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# COMMENTARY
# $GABA_A$ receptor diversity revealed in freeze-fracture replica (Commentary on Kasugai *et al.*)


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GABAergic inputs from at least 18 types of inhibitory interneurons regulate and coordinate the activity of pyramidal cells in the hippocampal area CA1 (Klausberger *et al.*, 2005), which in turn express at least 14 subunits of the $GABA_A$ receptor ($GABA_AR$) with varying affinity to GABA and other ligands (Persohn *et al.*, 1992; Wisden *et al.*, 1992; Sperk *et al.*, 1997; Ogurusu *et al.*, 1999). Thus, the subunit composition determines the local response of the $GABA_AR$ to synaptically released GABA. Elucidating the subunit composition of synaptic and extrasynaptic $GABA_AR$ is also crucial in understanding the phasic vs. tonic postsynaptic responses evoked by GABA.

In their elegant study, Kasugai *et al.* (2010) performed a series of double-labeling experiments to demonstrate for the first time that virtually all somatic inhibitory synapses in the rat hippocampal CA1 pyramidal cell contained $\alpha$1, $\alpha$2, and $\beta$3 subunits of $GABA_AR$. The authors developed a new antibody against the $\alpha$1 subunit to be used for a sensitive immunocytochemical method, freeze-fracture replica-immunogold labeling, that allows for quantitative analyses of transmembrane protein distribution (Fujimoto, 1995; Masugi-Tokita & Shigemoto, 2007). Their finding is consistent with previous post-embedding immunogold studies (Nusser *et al.*, 1996; Somogyi *et al.*, 1996). The presence of the three subunits in all somatic synapses does not imply that all synaptic $GABA_ARs$ consist of these subunits. However, this finding raises an interesting question regarding the relative proportion of the subunits at these synapses, because previous studies showed that $\alpha$1 and $\alpha$2 subunits preferentially mediate inputs from fast-spiking and regular-spiking basket cells, respectively (Pawelzik *et al.*, 1999; Thomson *et al.*, 2000; Nyiri *et al.*, 2001; Klausberger *et al.*, 2002).

Kasugai *et al.* (2010) also conducted single-labeling experiments to examine carefully the density of $\alpha$1, $\alpha$2 and $\beta$3 subunits in the synaptic and extrasynaptic plasma membrane of the pyramidal cells. Thirty to 50% of total labeling was found in synapses with 50–70% being extrasynaptic, suggesting that these subunits are well distributed between synaptic and extrasynaptic membrane. This is perhaps not too surprising as $GABA_ARs$ diffuse laterally in the plasma membrane (Bannai *et al.*, 2009). However, in light of previous studies suggesting that tonic inhibition is mediated by extrasynaptic $GABA_ARs$ containing the $\alpha$4, $\alpha$5, $\alpha$6 and/or $\delta$ subunits (Belelli *et al.*, 2009), one might wonder how $\alpha$1- or $\alpha$2-containing extrasynaptic receptors are different from the synaptic ones in their subunit composition, targeting and functions.

Elucidating the native subunit composition of $GABA_ARs$ at identified synapses and extrasynaptic membrane will require a combination of research tools, including subunit-specific antibodies against different epitopes (intracellular vs. extracellular), transgenic animals (with specific subunits deleted or mutated in specific neuronal populations) and subunit-specific allosteric modulators. Studies on the interactions between specific subunit and scaffolding proteins would also provide insight into the mechanisms that target and regulate $GABA_ARs$. The data from this study provide a solid anatomical substrate to understand the functions of $\alpha$1, $\alpha$2 and $\beta$3 subunits in synaptic and extrasynaptic plasma membrane. They begin to elucidate how a diverse population of $GABA_ARs$ and inhibitory interneurons may shape cortical network activity in health and disease.

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