in tonic conductance may affect the magnitude and duration of voltage responses to injected currents and increase the decrement of voltage with distance (Farrant and Nusser, 2005).

Synaptic and extrasynaptic GABA_A receptors are thought to belong to separate entities since they appear to be composed of

different subunits. However, the introduction of single molecule imaging technique has enabled measuring individual receptor movements in the plane of the plasma membrane (Triller and Choquet, 2005; Jacob et al., 2008; Luscher et al., 2011). This approach has revealed that receptors undergo lateral diffusion that allows them to continuously exchange between synaptic and extrasynaptic sites. Most receptors are delivered to extrasynaptic locations from where they can move and be trapped into synapses. GABA_A receptors trafficking and clustering is regulated by the scaffold protein gephyrin which, by anchoring GABA_A receptors to the cytoskeleton, exerts a stabilizing action.

The main focus of this review is on the contribution of “ambient” GABA in sculpting neuronal circuits at early developmental stages. We will first provide a brief overview of the depolarizing and excitatory action of GABA during embryonic and early postnatal life, emphasizing the role of this neurotransmitter in controlling cells proliferation, growth, migration, and differentiation during cortical neurogenesis as well as synaptogenesis immediately after birth. Then, we will discuss how a persistent tonic GABA_A-mediated conductance is instrumental in increasing cell excitability, thus contributing to trigger network-driven giant depolarizing potentials or GDPs in the immature hippocampus. GDPs are known to act as coincidence detectors for enhancing synaptic efficacy at emerging glutamatergic and GABAergic synapses. Finally, we will discuss how, immediately after birth, ambient GABA regulates cell excitability in other brain structures.

AT EARLY DEVELOPMENTAL STAGES GABA DEPOLARIZES AND EXCITES TARGETED CELLS VIA AN OUTFLOW OF CHLORIDE

GABAergic signaling is unique in that the polarity of its action largely depends on the intracellular chloride concentration $[Cl^-]_i$, leading in certain conditions to depolarizing and even excitatory effects. Neuronal $[Cl^-]_i$ is under the control of cation-chloride co-transporters (CCCs), intrinsic membrane proteins that transport Cl^- ions, together with Na^+ and/or K^+ ions, in an electroneutral manner due to the stoichiometric coupling and directionality of translocated ions. The two main CCCs which control chloride concentration inside the cell are the Na–K–2Cl importer NKCC1 and the K–Cl extruder KCC2. The low expression of KCC2 at birth leads to accumulation of chloride inside the cell and to the equilibrium potential for chloride (E_{Cl^-}) positive respect to the resting membrane potential (V_m). The progressive reduction in $[Cl^-]_i$ with age, due to the developmentally up-regulated expression of KCC2 and the concomitant down-regulated expression of NKCC1 (Yamada et al., 2004; Dzhalal et al., 2005), leads to relatively low $[Cl^-]_i$ (E_{Cl^-} close to V_m ; **Figure 1**; Rivera et al., 1999; Blaesse et al., 2009). Hence, while in adulthood GABA released from local interneurons opens GABA_A receptor channels causing a net flux of chloride inside the cells with consequent membrane hyperpolarization and reduction of cell firing (Cherubini and Conti, 2001), in the immediate postnatal period it depolarizes the membrane of targeted cells through an outwardly directed flux of chloride (Ben-Ari et al., 1989, 2012; Cherubini et al., 1991), thus enabling the membrane to reach spike threshold *via* amplification through a persistent non-inactivating sodium

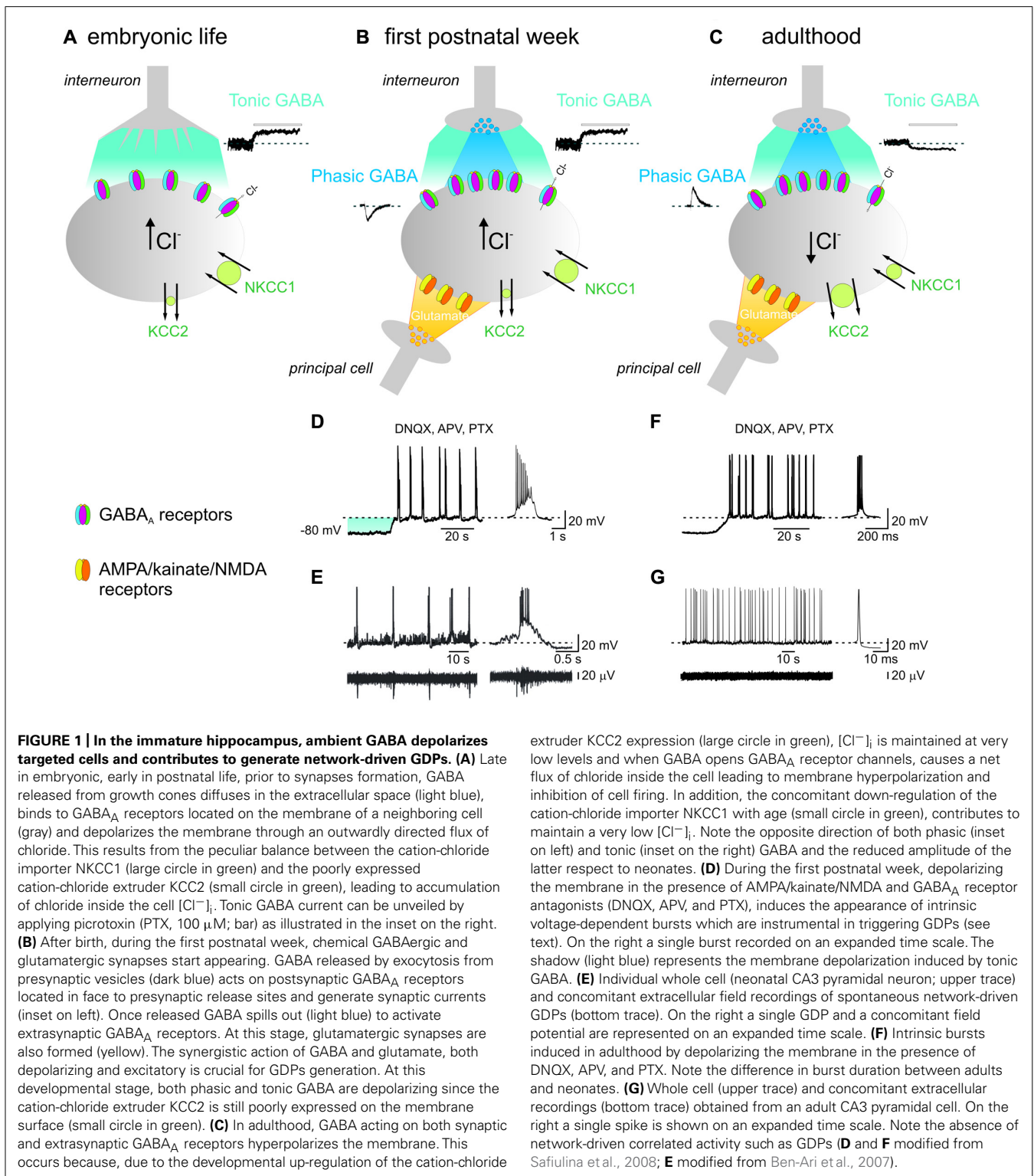
conductance (Valeeva et al., 2010; see also Song et al., 2011). It is worth noting that GABA can depolarize and still inhibit targeted cells *via* its shunting action (Mohajerani and Cherubini, 2005; Banke and McBain, 2006).

γ -aminobutyric acid-induced membrane depolarization activates voltage-dependent calcium channels and removes the voltage-dependent magnesium block from *N*-methyl-D-aspartic acid (NMDA) receptors leading to large calcium influx (Ben-Ari et al., 1997; Leinekugel et al., 1997). Calcium entry is crucial for the activation of second messengers, involved in a variety of developmental processes, from cell migration and differentiation to synaptogenesis.

Interestingly, GABAergic signals can be shifted in polarity by activity. This unique form of plasticity involves changes in the expression of CCCs able to convert GABA responses from hyperpolarizing to depolarizing and *vice versa* (Fiumelli and Woodin, 2007). While in adult hippocampal neurons, coincident detection of pre and postsynaptic signals alters the activity of KCC2 leading to changes in E_{Cl^-} and in the strength of inhibition (Woodin et al., 2003), in immature cells modifies the expression of NKCC1 causing E_{Cl^-} to shift toward more negative values (Balena and Woodin, 2008). Therefore, the dynamic regulation of intracellular chloride strictly depends on the ongoing activity generated by neurons and this may affect not only synaptic but also extrasynaptic GABA_A receptors.

Synapses start developing from birth following a well-defined sequence of events: GABAergic signals develop before glutamatergic ones whose operation correlates with the level of dendritic arborization (Tyzio et al., 1999; Ben-Ari et al., 2007). This occurs first in interneurons, and then in principal cells, indicating that GABAergic interneurons provide the early source of activity in otherwise silent networks (Gozlan and Ben-Ari, 2003).

Interestingly, prior to the formation of conventional synapses, GABA released from growth cones and astrocytes in a calcium- and soluble N-ethylmaleimide-sensitive-factor attachment protein receptor (SNARE)-independent way, diffuses away to activate in a paracrine fashion extrasynaptic receptors (**Figure 1**; Demarque et al., 2002). These are expressed at very early stages of development by neuronal precursors and by neurons in several brain areas giving rise to a tonic conductance (Nguyen et al., 2001). The existence of such conductance can be estimated by the shift in the baseline current obtained by blocking GABA_A receptors with selective antagonists. The tonic current persists after treatment with calcium channel blockers or botulin toxin which cleaves SNAP-25 (synaptosomal-associated protein 25), a SNARE protein that prevents vesicular release. In addition, a tonic current is present in Munc18-1 deficient mice lacking vesicular release (Demarque et al., 2002). It has been proposed that a relatively poor clearance system (but see Sipilä et al., 2004; Safiulina et al., 2006) enables GABA to accumulate in the extracellular space and reach a concentration sufficient to exert its depolarizing and excitatory effects on distal neurons making this form of intercellular communication very effective. In addition, since the main neuronal GABA transporter GAT-1 carries chloride along with GABA and sodium, the possibility that a high intracellular chloride may decrease the efficacy of GABA uptake cannot be excluded.



AMBIENT GABA REGULATES CELL MIGRATION AND SYNAPSES FORMATION

The construction of the cerebral cortex from a single sheet of neuroepithelium relies on a sequence of well-orchestrated developmental processes. Neurons must be generated in the correct

number, migrate to the proper position, and form connections with neighboring cells. Of the many cell-intrinsic and -extrinsic signals involved in neocortical development, ambient GABA plays a central role in these processes (Wang and Kriegstein, 2009).

During corticogenesis, endogenous GABA present in the extracellular space, binds with high affinity (higher than in post-migratory neurons) to extrasynaptic GABA_A receptors (relatively insensitive to desensitization), expressed on migrating neurons as well as on radial glia and causes a membrane depolarization and a transient increase in calcium *via* voltage-dependent calcium channels. This leads to inhibition of DNA synthesis and cell proliferation as assessed by the reduced number of progenitors incorporating BrdU (LoTurco et al., 1995; Owens and Kriegstein, 2002). Furthermore, tonic GABA exerts a chemotropic action on cell migration as demonstrated by the observation that blocking GABA_A receptors with bicuculline in organotypic hippocampal slices reduces the migration of neuroblasts (Manent et al., 2005). Ambient GABA may also regulate the speed of migration of young neurons in the rostral migratory stream (Bolteus and Bordey, 2004) and provide a stop signal for ending migration (Behar et al., 2000). The effect of GABA on cell migration has been recently questioned. Using *in utero* electroporation, Cancedda et al. (2007) have clearly demonstrated that the premature expression of the cation-chloride extruder KCC2, which eliminates the depolarizing action of endogenous GABA in a subpopulation of newly born cortical neurons, does not alter their migration while at P4–P6 severely affects their morphological structure (neurons exhibit few short dendrites). However, as highlighted in a recent study (Inoue et al., 2012), the ectopic expression of KCC2 is functional only in the postnatal but not in the embryonic brain. This is probably related to endogenous taurine, particularly abundant in the fetal brain that, *via* the with-no-lysine protein kinase 1 (WNK1) signaling pathway, exerts an inhibitory action on KCC2 activation.

Increasing evidence suggests that a tonic GABA_A-mediated membrane depolarization provides the first excitatory drive necessary for promoting neurite outgrowth and synapse formation. Unlike glutamate in fact, GABA depolarizes and at the same time, by clamping the membrane potential close to E_{Cl} (in immature neurons about -40 mV) exerts a shunting effect that would prevent an excessive calcium entry through voltage-dependent calcium channels with consequent excitotoxicity (Safulina et al., 2010). GABA-mediated membrane depolarization has been shown to regulate the formation of glutamatergic synapses in the developing cortex *in vivo*, an effect that needs the contribution of NMDA receptors. This will allow a proper balance between excitation and inhibition, essential for the correct functioning of neuronal circuits (Wang and Kriegstein, 2008). Cortical neurons begin to express NMDA receptors during migration to the cortical plate. However, these receptors do not conduct at rest because they are blocked by magnesium. By facilitating the relief of the voltage-dependent magnesium block, GABA *via* its depolarizing action renders these receptors conductive. The systemic blockade of early GABA-mediated depolarization during a critical period between E17–P7 in mice with bumetanide, a selective NKCC1 inhibitor, leads to lasting disruption of AMPA receptors-mediated glutamatergic transmission in the adult cortex and to an excitatory/inhibitory imbalance (Wang and Kriegstein, 2011). Morphological analysis of bumetanide-treated mice revealed reduced spines density and dendritic arborization in cortical neurons (Wang and Kriegstein, 2011). This is in contrast with

the data obtained by Pfeffer et al. (2009) from *Nkcc1*^{-/-} mice. Compensatory mechanisms responsible for the slightly depolarizing action of GABA in the genetic model may account for this discrepancy (Sipilä et al., 2009).

Interestingly, in the adult brain, GABA-mediated tonic excitation drives synaptic integration of newly generated neurons in pre-existing functional circuits suggesting that adult neurogenesis recapitulates the sequence of events occurring in immature cells at embryonic and early stages of postnatal development (Ge et al., 2006). As in postnatal development, conversion of GABA-induced depolarization into hyperpolarization in newborn dentate gyrus cells leads to significant defects in the formation of GABAergic and glutamatergic synapse as well as in dendritic arborization (Ge et al., 2006).

IMMEDIATELY AFTER BIRTH, A TONIC GABA_A-MEDIATED CONDUCTANCE CONTRIBUTES TO THE TRIGGERING OF NETWORK-DRIVEN GDPs IN THE HIPPOCAMPUS

In the neonatal hippocampus a GABA_A receptor-mediated tonic conductance has been well-characterized in both CA1 (Demarque et al., 2002; Marchionni et al., 2007) and CA3 regions (Marchionni et al., 2007; Sipilä et al., 2007). In these areas, principal cells exhibit a sustained tonic conductance (larger in the CA3 region; Marchionni et al., 2007) which plays a crucial role in enhancing cell excitability and neuronal firing, thus contributing to GDPs generation (Sipilä et al., 2005, 2009).

Tonic GABA_A-mediated currents are more pronounced in neonates than in adults (Figure 1; Stell and Mody, 2002; Semyanov et al., 2003; Stell et al., 2003; Caraiscos et al., 2004). This has been attributed to the relatively poor GABA clearance caused by the low expression of GABA transporters. However, at least GAT-1 has been found to be present and functional from birth as shown by the ability of NO-711, a selective GAT-1 inhibitor, to enhance the decay kinetics of GABA_A-mediated synaptic currents (Safulina et al., 2006) and the duration of GDPs (Sipilä et al., 2004), indicating that GABA uptake controls GABAergic transmission and limits the action of GABA on GDPs. It is worth mentioning that while NO-711 affects the shape of synaptic currents (Safulina et al., 2006), it only slightly alters tonic currents (Marchionni et al., 2007; Sipilä et al., 2007), suggesting that GAT-1, due to its preferential localization on axon terminals (Minelli et al., 1995), is more efficient at removing GABA from synapses than from the extracellular space.

In both CA1 and CA3 principal cells, “ambient” GABA may originate at least in part from spillover of the neurotransmitter from neighboring synapses during activity as shown by the reduction in the tonic current by tetrodotoxin (TTX) that blocks sodium currents and propagated action potentials (Marchionni et al., 2007; but see Sipilä et al., 2007 for the CA3 region). Interestingly, in contrast with adult guinea pigs (Semyanov et al., 2003) or rats (Marchionni et al., 2007), in neonatal animals stratum radiatum GABAergic interneurons fail to exhibit any sustained background conductance. This may be due either to a more efficient uptake system, able to maintain extracellular GABA at very low levels, or to a low expression of extrasynaptic receptors able to sense GABA. While the first possibility is unlikely since NO-711 did not modify the holding current, the second one remains

to be demonstrated. Whether other transporters, different from GAT-1 are actively involved in removing GABA from GABAergic interneurons cannot be excluded.

In immature CA1 and CA3 principal cells, the selective use of GABA_A receptor agonists and antagonist has unveiled the presence of various extrasynaptic receptor subunits. These include the $\alpha 1$, $\alpha 3$ (Sipilä et al., 2007), $\alpha 5$, $\beta 2$, $\beta 3$, and $\gamma 2$ (Marchionni et al., 2007). While $\alpha 5$ GABA_A receptor subunits are highly expressed in the neonatal hippocampus (Laurie et al., 1992; Didelon et al., 2000), δ subunits are relatively scarce (Didelon et al., 2000) and this may explain why allotetrahydrodeoxycorticosterone (THDOC), $\alpha \delta$ subunit selective neuroactive steroid (Korpi and Lüddens, 1997), was unable to modify the baseline current (Marchionni et al., 2007).

The $\gamma 2$, $\alpha 1$, and $\alpha 3$ subunits are crucial for zolpidem sensitivity. The observation that zolpidem potentiates tonic currents only in neonates (Sipilä et al., 2007) but not in adults (Semyanov et al., 2003) suggests a different expression of extrasynaptic GABA_A receptor subunits with age.

How could the tonic GABA_A-mediated conductance influence network activity immediately after birth? Thanks to the depolarizing and excitatory action of GABA at this stage of development the tonic conductance has been found to enhance pyramidal cells excitability and to reduce the threshold for action potential generation (Marchionni et al., 2007). Thus, in cell-attach experiments to maintain intact the intracellular chloride concentration, picrotoxin blocks synaptic and extrasynaptic GABA_A receptors leading to an increased firing in pyramidal cells but not in interneurons. This may enhance glutamate release from principal cells.

Giant depolarizing potentials, which represent a primordial form of synchrony between neurons, are generated by the synergistic action of glutamate and GABA, both of which are depolarizing and excitatory (Figure 1; Cherubini et al., 1991; Ben-Ari, 2002). In analogy with the synchronized activity generated in the disinhibited hippocampus (De la Prida et al., 2006), GDPs emerge when a sufficient number of cells fire and the excitability of the network attains a certain threshold within a restricted period of time. Although the entire hippocampal network possesses the capacity to generate GDPs, the CA3 area is centrally involved because: (i) in this area GABAergic interneurons with large axonal arborizations operate as functional hubs to synchronize large ensembles of cells (Bonifazi et al., 2009; Picardo et al., 2011; Allene et al., 2012); (ii) principal cells are connected by extensive glutamatergic recurrent collaterals; (iii) principal cells give rise to intrinsic bursts that drive other neurons to fire (Sipilä et al., 2005; Safiulina et al., 2008). In this context, GABA_A-mediated tonic inhibition provides the membrane depolarization needed for principal cells to reach the window where intrinsic bursts are generated (Figure 1; Sipilä et al., 2007; Ben-Ari et al., 2012).

Since GDPs are instrumental for enhancing synaptic efficacy at emerging glutamatergic and GABAergic synapses and for converting silent synapses into conductive ones (Kasyanov et al., 2004; Mohajerani et al., 2007), we can speculate that, immediately after birth, ambient GABA exerts a crucial role in synaptic wiring. Whether changes in synaptic efficacy are associated with structural modifications necessary to rewire local neuronal circuits remain to be demonstrated.

EARLY IN POSTNATAL DEVELOPMENT, TONIC GABAergic CURRENTS DIFFERENTLY REGULATES CELL EXCITABILITY IN VARIOUS BRAIN REGIONS

DENTATE GYRUS GRANULE CELLS

A powerful tonic GABAergic signaling has been described in granule cells of the dentate gyrus already at P3 (Holter et al., 2010). Using subunit-specific pharmacological modulators, it has been demonstrated that, as in adults (Glykys et al., 2008), this conductance is mediated by $\alpha 5$ and δ subunits containing extrasynaptic GABA_A receptors. The relative contribution of receptors containing $\alpha 5$ -subunits decreases with age, while that of receptors containing δ -subunits increases, indicating that as in principal cells the expression of δ subunits is developmentally regulated. The increased expression of δ subunits parallels that of tonic conductance that reaches its maximum during the second postnatal week and then declines (Holter et al., 2010). Surprisingly, unlike immature CA1 and CA3 principal cells (Marchionni et al., 2007), this conductance exerts mainly an inhibitory effect *via* a shunting inhibition. The inhibitory effect would prevent the occurrence of neonatal seizures (Holter et al., 2007).

NEOCORTICAL NEURONS

A GABA_A-mediated tonic conductance has been detected in layer V pyramidal cells of the somatosensory cortex of newborn animals (Sebe et al., 2010). This conductance, which is mainly mediated by $\alpha 5$ and δ subunits containing extrasynaptic GABA_A receptors, is very large immediately after birth and then decreases dramatically during the second postnatal week. The developmental change appears to be related to the enhanced clearance of GABA from the extracellular space (due to the increased activity of GABA transporters) and to the reduced expression of δ subunits (the $\alpha 5$ subunits are expressed also in adults; Yamada et al., 2007). In cell-attach recordings GABA has been found to exert opposite effects on immature cell excitability, decreasing or increasing cell firing in accord with the value of E_{Cl} relative to resting membrane potential (Sebe et al., 2010).

Unlike the somatosensory cortex, in the visual cortex tonic GABA_A-mediated currents increase with age (Jang et al., 2010). This may be related to the role of GABA in shaping neuronal circuits during a well-defined period of early postnatal development called critical period (Sale et al., 2010). In this period, the experience-dependent maturation of the visual acuity closely relies on the development of the intracortical GABAergic inhibition. Dark rearing, which causes a permanent loss of visual acuity reduces also GABAergic signaling mainly from parvalbumin positive interneurons (Hensch, 2005).

CEREBELLAR GRANULE CELLS

The cerebellum is the first brain structure where a persistent background GABA_A-mediated conductance has been characterized (Kaneda et al., 1995). This clearly exhibits a developmental profile, increasing progressively after P7. This may reflect the increasing number of GABAergic terminals within the glomerulus, a structure that limiting GABA diffusion may render the action of ambient GABA more efficient (Brickley et al., 1996; Wall and Usowicz, 1997). In addition, while at early developmental stages tonic GABA exerts a depolarizing and excitatory action, later it

exerts an inhibitory effect *via* shunting inhibition (E_{Cl} is close to the resting membrane potential; Brickley et al., 1996).

THALAMIC RELAY NEURONS

A similar sequence of events takes place in thalamic ventrobasal relay neurons, where GABA_A-mediated tonic current is developmentally regulated. This conductance can be detected from the first week of postnatal life (Belelli et al., 2005). It progressively increases with age. This effect is not related to changes in the expression of GABA transporters, since bicuculline-induced shift in baseline current does not change by blocking GABA uptake with selective inhibitors (Peden et al., 2008). The ontogenetic up-regulation of the tonic conductance depends on marked changes in subunit composition of extrasynaptic GABA_A receptors with increased expression of $\alpha 4$ and δ subunits, as assessed with electrophysiology and immunohistochemistry. While in adult neurons, the persistent activation of extrasynaptic receptors by ambient GABA leads to a membrane hyperpolarization needed to promote low threshold burst firing (Cope et al., 2005), in neonates its function is not clear since it depends on the hyperpolarizing or depolarizing action of GABA.

STRIATAL MEDIUM SPINY NEURONS

In GABAergic medium spiny striatal output neurons (MSNs), a tonic GABA_A-mediated conductance starts appearing toward the end of the second postnatal week (Ade et al., 2008; Kirmse et al., 2008). Before P14, it can be detected only in the presence of GAT-1 inhibitors suggesting that this transporter operates in a net uptake mode, able to efficiently remove GABA from the extracellular space. An increase in synaptic GABA release, due to an up-regulation of the glutamatergic drive to MSNs may account for the persistent activation of extrasynaptic GABA_A receptors with age. Ambient GABA originates mainly from action potential-dependent synaptic release, since TTX reduces the tonic current similarly to GABA antagonists (Ade et al., 2008).

The inhibitory GABA_A-mediated conductance is larger in MSNs expressing dopamine D2 receptors, which project to the globus pallidus respect to those expressing dopamine D1 receptors which project to the substantia nigra pars reticulata (Ade et al., 2008; Santhakumar et al., 2010). While the first are supposed to inhibit movements, the latter to facilitate them (Gerfen et al., 1990). The greater expression of $\alpha 5$ subunit containing GABA_A receptors in D2- respect to D1-positive MSNs may account for the observed effects (Ade et al., 2008). In addition, using conditional b3 subunit knock-out mice Janssen et al. (2011) have demonstrated that receptors containing the b3 subunit also contribute to tonic inhibition in D2-positive neurons. These data are in agreement with previous mRNA expression studies showing that young but not adult striatal tissue primarily expresses $\alpha 2$, $\alpha 5$, $\beta 3$, and γ GABA_A receptor subunits (Laurie et al., 1992). Differences in tonic currents between D1- and D2-positive cells observed in juvenile animals may be crucial for regulating MSNs excitability and motor output.

CONCLUSION

From the data reviewed here it is clear that a tonic GABA_A-mediated conductance plays an important role in brain development. This conductance is developmentally regulated and differs in various brain regions (see **Table 1**). While in some areas (i.e., hippocampus and somatosensory cortex) it diminishes with maturation, in others (i.e., thalamus, visual cortex, striatum, and cerebellum) it increases. This depends on several factors including stage of cell maturity, geometry of the synapses, interneuronal firing, neurotransmitter diffusion, expression and distribution of extrasynaptic GABA_A receptors, and/or GABA transporters. In addition, according to the direction of GABA signaling, accumulation of the neurotransmitter in the extracellular space may differently affect network excitability.

In spite progress in the field much remains to be known on how early activity or experience regulates receptors trafficking and

Table 1 | Tonic GABAergic currents in various brain regions during early development.

| Structure | Age | GABA _A R subunits | Direction of GABA action | Reference |
|---------------------------|-------------------------------------------------|---------------------------------------------------------------|--------------------------|----------------------------------------------------|
| CA1 region of hippocampus | Late embryonic, early postnatal life (1st week) | $\alpha 5$, $\gamma 2$ | Depolarizing | Demarque et al. (2002) Marchionni et al. (2007) |
| CA3 region of hippocampus | 1st postnatal week | $\alpha 1$, $\alpha 3$, $\alpha 5$, $\beta 3$, $\gamma 2$ | Depolarizing | Marchionni et al. (2007) Sipilä et al. (2007) |
| Dentate gyrus | Newborn cells in adulthood | Unknown | Depolarizing | Ge et al. (2006) |
| Somatosensory cortex | 1st postnatal week | $\alpha 5$, δ | Depolarizing | Sebe et al. (2010) |
| Cerebellum | 1st postnatal week | Unknown | Depolarizing | Brickley et al. (1996) |
| | 2nd postnatal week | Unknown | Hyperpolarizing | Wall and Usowicz (1997) |
| Thalamus (ventrobasal) | 1st postnatal week | $\alpha 4$, δ | Unknown | Peden et al. (2008) |
| Striatum | 2nd postnatal week | $\alpha 5$ | Unknown | Ade et al. (2008) |
| | 4th postnatal week | δ , $\beta 3$ | Unknown | Santhakumar et al. (2010) Janssen et al. (2011) |

exchanges between synaptic and extrasynaptic pools. While the interaction of scaffold proteins with synaptic receptors has been at least in part elucidated that related to extrasynaptic receptors is still poorly understood. Furthermore, it would be of interest to know how selectively silencing in embryonic or early postnatal life a particular extrasynaptic GABA_A receptor subunit may alter the computational properties of neuronal circuits and synaptogenesis. This would help to better understand the role of GABA as a developmental signal and its implications in neurodevelopmental disorders.

REFERENCES

- Ade, K. K., Janssen, M. J., Ortinski, P. I., and Vicini, S. (2008). Differential tonic GABA conductances in striatal medium spiny neurons. *J. Neurosci.* 28, 1185–1197. doi: 10.1523/JNEUROSCI.3908-07.2008
- Allene, C., Picardo, M. A., Becq, H., Miyoshi, G., Fishell, G., and Corsart, R. (2012). Dynamic changes in interneuron morphophysiological properties mark the maturation of hippocampal network activity. *J. Neurosci.* 32, 6688–6698. doi: 10.1523/JNEUROSCI.0081-12.2012
- Attwell, D., Barbour, B., and Szatkowski, M. (1993). Non vesicular release of neurotransmitter. *Neuron* 11, 401–407. doi: 10.1016/0896-6273(93)90145-H
- Balena, T., and Woodin, M. A. (2008). Coincident pre- and postsynaptic activity downregulates NKCC1 to hyperpolarize ECl during development. *Eur. J. Neurosci.* 27, 2402–2412. doi: 10.1111/j.1460-9568.2008.06194.x
- Banke, T. G., and McBain, C. J. (2006). GABAergic input onto CA3 hippocampal interneurons remains shunting throughout development. *J. Neurosci.* 26, 11720–11725. doi: 10.1523/JNEUROSCI.2887-06.2006
- Behar, T. N., Schaffner, A. E., Scott, C. A., Greene, C. L., and Barker, J. L. (2000). GABA receptor antagonists modulate postmitotic cell migration in slice cultures of embryonic rat cortex. *Cereb. Cortex* 10, 899–909. doi: 10.1093/cercor/10.9.899
- Belelli, D., Peden, D. R., Rosahl, T. W., Wafford, K. A., and Lambert, J. J. (2005). Extrasynaptic GABA_A receptors of thalamo-cortical neurons: a molecular target for hypnotics. *J. Neurosci.* 25, 11513–11520. doi: 10.1523/JNEUROSCI.2679-05.2005
- Ben-Ari, Y. (2002). Excitatory actions of GABA during development: the nature of the nurture. *Nat. Rev. Neurosci.* 3, 728–739. doi: 10.1038/nrn920
- Ben-Ari, Y., Cherubini, E., Corradetti, R., and Gaiarsa, J. L. (1989). Giant synaptic potentials in immature rat CA3 hippocampal neurones. *J. Physiol.* 416, 303–325.
- Ben-Ari, Y., Gaiarsa, J. L., Tyzio, R., and Khazipov, R. (2007). GABA: a pioneer transmitter that excites immature neurons and generates primitive oscillations. *Physiol. Rev.* 87, 1215–1284. doi: 10.1152/physrev.00017.2006
- Ben-Ari, Y., Khalilov, I., Kahle, K. T., and Cherubini, E. (2012). The GABA excitatory/inhibitory shift in brain maturation and neurological disorders. *Neuroscientist* 18, 467–486. doi: 10.1177/1073858412438697
- Ben-Ari, Y., Khazipov, R., Leinekugel, X., Caillard, O., and Gaiarsa, J. L. (1997). GABA_A, NMDA and AMPA receptors: a developmentally regulated ‘ménage à trois’. *Trends Neurosci.* 20, 523–529. doi: 10.1016/S0166-2236(97)01147-8
- Blaesse, P., Airaksinen, M. S., Rivera, C., and Kaila, K. (2009). Cation-chloride co-transporters and neuronal function. *Neuron* 61, 820–838. doi: 10.1016/j.neuron.2009.03.003
- Bolteus, A. J., and Bordey, A. (2004). GABA release and uptake regulate neuronal precursor migration in the postnatal subventricular zone. *J. Neurosci.* 24, 7623–7631. doi: 10.1523/JNEUROSCI.1999-04.2004
- Bonifazi, P., Goldin, M., Picardo, M. A., Jorquera, I., Cattani, A., Bianconi, G., et al. (2009). GABAergic hub neurons orchestrate synchrony in developing hippocampal networks. *Science* 326, 1419–1424. doi: 10.1126/science.1175509
- Bragina, L., Marchionni, I., Omrani, A., Cozzi, A., Pellegrini-Giampietro, D. E., Cherubini, E., et al. (2008). GAT-1 regulates both tonic and phasic GABA_A receptor-mediated inhibition in the cerebral cortex. *J. Neurochem.* 105, 1781–1793. doi: 10.1111/j.1471-4159.2008.05273.x
- Brickley, S. G., Cull-Candy, S. G., and Farrant, M. (1996). Development of a tonic form of synaptic inhibition in rat cerebellar granule cells resulting from persistent activation of GABA_A receptors. *J. Physiol.* 497, 753–759.
- Cancedda, L., Fiumelli, H., Chen, K., and Poo, M. M. (2007). Excitatory GABA action is essential for morphological maturation of cortical neurons in vivo. *J. Neurosci.* 27, 5224–5235. doi: 10.1523/JNEUROSCI.5169-06.2007
- Carascos, V. B., Elliott, E. M., You-Ten, K. E., Cheng, V. Y., Belelli, D., Newell, J. G., et al. (2004). Tonic inhibition in mouse hippocampal CA1 pyramidal neurons is mediated by alpha5 subunit-containing gamma-aminobutyric acid type A receptors. *Proc. Natl. Acad. Sci. U.S.A.* 101, 3662–3667. doi: 10.1073/pnas.0307231101
- Cavelier, P., Hamann, M., Rossi, D., Mobbs, P., and Attwell, D. (2005). Tonic excitation and inhibition of neurons: ambient transmitter sources and computational consequences. *Prog. Biophys. Mol. Biol.* 87, 3–16. doi: 10.1016/j.pbiomolbio.2004.06.001
- Cherubini, E. (2012). “Phasic GABA_A-mediated inhibition” in *Jasper’s Basic Mechanisms of the Epilepsies*, eds J. L. Noebels, M. Avoli, M. A. Rogawski, R. W. Olsen, and A. V. Delgado-Escueta (Oxford: Oxford University Press).
- Cherubini, E., and Conti, F. (2001). Generating diversity at GABAergic synapses. *Trends Neurosci.* 24, 155–162. doi: 10.1016/S0166-2236(00)01724-0
- Cherubini, E., Gaiarsa, J. L., and Ben-Ari, Y. (1991). GABA: an excitatory transmitter in early postnatal life. *Trends Neurosci.* 14, 515–519. doi: 10.1016/0166-2236(91)90003-D
- Cobb, S. R., Buhl, E. H., Halasy, K., Paulsen, O., and Somogyi, P. (1995). Synchronization of neuronal activity in hippocampus by individual GABAergic interneurons. *Nature* 378, 75–78. doi: 10.1038/378075a0
- Cope, D. W., Hughes, S. W., and Crunelli, V. (2005). GABA_A receptor-mediated tonic inhibition in thalamic neurons. *J. Neurosci.* 25, 11553–11563. doi: 10.1523/JNEUROSCI.3362-05.2005
- De la Prida, L. M., Huberfeld, G., Cohen, I., and Miles, R. (2006). Threshold behavior in the initiation of hippocampal population bursts. *Neuron* 49, 131–142. doi: 10.1016/j.neuron.2005.10.034
- Demarque, M., Represa, A., Becq, H., Khalilov, I., Ben-Ari, Y., and Aniksztejn, L. (2002). Paracrine intercellular communication by a Ca²⁺- and SNARE-independent release of GABA and glutamate prior to synapse formation. *Neuron* 36, 1051–1061. doi: 10.1016/S0896-6273(02)01053-X
- Didelon, F., Mladinic, M., Cherubini, E., and Bradbury, A. (2000). Early expression of GABA_A receptor delta subunit in the neonatal rat hippocampus. *J. Neurosci. Res.* 62, 638–643. doi: 10.1002/1097-4547(20001201)62:5
- Dzhala, V. I., Talos, D. M., Sdrulla, D. A., Brumback, A. C., Mathews, G. C., Benke, T. A., et al. (2005). NKCC1 transporter facilitates seizures in the developing brain. *Nat. Med.* 11, 1205–1213. doi: 10.1038/nm1301
- Farrant, M., and Nusser, Z. (2005). Variations on an inhibitory theme: phasic and tonic activation of GABA(A) receptors. *Nat. Rev. Neurosci.* 6, 215–229. doi: 10.1038/nrn1625
- Fiumelli, H., and Woodin, M. A. (2007). Role of activity-dependent regulation of neuronal chloride homeostasis in development. *Curr. Opin. Neurobiol.* 17, 81–86. doi: 10.1016/j.conb.2007.01.002
- Ge, S., Goh, E. L., Sailor, K. A., Kitabatake, Y., Ming, G. L., and Song, H. (2006). GABA regulates synaptic integration of newly generated neurons in the adult brain. *Nature* 439, 589–593. doi: 10.1038/nature04404
- Gerfen, C. R., Engber, T. M., Mahan, L. C., Susel, Z., Chase, T. N., Monson, F. J. Jr., et al. (1990). D1 and D2 dopamine receptor-regulated gene expression of striato-nigral and striato-pallidal neurons. *Science* 250, 1429–1432. doi: 10.1126/science.2147780
- Glykys, J., Mann, E. O., and Mody, I. (2008). Which GABA_A receptor subunits are necessary for

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Giada Cellot and Enrico Cherubini wrote the paper.

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- tonic inhibition in the hippocampus? *J. Neurosci.* 28, 1421–1426. doi: 10.1523/JNEUROSCI.4751-07.2008
- Gozlan, H., and Ben-Ari, Y. (2003). Interneurons are the source and the targets of the first synapses formed in the rat developing hippocampal circuit. *Cereb. Cortex* 13, 684–692. doi: 10.1093/cercor/13.6.684
- Hensch, T. K. (2005). Critical period mechanisms in developing visual cortex. *Curr. Top. Dev. Biol.* 69, 215–237. doi: 10.1016/S0070-2153(05)69008-4
- Holter, N. I., Zuber, N., Bruehl, C., and Draguhn, A. (2007). Functional maturation of developing interneurons in the molecular layer of mouse dentate gyrus. *Brain Res.* 1186, 56–64. doi: 10.1016/j.brainres.2007.09.089
- Holter, N. I., Zylla, M. M., Zuber, N., Bruehl, C., and Draguhn, A. (2010). Tonic GABAergic control of mouse dentate granule cells during postnatal development. *Eur. J. Neurosci.* 32, 1300–1309. doi: 10.1111/j.1460-9568.2010.07331.x
- Inoue, K., Furukawa, T., Kumada, T., Yamada, J., Wang, T., Inoue, R., et al. (2012). Taurine inhibits K⁺-Cl⁻ co-transporter KCC2 to regulate embryonic Cl⁻ homeostasis via with-no-lysine (WNK) protein kinase signaling pathway. *J. Biol. Chem.* 287, 20839–20850. doi: 10.1074/jbc.M111.319418
- Jacob, T. C., Moss, S. J., and Jurd, R. (2008). GABAA receptor trafficking and its role in the dynamic modulation of neuronal inhibition. *Nat. Rev. Neurosci.* 9, 331–343. doi: 10.1038/nrn2370
- Jang, H. J., Cho, K. H., Park, S. W., Kim, M. J., Yoon, S. H., and Rhie, D. J. (2010). The development of phasic and tonic inhibition in the rat visual cortex. *Korean J. Physiol. Pharmacol.* 14, 399–405. doi: 10.4196/kjpp.2010.14.6.399
- Janssen, M. J., Yasuda, R. P., and Vicini, S. (2011). GABAA receptor $\beta 3$ subunit expression regulates tonic current in developing striato-pallidal medium spiny neurons. *Front. Cell. Neurosci.* 5:15. doi: 10.3389/fncel.2011.00015
- Kaneda, M., Farrant, M., and Cull-Candy, S. G. (1995). Whole-cell and single-channel currents activated by GABA and glycine in granule cells of the rat cerebellum. *J. Physiol.* 485, 419–435.
- Kasyanov, A. M., Safulina, V. F., Voronin, L. L., and Cherubini, E. (2004). GABA-mediated giant depolarizing potentials as coincidence detectors for enhancing synaptic efficacy in the developing hippocampus. *Proc. Natl. Acad. Sci. U.S.A.* 101, 3967–3972. doi: 10.1073/pnas.0305974101
- Kirmse, K., Dvorzhak, A., Kirischuk, S., and Grantyn, R. (2008). GABA transporter 1 tunes GABAergic synaptic transmission at output neurons of the mouse neostriatum. *J. Physiol.* 586, 5665–5678. doi: 10.1111/jphysiol.2008.161943
- Klausberger, T., and Somogyi, P. (2008). Neuronal diversity and temporal dynamics: the unity of hippocampal circuit operations. *Science* 321, 53–57. doi: 10.1126/science.1149381
- Korpi, E. R., and Lüddens, H. (1997). Furosemide interactions with brain GABAA receptors. *Br. J. Pharmacol.* 120, 741–748. doi: 10.1038/sj.bjp.0700922
- Laurie, D. J., Wisden, W., and Seeburg, P. H. (1992). The distribution of thirteen GABAA receptor subunit mRNAs in the rat brain. III. Embryonic and postnatal development. *J. Neurosci.* 12, 4151–4172.
- Leinekugel, X., Medina, I., Khalilov, I., Ben-Ari, Y., and Khazipov, R. (1997). Ca²⁺ oscillations mediated by the synergistic excitatory actions of GABAA and NMDA receptors in the neonatal hippocampus. *Neuron* 18, 243–255. doi: 10.1016/S0896-6273(00)80265-2
- Liu, Q. Y., Schaffner, A. E., Chang, Y. H., Maric, D., and Barker, J. L. (2000). Persistent activation of GABAA receptor/Cl⁻ channels by astrocyte-derived GABA in cultured embryonic rat hippocampal neurons. *J. Neurophysiol.* 84, 1392–1403.
- LoTurco, J. J., Owens, D. F., Heath, M. J., Davis, M. B., and Kriegstein, A. R. (1995). GABA and glutamate depolarize cortical progenitor cells and inhibit DNA synthesis. *Neuron* 15, 1287–1298. doi: 10.1016/0896-6273(95)90008-X
- Luscher, B., Fuchs, T., and Kilpatrick, C. L. (2011). GABAA receptor trafficking-mediated plasticity of inhibitory synapses. *Neuron* 70, 385–409. doi: 10.1016/j.neuron.2011.03.024
- Manent, J. B., Demarque, M., Jorquera, I., Pellegrino, C., Ben-Ari, Y., Aniksztejn, L., et al. (2005). A non canonical release of GABA and glutamate modulates neuronal migration. *J. Neurosci.* 25, 4755–4765. doi: 10.1523/JNEUROSCI.0553-05.2005
- Marchionni, I., Omrani, A., and Cherubini, E. (2007). In the developing rat hippocampus a tonic GABAA-mediated conductance selectively enhances the glutamatergic drive of principal cells. *J. Physiol.* 581, 515–528. doi: 10.1113/jphysiol.2006.125609
- Minelli, A., Brecha, N. C., Karschin, C., DeBiasi, S., and Conti, F. (1995). GAT-1, a high-affinity GABA plasma membrane transporter, is localized to neurons and astroglia in the cerebral cortex. *J. Neurosci.* 15, 7734–7746.
- Mohajerani, M. H., and Cherubini, E. (2005). Spontaneous recurrent network activity in organotypic rat hippocampal slices. *Eur. J. Neurosci.* 22, 107–118. doi: 10.1111/j.1460-9568.2005.04198.x
- Mohajerani, M. H., Sivakumaran, S., Zacchi, P., Aguilera, P., and Cherubini, E. (2007). Correlated network activity enhances synaptic efficacy via BDNF and the ERK pathway at immature CA3–CA1 connections in the hippocampus. *Proc. Natl. Acad. Sci. U.S.A.* 104, 13176–13181. doi: 10.1073/pnas.0704533104
- Nguyen, L., Rigo, J. M., Rocher, V., Belachew, S., Malgrange, B., Rogister, B., et al. (2001). Neurotransmitters as early signals for central nervous system development. *Cell Tissue Res.* 305, 187–202. doi: 10.1007/s004410000343
- Owens, D. F., and Kriegstein, A. R. (2002). Is there more to GABA than synaptic inhibition? *Nat. Rev. Neurosci.* 9, 715–727. doi: 10.1038/nrn919
- Peden, D. R., Petitjean, C. M., Herd, M. B., Durakoglugil, M. S., Rosahl, T. W., Wafford, K., et al. (2008). Developmental maturation of synaptic and extrasynaptic GABAA receptors in mouse thalamic ventro-basal neurones. *J. Physiol.* 586, 965–987. doi: 10.1113/jphysiol.2007.145375
- Pfeffer, C. K., Stein, V., Keating, D. J., Maier, H., Rinke, I., Rudhard, Y., et al. (2009). NKCC1-dependent GABAergic excitation drives synaptic network maturation during early hippocampal development. *J. Neurosci.* 29, 3419–3430. doi: 10.1523/JNEUROSCI.1377-08.2009
- Picardo, M. A., Guigue, P., Bonifazi, P., Batista-Brito, R., Allene, C., Ribas, A., et al. (2011). Pioneer GABA cells comprise a subpopulation of hub neurons in the developing hippocampus. *Neuron* 71, 695–709. doi: 10.1016/j.neuron.2011.06.018
- Pouille, F., and Scanziani, M. (2001). Enforcement of temporal fidelity in pyramidal cells by somatic feed-forward inhibition. *Science* 293, 1159–1163. doi: 10.1126/science.1060342
- Rivera, C., Voipio, J., Payne, J. A., Ruusuvuori, E., Lahtinen, H., Lamsa, K., et al. (1999). The K⁺/Cl⁻ co-transporter KCC2 renders GABA hyperpolarizing during neuronal maturation. *Nature* 397, 251–255. doi: 10.1038/16697
- Safulina, V. F., Caiati, M. D., Sivakumaran, S., Bisson, G., Migliore, M., and Cherubini, E. (2010). Control of GABA release at mossy fiber-CA3 connections in the developing hippocampus. *Front. Synaptic Neurosci.* 2:1. doi: 10.3389/neuro.19.001.2010
- Safulina, V. F., Fattorini, G., Conti, F., and Cherubini, E. (2006). GABAergic signaling at mossy fiber synapses in neonatal rat hippocampus. *J. Neurosci.* 26, 597–608. doi: 10.1523/JNEUROSCI.4493-05.2006
- Safulina, V. F., Zacchi, P., Tagliatalata, M., Yaari, Y., and Cherubini, E. (2008). Low expression of Kv7/M channels facilitates intrinsic and network bursting in the developing rat hippocampus. *J. Physiol.* 586, 5437–5453. doi: 10.1113/jphysiol.2008.156257
- Sale, A., Berardi, N., Spolidoro, M., Baroncelli, L., and Maffei, L. (2010). GABAergic inhibition in visual cortical plasticity. *Front. Cell. Neurosci.* 4:10. doi: 10.3389/fncel.2010.00010
- Santhakumar, V., Jones, R. T., and Mody, I. (2010). Developmental regulation and neuroprotective effects of striatal tonic GABAA currents. *Neuroscience* 167, 644–655. doi: 10.1016/j.neuroscience.2010.02.048
- Schousboe, A. (2003). Role of astrocytes in the maintenance and modulation of glutamatergic and GABAergic neurotransmission. *Neurochem. Res.* 28, 347–352. doi: 10.1023/A:1022397704922
- Sebe, J. Y., Looke-Stewart, E. C., Estrada, R. C., and Baraban, S. C. (2010). Robust tonic GABA currents can inhibit cell firing in mouse newborn neocortical pyramidal cells. *Eur. J. Neurosci.* 32, 1310–1318. doi: 10.1111/j.1460-9568.2010.07373.x
- Semyanov, A., Walker, M. C., and Kullmann, D. M. (2003). GABA uptake regulates cortical excitability via cell type-specific tonic inhibition. *Nat. Neurosci.* 6, 484–490. doi: 10.1038/nn1043
- Semyanov, A., Walker, M. C., Kullmann, D. M., and Silver, R. A. (2004). Tonic active GABAA receptors: modulating gain and maintaining the tone. *Trends Neurosci.* 27, 262–269. doi: 10.1016/j.tins.2004.03.005

- Sipilä, S., Huttu, K., Voipio, J., and Kaila, K. (2004). GABA uptake via GABA transporter-1 modulates GABAergic transmission in the immature hippocampus. *J. Neurosci.* 24, 5877–5880. doi: 10.1523/JNEUROSCI.1287-04.2004
- Sipilä, S. T., Huttu, K., Soltesz, I., Voipio, J., and Kaila, K. (2005). Depolarizing GABA acts on intrinsically bursting pyramidal neurons to drive giant depolarizing potentials in the immature hippocampus. *J. Neurosci.* 25, 5280–5289. doi: 10.1523/JNEUROSCI.0378-05.2005
- Sipilä, S. T., Huttu, K., Yamada, J., Afzalov, R., Voipio, J., Blaesse, P., et al. (2009). Compensatory enhancement of intrinsic spiking upon NKCC1 disruption in neonatal hippocampus. *J. Neurosci.* 29, 6982–6988. doi: 10.1523/JNEUROSCI.0443-09.2009
- Sipilä, S. T., Voipio, J., and Kaila, K. (2007). GAT-1 acts to limit a tonic GABAA current in rat CA3 pyramidal neurons at birth. *Eur. J. Neurosci.* 25, 717–722. doi: 10.1111/j.1460-9568.2007.05342.x
- Song, I., Savtchenko, L., and Semyanov, A. (2011). Tonic excitation or inhibition is set by GABA(A) conductance in hippocampal interneurons. *Nat. Commun.* 2, 376. doi: 10.1038/ncomms1377
- Stell, B. M., Brickley, S. G., Tang, C. Y., Farrant, M., and Mody, I. (2003). Neuroactive steroids reduce neuronal excitability by selectively enhancing tonic inhibition mediated by delta subunit-containing GABAA receptors. *Proc. Natl. Acad. Sci. U.S.A.* 100, 14439–14444. doi: 10.1073/pnas.2435457100
- Stell, B. M., and Mody, I. (2002). Receptors with different affinities mediate phasic and tonic GABAA conductances in hippocampal neurons. *J. Neurosci.* 22, RC223.
- Triller, A., and Choquet, D. (2005). Surface trafficking of receptors between synaptic and extrasynaptic membranes: and yet they do move! *Trends Neurosci.* 28, 133–139. doi: 10.1016/j.tins.2005.01.001
- Tyzio, R., Represa, A., Jorquera, I., Ben-Ari, Y., Gozlan, H., and Aniksztejn, L. (1999). The establishment of GABAergic and glutamatergic synapses on CA1 pyramidal neurons is sequential and correlates with the development of the apical dendrite. *J. Neurosci.* 19, 10372–10382.
- Valeeva, G., Abdullin, A., Tyzio, R., Skorinkin, A., Nikolski, E., Ben-Ari, Y., et al. (2010). Temporal coding at the immature depolarizing GABAergic synapse. *Front. Cell. Neurosci.* 4:17. doi: 10.3389/fncel.2010.00017
- Wall, M. J., and Usowicz, M. M. (1997). Development of action potential-dependent and independent spontaneous GABAA receptor-mediated currents in granule cells of postnatal rat cerebellum. *Eur. J. Neurosci.* 9, 533–548. doi: 10.1111/j.1460-9568.1997.tb01630.x
- Wang, D. D., and Kriegstein, A. R. (2008). GABA regulates excitatory synapse formation in the neocortex via NMDA receptor activation. *J. Neurosci.* 28, 5547–5558. doi: 10.1523/JNEUROSCI.5599-07.2008
- Wang, D. D., and Kriegstein, A. R. (2009). Defining the role of GABA in cortical development. *J. Physiol.* 587, 1873–1879. doi: 10.1113/jphysiol.2008.167635
- Wang, D. D., and Kriegstein, A. R. (2011). Blocking early GABA depolarization with bumetanide results in permanent alterations in cortical circuits and sensorimotor gating deficits. *Cereb. Cortex* 21, 574–587. doi: 10.1093/cercor/bhq124
- Woodin, M. A., Ganguly, K., and Poo, M. M. (2003). Coincident pre- and postsynaptic activity modifies GABAergic synapses by postsynaptic changes in Cl⁻ transporter activity. *Neuron* 39, 807–820. doi: 10.1016/S0896-6273(03)00507-5
- Wu, Y., Wang, W., and Richerson, G. B. (2001). GABA transaminase inhibition induces spontaneous and enhances depolarization-evoked GABA efflux via reversal of the GABA transporter. *J. Neurosci.* 21, 2630–2639.
- Yamada, J., Furukawa, T., Ueno, S., Yamamoto, S., and Fukuda, A. (2007). Molecular basis for the GABAA receptor-mediated tonic inhibition in rat somatosensory cortex. *Cereb. Cortex* 17, 1782–1787. doi: 10.1093/cercor/bhl087
- Yamada, J., Okabe, A., Toyoda, H., Kilb, W., Luhmann, H. J., and Fukuda, A. (2004). Cl⁻ uptake promoting depolarizing GABA actions in immature rat neocortical neurons is mediated by NKCC1. *J. Physiol.* 557, 829–841. doi: 10.1113/jphysiol.2004.062471

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