Burst Firing Induces a Rebound of Synaptic Strength at Unitary Neocortical Synapses

MARIO GALARRETA AND SHAUL HESTRIN
Department of Anatomy and Neurobiology, University of Tennessee, Memphis, Tennessee 38163

INTRODUCTION

At many central synapses, synaptic strength is transiently reduced during high-frequency stimulation. This form of short-term synaptic plasticity is generally attributed to presynaptic mechanisms (Zucker 1989) and is thought to have significant computational and functional implications (Abbott et al. 1997; Galarreta and Hestrin 1998; Tsodyks and Markram 1997). Recently, it has been shown that high-frequency firing can additionally accelerate the rate at which synapses recover from depression (Dittman and Regehr 1998; Stevens and Wesseling 1998; Wang and Kaczmarek 1998). This process may operate as a frequency-dependent “boosting” of the synaptic strength (Dittman and Regehr 1998) and thus may play a role in how central synapses respond to complex patterns of stimulation.

We have studied the effects of high-frequency firing on the recovery from synaptic depression at neocortical excitatory synapses. These synapses exhibit a prominent frequency-dependent depression (Abbott et al. 1997; Castro-Alamancos and Connors 1997; Galarreta and Hestrin 1998; Stratford et al. 1996; Thomson et al. 1993; Tsodyks and Markram 1997) and are thought to sustain in vivo an ongoing activity. We used dual recordings to show that in the presence of ongoing activity bursts of high-frequency spikes are followed by a transient increase of synaptic strength.

METHODS

Cortical slice preparation and cell identification

Experimental procedures were similar to those previously described (Galarreta and Hestrin 1998). Sagittal cortical slices (30°, 300 μm thick) were obtained from 14- to 18-day-old Wistar rats. After dissection, slices were incubated at 32–34°C for 30 min and then at room temperature (20–22°C) until transferred to a submersion-type recording chamber. The extracellular solution bathing the slices contained (in mM) 125 NaCl, 2.5 KCl, 1.25 NaH2PO4, 1 MgSO4, 2 CaCl2, 26 NaHCO3, 20 glucose, and 0.4 ascorbic acid (pH 7.4, 315 mOsm) and was continuously bubbled with a gas mixture of 95% O2-5% CO2.

Neurons in layer five of the somatosensory cortex were visualized using infrared differential interference contrast video microscopy with an upright microscope (Axioskop, Zeiss, equipped with a ×40 water immersion lens). Pyramidal and fast-spiking cells, a subtype of GABAergic interneuron, were identified according to their morphological appearance and their pattern of firing in response to depolarizing pulses of current (Kawaguchi 1993). Biocytin was included in the recording pipettes and cell classification was confirmed morphologically. Similar results were obtained when the postsynaptic neuron was a pyramidal or a fast-spiking cell and data have been pooled together.

Recording and data analysis

We recorded simultaneously from pairs of synaptically connected neurons. Patch electrodes (3–4 MΩ) were filled with a solution containing (in mM) 95 K-methylsulfate, 40 KCl, 10 HEPES, 4 MgATP, 20 phosphocreatine(Na), 0.3 NaGTP, 0.2 EGTA, and 0.3% biocytin (pH 7.3, 295 mOsm). Experiments were performed at 32–33°C. We recorded from the presynaptic neuron under current-clamp mode while the postsynaptic cell was kept under voltage-clamp mode. Presynaptic action potentials were generated by injecting brief pulses (3–5 ms) of depolarizing current at the appropriate frequency. Experiments were interrupted whenever the access resistance to the postsynaptic neuron increased significantly. Signals were recorded using an Axopatch 200A and an Axopatch 200B amplifiers (Axon Instruments). The voltage and current output were filtered at 1–10 kHz and digitized at 16-bit resolution (National Instruments). The sampling frequency was 5 or 10 kHz.

The amplitude of individual unitary excitatory postsynaptic currents (uEPSCs) was measured as the difference between the peak of the uEPSC (minimum value in a 1-ms window at the peak) and the mean current at a 2-ms window before the onset of the synaptic current. In Fig. 2, baseline was obtained by averaging the amplitude of the last uEPSCs before the burst and the first response in the burst. Data are described as mean ± SE. Statistical analysis testing two-sample hypothesis was performed using unpaired, two-tailed Student’s t test. Differences were considered significant when \( P < 0.05 \).

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RESULTS

Dynamics of local excitatory synapses in neocortical slices (layer 5) were studied by recording simultaneously from presynaptic pyramidal neurons and their postsynaptic targets; either other pyramidal neurons \( (n = 6) \) or fast-spiking cells \( (n = 10) \). With this method we could precisely control the presynaptic spiking activity and avoid possible modulatory effects caused by the stimulation of other axons. Individual presynaptic action potentials were produced by injecting brief current pulses \( (3-5 \text{ ms}, 0.8-1 \text{ nA}) \), and the uEPSCs were recorded from the postsynaptic cell under voltage-clamp mode \( (V_h = -70 \text{ mV}) \).

It has been reported that in vivo cortical neurons exhibit ongoing firing rates at the range of 1–50 Hz (Hubel 1959; Mountcastle et al. 1969). We tested the effect on synaptic strength of steady activation at a frequency of 1 Hz (Fig. 1). Baseline response was obtained as the average amplitude when stimulating at 0.25 Hz. Increasing the stimulation rate to 1 Hz produced a depression of the uEPSC amplitude to 77.9 ± 2.7% of baseline value (Fig. 1, \( n = 4 \)). This result together with previous observations (Galarreta and Hestrin 1998; Thomson and West 1993; Tsodyks and Markram 1997; Varela et al. 1997) suggests that the ongoing activity in vivo produces some degree of depression.

Given that in vivo neocortical neurons fire bursts of high-frequency spikes superimposed on an ongoing single-spike activity (Bair et al. 1994; Cattaneo et al. 1981; Hubel 1959), we examined the response to burst firing when these two patterns are combined. Ongoing low-frequency spiking activity was generated by producing presynaptic spikes at a frequency of 1 Hz. In addition, a burst of 10 spikes at 100 Hz replaced an individual action potential every 15 s (Fig. 2A). High-frequency bursts produced a strong depression of the uEPSCs (Fig. 2A) and the amplitude of the last uEPSC in the burst was 19.3 ± 3.5% of the amplitude of the first response \( (n = 12 \text{ pairs}) \). In spite of this strong depression, we found that uEPSCs recorded 1 s after the burst were larger than those obtained at baseline stimulation rate (Fig. 2A and B; \( P < 0.01 \), t-test). The increase of synaptic strength was transient and the uEPSCs returned to baseline values 3 s after the burst. Similar results were obtained in 10 of 12 pairs. The average rebound of synaptic strength, measured 1 s after the burst, was 31.4 ± 7.8% over baseline (Fig. 2B, \( n = 12 \)).

It was important to test whether the rebound of synaptic strength observed after burst firing required an ongoing activity. To study this issue we compared, in the same pairs, the postsynaptic responses produced by identical bursts of high-frequency spikes with and without sustained spiking at 1 Hz (Fig. 3). In the absence of 1 Hz stimulation, bursts were not followed by a transient increase of synaptic strength (Fig. 3A). In five pairs, the average amplitude of an uEPSC obtained 1 s after the burst was 99.0 ± 7.8% of baseline value (Fig. 3A). In contrast, brief bursts superimposed on an ongoing low-frequency activity (1 Hz) were followed by a transient increase of uEPSC amplitude (Fig. 3B). In response to this protocol, the amplitude of the test response obtained 1 s after the burst was 140.6 ± 24.2% of baseline (Fig. 3B; \( n = 5 \text{ pairs}, P < 0.01 \)).
DISCUSSION

We have shown that at local excitatory neocortical synapses, the strong synaptic depression produced by bursts of high-frequency spikes is followed by a transient enhancement of synaptic strength. This rebound of synaptic efficacy occurred when synapses sustained an ongoing low level of activity, suggesting that synapses displaying activity-dependent depression may functionally facilitate in response to burst activity.

Previous work has shown that bursts of action potentials can accelerate the recovery from activity-dependent depression (Dittman and Regehr 1998; Stevens and Wesseling 1998; Wang and Kaczmarek 1998). In agreement with these data, we found that the uEPSC obtained 1 s after the burst (filled bar labeled “after”) are normalized in relation to the 1st response of the burst shown for visual comparison (open bar labeled “burst”).

In vivo experiments have revealed that burst activity in neocortical neurons is associated with sensory stimuli (Bair et al. 1994; Cattaneo et al. 1981; Livingstone et al. 1996), suggesting that brief periods of very rapid firing might play a role in encoding information (Lisman 1997). Because the patterns of stimulation we used are within the range of frequencies observed in vivo, we suggest that this rebound of synaptic efficacy may contribute to the readout of the information contained in complex patterns of spikes.

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Address for reprint requests: M. Galarreta, Dept. of Anatomy and Neurobiology, University of Tennessee, Memphis, 855 Monroe Ave., Memphis, TN 38163.

REFERENCES


FIG. 3. Increase of synaptic strength after burst firing depends on background activity. A: simultaneous recording from a presynaptic pyramidal neuron and a postsynaptic fast-spiking cell. A1: schematic description of the pattern of activity generated in the presynaptic neuron. Bursts of 10 spikes at 100 Hz (thick lines, arrow heads) were generated every 7 s and a single action potential was produced 1 s after each burst (thin lines). A2: uEPSCs obtained during the burst (left) and 1 s after the burst (right). Traces are the average of 50 trials. A3: uEPSCs obtained 1 s after the burst (filled bar labeled “after”) are normalized in relation to the 1st response of the burst shown for visual comparison (open bar labeled “burst”). B1: data from the same pair of cells as in A1 in response to the protocol shown in B2. B2: data from the same pair of cells as in A2 in response to the protocol shown in B3. Average uEPSC obtained 1 s after the burst was significantly larger than the 1st response in the burst (24%, P < 0.01). Each trace is the average of 40 trials. Protocol in B1 was tested before protocol in A1. B3: amplitude of the uEPSC obtained 1 s after the burst (filled bar labeled “after”) are normalized in relation to the 1st response of the burst shown for visual comparison (open bar labeled “burst”). Scale bars in A also apply to B.


