Example Simultaneous Recording in V1 and FEF

Fig. S1A shows the responses of an example frontal eye fields (FEF) neuron evoked by the easy, intermediate, and difficult stimuli, and Fig. S1B illustrates the simultaneously recorded responses of a primary visual cortex (V1) multiunit recording site. The receptive field of the FEF and V1 neurons fell on the same curve. We compared the activity evoked by the target curve (Fig. S1A, Top) to the responses evoked by the distractor (Fig. S1A, Bottom). The appearance of the stimulus in the receptive field (RF) evoked a strong visual response in FEF with a latency of 60 ms and also in area V1 with a latency of 52 ms (as determined with a curve-fitting method; see below). We examined how the strength and latency of the attentional response modulation depended on task difficulty. In the example FEF cell, the modulation index (MI) was 1.70 in the easy condition and 1.72 in the intermediate condition, and it decreased to 1.03 in the difficult condition. Attentional modulation was weaker at the V1 multiunit recording site with an MI of 0.2 in the easy condition that decreased to 0.06 and 0.05 in the intermediate and difficult conditions, respectively. It can be seen in Fig. S1 that V1 maintained a representation of the distractor curve, whereas the representation of this curve was largely suppressed in FEF.

For the FEF cell, the latency of the attentional modulation increased from 155 ms in the easy condition to 190 ms in the intermediate condition and to 280 ms in the difficult condition. The attentional modulation latency at the V1 recording site also increased with task difficulty, from 145 ms in the easy condition to 257 ms in the intermediate condition and to 309 ms in the difficult condition. Thus, in this example recording session, the onset of attentional modulation in area V1 and FEF was delayed if the task was more difficult with latency differences between −10 ms (V1 leading FEF) and +67 ms (FEF leading V1).

Comparison of Visual and Visuomovement Neurons in FEF and V1 Recording Sites

We focused on a population of FEF neurons that included two cell types, visual and visuomovement neurons (1). We have also examined the responses of these two classes of neurons separately. To classify the cells, we used a memory-guided saccade task (1) and applied criteria based on a previous study in FEF (2) (see below for a description of this task and classification criteria). Fig. S2 shows the responses of 16 visual and 23 visuomovement cells in the curve-tracing task. The latencies of attentional modulation of the visual cells were 123, 208, and 229 ms in easy, intermediate, and difficult conditions, respectively. Average modulation latencies of visuomovement cells were 126, 169, and 224 ms in the three difficulty conditions. When we compared these latencies with V1 latencies, we did not observe significant differences at any of the task difficulty levels (P > 0.05, Mann–Whitney u test). Therefore, our main finding that attentional response modulation occurs at approximately the same time in V1 and FEF also holds for these two classes of FEF neurons, if analyzed separately.

Comparison of Latencies Between Multiunit Activity in FEF and V1

Our main analysis compared the latencies of V1 MUA recordings to single-unit activity in FEF. Do we obtain the same results with multiunit responses in FEF? Fig. S3 shows the MUA responses of FEF and their latencies at the three difficulty levels. The population latency of FEF multiunit response was 122 ± 39 ms (±std), 167 ± 22 ms, and 177 ± 36 ms for the easy, intermediate, and difficult condition, respectively. We did not observe a significant difference between latencies of FEF and V1 neurons in these conditions (Ps > 0.05, Mann–Whitney u test). Therefore, our findings are not caused by the different methods for recording neuronal activity in FEF and V1.

Influence of Firing Rate on the V1–FEF Noise Correlation

We found that the noise correlations between V1 and FEF were stronger if the RFs in the two areas fell on the target curve than if they fell on the distractor. Is this difference in noise correlation genuine, or is it related to the higher firing rates elicited by the target curve? To investigate this question we carried out a stratification procedure in which the difference in firing rate between conditions was eliminated by equalizing the number of trials across the firing rate distributions. To this end, we defined three bins of equal size between the minimum and maximum responses for every V1 recording site and also three bins for the single unit activity in the FEF (pooling across difficulty levels). Every trial of the condition with the target curve in the RFs was assigned to one of nine bins, one bin for every combination of the firing rate classes in V1 and FEF, and the same binning procedure was also used to categorize trials with the distractor in the RFs. We then equalized the firing rate distributions in the two conditions by randomly removing surplus trials from the target or distractor condition until trial number was the same in every bin. On average, 30% of trials were discarded across all pairs (between 9% and 85%), and we excluded pairs where more than half of the trials were removed from the analysis. We then repeated our analysis in the stratified data set. The average noise correlation between the remaining FEF–V1 pairs (n = 95) was 0.05 for responses elicited by the target curve, which was significantly larger than the noise correlation elicited by the distractor (average = 0.006 ± P < 10^{-5}, Wilcoxon signed-rank test) (Fig. S4). Thus, the stratification analysis demonstrated that the higher noise correlation between FEF and V1 elicited by the attended curve was not caused by the stronger responses in the two areas.

SI Materials and Methods

Surgical Procedures. The details of the surgical procedures have been described elsewhere (3, 4). In short, a head-holder was implanted in a first operation which allowed head immobilization. During this first operation we also inserted a gold ring under the conjunctiva of one eye to allow the monitoring of eye position. In a second operation, arrays of 4 × 5 or 5 × 5 electrodes (Cyberkinetics Neurotechnology Systems Inc.) were chronically implanted in area V1 (right hemisphere in monkey G and left hemisphere in monkey J). In the third operation we implanted a recording chamber, on the same side as V1 arrays, above the frontal eye fields. Area FEF was localized before the surgery with MRI.

Details of the Curve-Tracing Task. The monkeys sat in a primate chair with their heads restrained, at a distance of 0.75 m from a screen. The stimuli (white curves on a black background; Fig. 1B) were back-projected onto the screen (70° of visual angle; 1024 × 768 pixels) by a video projector in combination with a Texas Instruments Graphics Architecture graphics board running at a frame rate of 72 Hz. A trial started as soon as the monkey’s eye position was within a 1–1.5° square window centered on a 0.2° fixation point (FP). After 300 ms, the stimulus appeared (Fig. 1A), but the monkey had to maintain steady fixation. Correct responses were rewarded with a drop of apple juice. If the monkey broke fixation before FP offset, the trial was
terminated and discarded. The monkeys were experienced in the curve-tracing task and received additional training with the varying luminance before the recording sessions.

**Memory-Guided Saccade Task.** This task was used to classify FEF neurons as visuomovement or movement cells. After a fixation epoch (300 ms), a probe stimulus (circle) appeared on the screen and stayed in view for 100 ms. Monkeys were required to maintain fixation during an additional 400 ms (delay period) after the disappearance of the stimulus. Thereafter, two saccade targets were displayed, one at the same location as the probe stimulus and the other at the mirror location relative to the fixation point. Monkeys made a saccade toward the remembered location of the probe. We either placed the probe stimulus in the neuron’s RF (as determined online by radial tuning and eccentricity tasks) or at the mirror location.

We measured visually driven activity in a time window between 50 and 150 ms after the stimulus onset and baseline activity in a 150-ms window before stimulus onset. Movement-related responses were measured in a time window between 100 ms before and 20 ms after the initiation of the saccade, whereas a presaccadic baseline was determined during the memory delay, between 350 ms and 200 ms before the initiation of the saccade. A neuron was classified as visual if the visual response was significantly greater than baseline activity (Wilcoxon signed-rank test, \( P < 0.05 \)) and if the movement response was not significantly greater than the presaccadic baseline activity (Wilcoxon signed-rank test, \( P > 0.05 \)). A neuron was classified as visuomovement if visual and movement responses were both significantly larger than their respective baselines (Wilcoxon signed-rank test, \( P < 0.05 \)). For 4 out of 43 neurons, we could not record the responses in a memory-guided saccade task, because the isolation of the single neuron was lost after recordings in the main curve-tracing task. These cells were not included in the analysis above that separated visual cells from visuomovement cells.

**Recording of Neuronal Activity, Eye Position, and Data Analysis.** The monkey’s eye position was monitored with a double magnetic induction technique (sampling rate of 1 kHz). We simultaneously recorded extracellular activity of neurons in area V1 and FEF. In area V1, spiking activity was recorded from chronically implanted multielectrode arrays with Tucker–Davis Technologies multichannel recording equipment. As in previous studies (5–9), the signals from the electrodes were amplified, band-pass filtered (500–5000 Hz), full-wave rectified, and then low-pass filtered at 500 Hz and sampled at a rate of 763 Hz. The MUA represents the pooled activity of a number of single units in the vicinity of the tip of the electrode. The population response obtained with this method is therefore expected to be identical to the population response obtained by pooling across single units. A recent study demonstrated that the MUA signal indeed provides a reliable estimate of the average single-unit response (10). After the postoperative recovery, we first measured the dimensions of the V1 receptive fields by determining the onset and offset of the visual response to a slowly moving light bar, for each of eight movement directions (10). The median area of V1 receptive fields was 0.35 deg² (range 0.11–4.2 deg²), and the eccentricity ranged from 1.05º to 5.46º with a median of 3º.

The responses of single neurons in area FEF were recorded with tungsten electrodes (FHC, impedance ~2 MΩ), which were lowered through the dura with a hydraulic microdrive (Narishtig). Spikes were detected if they crossed a threshold that was determined by the experimenter. Spikes were sorted offline using the MClust toolbox (MClust 3.4, A. D. Redish) in MATLAB. Upon isolation of a neuron, we first mapped its response field by presenting saccade targets at various directions and eccentricities, as described elsewhere (3). To monitor FEF activity in the curve tracing task, we positioned one of the circular saccade targets near the center of the neuron’s RF. The other circle was positioned outside the RF, at an angle of ~90º. One of the curves fell in the receptive fields of the V1 neurons. At the end of the recording session, we usually confirmed that the electrode penetration was in FEF with intracortical microstimulation (biphasic current pulses, 100-ms train duration, 200 Hz). The penetration was considered to be in FEF if a saccade could be triggered using currents that were <100 μA (usually <50 μA).

**Estimation of the Latency of the Visual Response and Attentional Modulation.** We estimated the latency of attentional modulation by fitting a function \( f(t) \) (Eq. 1) to the difference in response evoked by the target and distractor curve. For the fitting of the visual response we used a more complex curve (3) that exhibits a peak and then reaches a lower sustained level:

\[
\begin{aligned}
    f(t) = & d \cdot \exp\left(\mu t + 0.5\sigma^2 - \mu t\right) \cdot G(t, \mu + \sigma^2, \sigma) + c \cdot G(t, \mu, \sigma),
\end{aligned}
\]

The shape of this function is determined by five parameters, \( \mu, \sigma, a, c, \) and \( d; G(t, \mu, \sigma) \) is a cumulative Gaussian. The latency of the visual response was defined as the point in time where the fitted function reached 33% of its maximum. We computed a 95% confidence interval for the latency of the visual and attentional response modulation with a bootstrapping procedure. If there are \( N \) recording sites, we randomly selected \( N \) sites with replacement and determined the latency using the curve-fitting method described above. We repeated this procedure 10,000 times to estimate the 95% confidence interval.

**Computation of Noise Correlations.** Noise correlations between FEF and V1 neurons (interareal correlations) and between pairs of V1 recording sites (intraareal correlations) were computed as follows. We measured the response strength in each trial by counting the number of spikes in FEF and by measuring the amplitude of the MUA response in V1 in a time window from 200 to 500 ms after stimulus onset. To compute the noise correlation, we combined data of the different difficulty levels. We removed the possible influence of differences in firing rates between difficulty levels by first z-scoring the single trial responses within difficulty levels, and then we computed the Pearson correlation coefficient of the z scores pooled across difficulty levels (Fig. 5). We applied a Wilcoxon signed-rank test to measure the significance of differences in correlation between conditions.

3. Khayat PS, Pooresmaeili A, Roelfsema PR (2009) Time course of attentional modulation by fitting a function \( f(t) \) (Eq. 1) to the difference in response evoked by the target and distractor curve. For the fitting of the visual response we used a more complex curve (3) that exhibits a peak and then reaches a lower sustained level.
Fig. S1. Example simultaneous recording from FEF area V1. (A and B) Responses of an example FEF cell and a V1 recording site. (Top) The FEF and V1 RF relative to the stimuli. The PSTHs depict neuronal responses of the FEF cell (A) and the V1 neurons (B) evoked by stimuli at the three levels of difficulty. The red and blue traces show the responses evoked by the target and distractor curve, respectively. In both A and B, lower panels show the visual response (averaged across target and distractor curve (T + D)/2) and the attentional response modulation (T − D). Purple and green curves were fitted to estimate the latency of the visual response and the response modulation, respectively. On the right side of the graphs, PSTHs have been aligned on the saccade onset, defined as the moment when the monkey’s gaze left the fixation window (dashed line).
Fig. S2. Activity of visual and visuomovement neurons in FEF and comparison with area V1. The red and blue traces show the responses evoked by the target and distractor curve, respectively, for visual neurons (A) and visuomovement neurons (B) in FEF and recording sites in area V1 (C). In A–C, Lower panels show the attentional response modulation. Curves were fitted to estimate the latency of the response modulation (green). Rectangles on the x axis show the mean estimate ±s.d. (determined with bootstrapping). On the right side of the graphs, the PSTHs were aligned on the saccade onset.
Fig. S3. Population multiunit responses in FEF and area V1. Average responses of all FEF MUA recordings (A) and area V1 MUA recordings (B). All conventions are as in Fig. S2.
Fig. S4. Noise correlations between FEF and V1 after stratification to equalize response magnitudes. Bars show noise correlation between FEF and V1 evoked by the target curve (gray) and distractor curve (white). **$P < 0.0001$. 

N = 95