Regulation by hypocretin (orexin) of excitatory postsynaptic potentials in layer V pyramidal neurons of murine prefrontal cortex

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Hypocretin 1 and 2 or Orexin (Orx) A and B are neuropeptides synthesized by lateral hypothalamic neurons, most active during waking, and silent during sleep. By innervating subcortical arousal nuclei, Orx cells set the level of cortical activation necessary for exploratory goal-oriented behaviors, in response to physiological or emotional drives. Moreover, by innervating PFC, Orx neurons directly regulate cognitive tasks.

Orx actions are mediated by Orx receptor 1 (Ox1R) and 2 (Ox2R), but little is known about how Orxs regulate neocortical circuits. We studied the effect of OrxA on glutamate release in PFC frontal area 2 (Fr2), a premotor region implicated in goal-oriented behaviors.

In slices from 4 to 9 weeks old mice, OrxA (500 nM) increased the frequency of spontaneous excitatory postsynaptic currents (EPSCs) on layer V pyramidal neurons, from 14 ± 5.5 to 19.2 ± 6.1 Hz ($p < 0.05$; paired t-test, $n = 7$), with no effect on EPSC amplitude. The effect was also produced by 100 nM OrxA and was abolished by 1 μM SB-334867 (an antagonist of Ox1R).

Since OrxA could exert its excitatory effect on both pre- and postsynaptic sites, it has been tested in presence of TTX 1 μM to evaluate miniature EPSCs. Their mean frequency was increased by OrxA 50 nM in pro-excitatory conditions, suggesting a pre-synaptic action, as well as their median amplitude, pointing to some post-synaptic mechanism to be elucidated.

At last, OrxA was tested in combination with nicotine or acetylcholine, in order to assess the effect of these two wake-promoting neurotransmitters acting together. Orexinergic and nicotinic action proved to be excitatory and additive.

Our results support the notion that OrxA exerts its excitatory effect on layer V Py neurons through an Ox1R-dependent mechanism, which could contribute to sustain neocortex activity during waking. Moreover, considering the implication of pyramidal neuron firing in θ rhythms, our data suggest how an OrxA deficit could decrease EEG θ activity during wakefulness. The fact that nicotinic and orexinergic signalling are additive on the same pyramidal neurons also shows that their interaction in wakefulness regulation could exert single-cell effects as well as global ones on rhythmical activity on a whole-brain scale.