Effects of GABA and glutamate uptake blockers on global background synaptic activity and excitability in entorhinal cortical neurones in vitro

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Cortical neurones are subject to a continuous level of background synaptic activity resulting from spontaneous release of both glutamate and GABA from presynaptic terminals. The balance between the resultant excitation and inhibition is important in determining neuronal excitability and thus network responsiveness. Modification of background activity may therefore be of importance in the actions of CNS active drugs. We have been using a novel approach (Rudolph *et al*., 2004) to mathematically estimate total global background excitatory (E_{Bg}) and inhibitory (I_{Bg}) conductances in cortical neurones from fluctuations in membrane potential recorded intracellularly in rat entorhinal cortical (EC) neurones *in vitro* (Greenhill and Jones, 2007). In a previous communication, we showed that the anticonvulsant, lamotrigine, concurrently increases I_{Bg} and decreases E_{Bg}. Here, we have studied the effect of another anticonvulsant, tiagabine, whose clinical action is thought to result from potentiation of inhibitory transmission via blockade of GABA uptake. For comparison we have studied the effect of a glutamate uptake blocker, L-*trans*-pyrrolidine-dicarboxylate (PDC). Intracellular recordings were made from pyramidal cells in layer III in slices of EC obtained from male Wistar rats (50-60g), and Ebg and IBg derived as described previously (Greenhill and Jones, 2007). Cellular excitability was determined from spike threshold and the number of spikes evoked by a supra-threshold depolarizing current pulse.

Tiagabine (4 μM) caused a profound rise in I_{Bg} from 8.1±2.2 nS to 48.9±16.8 nS (mean± sem; $P<0.01$, paired t-test, n=6. Unexpectedly, however, E_{Bg} also increased (1.8± 0.5 to 3.9±1.0 nS; $P<0.05$). Overall, the I:E ratio increased heavily in favour of inhibition, from 4.5±0.3 to 12.6±4.5. Concurrently, spike threshold increased from 18.8±0.6 mV (positive to rest) to 25.0±0.6 mV ($P<0.001$) and the number of spikes evoked during a depolarizing current pulse fell from 3.5±0.4 to 1.3±0.2. Patch clamp recordings (not shown) actually showed a slightly decreased frequency of spontaneous inhibitory postsynaptic currents but an increase in amplitude and duration. Excitatory currents increased in frequency with little change in amplitude or duration. PDC increased both E_{Bg} (from 1.0±0.2 to 1.9±0.6 nS; $P=0.05$, n=6) and I_{Bg} (3.9±1.0 to 8.5 ±1.4 nS; $P>0.05$) with the I:E ratio remaining the same (4.0±0.5 *v* 3.9±0.9). Neither spike threshold (25±0.5 *v* 24.3±0.6 mV), nor number of spikes 3.5±0.4 *v* 4.1±0.5) were significantly altered. Patch clamp recordings showed a marked increase in frequency of spontaneous excitatory currents, whilst both amplitude and frequency of inhibitory currents was increased (not shown).

The results show that it is important to determine overall network effects when considering the effects of anticonvulsants. Although tiagabine caused an unexpected increase in background excitation, it decreased cellular excitability consistent with its effects on overall the balance between excitation and inhibition.

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