

Supporting Information

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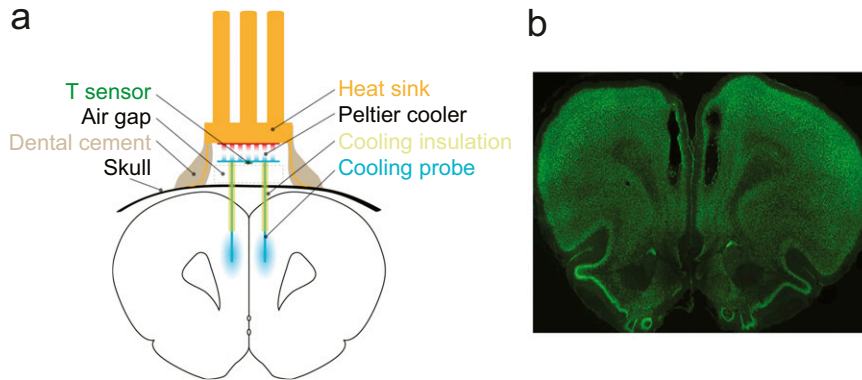


Fig. S1. (A) Schematic diagram of the cooling device used in current study. (B) Coronal section of the rat brain showing the track of cooling probes after the experiments.

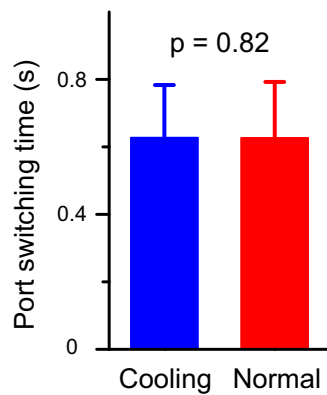


Fig. S2. Cooling of mPFC has no effect on general movement as measured by port-switching time.

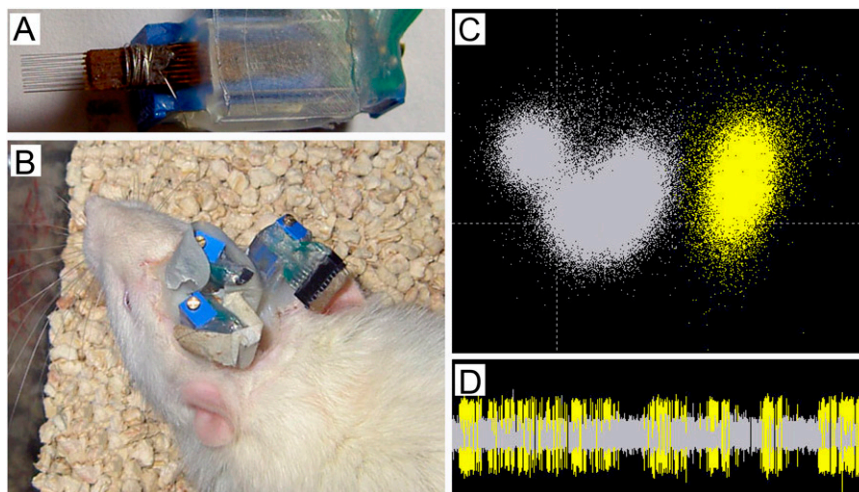
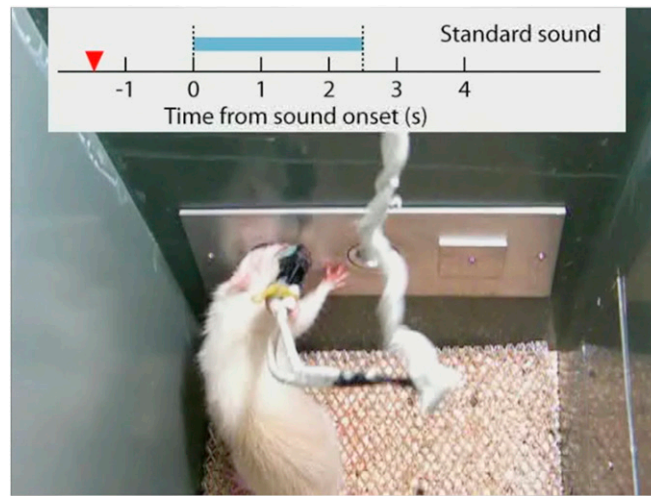


Fig. S3. Single-unit recording in free-moving rats. (A) The 32-channel microwire-array electrodes used for recording medial prefrontal cortex (mPFC) activities. (B) The rat with microwire-array electrodes implanted. (C and D) An example of the single unit (marked yellow) isolated manually by using principle component analysis-based clustering method.



Movie S1. Rat performing interval-timing task for 2.5-s time estimation, after it was well trained. The time of waiting port (on the right side) entry and exit are marked by “green” and “blue” triangles. In instructive trials, a standard sound (2.5-s white noise, instructive sound) was given (sound onset at $t = 0$). In test trials (randomly interleaved), a prolonged sound (3.5-s white noise, test sound) was given. The rat was required to exit the waiting port within a fixed window around 2.5-s (reward window, 2.25–3.5 s) to receive the water reward in the reward port (on the left side). Exit times for instructive and test trials were very similar (see Fig. 1 *B* and *C* for an example), indicating the rat was correctly making time estimation during the test trials.

[Movie S1](#)