Chapter 15 Web Text Box 2 and Video 15.2

Imaging the diffusion of calcium in a nerve cell



Calcium diffusion A

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In example 15.1 on book page 253 we show how brief depolarization of a pain receptor nerve cell causes the calcium concentration to increase around the edge of the cell as calcium ions enter through voltage gated calcium channels. The figures here show an experiment that investigated the spatial dynamics of calcium in these cells at higher spatial and temporal resolution. Panel A combines a transmitted light image of the nerve cell body, with its two axons, and an image of Hoechst 33342 fluorescence that reveals the location of the nucleus (see book page 6). The dark object entering the image from the bottom right hand corner is a patch pipette in whole-cell mode (see In Depth 14.1 on book page 230). The patch pipette contained the dye Oregon Green BAPTA, which fluoresces more brightly when calcium concentrations increase. The microscope was then set up to scan a laser spot across the line indicated by the yellow arrow at a wavelength that excites Oregon Green BAPTA.



Calcium diffusion B

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Panel B shows a line scan image. Each horizontal line was generated by one scan of the laser across the cell; the vertical axis is time. The brightness of the fluorescence has been coded on the "glow" scale shown at the left, with increasing fluorescence represented as increasingly "warm" shades of red, yellow and finally white. About a quarter of the way down the line scan image the cell was depolarized to +10 mV for 50 ms. One can tell that the fluorescence increases, but it is difficult to make out precisely what happens.



Calcium diffusion C

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In panel C the fluorescence at each point on the scanned line has been normalized by the fluorescence at that point at time zero (that is, during the first few scans). The normalized fluorescence is coded by color, as indicated on the scale at the right. We can now see clearly that immediately after the onset of depolarization calcium increases at the cell edges; a wave of increasing calcium then travels in towards the cell center. This is not a calcium wave driven by the release of calcium from the endoplasmic reticulum, as described in <u>Video 15.2 Calcium waves in glial cells</u>. Rather, it simply represents the diffusion of calcium through the cytosol (on the right) and nucleoplasm (on the left).

This data first appeared in Coatesworth and Bolsover. 2008. Calcium signal transmission in chick sensory neurones is diffusion based. Cell Calcium. 43(3):236-49; see there for more information.