Cell Reports Methods

CellPress
OPEN ACCESS

Preview

A signal EMerGes from the noise

Hongshen He[1] and Thomas J. McHugh[1,*]
[1]Laboratory for Circuit & Behavioral Physiology, RIKEN Center for Brain Science, 2-1 Hirosawa, Wako-shi, Saitama, Japan
*Correspondence: thomas.mchugh@riken.jp
https://doi.org/10.1016/j.crmeth.2023.100510

In this issue of *Cell Reports Methods*, Osanai et al. report an innovative approach to extract an electromyography (EMG) signal from multi-channel local field potential (LFP) recordings using independent component analysis (ICA). This ICA-based approach offers precise and stable long-term behavioral assessment, eliminating the need for direct muscular recordings.

Electromyography (EMG) is a powerful technique for capturing the electrical activity generated by skeletal muscles.[1] It has been widely used in clinical studies involving humans, such as for monitoring disease progression or assessing and identifying treatment strategies for motor diseases.[2,3] Because EMG is generated by muscle cells within the motor system, it is not only used as a diagnostic tool but also extensively employed in laboratory studies to monitor animals' behavioral status. It can be particularly valuable for detecting subtle behavioral changes that are often overlooked by video tracking methods. In parallel, many EMG experiments also record local field potentials (LFP), transient electrical signals generated within the brain, to characterize brain states and correlate with behavior. Consequently, the combination of LFP recordings and EMG signals has proved invaluable as a tool to accurately classify animals' behavioral states during free movement,[4] fear behaviors,[5] and across the sleep/wake cycle.[6] However, the surgical procedures for EMG recordings often involve cumbersome preparations, including separate wires and additional preamplifiers. These inconveniences, along with the risk of EMG wires breaking, can potentially disrupt an entire experiment.[7]

Research led by Osanai et al., published in *Cell Reports Methods*,[8] demonstrates a readily applicable method for extracting EMG from LFP signals without the need for direct muscle recordings. This was achieved using a blind source separation approach called independent component analysis (ICA). Unlike most conventional applications, which use ICA to remove "noise components" from field recordings, in this work the authors utilize this "noise" to reconstruct EMG signals. The study demonstrates that these EMG-reflective components can be applied to improve the accuracy and stability of behavioral quantification in animals.

The authors initially recorded LFP signals from multiple brain areas, using a skull screw placed above the cerebellum as an electrical reference; a key point, as this reference allows noise from the nearby muscles to be reflected in the data (Figure 1A). Simultaneously, they collected actual EMG signals from neck muscles to serve as a "ground-truth" comparison. Subsequently, ICA was applied to the LFP signals to extract a noise-like high-frequency component (Figure 1B). Interestingly, these noise-like components, which occurred synchronously across all channels, exhibited a similar peak frequency range (100–200 Hz) to the actual EMG signals and demonstrated a high amplitude correlation (>0.9) with the ground-truth EMG signals over time. Therefore, the noise component isolated through ICA, which the authors termed IC-EMG, could be considered a reliable proxy for EMG.

Next, the authors investigated the ability of the IC-EMG method to annotate animals' behavioral states across multiple experimental paradigms (Figure 1C). They first recorded LFPs and EMGs while a mouse traversed an open arena, using a video tracking camera to classify the animal's behavior. The authors demonstrated that IC-EMG not only accurately tracked diverse states, such as active movement, sleep, and quiet wakefulness (QAW), but also performed comparably to the real EMG signal. Next, the authors demonstrated that IC-EMG could serve as an indicator to detect fear-induced freezing, characterized by significantly lower signal amplitudes during freezing compared with non-freezing states. These experiments highlighted the strengths of this approach in annotating animal behaviors and its ability to preclude the additional surgical techniques required for traditional EMG recording.

To further demonstrate its capabilities, the authors tested whether IC-EMG could classify sleep stages. The conventional approach for classifying sleep cycles relies on a combination of LFP features and video tracking, a method that often struggles to separate QAW and rapid eye movement (REM) sleep accurately. Adding EMG signals has proven to be a more robust approach for scoring sleep stages.[9] The authors demonstrated that the IC-EMG-based method showed similar efficiency to the real EMG-based method, with fewer false-positive detections compared to the video-only approach. Altogether, these experiments underscore the advantages of IC-EMG in precisely and reliably annotating animal behavior, on par with real EMG signals. The method also holds potential for the re-evaluation of animal states in datasets previously acquired without EMG.

The authors also highlight two additional critical advantages of IC-EMG approach as a benchmarking technique for behavior quantification. Mechanically, the micro-drive used in their approach is considerably lighter in weight compared with typical methods for recording both neurophysiology and EMG. This reduction enables recording in younger animals with lighter body weights, expanding the scope of research possibilities. Moreover,

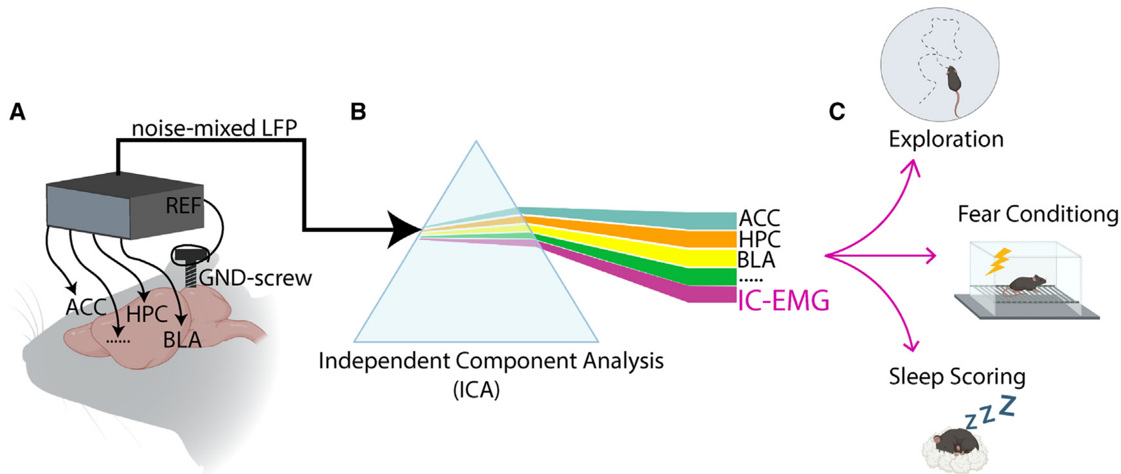

Figure 1. Schematic of an ICA-based framework for EMG extraction
(A) Experimental design: LFPs from multisite brain-area recordings are referenced to the skull screw.
(B) ICA framework: noise-mixed LFPs are fed into ICA algorithms to isolate the EMG component.
(C) Application of IC-EMG for behavior annotation.
ACC, anterior cingulate cortex; HPC, hippocampus; BLA, basolateral amygdala; GND, ground.

this approach eliminates the risk of wire breakage during vigorous behaviors, such as during foot shocks, ensuring stable measurements for long-term recordings. Experimentally, in addition to isolating EMG-like components, ICA also provides undistorted and noise-canceled LFP signals for further analyses. Finally, unlike typical reference estimate approaches such as current source density analysis, the IC-EMG approach does not require information about the spatial layout of the electrode sites and can be considered a smart way to reference the zero-activity location, avoiding signal distortion caused by nearby referencing.

Conducting safe and stable EMG recordings, particularly in laboratory rodents, poses various challenges. Inspired by the computational nature of ICA algorithms, Osanai et al. have provided an approach to isolate EMG-like signals without the need for additional surgical setups for EMG wire implantation. As such, this approach offers a protocol that is less stressful for the animal, which is particularly critical when studying specific disease mouse models characterized by high aggression levels[10] or motor syndromes resulting from muscle dysfunction.[11] Additionally, it offers a roadmap for integrating activity from both the central and peripheral systems to track physiological features for studying neurodevelopmental disorders and age-related changes throughout an animal's lifespan.

DECLARATION OF INTERESTS

The authors declare no competing interests.

REFERENCES

1. Buchthal, F. (1985). Electromyography in the evaluation of muscle diseases. Neurol. Clin. *3*, 573–598.
2. Benatar, M. (2006). A systematic review of diagnostic studies in myasthenia gravis. Neuromuscul. Disord. *16*, 459–467. https://doi.org/10.1016/j.nmd.2006.05.006.
3. Leblond, H., L'Esperance, M., Orsal, D., and Rossignol, S. (2003). Treadmill locomotion in the intact and spinal mouse. J. Neurosci. *23*, 11411–11419. https://doi.org/10.1523/JNEUROSCI.23-36-11411.2003.
4. Hadzipasic, M., Ni, W., Nagy, M., Steenrod, N., McGinley, M.J., Kaushal, A., Thomas, E., McCormick, D.A., and Horwich, A.L. (2016). Reduced high-frequency motor neuron firing, EMG fractionation, and gait variability in awake walking ALS mice. Proc. Natl. Acad. Sci. USA *113*, E7600–E7609. https://doi.org/10.1073/pnas.1616832113.
5. Steenland, H.W., and Zhuo, M. (2009). Neck electromyography is an effective measure of fear behavior. J. Neurosci. Methods *177*, 355–360. https://doi.org/10.1016/j.jneumeth.2008.10.020.
6. Weber, F., and Dan, Y. (2016). Circuit-based interrogation of sleep control. Nature *538*, 51–59. https://doi.org/10.1038/nature19773.
7. Whelan, P.J. (2003). Electromyogram recordings from freely moving animals. Methods *30*, 127–141. https://doi.org/10.1016/s1046-2023(03)00074-4.
8. Osanai, H., Yamamoto, J., and Kitamura, T. (2023). Extracting electromyographic signals from multi-channel LFPs using independent component analysis without direct muscular recording. Cell Reports Methods. *3*, 100482. https://doi.org/10.1016/j.crmeth.2023.100482.
9. Pace-Schott, E.F., and Hobson, J.A. (2002). The neurobiology of sleep: genetics, cellular physiology and subcortical networks. Nat. Rev. Neurosci. *3*, 591–605. https://doi.org/10.1038/nrn895.
10. Jager, A., Maas, D.A., Fricke, K., de Vries, R.B., Poelmans, G., and Glennon, J.C. (2018). Aggressive behavior in transgenic animal models: a systematic review. Neurosci. Biobehav. Rev. *91*, 198–217. https://doi.org/10.1016/j.neubiorev.2017.09.028.
11. Sartori, R., Romanello, V., and Sandri, M. (2021). Mechanisms of muscle atrophy and hypertrophy: implications in health and disease. Nat. Commun. *12*, 330. https://doi.org/10.1038/s41467-020-20123-1.