

## **Alcohol consumption impairs the ependymal cilia motility in the brain ventricles**

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### **Movie Legends**

**Movie 1. Type I ependymal cilia has a minimum beating frequency of 60 Hz and a maximum beating angle of 90°.** Type I ependymal cilia beating characteristics are recorded. Ependymal cells in the brain lateral ventricle are captured at 200 frames per second.

**Movie 2. Type II ependymal cilia has a beating frequency between 30 and 60 Hz and a beating angle between 90° and 135°.** Type II ependymal cilia beating characteristics are recorded. Ependymal cells in the lateral ventricle are captured at 200 frames per second.

**Movie 3. Type III ependymal cells has a maximum beating frequency of 30 Hz and a minimum beating angle of 135°.** Type III ependymal cilia beating characteristics are recorded. Ependymal cells in the lateral ventricle are captured at 200 frames per second.

**Movie 4. Type I ependymal cilia beating decreased following *ex vivo* ethanol treatment.** The brain slice was pre-treated with 0.25% ethanol and type I ependymal cilia beating in the brain lateral ventricle were captured at 200 frames per second.

**Movie 5. Type II ependymal cilia beating decreased following *ex vivo* ethanol treatment.**

The brain slice was pre-treated with 0.25% ethanol and type II ependymal cilia beating in the brain lateral ventricle were captured at 200 frames per second.

**Movie 6. Type III ependymal cilia beating decreased following *ex vivo* ethanol treatment.**

The brain slice was pre-treated with 0.25% ethanol and type III ependymal cilia beating in the brain lateral ventricle were captured at 200 frames per second.

**Movie 7. Type I ependymal cilia has a minimum beating frequency of 60 Hz and a maximum beating angle of 90° in the rat brain lateral ventricle.** Type I ependymal cilia beating characteristics are recorded. The control rat group was treated with water by oral gavage for seven days. Type I ependymal cilia beating in the brain lateral ventricle are captured at 200 frames per second.

**Movie 8. Type I ependymal cilia beating in the rat brain lateral ventricle decreased following *in vivo* ethanol treatment.** The treatment rat group was given 95% ethanol by oral gavage at 6g per kg. Type I ependymal cilia beating in the brain lateral ventricle are captured at 200 frames per second.

**Movie 9. Type II ependymal cilia has a beating frequency between 30 and 60 Hz and a beating angle between 90° and 135° in the rat brain lateral ventricle.** The control rat group was treated with water by oral gavage for seven days. Type II ependymal cilia beating characteristics are recorded. Type I ependymal cilia beating in the brain lateral ventricle are captured at 200 frames per second.

**Movie 10. Type II ependymal cilia beating in the rat brain lateral ventricle decreased following *in vivo* ethanol treatment.** The treatment rat group was given 95% ethanol by oral gavage at 6g per kg. Type II ependymal cilia beating in the brain lateral ventricle are captured at 200 frames per second.

**Movie 11. Type I ependymal cilia has a minimum beating frequency of 60 Hz and a maximum beating angle of 90° in the rat brain third ventricle.** Type I ependymal cilia beating characteristics are recorded. The control rat group was treated with water by oral gavage for seven days. Type I ependymal cilia beating in the brain third ventricle are captured at 200 frames per second.

**Movie 12. Type I ependymal cilia beating in the rat brain third ventricle decreased following *in vivo* ethanol treatment.** The treatment rat group was given 95% ethanol by oral gavage at 6g per kg. Type I ependymal cilia beating in the brain third ventricle are captured at 200 frames per second.

**Movie 13. Type II ependymal cilia has a beating frequency between 30 and 60 Hz and a beating angle between 90° and 135° in the rat brain third ventricle.** The control rat group was treated with water by oral gavage for seven days. Type II ependymal cilia beating characteristics are recorded. Type I ependymal cilia beating in the brain third ventricle are captured at 200 frames per second.

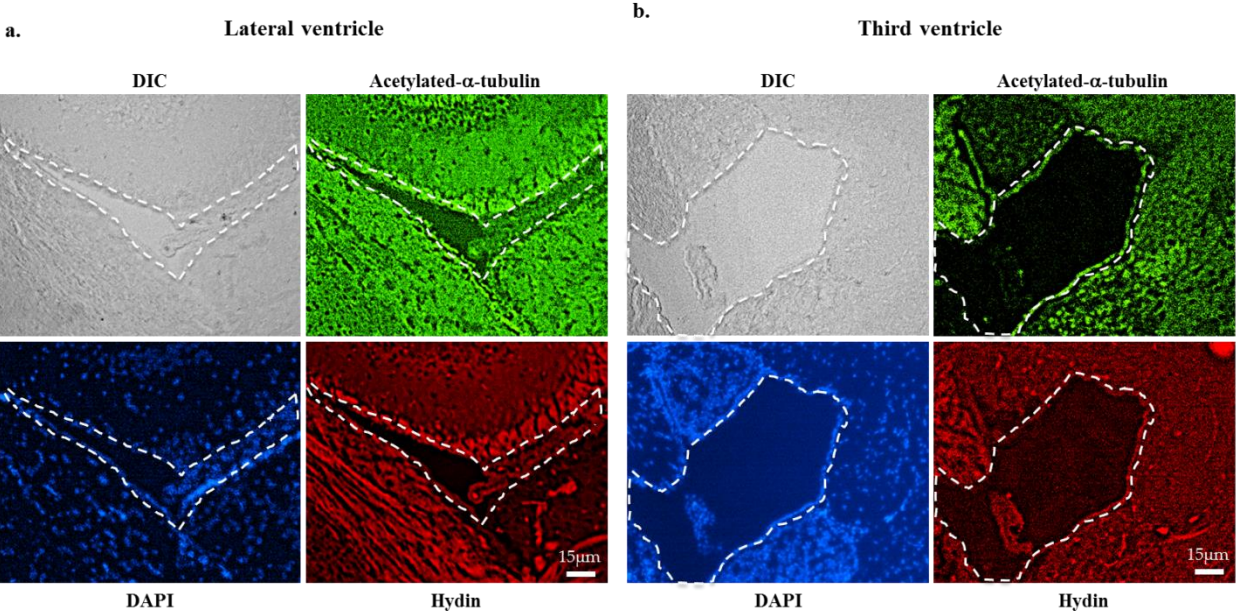
**Movie 14. Type II ependymal cilia beating in the rat brain third ventricle decreased following *in vivo* ethanol treatment.** The treatment rat group was given 95% ethanol by oral gavage at 6g per kg. Type II ependymal cilia beating in the brain third ventricle are captured at 200 frames per second.

### **Supplementary Figure Legends**

**Supplementary Figure S1. Ependymal cilia in the mouse brain line the lateral and third ventricles.** Shown here are ependymal cells from, **a.** the lateral and, **b.** the third ventricle of a mouse brain. The brain sections were stained with acetylated- $\alpha$ -tubulin, a ciliary marker; hydin, an axonemal protein and counter-stained with DAPI, a nucleus marker. Individual differential interference contrast (**top left panel**), and fluorescence (**other panels**) images shown are captured at low magnification (10x). Bar=30  $\mu$ m.

**Supplementary Figure S2. Ciliary structural and functional proteins are expressed in ependymal cilia of mouse brain.** Shown here are high-resolution fluorescent microscopy images of ependymal cells from the lateral ventricle of a mouse brain. **a.** The brain section was stained with acetylated- $\alpha$ -tubulin, a ciliary marker, and counter-stained with DAPI, a nucleus marker. **b.** The brain section was stained with acetylated- $\alpha$ -tubulin, a ciliary marker, hydin, a central axonemal marker and counter-stained with DAPI, a nucleus marker. **c.** The brain section was stained with acetylated- $\alpha$ -tubulin, a ciliary marker, polycystin-2, a mechanosensory ciliary protein and counter-stained with DAPI, the nuclear marker. The images were captured at 60, 100 and 150x magnifications. Bar=15  $\mu$ m.

Supplementary Figure S1



Supplementary Figure S2

