

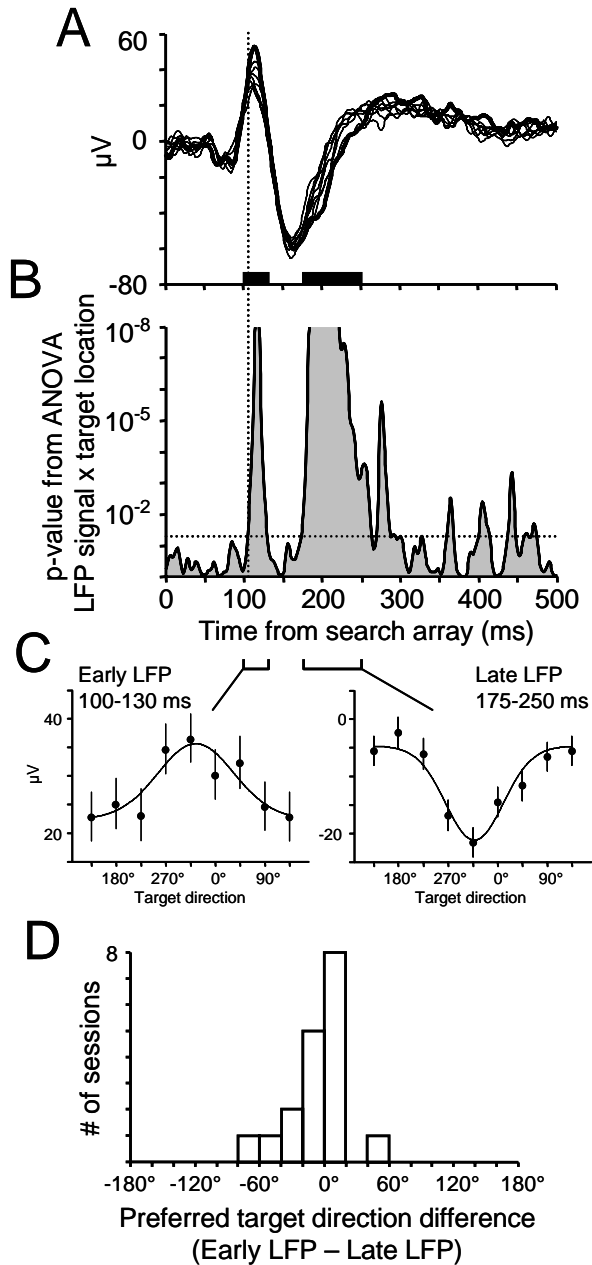
Measurements of Simultaneously Recorded Spiking Activity and Local Field Potentials Suggest that Spatial Selection Emerges in the Frontal Eye Field

Ilya E. Monosov, Jason C. Trageser, and Kirk G. Thompson

Supplemental Results

Spatial Tuning in Early vs. Late Time Periods in the Covert Visual Search Task. *Selection time* is defined as the first time that there was significant spatial tuning. In the covert visual search task, the earliest spatial tuning in the LFP from 18 of the recording sites was short-lived and positively tuned. In each of these 18 recordings there was also a later, longer lasting and negatively tuned response that was more significant statistically. The LFP shown in manuscript Figure 3 is one example of this pattern although the separation of the early and late time periods is not clear. A clearer example of this pattern is shown here in Supplemental Figure 1. Instead of attempting to combine two periods of spatial tuning that have opposite polarities, we chose to report the spatial tuning parameters of the later, more strongly tuned and therefore more reliable response. Here we show that there is a strong correlation between the spatial tuning in the two time periods. The spatial tuning is consistent throughout the LFP response and *selection time* measures the first time that this spatial tuning emerges in the LFP. For these 18 recording sites, the LFP selection times followed the spike selection times by an average of 13.1 ms.

The LFP shown in Supplemental Figure 1A was recorded in Monkey C performing the “identity” version of the covert search task (Figure 1B, lower task). The ANOVA analysis identified two separate time periods that exhibited responses that differed across target locations (Supplemental Figure 1B). Supplemental Figure 1C compares the spatial tuning of the LFP response during the early time period (100-130 ms) and the late time period (175-250 ms). The polarity of tuning is opposite and the responses during the early period are more variable than during the late period, but the preferred target directions are nearly identical. Across the population, there was a strong correlation between the preferred target directions from the early and late time periods ($p = 0.005$; Supplemental Figure 1D). The preferred target directions of 16 of the 18 LFPs (89%) were separated by less than the distance between adjacent target locations (60° for Monkey S and 45° for Monkey C).



Supplemental Figure 1.

Comparison of early and late spatial tuning in LFPs recorded during visual search.

(A) The superimposed average LFP response for each target position recorded in the FEF of Monkey C performing the “identity” version of the covert visual search task.

(B) The p-value (ANOVA) at each millisecond that estimates the probability that the LFP response did not vary across target locations. The selection time of the LFP response at this recording site is 105 ms. A second period of spatial selectivity begins at 174 ms.

(C) Spatial selectivity measured during the early (100-130 ms) and late (175-250 ms) time periods identified in the ANOVA analysis shown in (B). The parameters of the best fit Gaussian curve from the early time period (left) are $B = 22.48$, $R = 13.15$, $\Phi = -35.08^\circ$, and $T_\phi = 69.21^\circ$; and from the late time period (right) are $B = -4.72$, $R = -16.43$, $\Phi = -41.59^\circ$, and $T_\phi = 52.48^\circ$.

(D) The distribution of preferred target direction differences measured from the early and late time periods from the 18 LFPs that exhibited significant spatial tuning in two separate time periods in the visual search tasks. The distribution is peaked near 0° (Rayleigh test; $p < 0.001$), and a circular correlation analysis (Mardia and Jupp, 2000) showed that the preferred target directions are correlated ($p = 0.005$).

Signal Variability and the Reliability of Timing Measurements. The accuracy of the timing relationships between LFPs and spikes presented in this study depends on the reliability of the signals and analysis methods. If the amount of variability was different for spikes and LFP signals, then the results of the ANOVA analysis comparing the timing relationships between the spikes and LFPs are questionable. Therefore we compared the variability of the LFP responses and spiking activity at each recording site by calculating

a variability index (VI) for each signal, a ratio of two standard deviations that measures signal variability across trials relative to the variability across target positions. The numerator of the VI is the standard deviation across all trials measured at each millisecond and averaged over the first 50 ms following search array presentation, and the denominator is the standard deviation across the mean activity for each target position averaged over the same time period. The VI for spikes (6.8 ± 0.4) was not significantly different from the VI for LFPs (6.6 ± 0.4) (paired t-test; $p = 0.6$). The results were the same when the initial non-selective visual response was included by calculating VI from the time of search array presentation to the measured selection time. This indicates that the later selection times in the LFPs than in the spiking activity determined from the ANOVA analysis was not a false result caused by more variability in the LFP data than in the spike data.

Another indication that the later LFP selection times in the visual search task are not due to the quality of the LFP signal or the analysis methods is the result of the selection time analysis in the memory-guided saccade task. Overall, approximately 2.5 times more trials were included in the analysis of visual search data (an average of 290 trials per session) than were included in the analysis of the memory-guided saccade data (an average of 110 trials per session). Because variability affects an ANOVA more when there are fewer samples, it should have affected the selection time results from the memory-guided saccade task more than from the visual search tasks. But in the memory-guided saccade task, the selection times were on average 9.9 ± 2.5 ms earlier in the LFPs than in the spikes (Figure 6C). This shows that the later LFP selection times observed in the visual search task (Figure 6B) are unlikely due to the analysis method.

Effects of Signal Filtering. We were concerned that the signal filtering during data acquisition distorted the recorded LFP and artificially delayed the selection times measured in the LFPs relative to spikes (Nelson et al., 2006). One indication that this is not a serious problem in our data is the earlier LFP visual onset latencies measured in the visual search tasks (Figure 6A) and the earlier LFP selection times in the memory-guided saccade task (see above and Figure 6C). If signal filtering caused the delay of the selection times in the visual search LFP signals, it should have also caused a delay of selection times in the memory-guided saccade LFP signals. But to be sure, we directly tested whether signal distortions during data acquisition affected the results. This test is illustrated for two recording sessions in Supplemental Figure 2. First we appended 100 ms of a 40 Hz sine wave to the beginning and end of an actual LFP signal recorded in the covert visual search task. The appended sine waves were used to align temporally the original LFP signal to signals re-recorded in the same Plexon data acquisition system (Plexon Inc.) used to record the original LFP (Supplemental Figure 2A). The original LFP signals with the appended sine waves were converted into “.wav” sound files in Matlab (The MathWorks, Inc). We used a portable battery powered audio compact disc player (Panasonic Model No. SL-CT582V) to feed the .wav file signal into a beaker of 0.9% saline in which we placed a 1 M Ω metal electrode used in the neural recordings. The electrode was connected to the filtered (3.3 – 88 Hz) Plexon LFP input channel. An unfiltered signal was simultaneously recorded on a separate a/d card (National Instruments) in the Plexon system normally used to record eye position. Supplemental Figure 2A shows that the continuous voltage signal recorded on the Plexon analog input channel from the portable audio player effectively reproduced the original LFP signal.

(We tried playing the .wav file from various laptop computers, but the sound cards attenuated the lower frequencies.)

We replaced the actual LFP signal in the original data file with the re-recorded LFP signals and performed the same time course analysis. Supplemental Figure 2B shows the analysis performed on the .wav signal recorded on the unfiltered analog input channels. The original LFP signals recorded in the visual search task are shown in Figure 3 and in Supplemental Figure 1. The average stimulus related LFP signals for each target position are nearly identical to the originals, and the ANOVA time course analysis generated the exact same selection times as the originals. This shows that the .wav file and audio signals reliably reproduced the originally recorded LFP signals. Supplemental Figure 2C shows the analysis performed on the .wav signals recorded on the filtered LFP input through a 1 M Ω metal electrode in 0.9% saline. There were some distortions of the average stimulus related signals that are apparent when compared to the original LFP signals, and the significance levels of the ANOVA analysis were lessened. But the important result was that the measurement of selection time was not appreciably affected. We performed this test on the LFP signals from 5 separate recording sessions (3 from monkey S and 2 from monkey C). The selection times determined from the .wav file signals recorded on the LFP input channel ranged from 1 ms before to 2 ms after the selection times determined from the .wav file signal recorded on the analog input channel. In summary, the results were the same from all 5 sessions – the signal filtering during data acquisition somewhat altered the shape of the evoked LFP response, but did not affect the determination of selection time.

Supplemental Figure 2 (on next page).

Testing the effects of signal filtering on the determination of selection time.

(A) Voltage signals from the original LFP recording (red line) and audio .wav file signals re-recorded through an unfiltered analog input channel (dark blue) and the filtered LFP input channel (light blue). A 40 Hz sine wave was appended to the original before creating the .wav file to temporally align the re-recorded signals.

(B) The superimposed average .wav signals recorded through the analog input channels for two LFP signals from two different monkeys (top), and results of the ANOVA time course analysis (bottom). The selection times determined from these re-recorded signals are the same as from the original LFP signals (left – 142 ms, right – 105 ms).

(C) The superimposed average .wav signals recorded through the Plexon LFP input channel for the two LFP signals (top), and results of the ANOVA time course analysis (bottom). The selection times determined from these re-recorded signals are nearly the same as from the original LFP signals (left – 144 ms, right – 104 ms).

References

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Supplemental Figure 2

