

# LOCAL ETHANOL PROMOTES EXTRACELLULAR ACCUMULATION OF ADENOSINE IN THE OREXINERGIC PERIFORNICAL HYPOTHALAMUS

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Ethanol promotes sleep. However, the neuroanatomical site where ethanol acts to modulate sleep/wakefulness is unknown. The perifornical hypothalamus (PFH) contains wake-promoting orexinergic neurons. Studies from our laboratory and others have reported that adenosine may regulate sleep by modulating the activity of orexin neurons via A1 receptors. Previous studies performed in cell cultures suggest that ethanol inhibits adenosine uptake. Does ethanol block the uptake of adenosine in vivo, and increase extracellular levels of adenosine in PFH?

**Methods:** Male Sprague-Dawley rats were unilaterally implanted with guide cannula targeted toward the PFH. Following post-operative recovery, microdialysis probe was inserted and artificial cerebrospinal fluid (aCSF) was perfused (flow rate =0.7  $\mu\text{L}/\text{min}$ ). Following probe insertion recovery, the experiment was begun at the dark onset. aCSF was perfused for 80 min and 4x 20 min samples were collected. Subsequently, 3 doses of ethanol (30, 100 and 300 mM) were perfused. Each dose was perfused for 80 min and 4 x 20 min samples were collected. On completion, the animals were euthanized, brains removed and processed for orexin immunohistochemistry (IH) to localize the probe sites. Cresyl violet staining was done to evaluate potential neurotoxicity caused due to ethanol perfusion.

**Results:** Local ethanol perfusion increased the extracellular levels of adenosine in the PFH. (N=4; stats pending). Orexin IH revealed that all probe sites were localized in the orexinergic PFH. Cresyl violet staining did not show any neurotoxicity at perfusion sites.

**Conclusions:** Local perfusion of ethanol in PFH increases extracellular adenosine. Increased adenosine may mediate ethanol induced sleep promotion.