SFN 32nd Annual Meeting Satellite Symposium

Recent Advances in Network Electrophysiology Using Multi-Electrode Arrays

Tuesday, Nov. 5, 2002, 6:30-9:00pm
Orange County Convention Center, Room 206A

6:30 Opening Remarks
Makoto Taketani
Tensor Biosciences, Irvine, CA

6:40 Brain-on-a-Chip for Better Mood-Boosters
Michel Baudry
UC Irvine / Tensor Biosciences, Irvine, CA

7:10 Long-term Recording in Cerebellar Slice Cultures Using Multi-Electrode Array
Amy Arai
Southern Illinois University School of Medicine, Springfield, IL

7:40 Network oscillations using multi-electrode arrays
Ole Paulsen
University of Oxford, Oxford, U.K.

8:10 Biological clock on an MED probe; circadian rhythm analysis using long-term culture of the suprachiasmatic nucleus
Sato Honma
Hokkaido University School of Medicine, Sapporo, JAPAN

8:40 Discussion

Brain-on-a-Chip^{TM} for Better Mood-Boosters

Michel Baudry, Yousheng Jia,, Ken Shimono, Michael Lee, Makoto Taketani and Gary Lynch.
Dept. of Psychiatry & Human Behavior, UCI, Tensor Biosciences, 101 Theory Drive, Irvine, CA 92612.

A major problem with current approaches for CNS drug discovery is the limited predictive values of most commonly used experimental models. On one hand, psychoactive drugs influence brain function by modifying brain network activity while most screening assays evaluate the effects of drug candidates at the molecular or cellular levels. On the other hand, most animal models of psychiatric diseases do not predict effects in human behavior and cannot be used for large-scale screening. Tensor is developing Brain-on-a-Chip technology as a new tool to facilitate CNS drug discovery. Applications of this technology for 2 separate areas of pharmacology will be illustrated. The first one concerns the identification of new serotonergic drugs that could be used as anti-anxiety or anti-depressants. The approach takes advantage of the use of Multi-Electrode Array System (MED64) to routinely measure rhythmic activity in hippocampal networks of acutely prepared hippocampal slices. We found that 5-HT suppresses carbachol-induced rhythmic activity in a concentration-dependent manner through the activation of 5-HT1a receptors. Using this approach, new and potent 5_HT1a agonists have been identified. The second area of application will be illustrated by studies of co-cultures of septal and hippocampal slices for the identification of compounds facilitating the establishment of functional connections between cholinergic neurons and their target neurons in hippocampus. Septal and hippocampal slices were cultured directly on MED64 probes and spontaneous as well as evoked electrical responses were chronically recorded in hippocampus and septum. Cholinergic responses were pharmacologically identified and studied in various culture media. These results illustrate the wide range of questions that can be addressed with this new technology and stress the importance of network pharmacology to understand the mechanisms of psychoactive drugs.

Long-Term Recording In Cerebellar Slice Cultures Using Multielectrode Array.

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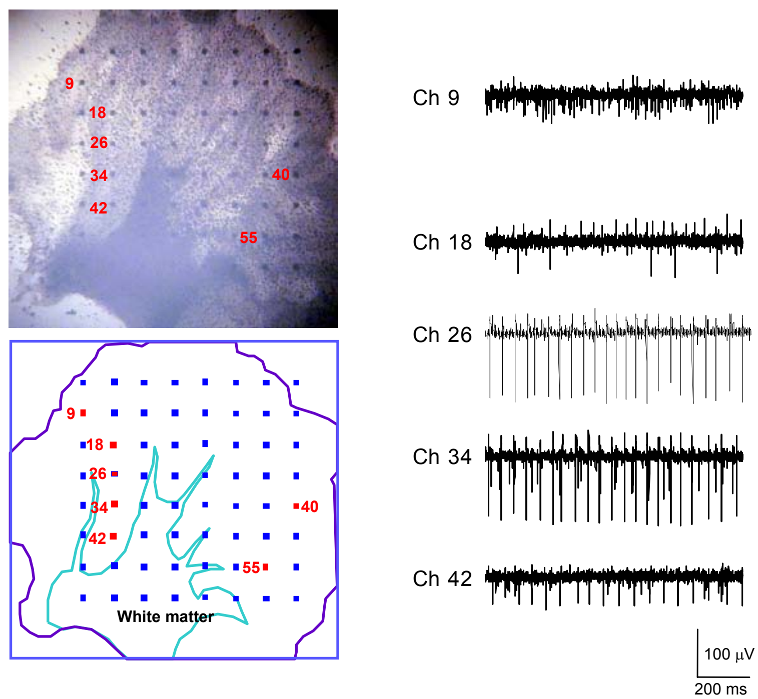
The combination of organotypic slice culture and multielectrode dish provides unique opportunities to record neuronal activity from brain slices for weeks to months. This allows us to study long-term consequences of drug treatment as well as changes after pathogenic challenges.

In the present study, we are using cultured cerebellar slices prepared from Sprague-Dawley rats of postnatal day 10-11. The slices remain viable for several months after being placed on the multielectrode dish and they contain various cell types, including Purkinje cells. The latter exhibit spontaneous firing that becomes very regular after blocking glutamatergic input, with spiking frequencies typically in the range of 10-40 Hz. These slices can thus readily be used to study factors which affect the activity of Purkinje cells, including the role of ion channels and the impact of various transmitter systems.

This presentation will mainly discuss the methods we use to prepare cultured cerebellar slices, to maintain them on the multielectrode dish, and to obtain stable long-term recordings.



Organotypic cerebellar slice culture grown on a multielectrode dish.
Left: Cerebellar slice culture on the multielectrode dish (top) and electrode arrangement (bottom).
Right: Representative example of spontaneous activity recorded from the same slice.

Network Oscillations Using Multi-Electrode Arrays

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Network activity in the brain is often oscillatory in nature. Recent *in vitro* models have successfully reproduced physiologically relevant network oscillations in acute brain slice preparations, which have been studied conventionally with one or two extracellular electrodes. Planar multi-electrode arrays provide a new, powerful tool to monitor the spatial and temporal characteristics of such network oscillations.

Network oscillations at gamma frequencies (~40 Hz) are readily induced in hippocampal slices by 20 μM carbachol or 10 μM (RS)-3,5-dihydroxyphenylglycine (DHPG). These oscillations are generated in the CA3 and propagate to the CA1 area. They are temperature dependent, increasing from ~10 Hz at room temperature (23-25 °C) to ~40 Hz at body temperature (37-38 °C). Both excitatory AMPA and inhibitory $GABA_A$ receptor mediated synaptic events are involved, as inhibition of either blocks the gamma-frequency network oscillation.

This presentation will discuss the recording and analysis of such network oscillations *in vitro*. We will primarily focus on the advantages offered by planar multi-electrode arrays compared to conventional extracellular recording techniques, and the analysis tools that are used to process the large amounts of data generated during these recordings. In the future, combining multielectrode recordings with other recording techniques, such as patch-clamping of individual neurons and fluorescence imaging, would open up new avenues in the study of network oscillations.

Biological Clock On An MED Probe; Circadian Rhythm Analysis Using Long-Term Culture Of The Suprachiasmatic Nucleus

Sato Honma

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Almost all organisms on earth express circadian rhythms in behavior, physiology, and cellular functions. The rhythm is generated by a circadian clock that, in mammals, is located in the suprachiasmatic nucleus of the hypothalamus (SCN). Since the circadian cycle length is very long compared to other oscillations in brain functions, analyzing its properties in neurons is extremely limited using traditional neurophysiological methods. The development of a multielectrode dish (MED) probe made it possible to monitor neuronal activity simultaneously from multiple neurons for weeks and months. We analyzed the circadian rhythms of spontaneous discharges from SCN neurons in both dispersed cell cultures (Fig. 1A) and organotypic slice cultures (Fig. 1B). In both cultures, single SCN neurons exhibited robust firing rhythms (Fig. 1C). Circadian periods of individual neuron firing were distributed in a wide range of 20-30 h in dispersed neuronal, and of 22-26 h in slice cultures. Cross-correlation analysis revealed that circadian firing rhythms were synchronized through synaptic interactions. The MED system also provided a powerful tool for the study of genetically modified animals, especially those with postnatal lethal defects. We recorded spontaneous firing of SCN neurons from *Clock* mutant mice and found that 77% in slices and 20% in dispersed cell cultures exhibited circadian firing rhythms with lengthened periods (Fig. 1D). These findings challenge the current hypothesis that the *Clock* mutation terminates cellular oscillation. The results also indicate that not all SCN neurons express an autonomous oscillator and that an intact assemblage of SCN neurons with sufficient cell-cell interactions is critical for sustaining circadian output.

References:

Honma, S. et al. *Neurosci. Lett.*, **250**, 157-160, 1998.
Honma, S. et al. *Neurosci. Lett.*, **294**, 113-116, 2000.
Shirakawa, T. et al. *Eur. J. Neurosci.*, **12**, 2833-3838, 2000.
Nakamura, W et al. *Eur. J. Neurosci*. 14: 666-674, 2001.
Nakamura, W et al. *Nature Neurosci*.5:399-400,2002.