Is burst activity in cortical slices a representative model for epilepsy?

Wim van Drongelen[1], Henner Koch[2], Charles Marcuccilli[1], Kurt Hecox[1], and Jan-Marino Ramirez[2]

[1] The University of Chicago, Department of Pediatrics, 5841 South Maryland Avenue MC 3055, Chicago, IL 60637.

[2] The University of Chicago, Department of Organismal Biology and Anatomy, 1027 E-57th Street, Chicago, IL 60637.

Summary

Synchronized population activity is one of the hallmarks of information processing in the mammalian central nervous system. Thalamocortical rhythms are characterized by bursts of activity in large populations of synchronized neurons. A fundamental property of this network activity is that the degree to which these neuronal populations are coordinated is state-dependent. The sleep state is characterized by a slow and highly coordinated activation of millions of neurons, the wake state is characterized by faster and much less coordinated activity of neurons. Synchronization can also be pathological. Synchronized hyperactivity in neural networks and high levels of cellular depolarization are processes frequently associated with seizures, e.g. [1], [3]. At a macroscopic level, clinical EEG and EcoG recordings demonstrate that during seizure onset,

synchronization between different locations increases. Although these activity patterns are fingerprints of seizures, one of the real challenges of clinical research is to discriminate synchronized activity occurring during an epileptic state from synchronized activity occurring during other physiological states of cortical activity. In the present study, we use a cortical slice preparation of mice to study different types of synchronized rhythmic activity.

Neonatal (P7-11) males or females of CD-1 mice were deeply anesthetized with ether, decapitated at the C3/C4 spinal level and the cortex isolated in ice-cold artificial cerebrospinal fluid (a-CSF). One hemisphere was then glued onto an agar block with its rostral end up and mounted into a vibrating tissue slicer. Thin slices were serially sectioned from rostral to caudal 1500μm from the frontal pole. Slices (500 μm thick) were sectioned serially from rostral to caudal encompassing the motor cortex of the mouse and stored in an oxygenated perfusion chamber. One slice taken at this region was immediately transferred into a recording chamber where it was submerged under a stream of a-CSF (temperature, 30^{o}C; flow rate, 10 ml/min) containing (mM): 118 NaCl, 3 KCl, 1.5 $CaCl_2$, 1 $MgSO_4$, 25 $NaHCO_3$, 1 NaH_2PO_4 and 30 D-glucose equilibrated with carbogen (95% O_2 and 5% CO_2) at 27^{o}C (pH 7.4). After 30 min the potassium

concentration was raised from 3 to 5 mM over another 30 min to obtain spontaneous rhythmic activity.

Population activity was recorded extracellularly with suction electrodes positioned onto the surface of the cortical layers 4/5. The signals were amplified 2000 times, filtered between 0.25 and 1.5 kHz, rectified and integrated using an electronic filter (time constant of 30-50 ms). Intracellular patch-clamp recordings were obtained from cortical neurons with the blind-patch technique. The patch electrodes were manufactured from filamented borosilicate glass tubes (Clarke GC 150TF), filled with a solution containing (mM): 140 K-gluconic acid, 1 $CaCl_2*6H_2O$, 10 EGTA, 2 $MgCl_2*6H_2O$, 4 Na_2ATP, 10 HEPES. Electrodes with a positive pressure of 35-50 mm Hg, were penetrated deep into the slice in 10 um steps using a piezo-driven micro-manipulator (Böhm, Germany). As the electrode approaches the cell, the measured electrode resistance increases. To obtain a Giga seal, we removed the positive pressure and applied negative suction. After obtaining a seal, somatic recordings are distinguished from intra-axonal recordings by the shape of the action potential and presence of synaptic activity. All recordings were low-pass filtered (0-2 kHz, Bessel 4-pole filter, -3dB).

Signal processing was applied to describe the entropy and information content in the signals during the different bursting patterns. The signals were averaged on the basis of the activity bursts in the population, at random, and on the basis of spike activity. In addition, spike activity in each of the trials in the average was determined. To study information content in the cellular activity, entropy was calculated using Shannon's formula for entropy:

$$H = -\sum_{i=0}^{inf} p(i) \cdot \log_2(p(i))$$

, where H is the entropy in bits and p(i) the probability of observing the ith activity pattern. We determined total entropy, noise entropy and their difference (a measure for information rate) with the method described by Reich and coworkers [2]. These metrics were computed for each type of averaged signal.

At concentrations between 3-5 mM K^+ in the a-CSF, the slice preparation generated rhythmic synchronized activity at the single cell and population level. This activity resembles synchronized activity randomly occurring under physiological conditions. Typically, the frequency of the population's activity bursts was around 0.2 Hz. Following the blockade of

GABAergic synaptic transmission with bicuculline (20uM), the synchronized activity changes significantly. Large amplitude bursts of synchronized population activity are generated in trains of 15 – 25 repetitive bursts at a frequency of about 0.5 Hz, followed by a phase of ongoing activity of repetitive bursts at a frequency of ~ 0.1 Hz. The regular activity after adding bicuculline, resembles seizure like activity. Using this model, we explore the ability of entropy of spike activities, noise entropy, and information content to distinguish between different types of bursting activity. The entropy of the cell's activity that relates to the burst activity in the population is at the same level before and after the dis-inhibition. However, the inter-trial variation in the action potential patterns is considerably reduced after adding the GABA-A blocking agent. The total entropy, representing the potential maximum information content, therefore is decreased after the onset of seizure-like bursts.

At this stage, we cannot produce a definite answer to the question in our title, however, our data supports that bursting patterns associated with a low information content are a fingerprint of, and a promising model for epileptiform activity.

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References

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