Simultaneous BOLD fMRI and fiber-optic calcium recording in rat neocortex

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Supplementary Figure 1
Properties of glial calcium signals in S1 evoked by forepaw stimulation

(a) Two-photon image of astrocyte-specific staining following surface application of the green fluorescent calcium indicator Fluo-4. Several astrocytes (a1-5), a cross-section of a blood vessel (bv), and a region for analysis ‘gliapil’ signal (gp) are indicated. (b) Fluo-4 calcium signals in 5 selected astrocytes and the gliapil evoked by 3 Hz forepaw stimulation for 40 s. Note the different onset times of somatic astrocyte signals. The corresponding percent increase in blood vessel perimeter for the vessel indicated in (a) is shown at the bottom together with an exponential onset fit. (c) Astrocytic calcium signals depended on the interval between stimulations. Fluo-4 signals upon 3-Hz forepaw stimulation for 40 s are shown as a function of the time interval elapsed since the previous stimulation cycle. Glia signals displayed a refractory period of a few minutes. (d) Same analysis as in (c) but for a Rhod-2 experiment with forepaw stimulation at 5 Hz for 20 s. Again Rhod-2 calcium signals required a few minutes to recover. (e) Dependence of FWHM of fiber-optically measured Rhod-2 calcium signals on duration of stimulation (3 Hz: n = 2, 5 Hz: n = 26, and 10 Hz: n = 10; data pooled). (f) Dependence of FWHM of fiber-optically measured Rhod-2 calcium signals on stimulation frequency (10-40 s stimulation durations pooled).
Supplementary Figure 2
Schematic of model simulations

(a) Shape and timing of the assumed elementary hemodynamic impulse response (IR) function in response to a single stimulus. (b) Assuming a uniform response model each stimulus contributes the exact same IR (grey traces, every 10th trace highlighted in red). Summation of the IR train and down-sampling in time to the acquisition rate of measured BOLD data (right). (c) Assuming adapting responses, each IR was scaled according to the measured fluorescence signal amplitude (black), generating a train of IR with variable amplitude which reflects the adaptation observed in the fiber-optic measurement. IR summation and down-sampling of simulated traces were performed as described in b (right). All blocks devoid of apparent glial component were taken into account to obtain the global scale factor for the respective experimental animal.