Stimulation of the α2a Adrenergic Receptor in the Ventrolateral Preoptic Area Promotes Arousal

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Introduction: Anesthetic induced hypnosis arises in part from specific actions of anesthetic drugs upon the endogenous circuits regulating sleep and wakefulness. We have previously demonstrated that isoflurane directly depolarizes sleep-promoting ventrolateral preoptic (VLPO) neurons. Neighboring non-sleep active VLPO neurons are not depolarized by general anesthetics. However, the behavioral significance of these effects has been called into question. We hypothesized that acute pharmacologic modulation of adrenergic signaling in VLPO would counteract anesthetic-induced hypnosis both ex vivo and in vivo.

Methods: Whole-cell current clamp recordings were conducted on 200µm VLPO-containing slices obtained from C57B6/J mice. Cells were categorized as putative sleep-promoting based upon a hyperpolarizing response to norepinephrine (NE). To determine the mechanism of adrenergic-induced hyperpolarization, the highly specific alpha2A agonist, dexmedetomidine was bath applied. In 7/7 NE hyperpolarized VLPO neurons, 100nM dexmedetomidine also elicited a hyperpolarization (-43±2.7mV to -50.0±2.3mV, p=0.0014). Multiplex RT-PCR performed on cytoplasmic aspirates from single neurons confirmed the presence of alpha2A, 2B, and 2C adrenoceptors in the dexmedetomidine-hyperpolarized neurons. Conversely, in 3/3 NE depolarized VLPO neurons dexmedetomidine did not significantly alter resting membrane potential.

Results: Having demonstrated that dexmedetomidine hyperpolarizes putative sleep-promoting neurons, we explored the effects of adrenergic ligands in vivo. Indwelling bilateral cannulae were used to deliver 25nl of adrenergic drugs into VLPO of 0.8% isoflurane-anesthetized mice or into mice with bilateral cannulae implanted 500um more caudally. Arousal state behavioral scores ranging from 0 (no movement) to 4 (full return of righting reflex) were assigned for the 10-minute period prior to drug and the 5-minutes following drug.

Conclusions: Dexmedetomidine infusion significantly increased arousal, while saline, α1 agonist phenylephrine, and NE did not ($p<0.05$). Dexmedetomidine failed to rouse animals exposed to a deeper state of isoflurane anesthesia. These results suggest that stimulation of α2A adrenergic receptors in VLPO promotes arousal.

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Trainee Award Eligible