

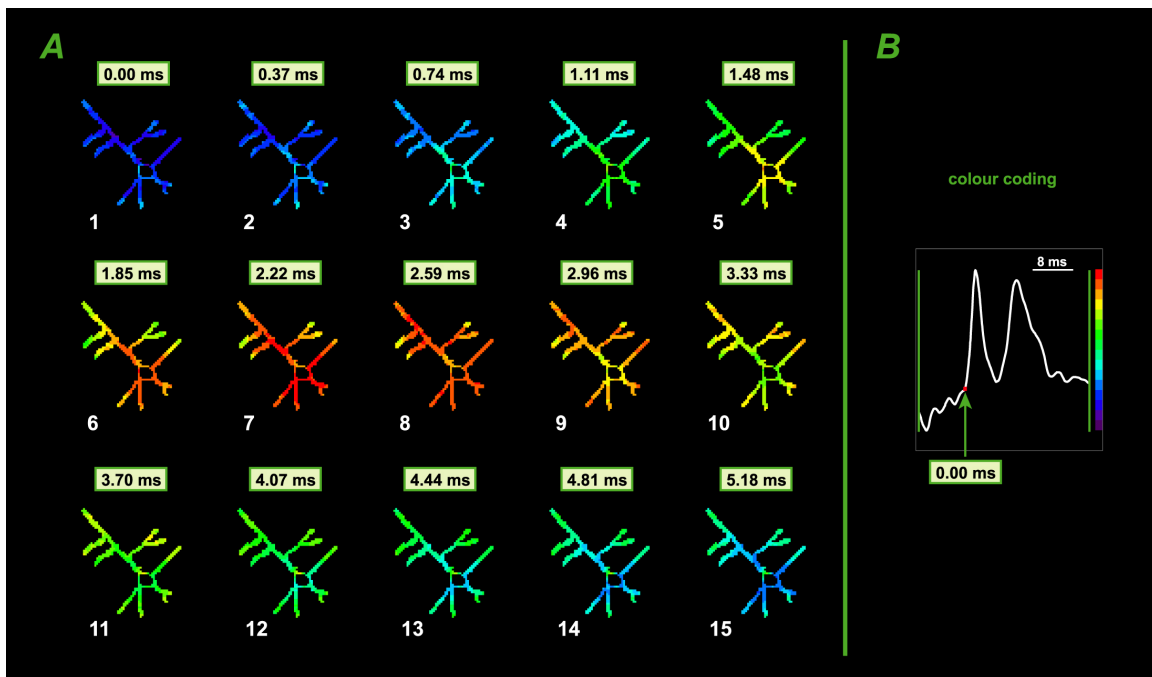
## Supplementary Material

### **Action potentials in basal and oblique dendrites of rat neocortical pyramidal neurons**

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#### **Three different ways to analyze the voltage-imaging data:**

1. One way to analyze voltage-imaging data is to choose one, or group of photo-detectors (pixels), and plot their output (signal amplitude) versus time, as was done in Figures 2, 3, 8, and 9.
2. An alternative way is to colour-code the signal amplitude and display the output of all pixels in the array simultaneously (full frame). Since one frame carries information about one instant in time, in order to follow the dynamics of the event it is necessary to display multiple frames as shown in Figure 10.



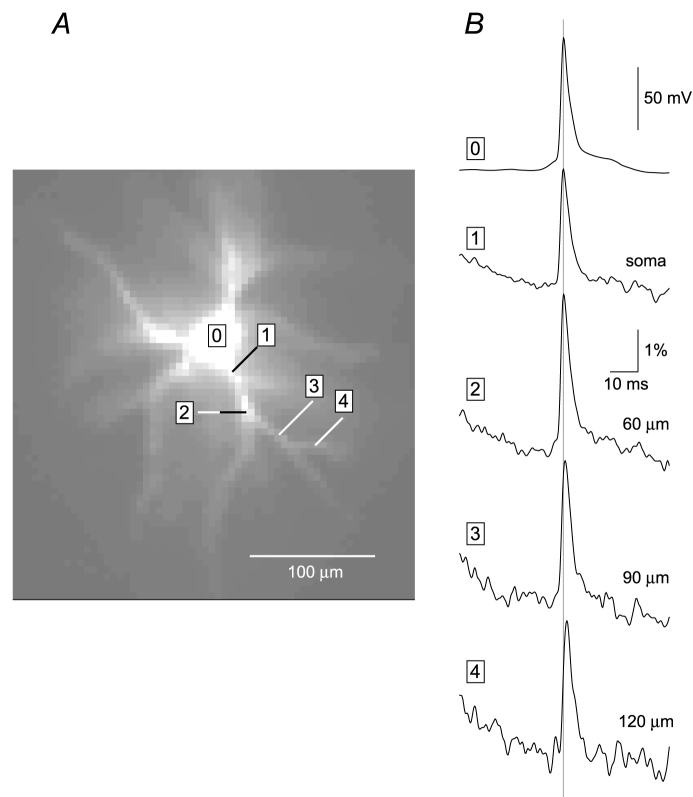
**Figure 10. Multiple-frame analysis of voltage-imaging data.**

*A*, A sequence of 15 frames, acquired during action potential backpropagation across the dendritic tree. Same data as in Figure 3. The neuron fired two action potentials upon direct current injection. A short sequence (~5 ms) that includes only to the first action potential is shown in *A*. Pixels, that do not receive in focus fluorescent light from the image of the neuron, are omitted. Somatic pixels, saturated by the high light intensity, are also omitted. Inter-frame interval is 0.37 ms. The timing of each frame is displayed above the frame. *B*, The optical signal from a single pixel versus time together with the color scale bar used for *A*. Red represents the peak of the first action potential. In *A* the output of each pixel is scaled separately (“variable” scale), such that the maximum signal for each pixel (peak of the first action potential) turns red. The sampling point indicated by the red dot, corresponds to the first frame (zero time). Zero time is set arbitrarily, just before the inflection of the action potential. The optical trace shown in *B* is the output of a single pixel positioned on the apical dendrite, at the middle branch point.

3. Finally, multiple frames can be played in the form of a short movie. Click [here](#) to play the data file from Figure 3 – backpropagation of two action potentials (video clip). The colour-coding is same as in Figure 10B.

### **Action potential propagation failure at branch points**

An action potential exhibits changes in the shape and velocity as it approaches a region of changing core conductor geometry, such as a branch point. Depending upon the amount of the geometric change (and the channel density), the action potential can fail to propagate beyond the branch point, or it can succeed with or without delay (Goldstein and Rall, 1974). In repetitive recordings from 36 second- and third-order basal dendrites ( $n = 13$  neurons) action potential propagation failure has never been observed. An example of such an experiment is shown in Figure 11.



### **Figure 11. Voltage-sensitive dye recordings from second- and third-order basal dendrites**

*A*, Microphotograph of a layer 5 pyramidal neuron filled with JPW3028. *B*, Whole-cell recording (0) is aligned with the action potential associated optical signal from the cell body (1), from a first-order basal dendrite (2), a second-order basal dendrite (3), and a third-order basal branch (4). The distance from the recording site to the center of the soma is displayed above each trace. The thin vertical line marks the peak of the somatic action potential. Average of 4 (trials).