Supplementary Material

Location of recorded striatal neurons

The range of locations of recorded neurons across the mediolateral and dorsoventral extent of the striatum is depicted in Fig. S1 below. This area corresponds to the striatal area receiving overlapping input from visual and sensorimotor areas of cerebral cortex (see: McGeorge and Faull, 1989; *Neuroscience* 29:503-537, their Fig. 17a).

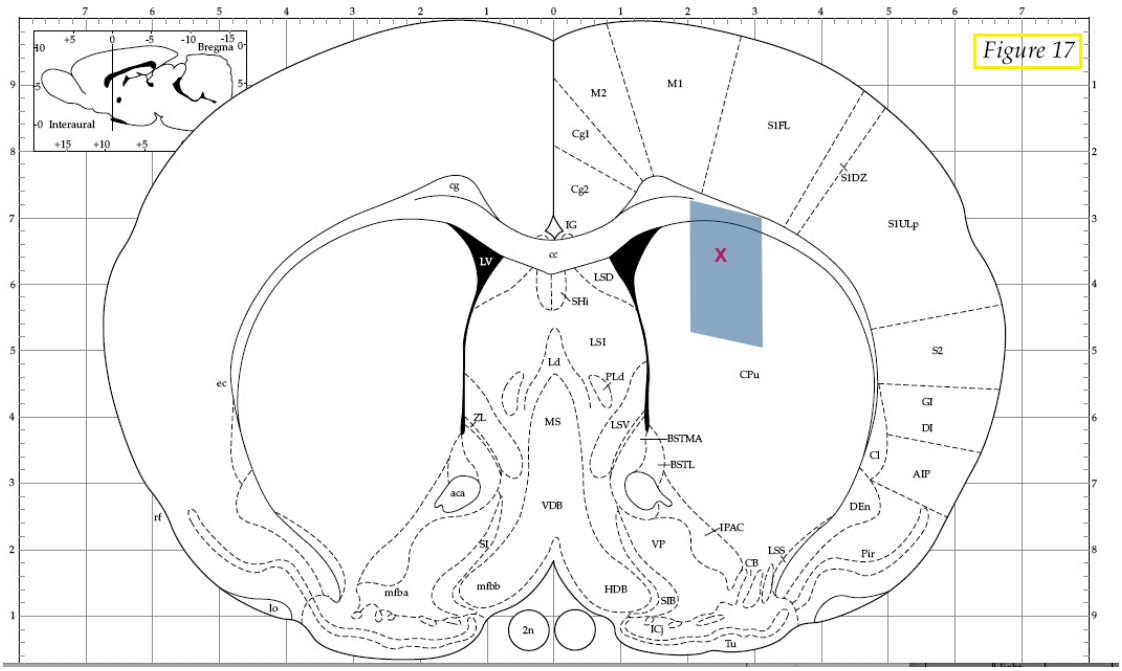

Figure S1. Location of recorded striatal neurons. The mediolateral and dorsolateral extent of the recording locations are within the grey area in this coronal section, drawn at the level of bregma, anteroposterior coordinate. The 'average' recording position is indicated by the red cross. CPu = striatum.
(Diagram adapted from the atlas of Paxinos and Watson, 2007).

Estimation of the spread of drug ejections

In separate experiments an estimate of the spread of bicuculline (BIC) ejected into the SC was made using c-Fos immunohistochemistry (see Fig. S2A). Referenced to baseline c-Fos immunoreactivity observed using the same anesthetic regimen and ejection of saline vehicle, it was apparent that c-Fos activity attributable to BIC ejection was limited to the deep and superficial layers of the SC, with little apparent direct spread to adjacent structures. Over a series of 18 experiments, we estimated the spread of BIC to be typically 2 mm^3.

Using two different markers applied in the same experiment, we also estimated the extent of drug spread when ejections were made onto the surface of the cortex or into the thalamus (n = 2 experiments), using the same ejection protocol employed to deliver muscimol and baclofen during the electrophysiological experiments. Fluorescently-conjugated cholera toxin B (C34776 Molecular Probes; 5 µl of a 0.1% solution in 0.9% saline) was ejected onto the cortex overlying the SC and cresyl violet (1 µl of a 5mM concentration solution) was ejected into the parafascicular nucleus of the thalamus. In Fig. S2B it can be seen that after six hours cholera toxin had spread across a large area of cortex, penetrating the superficial layers in places, without spread beyond the cortex to deep structures such as the superior colliculus. It is likely that this somewhat underestimates the penetration of the cortical layers by the tracer since this would be taken up by neuronal elements. Extent of staining after thalamic ejection was limited to 1 mm^3 and wholly restricted to the targeted parafascicular nucleus (Fig. S2C). Based on these experiments, it is highly unlikely that there was spread of inhibitory drugs beyond the desired target of the cortex or the thalamus.

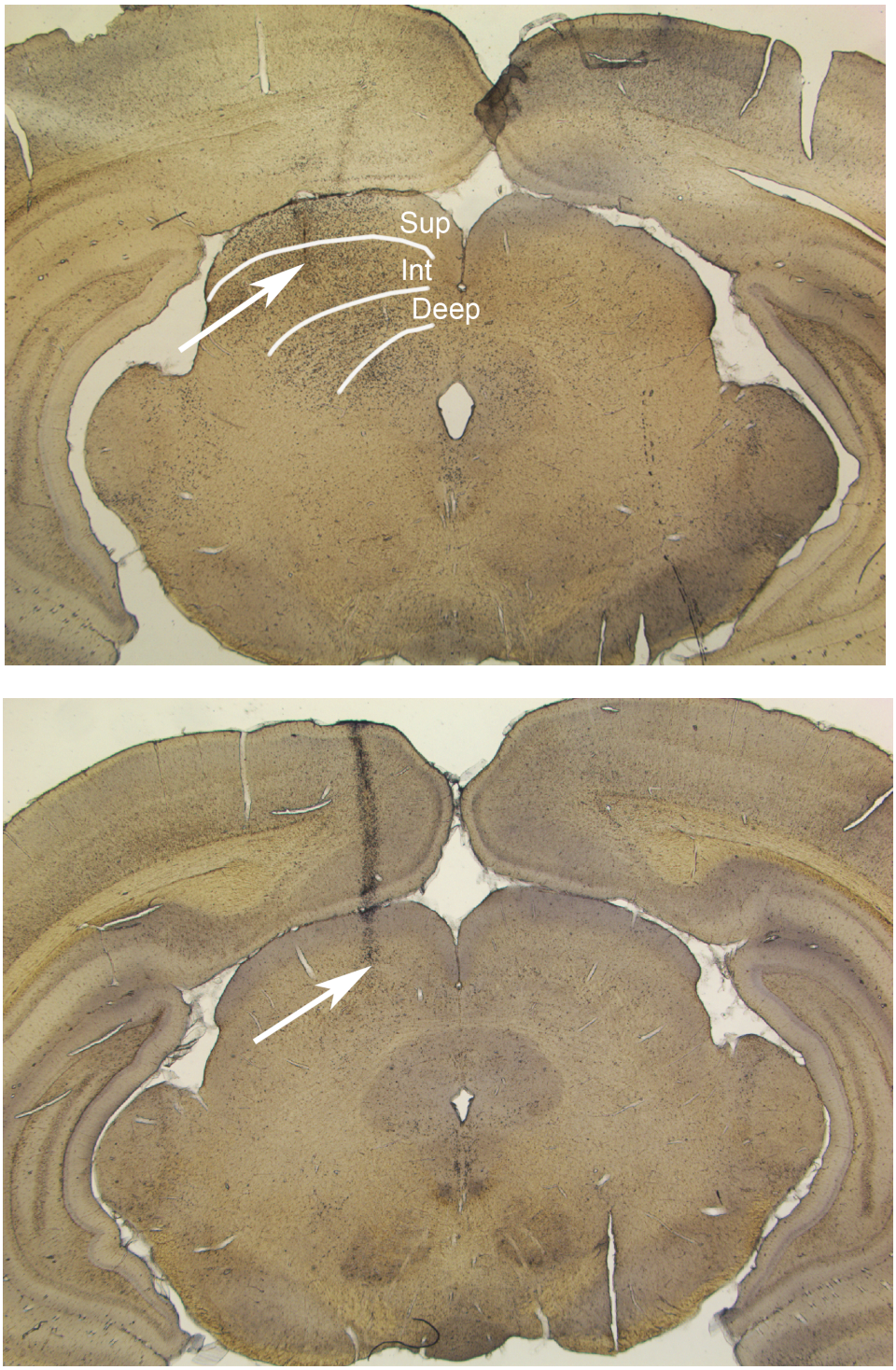

Figure S2A. c-Fos immunoreactivity induced by BIC ejection into the intermediate (int)/deep layers of the SC. Top: BIC ejection. c-Fos activation extends to the deep layers. Bottom: saline control ejection. Ejection pipette tips indicated by arrow.

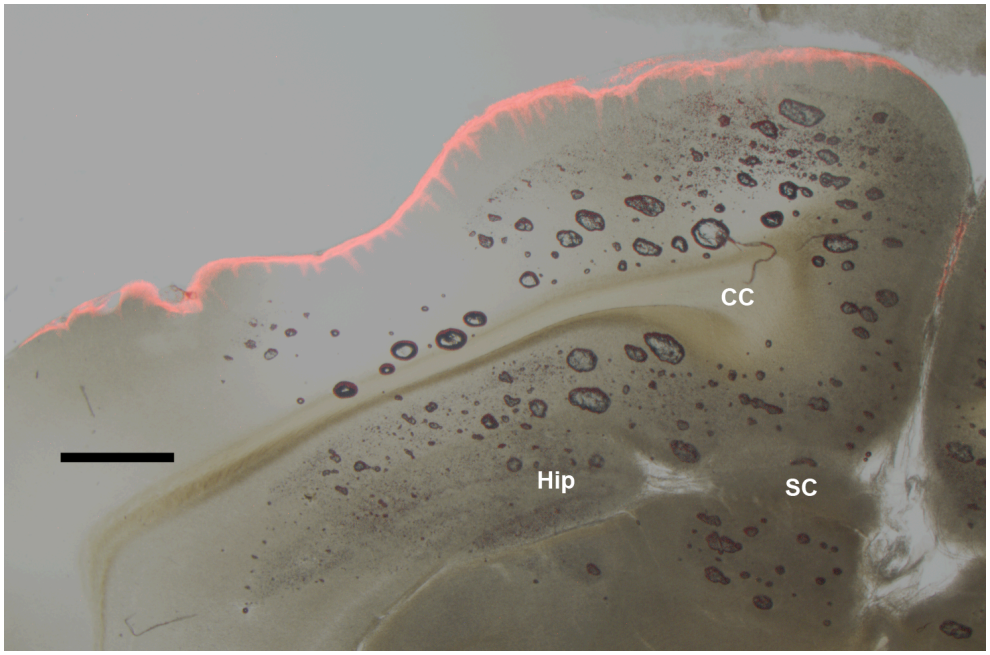

Figure S2B. Spread of fluorescently-labelled cholera toxin after ejection onto the superficial cortex overlying the superior colliculus (SC). Image is a composite of a confocal laser image and a light microscope image of the same coronal section (approx Bregma -5.5 mm). Note that this section is taken approx 2.5 mm more anterior than the site of drug ejection onto the cortex. 'Bubbles' are artifacts. Scale bar = 1mm. Hip = hippocampus. CC = corpus callosum.

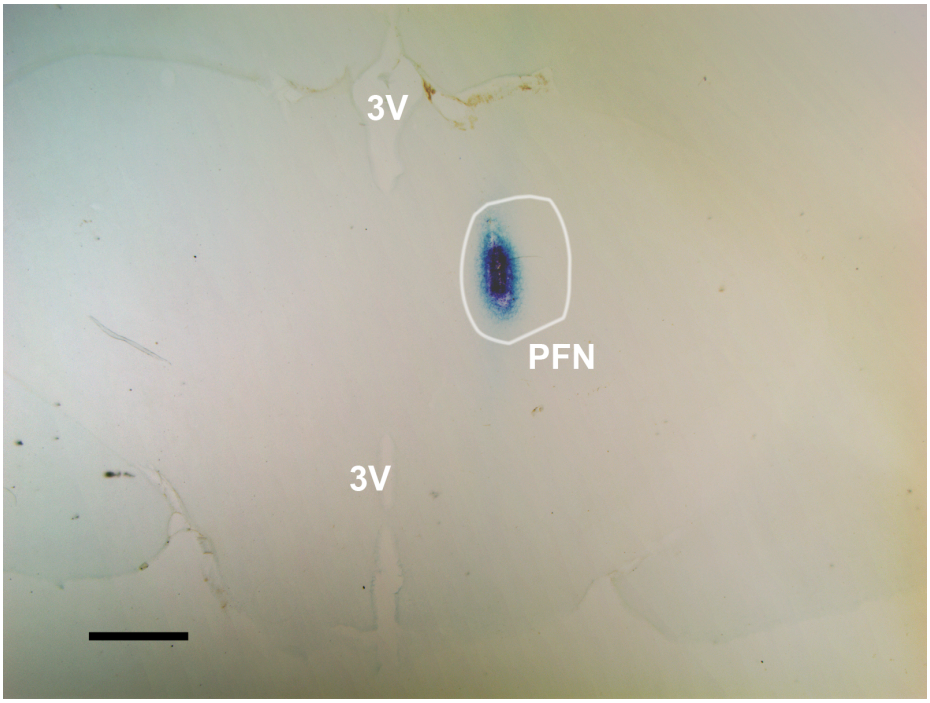

Figure S2C. Extent of spread of cresyl violet after ejection aimed at the parafascicular nucleus of the thalamus (PFN = approximately within white area, based on Paxinos and Watson, 1997). Image is a light microscope image of a coronal section (approx Bregma -4.2 mm). Scale bar = 1mm. 3V = 3rd ventricle.

Cortical VEPs

VEPs were separated from slow oscillations by the following procedure: The EEG signal was lowpass filtered using a 4^{th} order Butterworth filter with 45 Hz as cut-off frequency. The EEG signal was then downsampled from 10 kHz sampling rate (11.11 kHz for experiments from 2004) to 500 Hz sampling rate, so that a 7^{th} order Butterworth filter with 2.3Hz cut-off frequency could be used for efficient highpass filtering.

VEPs were assigned to one of the two following conditions according to the recording time with respect to the ejection of BIC. VEPs recorded less than 12 minutes post BIC were assigned to the condition 'VEPs with BIC'. Note that the other condition, referred to as VEPs without BIC, also included those VEPs which were recorded more than 12 min post BIC. For the effect of BIC on the cortical VEP, single trial VEPs were tested for differences between conditions for each experiment using the Wilcoxon rank sum test. The means of all recording sessions were tested again as a group for differences evoked by BIC ejection using the Wilcoxon signed rank test. In both analyses, data from an additional session was included, the intracellular data of which was rejected due to the low quality of the recording. Finally, the grand means were calculated separately for VEPs before and after BIC as the mean of the means of all recording sessions.

The amplitudes of the peaks N1, P2 and P3 were measured in single trials. The amplitude of N1 was computed as the difference between the baseline value at the start of the trial and the minimal EEG potential at 30 to 70 ms. Similarly, the amplitude of P2 and P3 were calculated with reference to the baseline value at trial

start and the maximal potential at 70 to 104 ms and at 106 ms to 150 ms, respectively. Times are given with respect to trial onset, i.e. 10 ms earlier than the onset of the light flash.

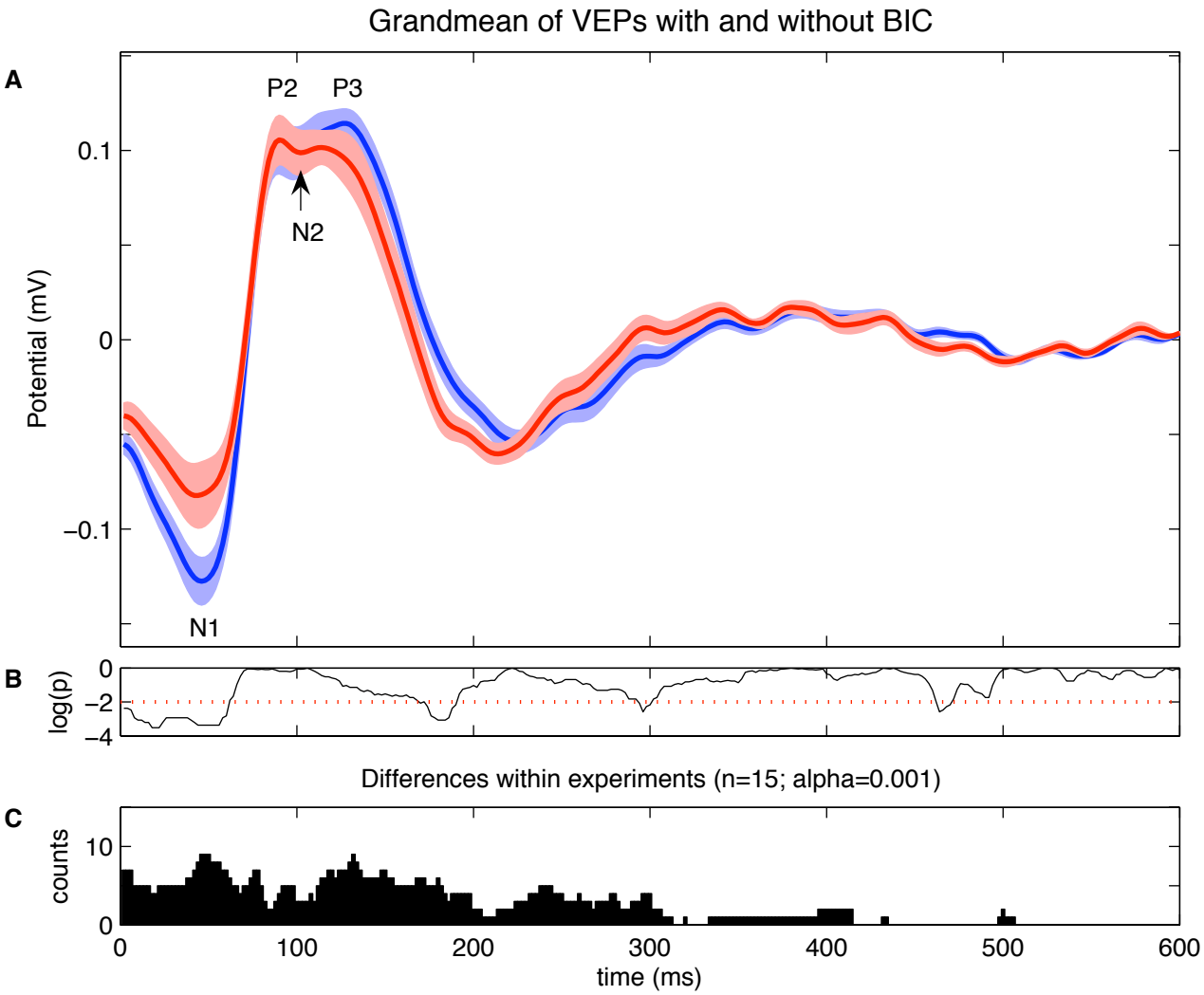

Figure S3. A) The grand mean VEPs without (blue) and with (red) BIC. Lighter colours indicate the standard error of the mean. The peaks are indicated in established nomenclature: the first negative peak, N1, at ~40 ms; the second positive peak, P2, at ~90 ms; N2 (arrow) at ~105 ms; P3 at ~125 ms; P1 was not visible. Light flash onset was at 10 ms, where a difference in VEPs is already apparent due to an offset (see text for details). **B**) Logarithmised p-values from Wilcoxon signed rank test for differences between means without BIC and with BIC. The significance level of 0.01 is indicated by the dotted red line. **C**) The number of recording sessions with significantly different VEPs shown for each time point. Significance level alpha was set at 0.001. In ten of fifteen sessions, there was a significant difference between VEPs with and without BIC at the time of N1. However, no component exhibited an increased amplitude indicating a sensitization to visual stimulation.

Thalamic VEPs

In four experiments, a glass pipette was advanced in multiple tracts through the thalamus and the LFP was recorded from various depths following light flash presentation. All light flashes were presented to the contralateral eye after disinhibiting the SC. Visual activation of the SC was monitored by another LFP electrode implanted adjacent to the BIC ejection pipette. Only episodes with a minimal amplitude of 0.5 mV for the SC VEP were included in the analysis. For each experiment, data from each tract was low-pass filtered (4^{th} order Butterworth filter, cut-off frequency = 45 Hz) and averaged (> 20 trials) for depth segments of 500 μm. Averaged data from all experiments was smoothed (3^{rd} order Savitzky-Golay of 65 ms window width), combined and plotted according to recording coordinates in order to identify areas that were activated by visual stimulation post BIC into the SC (Fig. S4 A and B). VEP amplitudes were measured as the difference between minimal and maximal potential during the first 400 ms. As a control, amplitudes were also measured for the last 400 ms of the 1.5 sec episodes when the influence of the light flash was presumably minimal. VEPs with amplitudes exceeding the 95-percentile of the control amplitudes (0.041 mV, from 53 smoothed VEPs) were considered to indicate visual activation (highlighted in black). The results from this analysis indicated that broad areas of the thalamus including coordinates potentially within the parafascicular nucleus were visually activated following light flash stimulation.

Additionally, the LFP signal adjacent from the MUS injection site was recorded in another four experiments involving intracellular recordings from striatal neurons and concomitant inhibition of the thalamus. Single trials were low-pass filtered (4^{th} order Butterworth filter, cut-off frequency = 45 Hz) and averaged. BIC ejection into the SC

allowed visual activation of the thalamus at short latencies of <50 ms (example in Fig. S4 C). Unfortunately, all neurons were lost before the ejection of MUS and the intracellular data from these experiments was not included in the current study. In these experiments, the recordings sites were histologically confirmed to be within or adjacent to the parafascicular nucleus (Fig. S4 D).

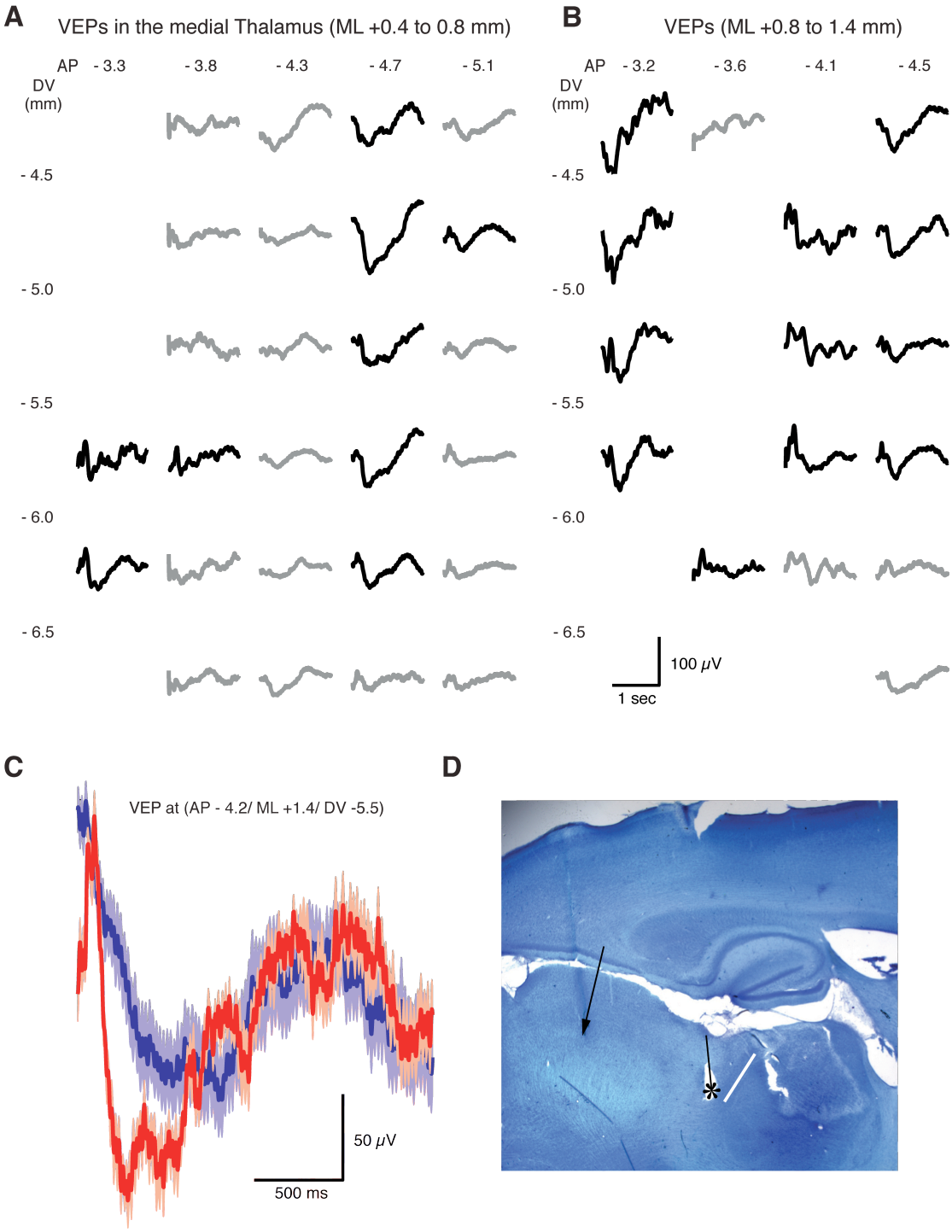

Figure S4. A+B) Mean VEPs recorded in the medial thalamus after disinhibition of the SC by local BIC injection. VEPs are arranged according to coordinates of recording sites in relation to Bregma; all coordinates are given in mm; AP: anterior-posterior; DV: dorsal-ventral; ML: medial-lateral. Black traces depict VEPs exceeding the 95-percentile of control amplitudes. Plot (A) includes more medial, Plot (B) more lateral recording sites. Note the broad distribution of visual activation within the thalamus. **C**) Mean VEPs before (blue) and after (red) BIC recorded from the posterior parafascicular nucleus during the recording of a striatal spiny neuron. Lighter colours indicate the standard error of the mean. Note the early positive and late negative component of the VEP post BIC. The light flash stimulation was

triggered by the onset of the Down state in the neuron's membrane potential (see Methods), hence the VEP recording before BIC exhibits slow wave activity. **D**) Nissl-stained sagittal brain slice from the same experiment with the location of the recording site (asterisk) in the thalamus located near the parafascicular nucleus (white line) and BIC ejection site in the deep layers of the SC (arrow) indicated.