Coherence of subthreshold activity in coupled inferior olivary neurons

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Introduction
Subthreshold membrane potential oscillations (STO) in the inferior olivary (IO) nucleus are believed to underlie synchrony and rhythmicity of complex spikes in the cerebellar cortex. Synchronous and rhythmic behavior of complex spikes is modulated either pharmacologically, by applying drugs directly to the IO nucleus (Lang 2001; Lang et al. 1996), or during motor behavior (Onuki 1998; Wadich et al. 1995). In the present study we examined the stability of STO using slice preparations of the IO nucleus.


Methods
Single and double patch recordings were performed in sagittal brain stem slices of 9-31 day old rats. The pipettes were filled with the intracellular solution (containing in mM: 4 NaCl, 10-3 CaCl2, 140 K-gluconate, 10-2 EGTA, 4 Mg-ATP, 10 Hepes, pH 7.2). Time-frequency (TF) plots (Palva et al. 2000) are color coded representations of changes in frequency of STO in time. The amplitude scale of TF plots is in arbitrary units comparable between plots. The amplitude of Fouriercoefficients is expressed in units of the correlation coefficient (rhc).

Optical imaging was performed in brain slices using voltage sensitive styryl dye RH-414.

Prevalence of STO
The frequency and amplitude distribution of STO in olivary neurons.
A. The spontaneous STO in olivary neurons appear at different frequency, pattern and amplitude. Traces were recorded from different neurons in different slices.
B. Distribution of average STO-frequency in 94 neurons. Note two populations of frequencies/geopardized at about 3.5 Hz.
C. Average frequency of STO, plotted as a function of age of the animal (A), shows significant positive correlation, while STO maximal amplitude and the age of the animal are not correlated (D).

Blocking spiking activity stabilizes the temporal pattern of STO
TTX stabilizes frequency and amplitude modulations of STO.
A. STO recorded under control conditions exhibited significant frequency and amplitude modulations.
B. Both application of 0.5 μM of TTX largely abolished spontaneous shifts.
C. Washout of the drug restored the control behavior of STO. TF representations of the traces on the right show a complex pattern of frequency modulation before TTX application and after the washout, and a regular frequency of about 5 Hz following TTX.

CNQX stabilizes the temporal pattern of STO
A. STO recorded before (trace 1) and 20 min after (trace 2) bath application of 40 μM of CNQX. TF representations of the traces on the right show a complex pattern of frequency modulation before CNQX application and a regular frequency of about 6 Hz following CNQX.
B. Second example of the effect of CNQX in a neuron recorded from a different slice.

Optical imaging of propagating oscillations
A. Each trace represents the change in fluorescence recorded by a single photodiode in its relative location as a function of time. Fourier analysis was performed on all diodes. All diodes where the amplitude of the main frequency component was >= 50% of that of the best diode are marked in red. All diodes where the amplitude of the main frequency component was between 10% and 30% of that of the best diode are marked in yellow. Note the central patch of high amplitude oscillations.
B. Color coded activity over the whole array, measured at times denoted under the same graph.
C. Red and blue traces on C represent averaged signals from diodes inside the red and blue frames, marked in both A and B. Fourier spectra and crosscorrelation of these averaged signals are shown in the middle and on the right, respectively.

Complex pattern of STO modulation in non-coherent pairs.
A. Simultaneous recordings from two intermittently oscillating neurons. Note synchronous modulation in amplitude.
B. Fourier analysis shows almost identical spectra.
C. The crosscorrelation peak is shifted to the left indicating about 20 msec phase difference.
D. TF representation of the traces shown in A. Note synchronous modulations in frequency in both cells.

Conclusions
Olivary oscillations exhibited unstable temporal patterns. This instability was significantly reduced following a block of synaptic transmission, implying that frequency and amplitude modulations are imposed on the olivary network by extrinsic synaptic inputs.

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