Neural interactions between motor cortical hemispheres during bimanual and unimanual arm movements

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Abstract
Cortico-cortical connections through the corpus callosum are a major candidate for mediating bimanual coordination. However, aside from the deficits observed after lesioning this connection, little positive evidence indicates its function in bimanual tasks. In order to address this issue, we simultaneously recorded neuronal activity at multiple sites within the arm area of motor cortex in both hemispheres of awake primates performing different bimanual and unimanual movements. By employing an adapted form of the joint peri-stimulus time histogram technique, we discovered rapid movement-related correlation changes between the local field potentials (LFPs) of the two hemispheres that escaped detection by time-averaged cross-correlation methods. The frequency and amplitude of dynamic modifications in correlations between the hemispheres were similar to those within the same hemisphere. As in previous EEG studies, we found that, on average, correlation decreased during movements. However, a subset of recording site pairs did show transiently increased correlations around movement onset (57% of all pairs and conditions in monkey G, 39% in monkey P). In interhemispheric pairs, these increases were consistently related to the mode of coupling between the two arms. Both the correlations between the movements themselves and the interhemispheric LFP correlation increases were strongest during bimanual symmetric movements, and weaker during bimanual asymmetric and unimanual movements. Increased correlations occurred mainly around movement onset, whilst decreases in correlation dominated during movement execution. The task-specific way in which interhemispheric correlations are modulated is compatible with the notion that interactions between the hemispheres contribute to behavioural coupling between the arms.

Introduction
Movements involving different joints or limbs can require precise coordination of the timing and magnitude of activation of multiple sets of muscles. How this is achieved by the nervous system remains an unresolved question. One particular example of motor coordination, movements requiring simultaneous use of both arms or hands (bimanual movements), has been intensively investigated. Movements of the two arms have the tendency to produce spatially (symmetric) and temporally (phase-locked) similar movements (Kelso et al., 1979; Kelso, 1984; Franz, 1997). On the other hand, humans can be trained to produce complex combinations of bimanual movements without these symmetries, such as piano playing. What is the neurophysiological substrate of the linkage connecting the arms, and how is it overridden in bimanual tasks that require their independence? A number of studies suggest that the neocortex and, in particular, callosal interconnections between the hemispheres, may be involved in coordination of the upper limbs. Interhemispheric interactions are enhanced during bimanual learning (Andres et al., 1999), and split-brain patients (lacking direct interhemispheric connections) have difficulties learning novel patterns of spatial bimanual coordination (Preilowski, 1972, 1975; Franz et al., 2000). They also have considerably fewer problems than normal individuals in producing simultaneous but very different movements of their two hands (Eliassen et al., 1999; Franz et al., 1996). Although some studies found intact temporal coupling in split-brain patients (Tuller & Kelso, 1989; Franz et al., 1996; for review see Donchin et al., 1999), a newer report also showed deficits in the temporal domain (Eliassen et al., 2000).

Aside from these observations, little is known about the physiological mechanisms underlying bimanual coordination and interhemispheric interactions. Early work using a finger-tapping task (involving only distal muscles) supported the classical view that activity in primary motor cortex (MI) reflects mainly contralateral movements and is hardly affected by whether the movement is unimanual or bimanual (Tanji et al., 1987, 1988). More recently, however, two groups investigated the physiology of bimanual movements of the whole arm (involving proximal muscles), and both found, in different paradigms, that neuronal activity in MI could not be accounted for by the activity evoked during their unimanual components (Donchin et al., 1998; Kermadi et al., 1998). Whilst these results support the hypothesis that MI is involved in coding bimanual arm movements, they do not provide insight into the interactions between concurrently planned movements of the two arms. The present study addresses these interactions. We simultaneously recorded electrical activity from multiple sites within the motor cortex of both hemispheres. Local field potentials (LFPs) and
A. The Setup

B. Unimanual Movement Types

Unimanual left

\[\uparrow \downarrow \uparrow \downarrow\]

Unimanual right

\[\uparrow \downarrow \uparrow \downarrow\]

C. Bimanual Movement Types

Monkey G

\[\uparrow \uparrow \uparrow \uparrow \downarrow \uparrow \downarrow \uparrow \downarrow\]

\[\uparrow \uparrow \uparrow \uparrow \downarrow \uparrow \downarrow \uparrow \downarrow\]

\[\uparrow \uparrow \uparrow \uparrow \downarrow \uparrow \downarrow \uparrow \downarrow\]

\[\uparrow \uparrow \uparrow \uparrow \downarrow \uparrow \downarrow \uparrow \downarrow\]

Monkey P

\[\uparrow \uparrow \uparrow \uparrow \downarrow \uparrow \downarrow \uparrow \downarrow\]

\[\uparrow \uparrow \uparrow \uparrow \downarrow \uparrow \downarrow \uparrow \downarrow\]

\[\uparrow \uparrow \uparrow \uparrow \downarrow \uparrow \downarrow \uparrow \downarrow\]

\[\uparrow \uparrow \uparrow \uparrow \downarrow \uparrow \downarrow \uparrow \downarrow\]

Fig. 1. The behavioural task. (A) The monkey moved two manipulanda in the horizontal plane. The position of each manipulandum was displayed as a cross-shaped cursor on a vertical screen in front of the monkey. Each trial began by presenting two origin circles in the middle of the display (circles with crosses). After the monkey placed the cursors into the origins and held them immobile for a constant delay, the origin circles went off and two target circles appeared at different locations. As an example, the display for a bimanual parallel movement of the same direction and amplitude is sketched. In unimanual trials, one circle appeared in the same location as the origin (for the nonmoved hand). The other circle was displaced from the origin in one of the eight directions shown in B. (C) Schematic representation of the bimanual movements used in our task. For monkey G, movements consisted of moving both arms to the front, or movements in which the two arms were moved in perpendicular directions. In addition, movements of different amplitudes were tested, and all possible combinations of short and long movements were used. In monkey P, we used only movements of the same amplitude. Directions were either the same, perpendicular or opposite. Upward arrows in B and C correspond to forward movements of the monkey, and downward arrows to backward movements towards the chest of the monkey.

Methods

Behavioural paradigm

Two female rhesus monkeys (Macaca mulatta; monkey G, 3.5 kg, and monkey P, 4 kg) were trained to move two separate manipulanda, one with each arm. The experimental setup and the principle of the task design are described in Donchin et al. (1998). A sketch of the monkey engaged in performance of the task is shown in Fig. 1A. Each manipulandum was a low-weight, low-friction, two-joint mechanical arm, moveable only in the horizontal plane. Movement of each manipulandum caused movement of a corresponding cursor on a vertically orientated 21-inch video screen located ≈50 cm in front of the monkey. The movement of each cursor was mapped to its corresponding manipulandum movement such that each millimeter of manipulandum movement caused one millimeter of movement of the cursor on the video display.

The time course of typical unimanual and bimanual trials was as follows. A trial began when the monkey placed both cursors within 0.8 cm diameter ‘origins’ (Fig. 1A) and held them still for 500 ms (monkey G) or 1000 ms (monkey P). For each arm, a target (also 0.8 cm diameter) could appear at a distance of 3 cm (monkey P) from the origin. For monkey G, targets could occur at long (5 cm) or short (2.5 cm) distances from the origins. If only one target appeared, signalling a unimanual trial, the monkey moved the appropriate arm and brought the corresponding cursor into the target, but did not move the other arm. If two targets appeared, signalling a bimanual trial, the monkey moved both arms such that the two cursors moved into the targets on the screen. Three types of bimanual movements were studied: parallel, opposite and perpendicular. The types of bimanual trials used during recordings are shown in Fig. 1C. Note that for monkey G the bimanual movements could also entail movements of different amplitudes, but opposite movements were not included. Unimanual movements comprised movements in eight different directions (in all four cardinal directions, plus those directions 45° from the cardinal directions; Fig. 1B). For monkey G we also included long unimanual movements which were components of the bimanual movement types.

The monkey’s reaction time was not restricted, but targets had to be reached within 1 s (monkey G) or 1.5 s (monkey P) from target appearance. For bimanual trials, the animal was additionally required to begin movement of the arms within a maximal interarm interval (IAI) of 200 ms and the targets had to be reached with an IAI of 400 ms. Following acquisition of the targets, the monkey held both arms still in the target circle for at least 500 ms. No intertrial interval was imposed. Every second or third successful trial was rewarded with a liquid reward and followed by a 2-s pause to allow for its consumption. In all sessions, trials were presented pseudo-randomly without any separation into blocks. In monkey P, we introduced an additional criterion for successful trials in order to limit spatial intertrial variability in movement execution. This criterion checked whether movement paths were within a narrow straight virtual ‘tunnel’ connecting the origin and the target circle. The width of this tunnel was approximately 2 cm, and the length was 20 cm.

Single-unit activity were recorded from each electrode (Mitzdorf, 1994). This paper focuses on the LFP signal, which represents the grand average of the synaptic input to the local cortical network in the vicinity of the recording electrode. The data analysis is aimed at evaluation of interactions between neuronal populations as reflected in the LFP signal and their relationship to coordination of bimanual movements. Part of this study has been published in abstract form (Cardoso de Oliveira et al., 2000).
path was the same as the target circle diameter. This criterion eliminated deviations in movement direction, particularly those leading to curved movements. Because successful performance was more difficult under these conditions, monkey P had lower success rates than monkey G (see Results).

In addition to this behavioural paradigm, we tested the monkeys’ hand preferences in a ‘raisin board’ task. The monkey sat in the primate chair and faced a rectangular Perspex board containing 10 wells of ≈ 3 cm in diameter, arranged in two rows of five wells with a raisin in each well. The board was positioned in the middle, ≈ 30 cm in front of the monkey’s chest. The monkey rapidly collected all raisins from the wells. The behaviour of the monkey in this task was taped on video, and we counted how often the monkey used each hand to retrieve the raisins.

**Surgery**

After training, we implanted a recording chamber (27 × 27 mm) above each hemisphere, and a head holder was attached to the occipital bone. The surgery was performed under general anaesthesia and aspecific conditions. Magnetic resonance images (taken in a Biospec Bruker 4.7-Tesla animal system, fast-spin echo sequence; effective TE = 80 ms and TR = 2.5 s, 13 coronal slices 2 mm wide) aided in placement of the chambers. In Monkey P, the position of the recording chambers and the orientation of two electrodes that were inserted through the chambers into the brain were verified by a second MRI session after surgery. The animals’ care and all surgical procedures were in accordance with the NIH Guide for the Care and Use of Laboratory Animals (rev. 1996) and all applicable Hebrew University regulations.

**Recording**

During recording sessions, the monkey was seated in a primate chair placed in a dark chamber with its head fixed. Single-unit activity and LFPs were recorded by eight glass-coated tungsten microelectrodes (impedance 0.2–0.8 MΩ at 1 kHz) in the two hemispheres (four electrodes in each hemisphere). Electrodes were driven into each hemisphere through eight tubes that were tightly fitted into a metal guide, with seven of the tubes forming a circle and one in the middle. The horizontal distance between the four electrodes we inserted could be either ≈ 350, ≈ 600 or maximally ≈ 700 μm, depending on the relative positions in the guide. In normal placements, only two electrodes were spaced at the minimal distance, and the others were spaced at the intermediate or maximal distance. Electrodes were individually driven to allow optimal isolation of spiking activity. Therefore, the difference in depth between electrodes could vary from one session to another. The electrode signals were amplified and filtered by a multichannel analogue data processor (MCP, Alpha-Omega, Nazareth, Israel). The raw wideband signal was subjected to two different ranges of band-pass filters in order to yield spike activity (filtered between 300 Hz and 8 kHz) and LFP signals (filtered between 1 and 150 Hz). The LFP data were sampled at 400 Hz and stored on disk. As reference we used a grounded metal screw, which was implanted into the skull and connected by conductive wires that were wrapped around all screws that held the two chambers. Pick-up of mechanical or electrical noise was minimized by mechanically attaching the drives and head stages to the implanted chambers.

Fifty Hertz noise caused by the a.c. power line was attenuated using a digital notch filter applied off-line (49.8–50.2, 99.8–100.2 and 149.8–150.2 Hz four-pole Butterworth filter, applied forward and backward to prevent phase shifts).

**Selection of recording sites**

At each penetration coordinate, four electrodes were advanced into the brain. After penetrating the dura, electrodes were separately advanced until well-isolated units were detected. Recording sites were not especially selected for activity related to movements, but for the occurrence of stable single-unit recordings (as judged by stability of spike amplitude and spike rate). LFP channels numbered 0–3 were always recorded by microelectrodes in the right hemisphere, and channels numbered 4–7 were recorded in the left hemisphere. At the end of each recording session, we tested for neuronal responses to passive manipulation and tactile stimulation of the limbs, tail, trunk and head. Evoked activity was evaluated by listening to the amplified spike signal passed directly into a loudspeaker. Finally, we applied intracortical microstimulation (ICMS) with 50-ms trains of 200-μs cathodal pulses at 300 Hz with an intensity of 10–80 μA (BPG-2 and BSI-2, BAK Electronics, Germantown, MD, USA). When ICMS evoked movements, we documented the movements evoked and the threshold stimulation intensity. Stimulation and passive manipulation were performed at the end of each recording session, as well as in dedicated mapping penetrations. For this study, we only included recording sites that were well within the arm representation, as determined by passive stimulation and microstimulation.

After completion of recordings, monkey G was killed with an overdose of Nembutal® and perfused transectadially by 0.9% saline and paraformaldehyde fixative solution (4% paraformaldehyde in 0.1 M phosphate buffer). The brain was removed and photographed, sunk in sucrose, sliced on a cryostat (50 μm section width) and Nissl-stained with Cresyl Violet. Penetration positions were confirmed to be in the motor cortex by mapping them onto the surface of the brain using the chamber coordinates and positions. All our recordings were from the exposed surface of the precentral gyrus. Most of the sites were in primary motor cortex, but some of the more anterior sites were located at the border to the caudal portion of dorsal premotor cortex. Monkey P is still participating in experiments. Therefore, the locations of the recording sites in monkey P were determined by magnetic resonance images. During imaging, we placed microelectrodes into the brain at known chamber coordinates. In this way, the anatomical positions of the microelectrodes could be confirmed as lying within the motor cortex.

**Data analysis**

All recording sites were assessed for intertrial stability of LFP signals and for the occurrence of excessive noise. Recordings with recurring artifacts, and those with strong 50 Hz noise persisting after off-line filtering, were excluded from analysis. We also excluded portions of the data in which the LFP recordings changed considerably across trials (e.g. baseline shifts or changes in noise level). No selection was made on the basis of whether or not sites displayed task-related activity.

All LFP traces were aligned upon the beginning of movement, determined by an off-line algorithm, and double-checked manually. The movement initiation detection algorithm (courtesy of A. Arieli) calculated the zero intercept of a line passing through two points on the velocity profile. One point was at 2/3 of the peak speed and the other was at 1/3 of peak speed. It also included corrections for different special cases of unusual velocity profiles. In order to determine movement duration, the end of movement was detected similarly. For purposes of alignment, the beginning of movement in bimanual trials was determined by the first arm to begin moving.

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call the average of all aligned LFP signals recorded during identical movement types the mean motor evoked potential (mEP; see Fig. 3B).

**Determining preferred directions (PDs) of LFPs**

In order to determine the preferred direction of the LFPs recorded from a given site, we measured the peak-to-peak size of the mEP by calculating the distance between the maximum and the minimum of the mEP. Out of the eight directions movements of the contralateral arm, we constructed a tuning curve of these peak-to-peak values (observed tuning). We fit a cosine to these eight points using a simple least-squares fit to the equation:

$$PTP(\theta) = A \cdot \cos(\theta - PD) + B$$

$$\theta = 0^\circ, 45^\circ, ..., 315^\circ (1)$$

\(\theta\) is defined such that 0° corresponds to movements to the right, and angles proceed in a counterclockwise direction. We extracted the direction of the maximum of the cosine function (PD) and the goodness of fit, as measured by \(r^2\) (the variance of the fit divided by the variance of the original model). If the \(r^2\) value exceeded 0.6, we considered the LFP to be significantly tuned.

**Cross-correlation analysis**

As a first approach towards studying correlations between LFPs, we calculated time-averaged cross-correlograms between LFP channels. We applied this analysis to two different epochs of the trial. The first was the interval between 750 and 250 ms before movement onset, during which the monkey held its hands stationary at the origins and waited for the target (or targets) to appear. We call this time window the hold period. The second window contained the time from 250 ms...
before to 1000 ms after movement onset, during which motor preparation and movement execution occurred. We call this time window the movement period. For all possible pair-wise combinations of simultaneously recorded LFPs, we calculated the correlograms obtained during these two time windows, using time delays between −200 and +200 ms, and a bin width of 2.5 ms, which was our sampling resolution. To this end, we used the Matlab xcorr function (MatLab, MathWorks, Natick, MA, USA), with correlation expressed as the correlation coefficient. The correlogram of a single trial is defined by the following equation:

\[
CC(\tau) = \frac{\sum_{i=1}^{T} [LFP1(t) - \bar{LFP1}] \cdot [LFP2(t+\tau) - \bar{LFP2}]}{\sqrt{\sum_{i=1}^{T} [LFP1(t) - \bar{LFP1}]^2 \cdot \sum_{i=1}^{T} [LFP2(t+\tau) - \bar{LFP2}]^2}},
\]

where \(\tau\) is the time delay between the signals, \(t\) is the time bin out of \(T\) total time bins and a bar above \(LFP1\) or \(LFP2\) indicates the average of each trial.

The correlograms obtained for all trials recorded under the same conditions (during the same movement type or during the hold period) were averaged to reduce noise. Examples for such correlograms, obtained from the hold period, are shown in Fig. 5. From each correlogram, we determined the maximum value. Negative correlations constituted only \(\approx 1\%\) of our total sample and were not analysed separately. For correlation peaks \(> 0.1\), we also determined the time lag of maximum correlation.

The conventional cross-correlation is affected both by similarities of the average signals (which, during the movement period, correspond to the mEPs) in the two electrodes and by possible trial-wise interactions between the single trial signals. A common way to distinguish between these two aspects is to calculate a ‘shift predictor’ to approximate the correlation between the averages, and then subtract it from the correlograms to derive an estimate of the ‘pure’ trial-wise correlation. We calculated the shift-predictor by correlating the \(i\)-th trial of one electrode with the \((i+1)-\)th trial of the other, and the last trial with the first trial.

The shift predictor was also used to define a confidence limit for oscillatory components in cross-correlograms. As a confidence limit for oscillatory correlation, we chose the mean (over all delays) of the predictor \(\pm 3\) SD (again, over all delays). Whenever we found at least one satellite peak (on each side of the main correlogram peak) which exceeded these values, we scored a correlogram as having a significant oscillatory component.

The joint peri-event time correlogram (JPETC)

To study the temporal aspects of the modification of correlation strength, we used a dynamic correlation method, based on the joint peri-stimulus time histogram (JPSTH), developed by Aertsen and coworkers for single-neuron data (Aertsen et al., 1989). We adapted this method to LFP signals and produced the JPETC. The JPETC is a matrix of the correlation coefficients of two analogue channels for all possible combinations of paired time lags relative to an external event. We calculated the JPETC in an epoch starting 750 ms before movement onset and continuing to 1000 ms after movement onset. Using a time resolution of 2.5 ms, there were 700 bins in each segment. The correlation coefficients were calculated using the equation:

\[
CC(t_1, t_2) = \frac{\sum_{n=1}^{N} \left[ LFP1_n(t_1) - \bar{LFP1(t_1)} \right] \cdot \left[ LFP2_n(t_2) - \bar{LFP2(t_2)} \right]}{\sqrt{\sum_{n=1}^{N} \left[ LFP1_n(t_1) - \bar{LFP1(t_1)} \right]^2 \cdot \sum_{n=1}^{N} \left[ LFP2_n(t_2) - \bar{LFP2(t_2)} \right]^2}},
\]

where \(t_1\) is the time bin from LFP1, \(t_2\) is the time bin from LFP2, and \(n\) is the \(n\)-th trial out of a total of \(N\). A bar over \(LFP1\) or \(LFP2\) in Eqn 3 indicates that the mean should be taken across trials (thus, \(\bar{LFP1}\) and \(\bar{LFP2}\) are mEP1 and mEP2). The result takes the form of a 700 × 700-bin matrix constituting all possible time delays between LFP1 and LFP2. For example, the values corresponding to the simultaneous (zero-delay) correlation are situated along the main diagonal of this matrix. Figures 2 and 6–8 give examples of JPETC matrices displayed using a colour-coded flat display. Time progresses from the bottom left to the top right corner such that the value of \(t_1\) (the time index of the first LFP) increases along the \(x\)-axis and the value of \(t_2\) (the time index of the other LFP) increases along the \(y\)-axis. Finally, averaging all diagonals from bottom left to top right yields a cross-correlogram. Because of differences in normalization, however, this correlogram is not identical to the one obtained by the classical cross-correlogram technique mentioned above. The bin-wise
significance of the correlation coefficients in the JPETC was determined by testing against the hypothesis that the correlation coefficient is 0, using a standard t-test.

Theoretically, the JPETC procedure should result in correlation values that are not contaminated by similarities in the evoked potentials of the two signals. We checked that this was the case using two different tests. First, we applied the JPETC algorithm on sham data with similar time-dependent mean waveforms and uncorrelated additive trial-by-trial noise. We created 103 trials of two such sets of sham data by randomly drawing, for each trial and each time bin, a value from a normal distribution with the same mean and SD, as observed in two sets of real data. As expected, this procedure resulted in JPETC-displays without any structure (Fig. 2A). Both the diagonal and the correlogram (calculated by averaging the diagonals) were flat. Whilst these random sham data replicated the similarities of the means over all trials (the mEPs), they did not have the same temporal structure within a trial as the real data.

As a second method, we calculated a shift-predictor for the JPETC by correlating signals between subsequent instead of simultaneous trials (and the last trial with the first trial). Figure 2B shows the result of this procedure. For comparison, the JPETC of simultaneous trials data are displayed in Fig. 7B. Although the evoked potentials are identical, as they must be, the shifted JPETC displays no correlation whatsoever, either in the JPETC itself or in the average cross-correlogram. In contrast, the original data do show a clear correlation along the main diagonal, which is strongly modulated around the time of movement onset.

Third, we addressed the possible concern that the JPETC may be sensitive to changes in signal variance. It has been observed that, during evoked potentials, the variance of cortical activity is usually reduced slightly by about 10–20% (A. Arieli, unpublished observations). In order to check whether a change in variability could lead to a change in correlation strength observed in the JPETC, we created sham data in which the correlation coefficient between the two signals was held constant but the variance of the individual signals was increased or decreased over time. We found that the JEPTEC faithfully reflected the correlation coefficients and was not influenced by a change in variance (Fig. 2C).

When analysing spike data with the JPSTH technique, it has been shown that intertrial covariations in neuronal excitability and response latency can produce patterns of correlations in the JPSTH (Brody, 1999; Baker & Gerstein 2001; Ben-Shaul et al., 2001). This intertrial covariability will not show up in the shift predictor. As a result, when interpreting the JPSTH one has to take into account that its features could be caused by either ‘real’ spike timing correlations or by intertrial covariability. We have tested the effect of intertrial covariations on the JPETC method of analogue data. To simulate covariations in signal amplitude, we created surrogate data by adding random, independent noise to two mEPs which were randomly scaled in each trial by a common factor. The JPETC of these data showed no features along the diagonal (not shown). The fact that we observed practically no effect of size covariability in our data may be related to the range of size scaling that we applied (we used an SD of 0.3, creating mEPs which varied in size between 0 and 2 times the original amplitude). This range is reasonable for LFP data, because the amplitude range of variabilities in the LFP does not exceed twice the amplitude of the mean signal. It is possible that the more profound impact on the JPSTH of spike data emerges due to the higher intertrial variability of the spike responses. We thus conclude that covariations in signal velocity do not seem to play a major role in the JPETC of LFPs.

Next, we assessed the possibility that covariability in signal timing could affect the JPETC. In sensory systems, variability in signal timing is caused by variations in response latency. In the motor system, variability in timing is related to a variable time delay between neural activity and the initiation of a movement. We created another set of surrogate data by, again, adding noise to the mEP. This time, for every trial, the mEPs of the two signals were shifted along the time axis in a correlated way (by the same amount of time). The time delay for each trial was randomly chosen from a normal
distribution. We tested random distributions with various widths (standard deviations; SD), ranging from 25 to 100 ms. Figure 2D shows the result of this procedure with a standard deviation of the time jitter of 100 ms. The correlated timing jitter in the evoked response does induce a pattern in the JPETC which is somewhat similar to the pattern observed for the real data (Fig. 7B). Smaller ranges of time jitter resulted in weaker structures in the JPETC. However, it should be noted that even with the perfectly correlated and large jitters, the strength of the maximal correlation change (as shown in Fig. 2D) was substantially weaker than the one observed in the original data (compare Fig. 7B). We conclude that correlated time jitters in the two signals may contribute to structures within the JPETC, but are clearly not the sole source of them.

In summary, the simulations demonstrate that it is unlikely that the JPETC is contaminated by the sizes or shapes of the mEPs or by overall changes in variability. It rather reveals correlations between the trial-by-trial fluctuations of the two LFP signals around the mEPs, but may also reflect covariations in signal timing. Both phenomena are interesting, because they reflect interactions caused by the underlying network architecture. Changes in correlation patterns can be interpreted as evidence for dynamic changes in the functional network structure, resulting in variable interactions between the local cortical activities at two sites. Covariations in signal timing can be seen as evidence for a common timing linking distant neuronal sites.

Because the strongest correlations and correlation changes occurred at zero time delay, we focused on the main diagonals of the JPETC in further analyses. This diagonal had a width of 1 bin, i.e. 2.5 ms, and also a temporal resolution of 2.5 ms. To test for movement-related changes in correlation, we compared the diagonals during the hold period and the movement period. In order to allow for statistical comparison, we transformed the correlation coefficients on each diagonal using Fischer’s $z$-transform. We then normalized the diagonals during the movement period by subtracting the mean correlation during the hold period (corresponding to the mean of the first 200 bins, i.e. the first 500 ms of the diagonal) and dividing by the SD within this time. We defined the confidence limit for significant changes as 3 and –3, respectively. These values correspond to deviations of $>3$ SD from the mean correlation in the hold period. Assuming a normal distribution, this corresponds to a rejection at $P < 0.002$. To assess the times during which significant correlation changes occurred most frequently, we counted, for each time bin, the

![Fig. 5. Mean trial-by-trial cross-correlations among all simultaneously recorded LFPs from one recording session, analysed (A) during the immobile hold period, and (B) during a bimanual symmetric movement to the front. Autocorrelations are included along the diagonal. Note that the correlations between the hemispheres are much smaller than those within the same hemisphere. The differences between corresponding correlograms in A and B are minor. In the right hemisphere, strong oscillations in the gamma range are present. In contrast, alpha-range oscillations predominate in the left hemisphere. Gray lines indicate confidence limits indicating the range of ± 3 SD around the mean of the shift predictor.](image-url)
number of JPETC main diagonals (for all pairs and all conditions) in which values $> 3$ (for increased correlations) or $<-3$ (for decreases in correlation) occurred.

**Results**

**Behaviour**
The monkeys were trained until they performed all movement types reliably and at a stable performance level. In monkey G, $>90\%$ of the unimanual movements were performed correctly and within the timing requirements. Bimanual movements in the same direction were executed with a performance of $>75\%$ correct movements, whilst bimanual movements differing $90^\circ$ in direction were performed with $>55\%$ correct movements. In monkey P, the average success rate was somewhat lower ($60\text{-}70\%$ in unimanual trials and $50\text{-}60\%$ in bimanual trials), probably because the requirements for successful performance were more strict (see Materials and Methods). Movements were initiated with an average reaction time (from target appearance to movement initiation) of $267 \pm 140$ ms in monkey G, and $366 \pm 42$ ms in monkey P. The average movement times (from movement initiation to target acquisition) were $529.6 \pm 143.8$ ms (monkey P, left arm), $607.0 \pm 193.8$ ms (monkey P, right arm), and $587.6 \pm 129.9$ ms (monkey G, left arm), $632.6 \pm 198.4$ ms (monkey G, right arm).

Fig. 6. Time-dependent functional interactions between LFPs during different movement types as revealed by the JPETC technique. This particular example was recorded from the left hemisphere. The plots are arranged in the same way as in Fig. 3. Diagonals have been smoothed by a seven-point averaging filter. In order to compare the time course of correlation changes to the mEPs, the mEPs are inserted in the plot of the diagonal in grey (note that the mEP sizes are scaled differently than the diagonals). Vertical dotted lines mark the time of movement onset and 0 delay correlation. JPETCs (A and B) during unimanual movements and (C and D) during bimanual movements in which both arms are moved simultaneously in the same directions as in A (right arm to the right) and B (left arm to the front). Note that the time scale for the JPETC and the correlogram are different, with the correlogram depicting only 200 bins around the main diagonal.
In bimanual movements, both monkeys started movements of their individual arms almost simultaneously. Although we allowed for a maximal IAI of 200 ms, the actually observed IAIs were considerably smaller (48.8 ± 53.8 ms in monkey G and 1.5 ± 37.6 ms in monkey P). In monkey G, the right hand began the movements before the left in 92% of the movements whilst in monkey P the right hand led in only 56% of the cases. Both monkeys preferred to use the right hand when picking up raisins from the wells in a Perspex board. Monkey G picked up 93% of the raisins with the right hand, and monkey P picked up 82% with the right hand (average over the last 3 days of testing).

Neuronal activity

LFPs were recorded at a total of 341 recording sites during 88 penetrations in the two monkeys. Out of these, 227 passed our criterion for recording quality (90 in monkey G, 137 in monkey P). All possible combinations of simultaneously recorded sites yielded 648 pairs, 162 within the right hemisphere (69 from monkey G, 93 from monkey P), 120 pairs within the left hemisphere (52 from monkey G, 68 from monkey P), and 366 pairs from different hemispheres (157 from monkey G, 209 from monkey P).

Oscillatory activity

LFP signals frequently contained oscillatory epochs in various frequency bands, in particular in the alpha (8–14 Hz) and gamma (30–55 Hz) range. In some cases, oscillatory activity was strong enough to be observed in the LFP traces of single trials. Significant oscillatory side bands (see Materials and Methods) were present in 34% of the autocorrelograms in monkey G and 33% of the autocorrelograms of monkey P. Crosscorrelograms between the LFPs recorded from different sites also contained oscillatory side bands (see, for example, Fig. 5) in 32% (monkey G) and 16%...
Contralateral to the moving hand, although a smaller ipsilateral unimanual movements, the mEPs were larger in the hemisphere mEPs recorded within the same hemisphere were similar. In movements, large mEP amplitudes were recorded in both hemispheres (Fig. 4B and C). In many cases, the mEP amplitude in bimanual movements to the front. Comparing parts A and B of the figure shows that correlations did not change substantially during movements (see also below, Table 1). Note that, in this particular example, there are strong correlated oscillations of different frequency in each of the hemispheres. The oscillation frequency in the right hemisphere was in the gamma range (= 40 Hz), whilst in the left hemisphere, both alpha (= 10 Hz) and gamma oscillations were visible. Both the intensity and the frequency range of oscillations varied from one recording site to another. There was no general preference for a certain frequency band in either hemisphere.

Figure 5 demonstrates the general finding that cross-hemispheric pairs were weakly correlated whilst intrahemispheric pairs were strongly correlated. Table 1 demonstrates this for all pairs in our sample. It presents the averaged peak sizes for all pairs, during performance of unimanual and bimanual movements. In both monkeys, the correlations between hemispheres were significantly lower than within the same hemisphere (Wilcoxon rank sum test, P < 0.001). Surprisingly, we found a difference in the intrahemispheric correlation strength of the two hemispheres. In both monkeys, correlations within the right hemisphere were significantly smaller than in the left hemisphere (Wilcoxon rank sum test, P < 0.001). The time delays associated with maximal correlation strength were centred around zero delay, with a mean of 0 ms (monkey G) and 2.5 ms (monkey P) for intrahemispheric correlations, and ~5 ms (indicating that the left hemisphere was leading, monkey G) and 2.5 ms (indicating that the right hemisphere was slightly leading, monkey P) for interhemispheric correlograms. In pairs situated in different hemispheres, the SD of peak delays was considerably higher than for those within the same hemisphere (31 as compared to 20 ms in monkey P, and 58 as compared to 32 ms in monkey G).

In monkey G, the level of interhemispheric correlations was constant and did not show any significant differences between movement types. In monkey P, all bimanual movements elicited slightly higher correlations between the hemispheres than did unimanual movements. Despite its small size, this difference turned out to be significant (Wilcoxon rank sum test, P < 0.001). No significant differences among bimanual movement types were found in either monkey. The analysis summarized in Table 1 was repeated after subtracting the shift predictor from each of the correlograms. The results were very similar, but the small difference in monkey P between bimanual and unimanual movements disappeared.

To summarize, the averaged cross-correlograms revealed stronger correlation within hemispheres than correlation between hemispheres, but failed to clearly distinguish among different types of bimanual movements.

### Dynamics of correlations: results of the JPETC-analysis

The cross-correlograms of Fig. 5 depict correlations between LFPs averaged over a long time. However, rapid modifications of correlation can easily be eliminated due to this averaging. We used the JPETC to test for such modulations. Figures 6–8 show examples of JPETCs of three pairs of sites for different movement types. The JPETC matrix is displayed in colour code in the centre of each figure.
that the two sites were correlated during all four movement types (red left hemisphere during different types of movement. The figure shows different hemispheres (Figs 7 and 8).

correlation strength could be found in JPETCs taken from pairs at zero delay (Fig. 6A, C and D, and Fig. 7B). In some cases, axes. The main diagonal of the JPETC (the diagonal row of bins from situated in the same hemisphere (Fig. 6), as well as those situated in relatively common to see negative dips around elevated correlations æ 2001 Federation of European Neuroscience Societies, European Journal of Neuroscience, 14, 1881–1896

We display the mEP of the two signals in each pair along the x and y axes. The main diagonal of the JPETC (the diagonal row of bins from the bottom left to the top right) represents the time course of zero-delay correlation and is displayed separately in the upper right corner of each figure. The time-averaged correlogram is derived by averaging the diagonals at a range of –200 to +200 ms delay.

It is worth emphasizing that the JPETC depicts the correlated activity after subtracting the mEP from each raw LFP single-trial signal (see Materials and Methods). The tests described in the methods section (Fig. 2) indicated that the correlations depicted by this procedure are not created by the patterns of mEPs. Rather, the correlation patterns reflect correlated intertrial variability of neuronal activity, which may arise from correlated noise at a given time, or from correlated time shifts of the neuronal signals.

In both monkeys, the majority of JPETCs contained significant changes along the main diagonal. In 57.4% (39.0%) of all diagonals from monkey G (monkey P), the JPETC revealed significant increases of correlation during the movement period as compared to the hold period. Significant decreases were observed in 62.6% (80.9%, monkey P) of the diagonals. Both increases and decreases could occur in the same JPETC. In many cases, additional features showed up on diagonals off the main diagonal. For instance, it was relatively common to see negative dips around elevated correlations at zero delay (Fig. 6A, C and D, and Fig. 7B). In some cases, oscillations with at least one additional positive side peak arose around movement onset (as in Fig. 7B). Profound changes of correlation strength could be found in JPETCs taken from pairs situated in the same hemisphere (Fig. 6), as well as those situated in different hemispheres (Figs 7 and 8).

Figure 6 shows four JPETCs from a pair of LFPs recorded in the left hemisphere during different types of movement. The figure shows that the two sites were correlated during all four movement types (red bins along the main diagonals, indicating positive correlations). However, comparing the plots of the JPETC diagonals, it becomes clear that differences existed in modulation strength. In the bimanual movements (Fig. 6C and D), a clear movement-related modulation of correlation is visible, consisting of a pronounced increase peaking at movement onset and followed by a small decrease. In unimanual movements, this correlation change was much weaker (Fig. 6A) or virtually absent (Fig. 6B). In the JPETC colour display of C and D, negative side dips and an additional oscillatory side peak appear around movement onset, corresponding to a frequency of just below 10 Hz. In contrast, strong synchronized oscillations in the gamma frequency are present during the hold period which are also visible in the averaged correlogram. These oscillations can be seen in the inset, depicting the area surrounded by the rectangle within Fig. 6A at an expanded scale. Note that the time scales of the averaged correlogram and the JPETC differ, so that the satellite peaks appear at a wider spacing in the correlograms and are thus more clearly discernible than in the JPETC. These gamma oscillations became much weaker during the reaction time (target onset was on average 270 ms before movement onset) and had totally disappeared when movements were initiated. The different movement types were presented in a random order, but at a fixed interval after origin onset. Thus, whilst the monkeys were able to predict the time when a new movement would be cued, they could not predict which specific movement would be instructed next. Therefore, the oscillations before target onset could be related to some nonspecific process of movement preparation or readiness, but not to the preparation of a specific movement. Note that, whilst the diagonals are different, the correlograms look quite similar in all conditions. Both LFP signals were significantly tuned (r² > 0.6) to movements to the right, with a difference in preferred direction of only 17°. The PD of electrode 4 was 351°, and the PD of electrode 6 was 8°.

We display the mEP of the two signals in each pair along the x and y axes. The main diagonal of the JPETC (the diagonal row of bins from the bottom left to the top right) represents the time course of zero-delay correlation and is displayed separately in the upper right corner of each figure. The time-averaged correlogram is derived by averaging the diagonals at a range of –200 to +200 ms delay.

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Table 2. Frequency of significant movement-related correlation changes

<table>
<thead>
<tr>
<th></th>
<th>Within right hemisphere</th>
<th>Within left hemisphere</th>
<th>Between hemispheres</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Incidence (Diagonals)</td>
<td>Incidence (Diagonals)</td>
<td>Incidence (Diagonals)</td>
</tr>
<tr>
<td>Increases in correlation Monkey P</td>
<td>All trial types</td>
<td>45.4 (1420)</td>
<td>38.9 (1331)</td>
</tr>
<tr>
<td></td>
<td>Unimanual right movements</td>
<td>42.5 (518)</td>
<td>44.5 (483)</td>
</tr>
<tr>
<td></td>
<td>Unimanual left movements</td>
<td>47.4 (519)</td>
<td>38.0 (487)</td>
</tr>
<tr>
<td></td>
<td>Bimanual movements</td>
<td>46.5 (383)</td>
<td>32.7 (361)</td>
</tr>
<tr>
<td></td>
<td>Monkey G</td>
<td>All trial types</td>
<td>52.5 (1563)</td>
</tr>
<tr>
<td></td>
<td>Unimanual right movements</td>
<td>56.7 (522)</td>
<td>63.0 (414)</td>
</tr>
<tr>
<td></td>
<td>Unimanual left movements</td>
<td>58.2 (495)</td>
<td>56.0 (393)</td>
</tr>
<tr>
<td></td>
<td>Bimanual movements</td>
<td>34.8 (678)</td>
<td>61.2 (435)</td>
</tr>
<tr>
<td>Decreases in correlation Monkey P</td>
<td>All trial types</td>
<td>77.1 (1420)</td>
<td>85.6 (1331)</td>
</tr>
<tr>
<td></td>
<td>Unimanual right movements</td>
<td>79.3 (518)</td>
<td>86.8 (483)</td>
</tr>
<tr>
<td></td>
<td>Unimanual left movements</td>
<td>78.2 (519)</td>
<td>89.3 (487)</td>
</tr>
<tr>
<td></td>
<td>Bimanual movements</td>
<td>72.6 (383)</td>
<td>79.2 (361)</td>
</tr>
<tr>
<td></td>
<td>Monkey G</td>
<td>All trial types</td>
<td>66.7 (1563)</td>
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<td>Unimanual left movements</td>
<td>64.4 (495)</td>
<td>70.2 (393)</td>
</tr>
<tr>
<td></td>
<td>Bimanual movements</td>
<td>70.1 (678)</td>
<td>75.6 (435)</td>
</tr>
</tbody>
</table>

Frequency with which a significant change in correlation strength occurred in the main diagonal of the JPETC during the movement period as compared to the hold period. For each pair of sites, each movement type was considered separately. The number of diagonals from which the percentages were derived are shown in brackets.
Similar movement-related increases and decreases in correlation were also found interhemispherically (Fig. 7). In this example, there is no significant correlation change during unimanual movements (Fig. 7A), but a strong, brief correlation with an additional side peak arises around movement onset in bimanual movements (Fig. 7B). The time delay of this side peak is \( \approx 100 \) ms, corresponding to a frequency of \( \approx 10 \) Hz (i.e. in the alpha band). Similar to the example of Fig. 6, weak synchronized gamma oscillations were present during the hold period. Because of their relative weakness and the temporal resolution of the JPETC, however, they are barely visible in the colour display of the JPETC (lower left corner of the JPETC in Fig. 7B). Both the alpha and gamma components can be seen in the averaged correlogram. The diagonal runs first through a negative side dip, whereas it later straddles the main, positive peak. Therefore, the diagonal shows an initial decrease in correlation, followed by a later increase. The mEP on electrode 1 was not significantly tuned, but was most strongly activated during rightward movements of the contralateral arm. The mEP on electrode 4 was significantly tuned and had a preferred direction which was also to the right (351°).

Another example, this time demonstrating decorrelation, is shown in Fig. 8. This pair of sites was situated in different hemispheres of monkey P. The JPETC shows decorrelation both for unimanual (Fig. 8A) and bimanual movements (Fig. 8B). During the hold phase, negative side bands are present at relatively long delays (\( \approx 500-700 \) ms), which disappear during the movement and the decorrelation. Neither site was significantly cosine-tuned to the direction of movement.

In general, both the largest correlation values and the strongest correlation changes in our sample were seen along the main diagonal, i.e. at zero delay. Therefore, in the following analyses, we quantified the correlation changes over time along these diagonals. First, we analysed the frequency and size of correlation changes, and examined any differences among the different bimanual movement types. Then, we assessed the time course of correlation changes and the net correlation change observed over all recording sites.

**Comparing the percentages of increased and decreased correlation**

To assess how frequently one observes dynamic modulation of correlations, we counted the number of main JPETC diagonals that showed statistically significant changes of correlation strength (as compared to the hold period, for each pair and condition). The results are summarized in Table 2. The table illustrates a number of remarkable points. (i) Under all conditions, decreases of correlation strength were observed more frequently than increases. (ii) Interhemispheric pairs showed correlation increases and decreases in percentages that were similar to those shown in intrahemispheric pairs. This is in contrast to the averaged correlations, in which intrahemispheric correlations were stronger than interhemispheric ones. (iii) Intrahemispheric changes were observed during unimanual (both ipsilateral and contralateral) and bimanual arm movements. (iv) Interhemispheric pairs underwent dynamic correlation changes not only during bimanual, but also during unimanual movements.

**The JPETC dynamics during different movement types**

Table 2 does not show any systematic differences in correlation changes during different movement types. However, Table 2 only counts the incidence of correlation changes. In order to also compare the magnitude of correlation changes along different diagonals, we normalized the correlation scores as discussed in the Materials and Methods section. Normalized scores with absolute magnitude >3 SD from the mean (indicating significant increase in a given bin, as compared to the hold period, with an error probability of 0.002) were considered significant. Figure 9A compares the average normalized correlation strength over all significant bins of all JPETC-diagonals during unimanual and bimanual movements. For interhemispheric and left hemisphere pairs of LFP signals (contralateral to the preferred right hand), bimanual movements elicited significantly stronger correlation increases than unimanual movements (Wilcoxon
We found that 27% of the LFPs in monkey G and 19% of the LFPs in monkey P were significantly cosine-tuned (\( r^2 > 0.6 \), see Materials and Methods). For those pairs in which both sites showed significant tuning, we determined the angles between the preferred directions and compared the distributions of these angles between pairs showing significant increases, decreases or no change in correlation (Table 3). We found no consistent differences between the distributions for these three groups. There was no relationship between the difference in directional tuning and the correlation changes.

Within the same hemisphere, the medians of the angles between preferred directions were small (with medians ranging between 12 and 18°), indicating that the strongly tuned sites within a distance of 350–700\( \mu \)m lateral distance usually had similar preferred directions. However, the fact that we also found pairs with considerably different PDs clearly suggests that the observed similarity cannot be explained solely on the basis of passive current spread. Pairs recorded from different hemispheres had consistently larger differences between preferred directions, with medians between 32 and 55°.

**Relationships between preferred directions and the incidence of correlation changes**

For all electrodes, we determined whether the mEPs were cosine-tuned. We found that 27% of the LFPs in monkey G and 19% of the LFPs in monkey P were significantly cosine-tuned (\( r^2 > 0.6 \), see Materials and Methods). For those pairs in which both sites showed significant tuning, we determined the angles between the preferred directions and compared the distributions of these angles between pairs showing significant increases, decreases or no change in correlation (Table 3). We found no consistent differences between the distributions for these three groups. There was no relationship between the difference in directional tuning and the correlation changes.

The time course of correlation changes

We analysed the time course of significant correlation changes separately for increases and decreases. For each 2.5-ms time bin, we determined whether significant correlation changes occurred and used these results to construct two indicator vectors for each pair of LFP signals and each trial type. The vectors contained zeros at those time bins where correlation had not changed significantly and ones at bins in which a significant increase (for the first vector) or decrease (for the second vector) had occurred. Figure 10 shows the sums of these vectors over all pairs, which represents the number of significant changes occurring for each time bin in the whole database. The sum of the increased-correlation vector is plotted upwards and the sum of the decreased-correlation vector is plotted downwards. Onset of both increases and decreases is similar at \( \approx 200 \) ms before movement, corresponding to a time when the targets had already appeared on the screen. Increases in correlation were sharply peaked around movement onset. In contrast, decreases in correlation were more broadly distributed over the movement. Although in monkey G decorrelations also show a sharp initial peak shortly before movement onset, most of the decorrelations in both monkeys occurred after movements were initiated. In monkey P, both curves seem to be delayed in time as compared to those of monkey G. This may be related to the fact that the movement times of monkey P were much longer, so that the relevant time scale may have been expanded in this monkey. Repeating this procedure separately for inter- and intrahemispheric pairs and for different movement types did not reveal any systematic differences.

**TABLE 3. Differences between preferred directions**

<table>
<thead>
<tr>
<th></th>
<th>Pairs with significant increases in correlation</th>
<th>Pairs with significant decreases in correlation</th>
<th>Pairs with no change in correlation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Same</td>
<td>Different</td>
<td>Same</td>
</tr>
<tr>
<td>Monkey G</td>
<td>16.3°</td>
<td>43.3°</td>
<td>15.6°</td>
</tr>
<tr>
<td>Monkey P</td>
<td>17.5°</td>
<td>35.3°</td>
<td>12.0°</td>
</tr>
</tbody>
</table>

Medians of differences (in degrees) between the preferred directions of significantly cosine-tuned local field potentials (LFPs). Same, values of pairs within the same hemisphere; Different, values of pairs in different hemispheres. For each pair, all movement types have been considered separately.
The net correlation change during movements

Table 2 shows that decreased correlations occurred more often than increases. To test whether the decorrelations outweighed the increases, we calculated the grand average of correlations between all recording site pairs as shown in Fig. 11, either expressed as the correlation coefficient (Fig. 11A) or as the significance of the correlation coefficient (Fig. 11B). Indeed, the figure shows that, on average, the correlation decreased during the movement. In monkey G, this decrease was smaller and, in addition, there was a short epoch around movement onset during which the correlation was on the average increased. We found no differences between the grand average correlation changes in different movement types and in intra- vs. interhemispheric pairs.

Discussion

This study tested whether interactions between neuronal populations, especially between the hemispheres, might play a role in coordinated bimanual arm movements. In order to investigate this question, we used correlational approaches to detect functional interactions and examine how they change over time and how they are related to behaviour.

Time-averaged LFP correlations between the hemispheres were significantly smaller than those within the same hemisphere, suggesting that interhemispheric interactions are less intense than intrahemispheric ones. This is in agreement with EEG studies, which have reported only weak interhemispheric correlations (Schoppenhorst et al., 1980; Andrew & Pfurtscheller, 1997, 1999).

Although weak, many correlations recorded between the hemispheres were still significant, confirming earlier reports that interhemispheric correlations of single cells and LFPs do exist and are not negligible (Munk et al., 1995; Murthy & Fetz, 1996).

Time-averaged correlations did not differ between the hold period and the different movement types (Fig. 5). In order to address the possibility that this was a result of time averaging, we sought to reveal time-dependent modulations of interactions. To this end, we modified the JPSTH technique developed by Aertsen et al. (1989). The JPSTH detects dynamic modulation of interactions in relation to specific events (Eggermont, 1994; Sillito et al., 1994; Vaadia et al., 1995; Prut et al., 1998). We adapted the procedure for use with analogue data and called it the JPETC. It offers a temporal resolution in the ms range and is applied here for the first time on LFP data. Previous analyses of time-resolved correlation or coherence of analogue signals focused on sliding-window techniques (e.g. Bressler et al., 1993; Roelfsema et al., 1997; Destexhe et al., 1999), which suffer from a limited temporal resolution. In contrast to event-related coherence measures, the JPETC can also detect nonperiodic correlations.

In the methods section, we demonstrated that the JPETC is not contaminated by the sizes or shapes of the mEPs, or by overall changes in variability. It rather reveals correlations between the trial-by-trial fluctuations of the two LFP signals around the mEPs, but may also reflect covariations in signal timing. Both phenomena are interesting, because they reveal functional links between distant sites within the motor cortex or even between hemispheres. The context-dependence of these functional links demonstrates the dynamics of this system and its relationship to behaviour. Covariations in signal timing can be seen as evidence for a common timing linking distant neuronal sites.

The JPETC revealed movement-related correlations in a majority of pairs, suggesting that most neural interactions are flexible and are regulated in relation to movements. Although the average levels of correlation differed, intra- and interhemispheric pairs showed similar fractions and strengths of movement-related correlation changes. This indicates that interactions between the hemispheres are as strongly modulated as those within the same hemisphere.

Changes in correlation could consist of increases or decreases, and a given pair could show different patterns of correlation changes for different kinds of movements. This raises the possibility that the strength of correlation may carry some information about the type of movement to be executed. Variable correlations, changing with behavioural conditions or events, have also been reported in single neurons from precentral areas (Vaadia et al., 1995) and motor cortex (Murthy & Fetz, 1996; Riehle et al., 1997; Hatsopoulos et al., 1998; Grammont & Riehle, 1999; Laubach et al., 2000). One group specifically claimed that the information contained in correlations improve coding of movement direction (Maynard et al., 1999). Our findings show that, although the LFP signal is certainly less specific than single cell-activity, LFP correlations (in addition to mEP amplitudes) may convey behaviourally relevant information.

Concerning the question of whether interhemispheric interactions aid bimanual coordination, the most striking result of this study was that interhemispheric correlations (but not intrahemispheric ones) were consistently related to the degree of bimanual coupling. Symmetric bimanual movements were accompanied by significantly stronger correlation increases than asymmetric bimanual or unimanual movements. At the same time, the correlations between the movements of the two arms themselves were also highest for bimanual symmetric movements of the same amplitude. This suggests that interhemispheric correlations contribute to interlimb coupling and aid in production of bimanually symmetric movements. By the same token, interhemispheric coupling may underlie the
difficulties we have in producing asymmetric movements. The significantly weaker correlation increases that we found during asymmetric movements may be the result of an active process that reduces coupling, and the residual correlations may be a neural correlate of our inability to completely decouple our arms. This idea is also in line with the finding that split-brain patients (in which the callosal connections have been destroyed) are better than normal individuals in strongly asymmetric bimanual tasks (Eliassen et al., 1999). Thus, our findings are consistent with the view that interhemispheric correlations are the functional basis of crosstalk between the motor plans for the two hands, and regulation of the strength of this crosstalk may determine the level of behavioural coupling between the arms.

The time course of correlation changes lends further support to this notion. Because modulations of correlation began before movement onset, it is feasible that they are related to movement programming or preparation rather than execution. This would be consistent with the observation that interlimb crosstalk occurs even when a movement is not actually executed, such as when it is only imagined (Heuer et al., 1998b), or when a limb has been amputated (Franz & Ramachandran, 1998). Like the increased correlations described in this study, crosstalk between two simultaneously planned movements occurs during a transient phase associated with the process of movement preparation (Heuer et al., 1998a).

Finally, it is noteworthy that pairs within the same hemisphere also showed movement-related correlation increases during bimanual movements. This raises the possibility that intrahemispheric interactions may also participate in bimanual control. Although this may seem odd, there are other recent results that support this hypothesis. There is growing evidence of significant ipsilateral activation of the motor cortex (Tanji et al., 1988; Donchin et al., 1998; Kermadi et al., 1998; for review, see Chen et al., 1997). Furthermore, firing rates of MI cells are different during bimanual and unimanual reaching, supporting the suggestion that neurons in each hemisphere are related to the movement of both arms (Donchin et al., 1998). Finding inter- and intrahemispheric interactions during bimanual movements is further evidence of the fact that both hemispheres seem to be involved in movements of both arms.

Besides the increases in LFP correlations that have been discussed above, we also observed decreases in correlation. However, whilst increases in LFP correlations occurred mainly during movement planning and initiation (around movement onset), decreases occurred mainly during movement execution. As a result of this timing difference, the net correlation change after movement initiation was a decrease. Decreased correlations were found relatively uniformly in all movement types, and are an oft-reported phenomenon in population activity (Sanes & Donoghue, 1993; MacKay & Mendonca, 1995; Baker et al., 1997; Donoghue et al., 1998; Pfurtscheller & Lopes da Silva, 1999). The different time course of increased and decreased correlations could explain the behavioural finding that bimanual movements are most closely coupled at their initiation and are progressively desynchronized during movement execution (Boessenkool et al., 1999; Fowler et al., 1991). The overall picture would then be that increases in LFP correlations are involved in crosstalk during movement planning and initiation, and decreased correlation serves to decouple movements during their execution.

Previously, we had reported that firing rates and mEP amplitudes in primary motor cortex are bimanually related and thus may contribute to the coding of coordinated bimanual movements (Donchin et al., 1998; Donchin et al., 2001). Now, we show that dynamic interactions between neuronal populations are also involved in this function. We conclude that flexible interhemispheric correlations may be the neuronal substrate controlling the level of behavioural coupling between the arms.

Acknowledgements

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Abbreviations

IAI, inter-arm interval; JPETC, joint peri-event time correlogram; LFP, local field potential; mEP, mean motor evoked potential; PD, preferred direction.

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Single-Unit Activity Related to Bimanual Arm Movements in the Primary and Supplementary Motor Cortices

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Donchin, O. A. Gribova, O. Steinberg, A. R. Mitz, H. Bergman, and E. Vaadia. Single-unit activity related to bimanual arm movements in the primary and supplementary motor cortices. J Neurophysiol 88: 3498–3517, 2002; 10.1152/jn.00335.2001. Single units were recorded from the primary motor (MI) and supplementary motor (SMA) areas of Rhesus monkeys performing one-臂 (unimanual) and two-臂 (bimanual) proximal reaching tasks. During execution of the bimanual movements, the task related activity of about one-half the neurons in each area (MI: 129/232, SMA: 107/206) differed from the activity during similar displacements of one arm while the other was stationary. The bulk of this “bimanual-related” activity could not be explained by any linear combination of activities during unimanual reaching or by differences in kinematics or recorded EMG activity. The bimanual-related activity was relatively insensitive to trial-to-trial variations in muscular activity or arm kinematics. For example, trials where bimanual arm movements differed the most from their unimanual controls did not correspond to the ones where the largest bimanual neural effects were observed. Cortical localization established by using a mixture of surface landmarks, electromyographic recordings, microstimulation, and sensory testing suggests that the recorded neurons were not limited to areas specifically involved with postural muscles. By rejecting this range of alternative explanations, we conclude that neural activity in MI as well as SMA can reflect specialized cortical processing associated with bimanual movements.

Introduction

Simultaneous movements of two hands in space, bimanual movements,1 present a special control problem for the CNS. Controlling two dissimilar hand trajectories simultaneously can be significantly more difficult than moving along the same trajectories sequentially. For example, a bimanual task may split attention if each hand, during reaching movements of the arm, must approach a separate target. A bimanual task may also require a unique postural set to manage the interaction forces generated by simultaneous movements of the two limbs. The particular demands of bimanual tasks have led to the prediction that fundamental differences exist between bimanual and unimanual motor control (Kelso 1984; Tsutsui et al. 1998). It is reasonable to hypothesize that the primary motor cortex plays a role in controlling bimanual arm movements. If so, we should expect its neural activity to reflect the differences that characterize bimanual tasks.

It has been known for some time that the supplementary motor area (SMA) is especially involved in bimanual motor control. Combined lesions of the SMA and surrounding areas interfere with bimanual task performance (Brinkman 1984). A number of electrophysiological (Benecke et al. 1985; Deecke et al. 1987; Lang et al. 1990; Uhl et al. 1996), brain imaging (Sadato et al. 1997; Stephan et al. 1999; Toyokura et al. 1999), and clinical (Bell et al. 1994; Laplane et al. 1977; Penfield and Welch 1951; Viallet et al. 1992) studies have also explored the role of SMA in bimanual tasks. Yet, questions remain regarding how much SMA activity is specific to bimanual movements and whether SMA is the only cortical motor area with such neural specificity (Kazennikov et al. 1998; Wiesendanger and Wise 1992).

Tanji and his coworkers provided convincing evidence that, except for a tiny zone near the face area (Aizawa et al. 1990), primary motor area (MI) does not specifically encode bimanual finger movements, but SMA and the premotor cortex do (Tanji and Shima 1996; Tanji et al. 1988). This distinction between the two motor areas has not held up for tasks involving more proximal musculature, despite the expectation that this difference would be found (Donchin et al. 1998; Kazennikov et al. 1999; Kermadi et al. 1998; Tanji and Shima 1996; Tanji et al. 1988; Wiesendanger et al. 1996). Both Donchin et al. (1998) and Kermadi et al. (Kermadi et al. 1998) report equally strong bimanual effects in MI and SMA. We have speculated (Donchin et al. 1999) that the lack of bimanual-specific MI activity in the finger pressing task of Tanji reflects a special case, because MI has a well-established specialized role in the control of distal forelimb muscles (Andersen et al. 1975; Asanuma and Rosen 1972; Rouiller et al. 1994). If our speculation is correct, MI activity during most bimanual tasks reflects a higher level of processing than simply organizing muscle synergies (Phillips 1975). Recent studies also seem to point to a role for MI in other aspects of motor processing. MI neurons can encode multiple parameters of movement (Fu et al. 1995; Kahlion and Lisberger 1999; Moran and Schwartz 1999). Populations of MI neurons may reflect motor imagery.
(Georgopoulos et al. 1989; Porro et al. 1996) serial ordering (Carpenter et al. 1999), and perhaps stimulus-response associations (Zhang et al. 1997) and elements of transitive inference (Acuna et al. 2002).

If cortical activity differences between unimanual and bimanual arm movements reflect the different requirements involved in processing bimanual tasks, then the differences observed must be more elaborate than shifts in axial muscle activity or other simple variations in postural set. Tanji and his coworkers avoided this problem of changes in postural set by training monkeys to inhibit postural EMGs (TANJI et al. 1988). They reported that postural EMGs were not completely eliminated but were small and not time-locked to finger movements. Tanji et al. thus created a rather special motor control problem for the animal, one requiring months of training to inhibit “unwanted” motor outputs. Because it is impossible to achieve this level of control over proximal EMGs, the present study looks at whole arm movements and allows for natural postural adjustments by the subjects. The potential contribution of postural adjustment to bimanual effects is addressed with a more analytical approach.

While the majority of studies do report bimanual specific activity in frontal motor areas, one group did not find any substantial bimanual specificity in either MI or SMA (Kazenikov et al. 1999). These researchers have suggested that “subtle differences in the parameters of movement execution” explains the bimanual specific unit activity observed by others. This possibility has not been ruled out in any but the distal button pressing task of Tanji (TANJI and Shima 1996; Tanji et al. 1988). In the present paper, we rule out this possibility for a proximal task by analyzing the neural sensitivity to small differences in movements. Additionally, we rule out the possibility that differences in neural activity during bimanual arm movements are created by simple linear combinations of neural activities in unilateral movements.

METH O D S

Behavioral paradigm

Two female rhesus monkeys (Macaca mulatta) (monkey F, 4 kg, and monkey G, 3.5 kg) were trained to operate two separate manipulanda, one with each arm. Each manipulandum was a low-weight, low-friction, two-joint mechanical arm, oriented in the horizontal plane. Movement of each manipulandum produced movement of a corresponding cursor on a vertically oriented 21” video screen located 50 cm in front of the monkey. The movement of each cursor was mapped to its corresponding manipulandum movement such that each millimeter of manipulandum movement yielded one millimeter of movement of the cursor on the video display. The angular origin, 0°, was to the monkey’s right, and 90° was away from the monkey for the manipulandum movement and toward the top of the screen of the display.

The time course of typical unimanual and bimanual task trials is schematized in Fig. 1. A trial began when the monkey aligned both cursors on 0.8-cm-diam “origins” and held them still (as defined using velocity thresholds described in detail below) for 500 ms. The centers of the two origins were located 16 cm apart. For each arm, one of eight peripheral target circles (0.8 cm diam) could appear at a distance of 3 cm from the origin (Fig. 2). The small movement amplitude was chosen to minimize postural adjustments in accomplishment of the movements. Movements taking the cursor from the origin to the target were primarily elbow and shoulder movements, although the monkey was free to engage its wrists and fingers to accomplish the task. If only one target appeared—signaling a unimanual trial—the monkey moved the appropriate arm and brought the corresponding cursor into the target but did not move the other arm (again, according to the definition of movement initiation given below). Examples of the layout are shown in Fig. 2 (unimanual left and right). If two targets appeared—signaling a bimanual trial—the monkey moved both arms, such that the two cursors moved into the target circles on the screen. There were two classes of bimanual movements that were tested in the recording sessions: parallel and opposite (Fig. 2). Parallel bimanual movements were made to targets that were located in the same direction from their origins for each arm. For opposite bimanual movements, the direction from origin to target for one arm differed by 180° from the direction for the other arm. Every fourth successful trial was rewarded with liquid and followed by a 2-s pause to allow for fluid consumption.

The monkey’s reaction time was not restricted per se, but targets had to be acquired within 1.2 s. For bimanual trials, the animal was
Data acquisition

During training in monkey G, electromyographic signals (EMG) were recorded differentially using pairs of 1-cm surface electrodes from nine muscles; each muscle was recorded bilaterally. These muscles were the rhomboid, latissimus dorsi, teres major, pectoralis major, deltoid, biceps brachii, triceps brachii, flexor carpi ulnaris, and extensor carpi ulnaris. Up to four muscles were recorded simultaneously. The EMG was amplified, filtered (140 Hz–4 kHz), and its root mean square (RMS) was computed with a frequency cutoff of 100 Hz (the RMS is a nonlinear filter that first rectifies and squares the signal and then smooths this squared signal to the cutoff frequency before it’s square root is taken). EMG and manipulandum position were sampled by data acquisition boards (DAP-3200e, Microstar Laboratories, Bellevue, WA) at 400 Hz and stored for off-line analysis. Both signals were smoothed off-line with a low-pass 4 pole Butterworth filter with a corner frequency of 10 Hz, using a zero-phase smoothing algorithm. EMGs were recorded in monkey F after the end of recording in that monkey. Unfortunately, recording ended in monkey F as a result of cortical insult that caused transient hemiparesis and EMGs were recorded only after the monkey recovered. While the EMG results for the two monkeys were similar, we could not exclude the possibility that EMGs in Monkey F do not accurately reflect the muscular activity during recording of neuronal activity. Thus only the results for monkey G are presented.

We used MRI (Biospec Bruker 4.7 Tesla animal system; fast-spin echo sequence; effective echo time [TE] = 80 ms and repetition time [TR] = 2.5 s, 13 coronal slices 2 mm wide) to help locate the stereotaxic coordinates of the central and arcuate sulci. With the MRI pictures as a guide, two recording chambers (27 × 27 mm) were surgically implanted above the right and left hemispheres, and a head holder was attached to the occipital bone. The surgery was performed under isoflurane anesthesia in aseptic conditions. The animals’ care and surgery procedures were in accordance with The National Institutes of Health Guide for the Care and Use of Laboratory Animals and all applicable Hebrew University regulations.

During recording sessions, the monkeys were seated in a primate chair placed in a dark room and the head was fixed. Single-unit activity was recorded by eight individually driven glass-coated tungsten microelectrodes (impedance 0.2–0.8 MΩ at 1 kHz) in the two hemispheres (4 electrodes in each hemisphere). Electrodes were introduced into the SMA at an angle of 30° to the sagittal plane. Neurons were selected for recording on the basis of the isolation quality of their spike waveforms and stability of their firing rates. Units with very low firing rates were not recorded, but no effort was made to select units for their “task-related” behavior. The electrode signals were amplified, filtered, and sorted (MCP and MSD, Alpha-Omega, Nazareth, Israel). The MSD performs spike sorting based on an eight-point template-matching algorithm that allows two isolated neurons to be recorded from most electrodes (from some electrodes it is possible to record 3 isolated neurons and occasionally only 1 neuron can be isolated). In addition, the MSD indicates every time that the signal crosses a user-determined threshold but does not match any of the templates currently being isolated. Spike arrival times, threshold crossings, and timing of behavioral events were recorded with a resolution of 24 kHz, but were down-sampled off-line to a resolution of 400 Hz. The waveforms of all detected spikes and all the waveforms surrounding all unclassified threshold crossings were also sampled at 24 kHz allowing off-line confirmation of spike sorting.

During selected neural recording sessions for monkey G, EMG was collected from two muscles bilaterally: right and left flexor carpi
ulnaris and right and left deltoid. We chose to record those muscles that seemed to us most different in unimanual and bimanual movements on the basis of the EMG results during training.

Following surgery in each animal, a number of penetration sessions were devoted to mapping the cortical area that had been exposed. During these penetrations, unit receptive fields were tested with passive manipulation of each individual limb separately and of the tail. While two researchers worked directly with the monkey to isolate the movement of individual joints, a third researcher evaluated neuronal response using the amplified signal from each electrode in turn passed directly into a loudspeaker. In addition to manipulation of the limb, we tested the neuronal response to superficial and deep somatic stimulation on the arms, legs, back, trunk, tail, stomach, face, and neck. Cases where somatic stimulation produced neuronal activity were noted. We also tested for visual and oculomotor responses by moving interesting stimuli within the monkey’s field of view. Finally, we applied intracortical microstimulation (ICMS) with trains of 200-µs cathodal pulses at 300 Hz with an intensity of 10–80 µA (BPG-2 and BSI-2, BAK Electronics, Germantown, MD). Typical train durations were 50 ms for MI and 100 ms for SMA. Passive manipulation was tested at these sites just before stimulation. Movements were assessed following ICMS by two researchers in the recording room. They ascertained that the monkey was relaxed and completely still before stimulation. When ICMS evoked movements, current was reduced until it was possible to ascertain the smallest activation of a joint (or joints) possible at that site and the type of movement evoked. We documented the evoked movements and the stimulation intensity. Movements were classified as lower limb or tail movements if they caused movement of the lower limb or tail. Movements were classified as proximal movements if they involved movement of the spine, contraction of musculature in the back, or translation of the shoulder. Movements were classified as upper limb proximal movements if they involved movement of the elbow or a rotation of the shoulder joint. Movements were classified as upper limb distal movements if they involved movement of the wrist or the fingers. In addition to these initial mapping sessions, additional corroborations of recording locations were acquired at the end of most recording sessions. After recording for the day was completed, passive manipulation and ICMS were tested at the recording site following the procedures outlined above. Little effort was made in these instances to optimize stimulation depth or to test the precise threshold of activation.

**Histology**

Monkeys were given an overdose of pentobarbital, and then perfused transcardially with 0.9% saline followed by 4% formaldehyde in 0.1 M phosphate buffer. After fixation, in one monkey, pins were inserted in defined locations to allow reconstruction of chamber coordinates. The brains were photographed. Blocks of tissue were sectioned coronally in a freeze-dry microtome (section width = 50 µm). Alternate sections were stained with cresyl violet (0.1%). Surface penetration maps for both monkeys are shown in Fig. 3. Note that SMA penetrations are marked at the point of electrode insertion. Penetrations into the SMA were angled at 30° to the sagittal plane and advanced from the point of insertion until they reached the medial cortex. Generally, this meant that the electrodes were advanced through cortex into white matter and through that white matter before reaching SMA. The pattern: units through cortex into white matter and through that white matter before cortex. Generally, this meant that the electrodes were advanced from the point of insertion until they reached the medial cortex. Penetrations into the SMA were angled at 30° to the sagittal plane and advanced from the point of insertion until they reached the medial cortex. Generally, this meant that the electrodes were advanced through cortex into white matter and through that white matter before reaching SMA. The pattern: units—white matter—units was used as an indicator of recording location. Additionally, more lateral penetrations to the SMA involved crossing a greater extent of white matter and through that white matter before cortex. Generally, this meant that the electrodes were advanced through cortex into white matter and through that white matter before reaching SMA. The pattern: units through cortex into white matter and through that white matter before cortex. Generally, this meant that the electrodes were advanced from the point of insertion until they reached the medial cortex. Penetrations into the SMA were angled at 30° to the sagittal plane and advanced from the point of insertion until they reached the medial cortex.

**Data analysis**

All recorded units were assessed for stability of firing rate and responses before further analysis was performed. Units were selected for analysis if the stable period included ≥6 trials for each type of movement. No selection was made on the basis of responsiveness or task-related activity. (However, Table 2 shows that most recorded units—81% in MI and 76% in SMA—showed task-related activation.) Standard raster displays and peri-stimulus time histograms (PSTH) were computed and examined. PSTHs were constructed with a binwidth of 2.5 ms and smoothed for display purposes with a digital low-pass 4-pole zero-phase Butterworth filter with a cutoff of 100 Hz. All PSTHs were aligned on movement onset, which was determined by an off-line algorithm (A. Arieli, unpublished data) and then confirmed manually. For purposes of alignment, the beginning of movement in bimanual trials was determined by the first arm to begin moving; for reaction times, the beginning of movement for each arm was calculated separately. End of movement was determined with the same algorithm used for determining movement onset. End time was determined separately for the right and left arms, and movement times for each arm were generated independently.

The onset of neural activity changes was determined for each PSTH using the CUSUM algorithm (Davey et al. 1986; Ellaway 1977). Onsets were limited to the time from target appearance to 400 ms after movement initiation. The trial-by-trial firing rate of the cell was averaged from activation onset until 500 ms after activation onset (termed the activation epoch). The firing rate during this epoch is termed the evoked activity. This was compared with a baseline firing rate taken from 350 ms before activation onset to 100 ms before activation onset (the baseline epoch). While this method, in principle, includes part of the reaction time, the algorithm guarantees that the neural activity is unchanged prior to response onset and therefore our results are insensitive to the precise timing of the baseline epoch. Generally, this was a period during which the monkey’s arms were motionless at the origin position, and we averaged activity in this period for each neuron across the different types of movement. In cases with no response onset, as might occur for example in nonpreferred movement directions, we arbitrarily selected a default 500-ms period from 100 ms before movement initiation (the average activation onset across responsive units) to 400 ms after movement initiation.

To allow data from cells recorded during two-direction sessions to be combined with data from cells recorded during eight-direction sessions, we limited our current analysis of the eight-direction sessions to two directions. Combining the two-direction sessions data with eight-direction sessions is possible because each two-direction session includes a subset of the trial types performed in the eight-direction sessions. This was done using the following procedure. For eight-direction sessions, we used the firing rate in the activation epoch above to determine the primary direction to use for each cell. For each of the movement types—unimanual left, unimanual right, bimanual parallel, and bimanual opposite—we calculated the mean directional activity for the cell (Mardia 1972) and then combined these means to arrive at a single direction for each cell. This was taken to be the cell’s primary direction, and its secondary direction was simply the primary
direction plus 180°. Although there may have been some difference in the power of the tests applied to the cells that showed significant responses in two-direction and eight-direction sessions, no such statistical difference was observed in the actual data. Similarly, the strength of the bimanual-related effect in two-direction and eight-directions sessions was comparable (note, however, that the statistical significance of the results was affected by differences in the number of trials per movement type, as discussed in the following text). In general, statistical tests of bimanual-related activity were performed on different types of trials using data from a single cell, so data from eight-direction sessions and two-direction sessions were only combined into a single statistical test when the population distributions are being tested.

Lateral preference

The Mann-Whitney rank statistic—calculated on the trial-by-trial firing rate during the activation epoch and the baseline epoch—was used to evaluate statistical significance in all comparisons of neuronal activity. The statistical significance of the cells activation was evaluated by comparing the baseline epoch to the activation epoch; neurons were considered significantly activated if there was a statistically significant difference between baseline and evoked activity in at least one trial type. Contralateral preference of the neurons was determined by comparing the maximal evoked activity during unimanual contralateral movements to the maximal evoked activity during unimanual ipsilateral movements. The strength of the arm preference, termed the laterality index, was normalized by the summed evoked activity (EA)

\[
\text{Laterality Index} = \frac{\text{contralateral EA} - \text{ipsilateral EA}}{\text{contralateral EA} + \text{ipsilateral EA}}
\]

This index will be 1 for a neuron that responds only contralaterally, -1 for a neuron that responds only ipsilaterally, and 0 for a neuron with exactly the same response in ipsilateral and contralateral movements.

**FIG. 3.** Penetration maps. Penetration maps for both hemispheres of 2 monkeys. Inset: brain photograph indicating the location of the penetration map. As described in METHODS, proximal refers to shoulder-and elbow-related sites; distal refers to forearm-, wrist-, and finger-related sites. Sites in which microstimulation produced movements are marked with a colored horizontal bar. Sites in which touch or passive manipulation of the monkey’s limbs produced clear neural activity are marked with a colored vertical bar. Sites where we found both microstimulation and passive manipulation effects are marked with a plus formed by the overlap of horizontal and vertical bars. Sites from which units were recorded during the task are marked with a gray dot. Each dot represents a 4-electrode penetration in either primary motor (MI) or supplementary motor (SMA) areas. On some penetrations, fewer than 4 electrodes recorded isolated neurons. Penetrations are marked at the coordinates of insertion into the cortex. SMA penetrations were inserted into dorso-lateral cortex angled at 30° to the sagittal plane and advanced until they reached the medial cortex.
Bimanual-related activity

To compare evoked activity during bimanual movements to evoked activity during unimanual movements, it is necessary to choose which unimanual activity the bimanual activity will be compared with. Clearly, the bimanual activity should be compared with activity during one of the unimanual movements that compose it (although it could also be compared with some sort of combination of the activities during the 2 unimanual movements that compose it; this issue is addressed below). The question is, which of the two unimanual movements represents the appropriate comparison. One possibility is to always compare activity during bimanual movements to activity during a unimanual contralateral movement. However, this choice ignores the relatively large proportion of neurons with an ipsilateral preference in unimanual movements. We chose to compare the neural activity during bimanual movements to the neural activity in the unimanual movement that evoked a stronger response. In this way, we end up asking whether there is a difference between maximal activation in bimanual movements and maximal activation in unimanual movements. For example, in Fig. 7, the bimanual evoked activity in B would be compared with the ipsilateral evoked activity.

However, since there are four different bimanual movements performed by the monkey—two bimanual parallel movements and two bimanual opposite movements—this still leaves us with four different comparisons. These correspond to the four rows in each of our figures illustrating the activity of a neuron (Figs. 8 and 9). At this point we applied the logic that any difference between unimanual activation and bimanual activation represented an interesting effect from our point of view. Therefore we focused our attention on the comparison where the difference between unimanual and bimanual was largest.

Translating the logic of the preceding paragraphs into mathematical language, we performed four Mann-Whitney tests comparing the bimanual evoked activity to the unimanual evoked activity in each type of bimanual movement. The significance of the bimanual-related effect was taken to be the maximum significance over the four tests, and the criterion for significance (threshold at which \( P \) was deemed to be significant) was divided by 4 to correct for the compounded tests (a technique called the Bonferroni procedure).

The strength of the bimanual-related effect was quantified using a measure analogous to the laterality index

\[
\text{Bimanual-Related Effect} = \frac{\text{bimanual EA} - \text{unimanual EA}}{\text{bimanual EA} + \text{unimanual EA}} \quad (2)
\]

where bimanual EA is the evoked activity during the bimanual movement, and unimanual EA is the evoked activity during the unimanual movement to which it is being compared. The bimanual evoked activity was compared with the same unimanual evoked activity used in assessing statistical significance. Many other normalizations for the strength of the bimanual-related effect are possible. We examined several other measures of the effect (including subjective ranking by members of the laboratory) without uncovering any instability in the results. Note that the bimanual-related effect is not influenced by the baseline firing rate; it represents a direct comparison of the firing rates in the activation epochs of unimanual and bimanual movements.

Linear summation

One possible explanation for the existence of statistically significant bimanual-related effects is that evoked activity during bimanual movements may be a sum of the evoked activity during unimanual movements. While it is possible that absolute firing rates sum linearly, we thought that it was more likely that the evoked activity (the change from the baseline firing rate) in unimanual movements would be summed to give the evoked activity in bimanual movements. Therefore we normalized the evoked activity by subtracting the baseline activity [we call this the normalized evoked activity (NEA)].

First, we tested if NEA during bimanual movements is explained by a simple linear summation of the unimanual movements that compose it. Here again, we require that the linear summation hold true for all four bimanual movements. Therefore the deviations from linearity in each type of bimanual movement were combined to produce a statistic that should distribute like \( \chi^2 \) with 3 degrees of freedom (specifically, we calculated the sum of the squared differences between bimanual NEA and the sum of the unimanual NEAs divided by the combined variance of the bimanual and unimanual NEAs). We also tested for the possibility that NEA in bimanual movements is equal to NEA during contralateral movements and for the third possibility that it is equal to NEA during ipsilateral movements. If we could reject all three of these null hypotheses at \( P < 0.05 \), we determined that the bimanual activity of this neuron was not explained with the hypothesis of linear summation. Note that our failure to correct for the multiple statistical tests effectively increases the significance level since we are requiring that all three null hypotheses be rejected rather than requiring that only one of the three null hypotheses be rejected.

A more general possibility is that NEA during bimanual movements is some nontrivial linear combination of unimanual NEAs. To test this possibility we used a linear model of the form

\[
B_{pi} = \alpha C_{1i} + \beta I_{1i} \\
B_{p2} = \alpha C_{2i} + \beta I_{2i} \\
B_{u1} = \alpha C_{1i} + \beta I_{1i} \\
B_{u2} = \alpha C_{2i} + \beta I_{2i} \quad (3)
\]

where the \( B_s \) represent NEA during bimanual movements, the \( C_s \) represent NEA during unimanual movements, and the \( I_s \) represent NEA during ipsilateral movements. We used a constrained linear fit to generate \( \alpha \) and \( \beta \), and we restricted \( \alpha \) and \( \beta \) to positive values (Matlab 5.3, Mathworks, lsqlin function). Goodness of fit was assessed using an \( F \) test.

Analysis of behavioral controls

We tested movement trajectories, velocity profiles, and the EMG for differences between bimanual and unimanual movements. To simplify quantitative analysis of these behavioral variables, we parameterized each variable with a single number for each movement. For the movement trajectories, we calculated the average deviation (from 50 to 450 ms after movement initiation, the movement epoch) of each movement from the grand mean of all movements in that direction. We call this the trajectory deviation. For the velocity, we calculated the peak velocity of each movement during the movement epoch, and call it the peak velocity. For the EMG, we calculated the integral of the RMS of individual EMG traces recorded during the movement epoch (150 ms before movement initiation to 350 ms after movement initiation). This we called the integrated EMG.

Three different movements could involve a left arm movement to 45°. The left arm could move to 45° in a unimanual movement; it could move to 45° as part of a bimanual parallel movement in which the right arm also moved to 45°; and, it could move to 45° as part of a bimanual opposite movement in which the right arm moved to 225°. We examined plots of all three behavioral variables that allowed comparison of these three different movements. In addition, we applied an analysis similar to the one applied to the neural data, using the same measure of bimanual-related effect (Eq. 2). For the trajectories and velocity profiles, we also correlated the strength of this effect to the strength of the bimanual-related effect in the neurons, comparing the neuronal bimanual-related effect to a behavioral bimanual-related effect calculated on the same trials exactly.

Since much of the EMG was not recorded simultaneously with neuronal activity, it was not possible to correlate the bimanual-related effect of the integrated EMG with the neural effect as we did with the trajectory deviations and peak velocities. Instead, we analyzed the integrated EMG separately for each muscle and for each of the four primary directions (0°, 45°, 90°, and 135°). Like with the neuronal...
We used the formula for bimanual and unimanual evoked activity. To quantify this comparison, we compared the differences in activity in trials where the behavior is different. The index should be close to 0 if separation has no effect, meaning that the bimanual effect is not well explained by variations in the movement parameter. Using bootstrap techniques, we estimated the distribution of the index under the null hypothesis of no effect and used this estimate to generate stringent (P = 0.001) and permissive (P = 0.15) confidence limits for the index around 0. We considered the value to be significantly different from 0 if it lay outside the stringent confidence limits (P < 0.001) and relatively close to 0 if it lay within the permissive limits (P > 0.15). We also compared the distribution of separation indexes to a distribution of randomly generated separation indexes and tested the fit using the Kolmogorov-Smirnov test.

**RESULTS**

**Task performance**

The analysis of the movement initiation and offset in all recording sessions (Table 1) showed that in bimanual trials, the arms typically started to move together with an average inter-arm interval (IAI) of 40 ms and reached the targets with comparable accuracy. On average, the right arm began movement before the left and finished movement after the left in both monkeys, a trend that was significant in some cases (see Table 1). Successful performance of the trial could be achieved with an IAI of ≤300 ms, and the actual performance of the monkey varied from case to case.

**TABLE 1.  Movement times, reaction times, and interarm intervals**

<table>
<thead>
<tr>
<th></th>
<th>Monkey F</th>
<th></th>
<th>Monkey G</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Unimanual</td>
<td>Parallel</td>
<td>Opposite</td>
<td>Unimanual</td>
</tr>
<tr>
<td>RT Left</td>
<td>289 ± 36.4</td>
<td>280 ± 42.8</td>
<td>268 ± 29.9</td>
<td>277 ± 29.7</td>
</tr>
<tr>
<td>Right</td>
<td>238 ± 14.4</td>
<td>247 ± 18.6</td>
<td>248 ± 13.4</td>
<td>233 ± 16.6</td>
</tr>
<tr>
<td>MT Left</td>
<td>576 ± 72.1</td>
<td>550 ± 79.1</td>
<td>546 ± 73.4</td>
<td>622 ± 48.3</td>
</tr>
<tr>
<td>Right</td>
<td>647 ± 54.8</td>
<td>631 ± 49.7</td>
<td>597 ± 53.6</td>
<td>596 ± 58.7</td>
</tr>
<tr>
<td>IAI Start</td>
<td>38 ± 46.0</td>
<td>25 ± 21.5</td>
<td></td>
<td>29 ± 28.3</td>
</tr>
<tr>
<td>IAI End</td>
<td>−42 ± 70.7</td>
<td>−27 ± 48.3</td>
<td></td>
<td>−14 ± 40.8</td>
</tr>
</tbody>
</table>

Means ± SD over all recording sessions of the reaction time (RT), movement time (MT), and inter-arm interval (IAI) for both monkeys (all data in milliseconds). Reaction time and movement time are calculated separately for the left and right arm in bimanual trials. Interarm interval is calculated separately for start and end of movement and is positive when the right hand leads the left hand. Numbers in bold for the RT and MT of bimanual movements indicate values significantly different from the unimanual values (Mann-Whitney U, P < 0.01). Numbers in bold for IAI s indicate values significantly different from zero (sign test, P < 0.01).

**TABLE 2.  Activation of cells in MI and SMA**

<table>
<thead>
<tr>
<th>Area</th>
<th>Monkey</th>
<th>Total Number of Neurons</th>
<th>During any movement</th>
<th>During unimanual movements (but not bi)</th>
<th>During both bimanual and unimanual</th>
<th>During bimanual movements (but not uni)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MI</td>
<td>F</td>
<td>100</td>
<td>90 (90)</td>
<td>8 (8)</td>
<td>73 (73)</td>
<td>9 (9)</td>
</tr>
<tr>
<td></td>
<td>G</td>
<td>132</td>
<td>97 (73)</td>
<td>13 (10)</td>
<td>55 (42)</td>
<td>29 (22)</td>
</tr>
<tr>
<td></td>
<td>Both</td>
<td>232</td>
<td>187 (81)</td>
<td>21 (9)</td>
<td>128 (55)</td>
<td>38 (16)</td>
</tr>
<tr>
<td>SMA</td>
<td>F</td>
<td>83</td>
<td>75 (90)</td>
<td>12 (14)</td>
<td>58 (70)</td>
<td>5 (6)</td>
</tr>
<tr>
<td></td>
<td>G</td>
<td>123</td>
<td>82 (67)</td>
<td>19 (15)</td>
<td>46 (37)</td>
<td>17 (14)</td>
</tr>
<tr>
<td></td>
<td>Both</td>
<td>206</td>
<td>157 (76)</td>
<td>31 (15)</td>
<td>104 (50)</td>
<td>22 (11)</td>
</tr>
</tbody>
</table>

Numbers in parentheses represent percent of neurons with significant differences between baseline activity and activity during the activation epoch and comparison of the number of neurons activated in any of the different movements with the number of neurons activated in at least one unimanual movement (and also, possibly, in bimanual movements). Significance of activation was determined with the Mann-Whitney test (P < 0.001).
monkey was more simultaneous than required. The IAIs are also much shorter than the reaction time and movement time. The average reaction time was approximately 250 ms and the average movement time was approximately 600 ms.

We performed Mann-Whitney tests to compare reaction time and movement time in unimanual and bimanual movements. For monkey F, only the comparison of bimanual opposite movement times to those unimanual movement times showed a difference that was marginally significant ($P < 0.01$; note, however, that no correction has been made here for multiple comparisons). However, in monkey G, we found that unimanual movement times were usually slower than bimanual movement times ($P < 0.01$, except for bimanual parallel movements).

**Neuronal population**

In each session we made two simultaneous penetrations of four electrodes in each hemisphere, and recorded the activity of 8–16 isolated neurons. The proximal arm areas of SMA and MI were identified based on neuronal activity (during task performance, during somatosensory stimulation, and during passive limb movements), the effect of ICMS and the anatomy of the sulci and gyri determined by MRI and postmortem. A total of 665 neurons were recorded from the two monkeys during 82 penetrations (328 electrode tracks) in SMA and MI (see Fig. 3). Of these, 572 passed our criterion for waveform isolation, and of these, there were 438 for which isolation was maintained for $\geq6$ trials in each movement condition. Thus our analysis was performed on 438 cells, 232 from MI, and 206 from SMA. For all of our analyses, we tested the results on the right and left hemispheres of the monkeys separately. However, since no significant differences were found, data from both hemispheres of each monkey are presented together throughout the paper. Average activation onset for all units was $-119 \pm 183$ (SD) ms, and no significant difference in activation time was found between MI and SMA units.

Table 2 shows the number of units whose activity varied significantly during performance of the task. As can be seen, activity of 81% (187/232) of neurons recorded in MI and 76% (157/206) of neurons recorded in SMA were significantly modulated during performance of the task, despite the fact that no selection was made on this basis during the recording sessions. Table 2 also demonstrates that about one-half of the neurons in both MI and SMA were significantly activated during both unimanual and bimanual movements. The number of units active only during unimanual movements is approximately equal to the number of units active only during bimanual movements. A $\chi^2$ analysis applied to the combined data of the two monkeys shows a marginally significant difference between the areas ($P < 0.047$). The interpretation of this result is problematic, since the nominal significance level we have used for behavioral data are $P < 0.01$. Thus we are unable to conclude that the two areas have the same distribution of activation, but we are also unable to reject the hypothesis.

**Neural activity during unimanual movements**

Figure 4 shows the activity of two neurons recorded from left MI of monkey F during unimanual movements of both the right and the left arm. The neuron in Fig. 4A is strongly modulated during right-handed (contralateral) movements (laterality index of 0.59, Eq. 1), while the neuron B is strongly modulated during ipsilateral movements (laterality index of $-0.77$). Table 2 compares the number of neurons with significantly evoked activity during unimanual movements to the number with such activity in both unimanual and bimanual movements, while Table 3 compares the number of significantly activated neurons during unimanual contralateral movements with the number during unimanual ipsilateral movements. The latter table shows a mild contralateral preference in...
Neuronal activation during bimanual movements does not affect categorization on this table. Significant exclusive categories: contralaterally but not ipsilaterally activated, activated both contralaterally and ipsilaterally, or ipsilaterally but not contralaterally activated.

The table breaks down the neuronal activation during unimanual movements according to the side being activated creating three mutually exclusive categories: contralaterally but not ipsilaterally activated, activated both contralaterally and ipsilaterally, or ipsilaterally but not contralaterally activated. Neuronal activation during bimanual movements does not affect categorization on this table. Significance of activation was determined with the Mann-Whitney test ($P < 0.001$).

Both MI and SMA. In both recording areas, approximately one-third of the neurons are activated only during contralateral movements while approximately one-fifth of the neurons are activated only ipsilaterally. A chi-squared analysis of the data in this table (combined across the 2 monkeys) revealed no significant differences between MI and SMA. These findings are strengthened by Fig. 5, which shows the distribution of the lateralization index in MI and SMA. The figure shows that a large proportion of the cells have no significant difference in maximal activation during contralateral and ipsilateral movements, and that many neurons in both MI and SMA are more strongly activated during ipsilateral movements. Nevertheless, there is a slight contralateral preference, and a tendency for neurons in MI to be more contralateral than neurons in SMA.

The number of neurons in monkey F with significant lateralization of activity is larger than in monkey G. This is because monkey F performed more trials in each type of movement than monkey G, improving the power of the statistical tests performed. In monkey F, most sessions were two-direction sessions, while in monkey G most sessions were eight-direction sessions. This led to a difference in the number of trials performed in each direction.

Neural activity during bimanual arm movements

The comparisons of the cells’ activity in unimanual, bimanual parallel, and bimanual opposite trials revealed significant bimanual-related effects that are demonstrated in Figs. 6 and 7. Figure 6 shows activity of a left MI neuron during unimanual and bimanual movements. While there is slight modulation of activity during movements of the right (contralateral) arm, the neuron is strikingly active during one specific type of bimanual movement (bimanual parallel movements in which both arms move to 180°, i.e., to the left). The strength of the bimanual-related effect (Eq. 2) in this neuron is 0.63. Figure 7 shows a cell from the right SMA that shows evoked activity only in unimanual movements of the contralateral arm. This activity would normally be described as “classic motor-related” activity. Nevertheless, the cell has a strong bimanual-related effect. The clear, directionally selective, activity evoked during unimanual movements of the left (contralateral) arm disappears during all bimanual movements, and is replaced by a reduction in the firing rate of the neuron. The strength of the bimanual-related effect in this neuron is −0.84. Dramatic examples of the bimanual-related effects, in MI as well as SMA, can also be found in Fig. 14 of this manuscript and in Donchin et al. 1998.

Muscular activity in unimanual and bimanual arm movements

The monkey performed short movements (3 cm) that did not require noticeable postural adjustment. Indeed, observation of the monkey during task performance (aided by video recordings) revealed no postural adjustments or other differences that distinguished movements during bimanual and unimanual trials, and examination of the EMG of the axial muscles (rhomboids and latissimus dorsi) showed very little activity during performance of the task (Fig. 8). The figure allows a compar-

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### Table 3. Lateralized activation of cells in MI and SMA

<table>
<thead>
<tr>
<th>Area</th>
<th>Monkey</th>
<th>Cells With Significant Activation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>During any unimanual movement</td>
</tr>
<tr>
<td>MI</td>
<td>F</td>
<td>81</td>
</tr>
<tr>
<td></td>
<td>G</td>
<td>68</td>
</tr>
<tr>
<td></td>
<td>Both</td>
<td>149</td>
</tr>
<tr>
<td>SMA</td>
<td>F</td>
<td>70</td>
</tr>
<tr>
<td></td>
<td>G</td>
<td>65</td>
</tr>
<tr>
<td></td>
<td>Both</td>
<td>135</td>
</tr>
</tbody>
</table>

Numbers in parentheses represent percent of neurons with significant differences between baseline activity and activity during the activation epoch in unimanual trials. The table breaks down the neuronal activation during unimanual movements according to the side being activated creating three mutually exclusive categories: contralaterally but not ipsilaterally activated, activated both contralaterally and ipsilaterally, or ipsilaterally but not contralaterally activated. Neuronal activation during bimanual movements does not affect categorization on this table. Significance of activation was determined with the Mann-Whitney test ($P < 0.001$).

![Graph](https://via.placeholder.com/150)

**Fig. 5.** Contralateral preference in MI and SMA. The 2 histograms show that only a small difference exists in the laterality index (Eq. 1) of neurons in MI and SMA, and that in both MI and SMA there are many cells that are more strongly activated during unimanual ipsilateral movements than during unimanual contralateral movements. For neurons below the dotted line, there is a significant difference between the maximal activation during contralateral and ipsilateral movements ($P < 0.001$). Binwidth = 0.1.

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ison of the activity of all the different muscles from which data were collected in one particular combination of unimanual and bimanual movements. The rightmost two columns show EMG activity of nine muscles, recorded bilaterally from the left and right sides of the body, during a unimanual right movement to 45°. The middle two columns show activity of the same muscles during a unimanual left movement to the same direction and the leftmost two columns show the activity of those muscles when both hands are moving together in parallel to 45°. It is clear from this figure that there is very little contralateral EMG activation during unimanual movements. Similarly, it can be seen that the overall picture of EMG activation in the bimanual movement is similar (although not identical) to that in the two unimanual movements. Analysis of the arm endpoint trajectories and velocity profiles also indicate similarity between bimanual and unimanual movements (see Extending earlier results as well as Figs. 10–12).

Extending earlier results

To this point, the results described mirror those of our earlier report: both MI and SMA have significant proportions of neurons with ipsilateral and contralateral preference and both areas have neurons with dramatic bimanual-related activity (Donchin et al. 1998). Now we extend these findings by more carefully quantifying the bimanual related activity and examining the hypothesis that subtle differences in the EMG of axial and arm muscles or changes in trajectories and velocity profiles suffice to explain the bimanual-related effect.

Distribution of bimanual related cells in MI and SMA

The percentage of cells that exhibited significant bimanual-related effects was high in both MI and SMA: 55% (129/232) in MI and 52% (107/206) in SMA. Figure 9 shows the strength of the bimanual-related effect in the population of analyzed cells. The histograms are separated into two by a dotted line that distinguishes cells found to be significantly “bimanually related” (below the line) from others. From the histograms, one can see that evoked activity is stronger in unimanual movements (as in Fig. 7) at least as often as it is increased during bimanual movements (as in Fig. 6). The figure also shows that the distribution of strengths is similar.
in MI and in SMA. This can be verified by a Kolmogorov-Smirnov statistic that shows no significant difference between the distributions ($P > 0.1$). Interestingly, there is an interaction between lateralization and the sign of the bimanual-related effect. For neurons with a contralateral preference, the bimanual-related effect is positive as often as it is negative. However, for neurons preferring the ipsilateral arm (negative values in Eq. 1), in both MI and SMA, nearly all neurons show a reduction in activity during bimanual movements (results not shown).

Again, a slightly smaller proportion of neurons from monkey G are significantly "bimanually related." In this case, as with the contralateral preference, the smaller number of trials performed by monkey G in each movement type reduced the power of the statistical tests.

Linear combinations of unimanual activity

We tested the NEA during bimanual movements against three null hypotheses: that bimanual NEA is equal to contralateral NEA, that it is equal to the ipsilateral NEA, or that it is equal to a sum of the two. Table 4 shows that for most of the bimanual-related neurons (approximately 80%), all of these hypotheses could be rejected at $P < 0.05$. In contrast, for neurons that were not bimanual-related, 60% of the neurons in MI and 72% of the neurons in SMA failed to reject one or more of the hypotheses at this level—namely, their responses might be explained by a linear combination of the unimanual responses. In an additional analysis, we fit the neuronal activity with a model that attempts to explain bimanual NEA using a general linear combination of unimanual NEAs (Eq. 3). While this model fits 26% of the bimanual-related neurons in MI and 19% of the bimanual-related neurons in SMA (Table 4), the parameters of the fit for different neurons were not clustered in any way. Note that, when the variance in neuronal activity was large, a neuron could fit several of the models tested. However, to be as strict as possible with our results, we did not perform any corrections for the repeated tests. In sum, the majority of the bimanual-related neurons did not admit any linear explanation of their bimanual activity.
Analysis of behavioral controls

As mentioned above, our preliminary analysis and visual inspection of movement trajectories, velocity profiles, and EMG, revealed that, while all of these measures were quite similar in all movements types, they were not identical. The mean and SDs of the trajectories from one recording session are shown in Fig. 10. Velocity profiles for the same recording session are shown in Fig. 11, and examples of EMGs (recorded at the end of training) are shown in Fig. 12. The largest bimanual-related effect (0.067) for the movement trajectories shown in Fig. 10 is in the comparison of unimanual left hand movements toward 45° with movements of the left hand during a bimanual opposite movement in the same direction. This difference is among the largest bimanual effects seen in the movements (in the 90th percentile). The largest bimanual-related effect in the velocity profiles of Fig. 11 is larger than the bimanual-related effects in the movement trajectories on this day. The bimanual-related effect for the difference between unimanual right hand movements to 270° and bimanual opposite movements in the same direction is 0.187. This difference is in the 80th percentile of bimanual-related differences in velocity. Figure 12 shows the activity of the left and right deltoid during performance of the task. These are two of the four muscles that were chosen for simultaneous recording with the neural activity on the basis of apparent differences in the activity during unimanual and bimanual movements of our monkeys. The largest bimanual-related effect in these two muscles is −0.108, which is obtained in the comparison between unimanual right handed movements to 135° and bimanual parallel movements in the same direction.

Figure 13 summarizes the relationship between the strength of the bimanual-related effect in neurons and the strength of the bimanual-related effect in the behavioral variables. Figure 13, A and B, shows scatter plots of the neuronal and kinematic effects. Figure 13C shows a histogram comparing the distribution of strengths of effect in evoked activity in MI and SMA to the distribution of the effect in integrated EMG for all the muscles we recorded. Since the muscles were recorded separately from the neurons, no scatterplot can be shown. We did, however, repeat the statistical analysis applied to the units to determine how much of the EMG activity showed a significant bimanual-related effect. Across all muscles from both sides in all four primary directions (a total of 72 data points), only 19 cases (26.4%) showed a significant bimanual effect. A binomial test shows that this number is significantly less than would be predicted by the fraction of neurons in MI that showed bimanual effects (55.6%). The bimanual-related effect is clearly stronger in the neurons than in any of the behavioral variables we analyzed. Moreover, where tested, there is no correlation between the strength of the bimanual-related effect.
where the difference in the behavioral parameter in bimanual and unimanual trials is small. The “different” group, shown in the bottom row of plots, contains trials where the difference in the behavioral parameter is large. Figure 14A demonstrates this analysis applied to the trajectory deviations. The bimanual-related neuron shown is more active during unimanual movements (bimanual-related effect of \(-0.76\)). The bimanual-related effect is preserved whether or not the trajectory deviations are similar or different. The separation index (Eq. 4) for the separation of the neuronal activity is \(-0.15\) (different from 0, \(P > 0.15\)), while the separation index for the movement trajectories is 4.88 (different from 0, \(P < 0.001\)).

Figure 14B applies the same analysis to the peak velocity for one “bimanual related” neuron. The neuron has a “bimanual related” effect of 0.22, and it is more active in bimanual movements than in unimanual movements. The separation index for the neural activity of the neuron is 0.3 (not different from 0, \(P > 0.15\)). The separation index for the velocity is 3.66 (different from 0, \(P < 0.001\)). For this example, as well, the bimanual-related effect is preserved despite the separation. Finally, Fig. 14C shows an example of the separation analysis applied to the integrated EMG. The strength of the bimanual-related effect for the neuron in Fig. 14C is 0.91. The separation index for the neuronal activity in this analysis is \(-0.11\) (not different from 0, \(P > 0.15\)) while the separation index for the integrated EMG is 2.04 (different from 0, \(P < 0.001\)). As before, the bimanual-related effect is preserved.

Figure 15 shows that the examples in Fig. 14 are quite typical. The results of the separation analysis applied to trajectory deviation and peak velocity in all significantly bimanual-related cells is shown in Fig. 15, A and B. We compared the resulting distribution of separation indexes to a distribution of the analysis applied to the same neurons, but in which division into the “similar” and “different” groups was performed at random. The plots show the relationship between the actual distribution of separation indexes (gray histogram) and the random distribution (black line). A Kolmogorov-Smirnov test for the similarity of two distributions fails to find differences between the measured and random distributions of the separation index for the trajectory deviations (Fig. 15A, \(P > 0.1\)). It does reveal a difference for the distributions generated using the peak velocity (Fig. 15B, \(P < 0.01\)), indicating that for a few cells it may be possible to explain the bimanual-related effect as a reflection of differences in the kinematics of unimanual movements (bimanual-related effect of \(0.22\)).

**Separation analysis**

For many of the neurons, a large number of trials were collected in each condition. This permitted an analysis of the relation between the behavioral variables and the neural activity as illustrated in Fig. 14. The figure depicts the activity of bimanual-related units during performance of unimanual trials (left column, in red) and bimanual trials (right column, in blue). In each of the figure’s three sections, the top row of plots shows the “similar” group (see METHODS) containing trials in a neuron and the strength of the effect in the behavioral variable.

**TABLE 4. Categorization of cells according to the linear model**

<table>
<thead>
<tr>
<th>Area</th>
<th>Total</th>
<th>Ipsi Only</th>
<th>Contra Only</th>
<th>Sum</th>
<th>None</th>
<th>General Model</th>
</tr>
</thead>
<tbody>
<tr>
<td>MI</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Not bimanual</td>
<td>103</td>
<td>41 (40)</td>
<td>47 (46)</td>
<td>30 (29)</td>
<td>41 (40)</td>
<td>19 (18)</td>
</tr>
<tr>
<td>Bimanual-related</td>
<td>129</td>
<td>5 (4)</td>
<td>13 (10)</td>
<td>2 (2)</td>
<td>110 (85)</td>
<td>33 (26)</td>
</tr>
<tr>
<td>All cells</td>
<td>232</td>
<td>46 (20)</td>
<td>60 (26)</td>
<td>32 (14)</td>
<td>151 (65)</td>
<td>52 (22)</td>
</tr>
<tr>
<td>SMA</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Not bimanual</td>
<td>99</td>
<td>47 (47)</td>
<td>56 (57)</td>
<td>37 (37)</td>
<td>28 (28)</td>
<td>29 (29)</td>
</tr>
<tr>
<td>Bimanual-related</td>
<td>107</td>
<td>8 (7)</td>
<td>12 (11)</td>
<td>4 (4)</td>
<td>87 (81)</td>
<td>20 (19)</td>
</tr>
<tr>
<td>All cells</td>
<td>206</td>
<td>55 (27)</td>
<td>68 (33)</td>
<td>41 (20)</td>
<td>115 (56)</td>
<td>49 (24)</td>
</tr>
</tbody>
</table>

The numbers in parentheses represent percent of cells for which one may not reject (\(P > 0.05\)) the hypothesis that particular summations explain their activation during bimanual movements. Three alternative null hypotheses were explored, and if all three could be rejected (\(P < 0.05\)) for a given neuron, it was counted in the “None” column. The percentages do not add up to 100% because the categories are not exclusive (except for the “None” column). The significance criterion for bimanual-related is \(P < 0.001\). These hypotheses seem less appropriate for the bimanual-related neurons than for those that are not bimanual. A general linear model was also tested in which the bimanual activity could be any arbitrary linear combination of the ipsilateral and contralateral activity. In this model, also, most neurons did not fit the model, and there was no clustering of the parameters of the fit.

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and bimanual movements. However, for the majority of neurons, the separation indexes measured are completely consistent with those generated by chance. In general, therefore these results are not consistent with an explanation of the bimanual-related effect purely on the basis of the differences either in the movement paths or in the velocities.

We also applied the analysis to 59 bimanual-related neurons recorded simultaneously with EMG (Fig. 15C). We used a lower level of significance for the bimanual-related effect than in our other analyses ($P < 0.01$) to increase the size of the sample analyzed for this purpose. The muscles recorded were the anterior deltoid and the flexor carpi ulnaris on both the left and right side of the body. For each neuron, we performed a separation analysis separately with each of the four muscles recorded, but then discarded all but the most significant of these analyses (as determined by bootstrapping). This is because we were interested in finding the muscle to which the neuron was most strongly related. Figure 15C shows the distribution of the separation indexes we calculated (gray histogram) and the distribution generated by creating four separation indexes through random selection of trials and discarding all but the most significant (black line). There is no significant difference between these two distributions (Kolmogorov-Smirnov, $P > 0.1$). This analysis shows that the bimanual-related

![FIG. 10](image10.png) Comparison of movement trajectories in different trial types. Plot of the movement trajectories for unimanual and bimanual movements. Mean (shown by the trajectory) and SD (shown by the lines perpendicular to the trajectory) of the movements are shown. SD bars are drawn every 50 ms. Data are from 1 day of recording in monkey G. Strength of the bimanual-related effect is strongest between unimanual movement of the left hand to $45^\circ$ and bimanual opposite movements in the same direction. For this comparison, the bimanual-related effect is 0.067.

![FIG. 11](image11.png) Comparison of velocity profiles in different trial types. Plot of the velocity profiles for unimanual and bimanual movements. Velocity profiles shown are averages of repetitions aligned on beginning of movement. Data are from the same day of recording in monkey G as the movement trajectories shown in Fig. 10. The bimanual-related effect is strongest between unimanual movements of the right hand to $270^\circ$ and bimanual opposite movements in the same direction. Strength of this effect is 0.187.

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effect is not related to the activity in the muscles we recorded beyond chance levels. Figure 16 shows the relationship between the results in the separation analysis and the bimanual-related effect for all three behavioral parameters. The lack of correlation between “bimanual relatedness” and the separation indexes is indicated by the \( r \) values shown in the top right corner of each plot. Thus the population of neurons in our study taken as a whole does not seem to show a strong relationship between variations in the kinematics and dynamics of the movements and variations in the neural activity associated with bimanual movements. The failure to find a relationship of this sort undermines the argument that the bimanual-related effect results solely from differences in the performance of bimanual and unimanual movements.

**DISCUSSION**

This study demonstrates substantial differences between the cortical activity associated with unimanual movements and that associated with bimanual arm movements. The differences seen between these two movement classes are as robust in MI as they are in the SMA. Some of the MI activity differences could be variations in the subtle aspects of the movements, both kinematic and dynamic. Other activity could be due to differences in postural adjustments or postural set between the two tasks. And, some of the observed bimanual activity might simply be linear combinations of unimanual activity. But, these various contributions do not account for the bulk of the observed unit activity differences between unimanual and bimanual arm movements.

In general, both kinematics and dynamics are poor predictors of changes in neural activity specific to the difference between unimanual and bimanual arm movements. Two pairs of analyses reinforce this view. In the first pair, we establish 1) that changes in unit activity are generally far greater between the two task conditions than the similarly computed changes in kinematics or dynamics (summarized in Fig. 13) and 2) that there is no correlation between the bimanual-related effect and changes in kinematics or dynamics (also demonstrated by Fig. 13). This pair of analyses is strong evidence that the bimanual effects are not a result of differences in movement trajectories, velocity profiles, or details of EMG activation. However, averaging trials together might obscure the effects of movement variations. The second pair of analyses applies a trial-by-trial approach to validate the use of mean values by checking whether different subsets of movements contribute differently to the mean activation. This pair of analyses includes 1) looking at cortical activity when behavioral parameters are similar and comparing that to cortical activity when they are different to see if more extreme movements are associated with a greater “bimanual effect” and 2) looking for correlations between the degree of movement deviation from average and the strength of the bimanual effect. In the case of movement paths, we compare cortical activation during movements with a small trajectory deviation to cortical activation during movements with large trajectory deviations (Fig. 14A). The results of this comparison indicate that the observed bimanual effect did not differ significantly between “typical” trials and trials with an extreme trajectory (Fig. 14). The second pair of analyses is completed by showing that, in cases where the typical trials and the extreme trials are particularly distinct, the bimanual effect is no greater than in cases when extreme trials are more typical (Fig. 16). Equivalent analyses are carried out for velocity profiles and EMG activation (Figs. 15 and 16). In the analysis of the velocity profiles, a significant number of neurons was found whose bimanual-related effect could be, in part, explained by variations in velocity. However, for most neurons this was not the case. This battery of tests does not support the proposal by Kazennikov et al. (1999) that trial-to-trial variations in the
Indeed, it is true that a fraction of muscles showed a significant effect when switching between unimanual and bimanual tasks. Related effects as a result of postural adjustments might occur when asked to switch between the two experimental conditions. Our limited survey of EMG activity was carried out with surface recording electrodes (see METHODS) using electrodes that are more likely to oversample by including nearby muscles, rather than undersample, e.g., record from only a limited region of the muscle. One of the striking aspects of the seated posture of the monkey and the mechanics of the arm manipulanda is that very little axial muscle effort is required to displace either hand. Thus it was not surprising that relatively little axial muscle activity was found during the survey. The axial muscles that were active showed little or no modulation with the task. The lack of modulation of axial activity contrasts sharply with the strong modulation of both the proximal arm muscles (Fig. 8) and the single units.

It is still likely that some axial muscles had EMG modulation correlated with the task and some MI modulation varied systematically between unimanual and bimanual tasks. This contribution to the overall recorded neuronal population would necessarily be small, however. The most pronounced feature of the MI topography is that postural muscles have a dramatically small motor representation (Craggs et al. 1976; Woolsey et al. 1952). Muscles with such a small motor representation would not dominate the results unless the recordings were selected for axial muscle activity, accidentally concentrated in one tiny region of MI, or altogether out of the arm area. Figure 3 shows that the recorded units in this study were far too widespread in MI to be dominated by such a small motor representation. Evidence of clear arm and shoulder related activity seen in our recordings, often localized to a single joint, as well as the microstimulation sites that produced frank single-joint arm movements confirmed that our recording sites in MI were in the well-established proximal arm and shoulder areas explored by other investigators (Georgopoulos et al. 1983; Kalaska and Crammond 1992). Subsequent (Steinberg et al. 2002) recordings from the same chamber of monkey G on the directional tuning of MI neurons during unimanual and bimanual movements showed tuning in most neurons in both unimanual and bimanual movements, further demonstrating that our sample was from the forelimb motor area.

We explored the possibility that differences in cortical activation during bimanual arm movements are simply the result of some linear combination of unimanual activities. In this study, we examine three simple models that might predict bimanual activity: left limb activity alone, right limb activity alone, and an equally weighted sum of left and right limb activities. We reject all three models for more than one-half of the units in both MI and SMA (Table 4). Even a more general version of the third model, one that allows independent weighting of the contributions from each limb, cannot explain the activity of most units. Moreover, units with a significant bimanual effect fit the linear model less often than others, further reducing the likelihood that the bimanual effect is explained by a linear model.

Given the clear difference in activity during unimanual and bimanual movements, and given that these differences are neither the result of variations in the movement parameters, nor simply the combination of individual limb-related activations, we are led to conclude that there are signals in MI and SMA...
that specifically reflect bimanual arm movements. Similar analyses were not possible in prior studies because movements were either too restricted to provide useful trajectory information (Tanji et al. 1988: button pressing task) or the movements were not continuously measured (Kazennikov et al. 1999; Kermadi et al. 1998: food retrieval task). The two more recent studies, both using very similar behavioral paradigms, draw contradictory conclusions. Kermadi et al. reported essentially the same results as reported here: bimanual related units were common in MI (48%) and only slightly less so in SMA (44%). In contrast, Kazennikov et al. argued that their data did not support bimanual specificity in either MI or SMA, based on an unusually restrictive definition of bimanual specificity (Tanji et al. 1988; the methods section of this paper; compare with Kazennikov et al. 1999; Kermadi et al. 1998). When we apply our definition to their published data by combining the units in

FIG. 14. Separation analysis. Figure shows an example of the separation analysis applied to each of the behavioral parameters. For A, B, and C, trials from the “similar group” are in the top row, and trials from the “different” group are in the bottom row. Left column: PSTH for the neuron during 1 type of unimanual trial (red). Right column: PSTH for the neuron during 1 type of bimanual movement (blue). Middle column: behavior, where trials in red are unimanual trials and trials in blue are bimanual trials. A: separation analysis of the trajectory deviation for a “bimanual-related” neuron from left MI. For this neuron, evoked activity is greater during unimanual movements. During movements of the right hand, activity of the neuron was at baseline. Number of trials for each histogram: 18. B: separation analysis of the peak velocity for a “bimanual-related” neuron from left SMA. This neuron is significantly less active during unimanual right-handed movements than during bimanual opposite movements. Number of trials for each histogram: 41. C: separation analysis applied to the integrated EMG for a “bimanual-related” neuron from right MI. For this neuron, evoked activity is greater during bimanual movements than it is during unimanual movements. The neuron is not activated during movements of the ipsilateral arm. Number of trials per histogram: 5.
subclasses b, c, and e (Kazennikov et al. 1999, Table 1, under the assumption that “moderate” differences reported between bimanual and unimanual activation in subclass b and c are statistically significant), we find that 46% of units in MI and 48% of units in SMA have activity specific to bimanual arm movements. This interpretation is in agreement with Kermadi et al. as well as the present report.

The Tanji et al. (1988) study is unique in finding a substantial difference between MI and SMA in a bimanual task. In their study, the activity of units in MI and SMA was recorded during the performance of left handed, right handed, and bimanual finger presses. The monkeys were carefully trained, using EMG activity recorded during training and feedback from force transducers, to minimize undesired muscle activation. Undesired muscle activation included proximal muscle activity, activity in the contralateral muscles during unimanual movements, and differences in the activity of the ipsilateral muscles in bimanual and unimanual movements. Following this extensive training, most neurons in MI responded similarly during bimanual and contralateral unimanual movements. In contrast, many units in SMA were activated during contralateral (unimanual) movements but not during bimanual movements or vice versa.

We offer two alternative explanations for the observation that the button-pressing task of Tanji et al. rarely produced MI activity specific to bimanual movements, while our planar tracking task and the food retrieval task (Kazennikov et al. 1999; Kermadi et al. 1998) often produced such activity. One explanation, discussed in detail elsewhere (Donchin et al. 1998, 1999), is that MI control of distal hand movements may be different from MI control of more proximal movements (including movements at the elbow). In this view, MI representation of proximal movements is predominantly bilateral, whereas distal movements are represented as more or less simple combinations of unimanual movements. The other ex-
planation suggests that different training requirements in the two tasks caused the difference in the results. The extensive training required for suppression of disallowed muscle activation in Tanji’s study may indicate that bilateral suppression of muscle activity was a major task constraint. This constraint would be largely the same in both unimanual and bimanual button pressing. This could explain why MI activation was similar in both the bimanual and unimanual conditions. SMA, whose task representation may be more abstracted from muscular constraints, could still distinguish the task conditions. In our study, the major training hurdle was to achieve simultaneous onset and offset of the arm movements in bimanual movements and immobility of one arm in unimanual movements. These requirements called for a training period of several months and were clearly very different for bimanual and unimanual trials. We suggest that this type of extensive training shaped very different cortical activity patterns for the different conditions of the bimanual task, in contrast with the button pressing task.

Note that we differentiate “task requirements” from “variations in task performance.” A prediction that follows from the second explanation above is that the differences required by the task are critical to the development of bimanual related cortical activity. Execution differences that have no substantial impact on the learning would not be expected to influence the strength of the bimanual effect. Nudo recently showed that dynamic changes in the MI motor map have a similar dependence on task requirements (Plautz et al. 2000).

It is still impossible to say whether the conjectural “learning-based” explanation of the bimanual effect is the correct explanation. The possibility that fundamental differences between proximal and distal motor control explains the dichotomy of results begs further investigation. Tasks that compare proximal and distal bimanual movements could be helpful in this regard, as would a study of the development of the bimanual effect through the course of the training procedure. However, one conclusion that emerges from the present work is that bimanual arm movements can be represented by cortical activity that is distinct from the representation used for unimanual movements. This finding challenges our understanding of the relationship of the motor control system, and our knowledge regarding the underlying basis for the representation of movements in the cortex.

We thank Y. Donchin for help with the surgical procedures and G. Goelman for help with the MRI. We also thank S. P. Wise and R. Paz for comments on earlier versions of the manuscript.

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REFERENCES


pairs of pretraining trials. The procedure during pretraining trials differed from visuospatially distinct by a surrounding structure of Lego bricks. Access to plastic ice-cube trays (2 x 3 x 1 cm) as storage sites that were unique and discriminable on every trial for each bird, the start of each caching phase, a storage tray was placed in the bird’s home cage. At the beginning of each caching phase, peanuts were placed in the same ice-cube molds in which they had been cached. To minimize observer bias, different observers performed tasks that required coordination between the two limbs. The order of the 4-h and 124-h training and test trials was counterbalanced. None of the caches was returned to the storage tray before the recovery phase. Furthermore, before all recovery trials were omitted from the analysis.

Subjects.

Adult, hand-raised scrub jays, which were allocated randomly to either the non-pilfered or pilfered group. All birds had previously participated in experiments that involved caching trials, on which the most recent training and test trials were completed at least 14 days before the start of the current experiment. For each group, six birds were assigned to each of the three caching phases of the experiment. Before the training and test trials, all birds received at least four pretraining trials. Each caching phase consisted of four 15-min trials, on all of which, peanuts and worms were cached. To minimize observer bias, different observers performed tasks that required coordination between the two limbs. The procedure was the same on the test trials, except that none of the caches was returned to the storage tray before the recovery phase. Location of caches. The procedure was the same on the test trials, except that none of the caches was returned to the storage tray before the recovery phase. Location of caches.

Methods

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Examples of the four main types of trials (the open circles are not visible to the location of the manipulanda. Each cursor appears in the corresponding origin. All target locations are shown as grey circles surrounding each origin.

A monkey performed these four types along with complementary four types of movement (53 of 72 in both arms, modulation in contralateral movements was stronger in the right SMA and 22 of 44 from monkey G) and SMA (73 from monkey F). The monkey sat in a primate chair and faced a video screen. Two cursors (+) indicate the holding two manipulanda and facing a video screen. Two cursors (+) indicate the holding two manipulanda and facing a video screen.

We recorded single-unit activity from homologous sites in the right SMA. It was inactive in unimanual movements towards 45 or 225 when the two arms moved in parallel towards 225. It was less active in one other kind of bimanual trial (Fig. 2c) and failed to respond in the rest (Fig. 2a, d).

The neuron in Fig. 3A is from right MI, and fires before movement 8. The neuron had no contralateral activity towards 45 or 225 or ipsilateral activity when the two arms moved in parallel towards 225. It was inactive in unimanual trials (right column is unimanual right; middle column is unimanual left). Trials are aligned on the beginning of the movement (of the first arm) and graphed above the graph by arrows or a black circle if the arm does not move. The cell had almost completely absent when the monkey performed similar unimanual movements.

Many cells exhibited bimanual-related activity: 69% (53 of 72 in monkey F) and 47 of 73 in monkey G) in MI and 64% (47 of 73) in SMA. This indicates that bimanual-related activity is at least as common in MI as in SMA. Note that these cells outnumber 'directionally selective' cells, suggesting that even classically 'unresponsive' neurons may participate in bimanual coordination.
contralateral activity (component, at the limit of significance. The bimanual-related component appears only when the contralateral arm moves in the non-preferred direction (demonstrates a weak bimanual-related component at the limit of significance). Figure 3 and 3A are significant at a level of 0.001, and no significant effect was found in the analysis of variance (ANOVA) performed on the activity of the cell located between the origin and target. Examination of the monkeys during task performance did not reveal postural adjustments that distinguished movements during bimanual parallel movements is on the left (red), and during unimanual left movements is on the right (blue). Middle plots show the movement paths of the trials in which the movements were not similar are shown underneath. Figure 4 shows that selection of trials with similar trajectories in unimanual and bimanual conditions did not lessen the temporal pattern of bimanual-related MI neuron. Trials in which unimanual and bimanual trials. Visual inspection of the recorded EMG patterns similar to the one performed on the neural activity of single neurons in MI and SMA, which demonstrate that neurons can be activated in relation to the specific nature of the bimanual coordination (Fig. 2). We find that bimanual-related responses occur at least as frequently in MI as in SMA, whereas the previous work found that a far greater proportion of units in the SMA respond to bimanual and unimanual or exclusively to the bimanual movements. This difference may lie in the fact that our monkeys used their entire arm. We performed similar analyses on velocity profiles and EMG. For example, an analysis of one bimanual-related MI neuron. Trials in which unimanual and bimanual movements were similar are shown in the top displays; trials, although there were differences. For example, an analysis of the bimanual-related activity of single neurons in MI and SMA, which reveals that neurons fire specifically in the bimanual task. We extend these findings to different types of bimanual movements, and demonstrate in Fig. 5 that the bimanual-related activity is not solely dependent on precise kinematics or muscular activity. However, the discovery of bimanual-related activity in MI underlines the role of the SMA in bimanual movements. We find that a far greater proportion of units in the SMA respond to bimanual and unimanual or exclusively to the bimanual movements.
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Timing of bimanual movements in human and non-human primates in relation to neuronal activity in primary motor cortex and supplementary motor area

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Abstract This study investigates the timing of bimanual movements in a combined behavioral and physiological approach. Human subjects and rhesus monkeys performed the same bimanual task. In monkeys, we simultaneously recorded neuronal activity in the two hemispheres of primary motor cortex (MI) or supplementary motor area (SMA), and related it to bimanual coordination in the temporal domain. Both for monkeys and humans, the reaction times of bimanual movements never significantly exceeded the reaction times of the slower arm in unimanual movements. Consistent with this, the longest delay between neural activity onset in SMA and MI and movement initiation was observed in unimanual movements of the slower arm and not in bimanual movements. Both results suggest that the programming of bimanual movements does not require more processing time than unimanual movements. They are also consistent with the view that bimanual movements are programmed in a single process, rather than by combining two separate unimanual movement plans. In both humans and monkeys, movement initiation was highly correlated between the arms. However, once movements began, the temporal correlation between the arms progressively declined. Movement decorrelation was accompanied by a net decorrelation of neuronal population activity in MI and SMA, suggesting a functional connection between neuronal interactions and the level of bimanual coupling and decoupling. The similarity of neuronal activities in MI and SMA is consistent with the idea that both areas are involved in the temporal coordination of the arms.

Keywords Bimanual coordination · Temporal coordination · Motor psychophysics · SMA · MI

Introduction

Most natural movements involve the activation of a multitude of muscles, acting around several joints or even different limbs. For the neuronal control system, this immediately poses the question of how the different elements of a complex movement pattern are coordinated with each other. Do complex movements have their own representations, or are they constructed by combining individual representations of simple movements? Once a movement has started, how does the system ensure that spatial and temporal coordination of individual components is maintained? A popular example of motor coordination is bimanual coordination between the arms. It has been shown that in discrete bimanual movements the two arms tend to start moving together (Kelso et al. 1979, Marteniuk et al. 1984; Fowler et al. 1991; Boessenkool et al. 1999) and that the level of temporal correlation between the arms is regulated according to the demands of the task (Kazennikov et al. 1994; Weiss et al. 1997).

This paper investigates possible neuronal correlates of this bimanual coordination in the temporal domain. By using the same behavioral paradigm in both humans and
monkeys, we can relate our results to the existing research in the psychophysics of bimanual control in humans and explore the underlying neural basis of the behavior.

We first address the question of whether bimanual movements are generated by combination of two separate motor plans or by a single common plan. The first possibility would require an additional process (e.g., a “coordinating schema,” originally suggested for the interaction of reach and grasp; Hoff and Arbib 1993) that coordinates between two independent unimanual motor plans. This would imply that execution of bimanual movements may require additional cognitive resources and therefore predicts increased reaction times for bimanual movements as compared to the unimanual movements that compose them.

Alternatively, some have taken our tendency to produce spatially and temporally coupled movements as support for a single bimanual movement plan (e.g., a “generalized motor program” or GMP, Schmidt 1975; Schmidt et al. 1979). As a variant of this idea, the term “coordinative structure” was coined for collectives (synergies) of muscles that can be controlled jointly as a single functional unit (Kelso et al. 1979; Easton 1972; Turvey 1977). It has been argued that a single plan enhances coding efficiency by reducing the degrees of freedom (Bernstein 1967; Turvey 1977). In this framework, a bimanual movement, just like a unimanual movement, could be programmed as one motor pattern. As a result, no differences in preparation time would be expected. The existing experimental evidence about reaction times of bimanual movements is equivocal. In some conditions longer bimanual reaction times were reported, while in others bimanual reaction times were similar or shorter than unimanual ones (Kelso et al. 1979; Di Stefano et al. 1980; Agioti et al. 1993; Anson and Bird 1993; Garry and Franks 2000; Taniguchi et al. 2001). In this study we reexamine reaction times in bimanual reaching movements and compare them to neuronal processing times at a high level of the motor system, in the cortical motor areas MI and SMA.

Even if bimanual movements are planned by one motor plan, it is evident that this hypothetical plan must drive two independent effectors that drive each arm. Therefore, under any of the hypotheses above, the movements of the arms must be coordinated during the execution phase. The second question of our study concerns the mechanism of maintenance and regulation of temporal motor coordination during movement execution. One possibility is that during ongoing movements the motor commands of both arms interact, resulting in crosstalk effects (Marteniuk et al. 1984; Heuer et al. 2001). Recent studies have suggested that for bimanual movements, at least some of the crosstalk between the movements decays with increasing processing time (Heuer et al. 1998). Other studies also suggest that temporal coupling is strongest at the time when movements start (Boessenkool et al. 1999), and that coupling may deteriorate with time (Fowler et al. 1991). If this decoupling is a result of crosstalk between the motor codes of the two arms, one may expect to find signs of these interactions within the nervous system, at the level where the motor codes are generated. In a previous study, we found that correlations between the motor cortices of the two cortical hemispheres were related to the mode of spatial coordination between the arms (Cardoso de Oliveira et al. 2001). This result confirms previous clinical findings suggesting that the corpus callosum is involved in spatial bimanual coupling (Fuller and Kelso 1989; Franz et al. 1996; Eliassen et al. 1999; Preilowski 1972, 1975). Other studies suggest that the corpus callosum may also contribute to temporal coupling (Stephan et al. 1999; Eliassen et al. 2000). This study tests this hypothesis by relating interhemispheric interactions between cortical areas to temporal aspects of bimanual coordination. To that end, we present behavioral data from human subjects and monkeys performing an identical task. In the monkeys, we test neuronal activity in SMA and MI. Many early studies have suggested that the SMA is more involved in bimanual coordination (Brinkman 1984; Tanji et al. 1987, 1988; Kazennikov et al. 1999), but more recent work provides evidence that MI may be involved in this task to the same extent (Donchin et al. 1998, 2001; Kermadi et al. 1998; Cardoso de Oliveira et al. 2001).

### Materials and methods

#### Behavioral paradigm

The same experimental setup was used for experiments with humans and monkeys. The experimental setup and the principle of the task design have already been described in Donchin et al. (1998) and Donchin et al. (2001). A sketch of a monkey engaged in performance of the task is shown in Fig. 1A. Subjects moved simultaneously two separate manipulanda, one with each arm. Each manipulandum was a low-weight, low-friction, two-joint mechanical arm, moveable only in the horizontal plane. Movement of each manipulandum caused movement of a corresponding cursor on a vertically oriented 21” video screen located ~50 cm in front of the subject. The movement of each cursor was mapped to its corresponding manipulandum movement such that each millimeter of manipulandum movement caused 1 mm of movement of the cursor on the video display.

The time course of typical unimanual and bimanual trials was as follows. A trial began when the subject placed both cursors within 0.8-cm-diameter “origins” (Fig. 1A) and held them still during a hold period (of 500 ms in monkeys G and F, or 1,000 ms in monkey P and humans). For each arm, a target (also 0.8 cm in diameter) could appear at a distance of 3 cm (monkey P and monkey F) from the origin, and in one of eight different directions. For humans, targets could occur at long (5-cm) or short (2.5-cm) distances from the origins. If only one target appeared – signaling a unimanual trial – the subject moved the appropriate arm and brought the corresponding cursor into the target, but did not move the other arm. If two targets appeared – signaling a bimanual trial – the subject moved both arms, such that the two cursors moved into the targets on the screen. Three types of bimanual movements were studied: parallel, opposite and perpendicular. Unimanual movements comprised movements in eight different directions (in the four cardinal directions, plus those directions 45° from the cardinal directions, Fig. 1C). Monkeys F and G performed parallel and opposite movements in eight directions. Monkey P also performed perpendicular movements, in which the directions of arm move-
A. Behavioral setup  B. Raisin Task

C. Unimanual Movement Types

D. Bimanual Movement Types

Fig. 1A–D The behavioral task. A The monkey moved two manipulanda in the horizontal plane. The position of each manipulandum was displayed as a cross-shaped cursor on a vertical screen in front of the monkey. Each trial began by presenting two origin circles in the middle of the display (circles with crosses). After the monkey placed the cursors into the origins and held them immobile for a constant delay, the origin circles were replaced by two target circles at different locations. In unimanual trials, one of the target circles appeared in the same location as the origin (of the non-moving hand). The other circle was displaced from the origin in one of the eight directions shown in C. B Sketch of the raisin boards used. A raisin was placed in each well and the monkey merely had to retrieve them at its own pace, with either hand. The board displayed on top tested simple reaching, while the one displayed below required finger coordination to produce a precision grip. C Unimanual movements could be in one of the eight directions shown here. D Schematic representation of the bimanual movements used in our task. Upward arrows in C and D correspond to forward movements of the monkey’s arm, and downward arrows correspond to backward movements towards the chest of the monkey.

Monkeys

Three female rhesus monkeys (Macaca mulatta) (monkey F, 3.5 kg, monkey G, 3.5 kg, and monkey P, 4 kg) were trained in the task. They were familiarized with the task in a step-by-step procedure lasting for 6–10 months. The behavioral data presented here were collected after extensive training, and after the monkeys reached a stable level of performance in all movement types. During the sessions used for this study, neuronal activity was also recorded (see below).

In order to test the hand preference in monkeys, we used two “raisin board” tasks (see Fig. 1B). The first consisted of a 12.5x34-cm Perspex board into which nine wells of 4 cm diameter were drilled. This arrangement allowed fast retrieval of food reward out of the wells by whole-arm reaching and grasping. The second was a 10x20-cm Perspex board with 15 elongated slots (15 mm long, 6 mm deep and 6 mm wide) oriented randomly at either 0, 45, 90, 135 or 180 degrees. The spatial dimensions of the wells required retrieval of food by opposing thumb and index finger in a precision grip (Brinkman 1984). Each well was loaded with a raisin by the experimenter. The monkey sat in the primate chair and was presented the board, which was positioned in the middle, ~30 cm in front of the monkey’s chest. The monkey rapidly collected all raisins from the wells. The behavior of the monkey was videotaped, and hand preference was determined by counting how often the monkey used either hand to retrieve the raisins.

Human subjects

All human subjects (age 23–32 years, two female, four male, five right-handed, two left-handed, according to a modified Edinburgh Inventory questionnaire (Oldfield 1971) gave their informed consent and were naive to the purpose of the experiment. None of the subjects was aware of any neurological or neuromuscular abnormalities. The experimental procedures were in agreement with the Declaration of Helsinki (Ethical Principles for Medical Research Involving Human Subjects, adopted by the 18th WMA General Assembly, Helsinki, Finland, June 1964, and continuously amended, last by the 52nd WMA General Assembly, Edinburgh, Scotland, October 2000), and were approved by the ethics committee of the Psychology Department at the Hebrew University. Each human subject performed three to six sessions on different days. Only sessions with at least 60% successfully performed trials were included in this study. In some subjects, the first session did not conform to this criterion and was discarded, but all other sessions were performed to criteria by all subjects.

Recording of neuronal activity in monkeys

After completing the training period, two recording chambers (exposing the primary motor cortex) and a head holder were implanted on the skull. Surgery was performed as described before (Donchin et al. 1998, 2001; Cardoso de Oliveira et al. 2001). The animals' care and all surgical procedures were in accordance with the NIH Guide for the Care and Use of Laboratory Animals (rev.
monkey P is still participating in experiments, the locations of the within the recording areas described in Donchin et al. (1998). Since SMA proper. All recordings of monkeys F and G were located in the caudal portion of dorsal premotor cortex. All SMA recordings were situated within the medial bank of the cortex and in the exposed part of primary motor cortex using the chamber coordinates and positions. Most of our MI recordings were in the exposed part of primary motor cortex, as judged by stability of spike amplitude and spike rate. At the end of each recording session, we tested for neuronal responses to passive manipulation and tactile stimulation of the limbs, tail, trunk and head. Evoked activity was evaluated by listening to the amplified spike signal passed directly into a loudspeaker. Finally, we applied intracortical microstimulation (ICMS) with 50-msec trains of 200-μs cathodal pulses at 300 Hz with an intensity of 10–80 μA (BP-2 and BSI-2, BAK Electronics, Germantown, MD). When ICMS evoked movements, we documented the movements evoked and the threshold stimulation intensity. Stimulation and passive manipulation were performed at the end of each recording session, as well as in dedicated mapping penetrations. For this study, we only included recording sites that were within the arm representation of either SMA or MI, as determined by passive stimulation and ICMS.

Neuronal data

All cell recordings were assessed for intertrial stability, and only stable portions were analyzed. To compare the onsets of movements to the beginning of significant neuronal activation, we determined the onsets of neural activity changes for each cell using the CUSUM algorithm (Ellaway 1977; Davey et al. 1986). This procedure was performed on the basis of the average firing rate within a time window of 200 ms before to 300 ms after movement onset (as defined by the offline detection algorithm). We used two thresholds, a stricter one (4 times the confidence limit as defined in Ellaway 1977) and a less strict one (1.5 times the confidence limit). Onsets were limited to the time from target appearance to 400 ms after movement initiation. The distribution of activity onsets of each bimanual movement type was compared to the one of the
corresponding unimanual movements. The significance of the difference was tested using the non-parametric Mann-Whitney test.

We next sought to compare the time course of movement correlations to the correlations between neural activities within the cortical hemispheres. To that end, we analyzed the LFP signals of each pair of electrodes during each movement type within several time windows corresponding to the behavioral landmarks defined above. These windows were from \(M_{\text{on}}\) to the averaged TTP, from the averaged TTP to the averaged movement onset (\(M_{\text{off}}\)), and from \(M_{\text{off}}\) to 200 ms after \(M_{\text{off}}\). In addition, two windows of 200 ms duration were analyzed before \(M_{\text{on}}\) (–400 to –200 ms before \(M_{\text{on}}\)) and –200 ms to \(M_{\text{on}}\). The average \(M_{\text{on}}\), TTP and \(M_{\text{off}}\) were determined from the same movement type, and were always taken from the first hand to move. Mean interhemispheric correlations within these windows were calculated by averaging the synchronous LFP correlations as revealed by the JPETC method (for a detailed description of this technique, see Cardoso de Oliveira et al. 2001). Briefly, the JPETC method is an adaptation of the JPSTH method (Aertsen et al. 1989) and yields time-resolved correlations with the same temporal resolution as the sampling rate. Here, we restricted the analysis to synchronous correlations, corresponding to the diagonal of the JPETC. After determining the average correlation in these windows for each pair and movement type, we summarized the data by averaging over all pairs and movement types.

Other aspects of parts of the physiological data set underlying this study have been analyzed in separate manuscripts. Differences between cell activities and LFPs in unimanual and bimanual movements have been presented in Donchin et al. (1998, 2001; data from monkeys G and F). The predictive value of population vectors for bimanual movements has been assessed by the raisin board task and by performance in the bimanual task. Figure 2A demonstrates that all three monkeys preferred to pick up raisins with the right hand. In bimanual movements, the right arm led the movement more often than the left arm. In addition, in Fig. 2A, B Hand preference in monkeys. A The left column of the figure shows the percentage of usage of the right (black bars) and left (white bars) hand in the two raisin tasks (average values over 10 days for monkey G and 23 days for monkey P), and the percentage of movements in which each hand began to move first in the bimanual movements. In monkey F, the raisin task was not done. B The right column of the figure shows the average RT for each hand in unimanual and bimanual movements (white bars for the left hand, black bars for the right). Error bars display standard deviations and the asterisks indicate a significant difference between the hands (\(P<0.01\), Mann-Whitney test).

Results

Data

Data were recorded during 76 recording sessions in 3 monkeys (17 from monkey F, 37 from monkey G and 22 from monkey P) and 3 to 6 sessions in human subjects. The monkeys performed between 1,000 and 3,000 movements per daily session, and humans performed between 300 and 500 movements, resulting in 30–200 (monkeys) and 10–20 (humans) repetitions of each movement type. The neural activity analysis includes data from the three monkeys recorded during performance of the center-out task. In MI, we recorded from 877 neurons (129 from monkey F, 262 from monkey G, and 486 from monkey P), and LFPs from 278 sites (59 from monkey F, 82 from monkey G, and 137 from monkey P). In SMA, we recorded from 240 neurons (79 from monkey F, 161 from monkey G), and LFPs from 74 sites (39 from monkey F and 35 from monkey G).

Hand preference

All but two human subjects were right handed as judged by a modified Edinburgh Inventory questionnaire (Oldfield 1971). The hand preference of monkeys was determined by the raisin board tasks and by performance in the bimanual task. Figure 2A demonstrates that all three monkeys preferred to pick up raisins with the right hand. In bimanual movements, the right arm led the movement more often than the left arm. In addition, in Fig. 3 Comparison of RTs in bimanual and unimanual movements. For each subject, we show the average RTs of the non-dominant (white bars) and dominant (black bars) hand during unimanual movements and the RT of the faster hand during bimanual movements (gray bars). Vertical lines indicate standard deviations.

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### Table 1 Summary of RTs, MTs and TTPs of both arms in unimanual and bimanual movements for all monkeys and human subjects. The first number is the mean value, followed in brackets by the median value and the standard deviation. Outliers have been eliminated prior to averaging as follows: RTs and MTs smaller than 50 ms or bigger than 1,500 ms, and TTPs smaller than 50 ms and bigger than 500 ms, were discarded. The two rightmost columns indicate probabilities of type I errors estimated using a Mann-Whitney U-test on values during unimanual movements and movements of the same arm in bimanual movements.

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<td>Monkey F, 11 sessions</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RT</td>
<td>238 (234, 46)</td>
<td>241 (239, 45)</td>
<td>271 (259, 64)</td>
<td>280 (267, 72)</td>
<td>3×10⁻⁷</td>
<td>2×10⁻¹⁶</td>
</tr>
<tr>
<td>MT</td>
<td>380 (379, 84)</td>
<td>387 (376, 97)</td>
<td>386 (386, 90)</td>
<td>414 (411, 94)</td>
<td>0.0177</td>
<td>2×10⁻⁷³</td>
</tr>
<tr>
<td>TTP</td>
<td>274 (267, 66)</td>
<td>263 (249, 66)</td>
<td>227 (217, 62)</td>
<td>257 (244, 70)</td>
<td>1×10⁻³⁷</td>
<td>1×10⁻¹⁷³</td>
</tr>
<tr>
<td>Monkey G, 26 sessions</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RT</td>
<td>234 (227, 48)</td>
<td>254 (247, 49)</td>
<td>279 (269, 63)</td>
<td>280 (264, 78)</td>
<td>0</td>
<td>6.6×10⁻⁶</td>
</tr>
<tr>
<td>MT</td>
<td>492 (484, 121)</td>
<td>506 (496, 138)</td>
<td>478 (471, 146)</td>
<td>519 (524, 148)</td>
<td>1×10⁻¹⁴</td>
<td>1×10⁻¹²¹</td>
</tr>
<tr>
<td>TTP</td>
<td>231 (217, 68)</td>
<td>211 (197, 65)</td>
<td>222 (204, 72)</td>
<td>262 (247, 81)</td>
<td>1×10⁻¹⁴⁷</td>
<td>0</td>
</tr>
<tr>
<td>Monkey P, 22 sessions</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RT</td>
<td>343 (326, 83)</td>
<td>335 (319, 81)</td>
<td>340 (321, 87)</td>
<td>348 (334, 87)</td>
<td>6.5×10⁻⁷</td>
<td>5.6×10⁻⁸</td>
</tr>
<tr>
<td>MT</td>
<td>729 (728, 187)</td>
<td>612 (594, 173)</td>
<td>632 (621, 151)</td>
<td>677 (673, 169)</td>
<td>3×10⁻¹⁸³</td>
<td>2.1×10⁻⁶⁰</td>
</tr>
<tr>
<td>TTP</td>
<td>262 (252, 82)</td>
<td>190 (169, 63)</td>
<td>199 (189, 58)</td>
<td>264 (252, 72)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Human subjects</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Subject I, 4 sessions</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RT</td>
<td>291 (280, 64)</td>
<td>292 (284, 73)</td>
<td>295 (292, 68)</td>
<td>297 (282, 79)</td>
<td>0.4655</td>
<td>0.4589</td>
</tr>
<tr>
<td>MT</td>
<td>594 (559, 169)</td>
<td>610 (589, 163)</td>
<td>600 (569, 164)</td>
<td>560 (531, 160)</td>
<td>0.009</td>
<td>2×10⁻⁶³</td>
</tr>
<tr>
<td>TTP</td>
<td>240 (237, 62)</td>
<td>249 (244, 67)</td>
<td>238 (232, 66)</td>
<td>219 (212, 52)</td>
<td>0.0071</td>
<td>2×10⁻⁸</td>
</tr>
<tr>
<td>Subject D, 4 sessions (left-handed)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RT</td>
<td>263 (252, 59)</td>
<td>267 (259, 59)</td>
<td>281 (272, 56)</td>
<td>290 (279, 63)</td>
<td>0.0339</td>
<td>0.0122</td>
</tr>
<tr>
<td>MT</td>
<td>583 (534, 148)</td>
<td>649 (624, 159)</td>
<td>609 (581, 152)</td>
<td>539 (511, 145)</td>
<td>1×10⁻¹⁶</td>
<td>1×10⁻¹⁸³</td>
</tr>
<tr>
<td>TTP</td>
<td>257 (247, 55)</td>
<td>276 (272, 66)</td>
<td>271 (267, 69)</td>
<td>245 (234, 65)</td>
<td>3×10⁻³⁸</td>
<td>1×10⁻¹⁴⁴</td>
</tr>
<tr>
<td>Subject A, 3 sessions</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RT</td>
<td>293 (272, 96)</td>
<td>302 (287, 86)</td>
<td>313 (299, 90)</td>
<td>301 (277, 93)</td>
<td>0.0034</td>
<td>8×10⁻⁹</td>
</tr>
<tr>
<td>MT</td>
<td>556 (541, 168)</td>
<td>618 (584, 189)</td>
<td>578 (554, 186)</td>
<td>570 (551, 154)</td>
<td>9×10⁻⁸</td>
<td>0.2166</td>
</tr>
<tr>
<td>TTP</td>
<td>275 (264, 68)</td>
<td>291 (283, 70)</td>
<td>266 (254, 76)</td>
<td>267 (247, 79)</td>
<td>2×10⁻⁴</td>
<td>0.4455</td>
</tr>
<tr>
<td>Subject R, 3 sessions</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.256</td>
<td>5×10⁻⁹</td>
</tr>
<tr>
<td>RT</td>
<td>347 (326, 83)</td>
<td>351 (338, 92)</td>
<td>322 (314, 87)</td>
<td>335 (324, 77)</td>
<td>0.0337</td>
<td>0.3823</td>
</tr>
<tr>
<td>MT</td>
<td>649 (566, 236)</td>
<td>600 (570, 319)</td>
<td>653 (620, 274)</td>
<td>653 (619, 198)</td>
<td>0.0115</td>
<td>0.3015</td>
</tr>
<tr>
<td>TTP</td>
<td>313 (294, 87)</td>
<td>325 (326, 86)</td>
<td>309 (309, 79)</td>
<td>310 (294, 85)</td>
<td>0.0747</td>
<td>5×10⁻⁷</td>
</tr>
<tr>
<td>Subject M, 3 sessions (left-handed)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RT</td>
<td>221 (227, 32)</td>
<td>230 (224, 51)</td>
<td>240 (233, 65)</td>
<td>219 (217, 39)</td>
<td>0.0189</td>
<td>0.0994</td>
</tr>
<tr>
<td>MT</td>
<td>460 (489, 136)</td>
<td>495 (474, 161)</td>
<td>479 (444, 156)</td>
<td>484 (504, 133)</td>
<td>3×10⁻⁹</td>
<td>0.35</td>
</tr>
<tr>
<td>TTP</td>
<td>189 (179, 54)</td>
<td>222 (217, 55)</td>
<td>205 (198, 45)</td>
<td>204 (197, 45)</td>
<td>7×10⁻¹⁸</td>
<td>1×10⁻⁴³</td>
</tr>
<tr>
<td>Subject E, 6 sessions</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RT</td>
<td>295 (289, 58)</td>
<td>282 (277, 57)</td>
<td>307 (302, 64)</td>
<td>333 (324, 70)</td>
<td>1×10⁻⁵</td>
<td>0.0303</td>
</tr>
<tr>
<td>MT</td>
<td>486 (482, 81)</td>
<td>519 (516, 82)</td>
<td>526 (521, 87)</td>
<td>532 (529, 80)</td>
<td>1×10⁻⁵</td>
<td>0.0303</td>
</tr>
<tr>
<td>TTP</td>
<td>251 (242, 61)</td>
<td>263 (257, 63)</td>
<td>266 (257, 65)</td>
<td>277 (269, 67)</td>
<td>1×10⁻¹²</td>
<td>8×10⁻¹⁰</td>
</tr>
</tbody>
</table>
two out of the three monkeys, the reaction times of the right arm were significantly smaller than those of the left arm, during both unimanual and bimanual movements (Fig. 2B, monkeys G and F, Mann-Whitney test, \(P<0.05\)).

**Reaction times, times to peak velocity and movement times**

Figure 3 compares the RTs of both hands in unimanual movements and the RTs of the leading hand in bimanual movements. Here, unlike in Fig. 2, we specifically compared bimanual movements to those unimanual movements that composed them. In Table 1, we present the reaction times of both hands in bimanual movements separately, and present the results of a Mann-Whitney test between the reaction times of each hand in unimanual and bimanual movements. Significance thresholds were \(P<10^{-6}\) for monkeys, and \(P<0.01\) for humans (the reason for such a strict threshold in monkeys was the large number of observations). In all monkeys and human subjects, the RTs of the leading hand in bimanual movements were not significantly longer than the RTs of the slower arm in unimanual movements (Fig. 3).

Bimanual RTs were generally between the two unimanual ones or sometimes even faster than both. In two monkeys (P and F) and two human subjects (R and E), bimanual RTs of the subdominant arm were even faster than unimanual ones, indicating that bimanual performance can accelerate reactions of the non-dominant arm. Monkey P and subject E showed also an acceleration of the dominant arm during bimanual movements. We did not find any significant difference between the different movement types. Usage of the online movement onset definition gave similar results, with a tendency of RTs to be even shorter in bimanual movements than in unimanual ones.

Comparing MT and TTP values in bimanual movements to corresponding unimanual movements revealed opposite trends in humans and monkeys (Table 1). In monkeys, bimanual movement durations were generally shorter than unimanual durations. In humans, in contrast, when significant differences existed, bimanual movement times were longer. Similarly, the duration of the accelerating phase of the movement (the TTP value) in monkeys was always shorter during bimanual movements (both hands), but it was longer in five of our human subjects. The difference between monkeys and humans in the relative duration of bimanual movements is the only clear difference between species noted in this study. It may be related to the extensive training of the monkeys who were overtrained in the task.

**Delays between onsets of neural activity and movement initiation**

In the single-unit activity recorded from MI and SMA of the three monkeys, we determined the onsets of evoked activity prior to movement initiation. Figure 4A, B shows examples of neuronal activities and the times of detected activity onsets. The algorithm for detection of activity onsets was based on a cumulative sum of activity, subtracted by the baseline activity during 500 ms before target onset (see “Materials and methods” for details).
This algorithm proved to be very sensitive and reliably detected the very first beginning of increase in neuronal activity. We were interested to relate the onsets of neuronal activities to the reaction times described before (see Fig. 3). Therefore, only onset times below 0 (i.e., onsets starting before movement onset) were taken into account for this analysis. On average, the delays between the onset of neuronal activity and movement onset were ~90 ms shorter than the respective reaction times. As with reaction times, we compared the activity onset times during bimanual movements to unimanual movements in a Mann-Whitney U-test. Since the delays did not differ between the different bimanual or unimanual movement types, we combined the data of all unimanual and all bimanual movement types. Because of the high number of observations, the significance limit could be set to 0.001. Figure 4C displays a box plot of the onset distributions for bimanual and unimanual movements. The box-plot representation of the data (supplied by the “boxplot” function of the Matlab software package) was chosen since the distributions of onset times were clearly non-gaussian. The upper and lower borders of the boxes correspond to the upper and lower quartile of the data, the middle line depicts the median value, and the notches give confidence intervals for the median value. In all three monkeys, and in both SMA and MI, unimanual left movements were associated with a highly significant longer delay between activity onset and movement initiation than bimanual movements \((P<0.001)\). In contrast, the delays associated with unimanual right movements did not differ statistically from bimanual movements at this significance level. However, in some cases (MI of monkeys G and P), unimanual right movements were accompanied by significantly longer delays than bimanual movements at a lower significance level \((P<0.01)\). The results of neuronal onset times thus consistently revealed that bimanual movements were not accompanied by longer neuronal activation than unimanual movements. This finding is also illustrated in the examples shown in Fig. 4A, B.

We also compared the onset times between MI and SMA. For this comparison, all onset times were taken into account (also onsets after movement onset). Although the absolute onset delays tended to be slightly longer in SMA than in MI (monkey G: difference in median: 25 ms; monkey F: difference in median: 5 ms), this difference failed to reach significance (Mann-Whitney test, \(P>0.01\)).

Temporal correlations between the movements of the two arms

To unravel temporal coordination between the arms, we analyzed the temporal relations between the movements of the two arms, and how these relations changed over the time course of the movements. These temporal relations were determined by calculating the cross-correlations
between five different landmarks within the velocity profiles of the movements of both arms. These landmarks were: reaction time (RT), the times elapsed from RT until velocity had risen to 1/3 ($t_1$) and 2/3 ($t_2$) of the peak velocity, the time until the peak velocity was reached ($t_3$), the times until velocity dropped to 2/3 ($t_4$) or 1/3 ($t_5$) of the peak velocity, and, finally, the time until the end of the movement (MT). Figure 5A shows the mean correlations of these landmarks for each subject separately, and the mean over all subjects. In all subjects, correlations decreased over time. Among the monkeys, this effect was least pronounced in monkey F. For the mean over all subjects, we calculated the slope of a regression line fit to it and the significance of the linear regression coefficients ($s$ and $P$ values in the top right corner of each graph). Both slopes were negative and $P$ values of the linear regression were highly significant ($P<0.01$).

In order to test whether the decorrelation was the result of a cumulative effect of errors during the movement, we also correlated the lengths of the time windows between two subsequent time steps of the above analysis [the time differences between RT and the time at which 1/3 of the peak velocity was reached ($w_1$), the time from 1/3 of the peak velocity and 2/3 of the peak velocity ($w_2$), and so on]. Figure 5B shows the result of this analysis. The correlations decreased also in this case, indicating that the decrease was not an accumulating effect, but correlations indeed became markedly weaker during movements. Again, the slope of the linear fit was negative and the linear fit was highly significant ($P<0.01$). Repeating the analysis for all movement types separately did not reveal any consistent relation between the slopes of the regression and the different types of bimanual movements.

Figure 5 also shows that the average level of correlations between the arms was insignificantly higher in monkeys than humans (Mann-Whitney test, $P=0.21$). The negative slopes of the regression lines were very similar. We interpret this to suggest that progressive decorrelation is common to humans and monkeys.

In order to further investigate the decrease in correlation shown in Fig. 5, we analyzed the interarm intervals (IAIs) between RTs, MTs, TATs of the two hands and their variability (Fig. 6A). Interestingly, in all monkeys and most subjects, the mean values of RT, MT and TAT-IAIs were close to zero and not significantly different from each other. However, the standard deviations of the IAIs were close to zero and not significantly different from each other. In contrast, the mean values of RT- and MT- and TAT-IAIs were not significantly different, nor were they significantly different from zero. In B, we show the standard deviations of the IAIs at the temporal landmarks used in Fig. 5. For all subjects, there was a substantial increase in the standard deviations of the IAIs between corresponding landmarks in the two arms’ movements. Like in Fig. 5, a linear regression line was calculated for the means over all subjects, and the $P$ and $s$ values are given.

Fig. 6A, B Interarm intervals (IAI) of RT, MT and TAT in monkeys and humans. A Means (big bars) and standard deviations (thin bars) of RT differences (black bars), MT differences (gray bars) and TAT differences (white bars) between the two arms in bimanual movements. For all monkeys and humans, the standard deviations of RT differences were significantly lower than the MT or TAT differences ($P<0.05$, Wilcoxon signed rank test). In contrast, the mean values of RT-, MT- and TAT-IAIs were not significantly different, nor were they significantly different from zero. In B, we show the standard deviations of the IAIs at the temporal landmarks used in Fig. 5. For all subjects, there was a substantial increase in the standard deviations of the IAIs between corresponding landmarks in the two arms’ movements. Like in Fig. 5, a linear regression line was calculated for the means over all subjects, and the $P$ and $s$ values are given.

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Both slopes were negative and $P$ values of the linear regression were highly significant ($P<0.01$).

In order to test whether the decorrelation was the result of a cumulative effect of errors during the movement, we also correlated the lengths of the time windows between two subsequent time steps of the above analysis [the time differences between RT and the time at which 1/3 of the peak velocity was reached ($w_1$), the time from 1/3 of the peak velocity and 2/3 of the peak velocity ($w_2$), and so on]. Figure 5B shows the result of this analysis. The correlations decreased also in this case, indicating that the decrease was not an accumulating effect, but correlations indeed became markedly weaker during movements. Again, the slope of the linear fit was negative and the linear fit was highly significant ($P<0.01$). Repeating the analysis for all movement types separately did not reveal any consistent relation between the slopes of the regression and the different types of bimanual movements.

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Both slopes were negative and $P$ values of the linear regression were highly significant ($P<0.01$).

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Taken together, the results shown in Figs. 5 and 6 suggest that the decrease in interhand correlations is a result of increased IAI variabilities of corresponding landmarks in the velocity profiles of the two arms.
LFP correlations

Can the behavioral correlations between the arms be explained by neuronal interactions in motor cortical areas? We addressed this question by calculating the time course of correlations between LFP signals (examples of which are shown in Fig. 7A) in MI and SMA, and within and between the hemispheres. We averaged the LFP correlations over longer time windows than those in Fig. 5, separating two windows of 200 ms before movement onset, an acceleration phase (from $M_{on}$ to TTP), a deceleration phase (TTP to $M_{off}$), and 200 ms after $M_{off}$. Since all movement types showed a consistent time course, we averaged over all movement types and pairs (Fig. 7B). In two monkeys (monkeys G and P), and both within and between hemispheres, there was a decrease in correlation over time. A similar decrease was also seen with unimanual movements (not shown). In monkey G, this decrease in correlation was present in both MI and SMA. In one monkey (monkey F), we observed no significant change in correlation, neither in SMA nor in MI, and neither within nor between hemispheres. This monkey, however, showed also only a very mild decrease in correlation of behavior (see Fig. 5, uppermost line in the monkeys’ plot).

Discussion

In this paper, we describe temporal aspects of bimanual coordination both in human and in non-human primates engaged in the same bimanual task. By recording neuronal activities in monkeys during task performance, we could identify consistent relations between the timing of bimanual movements and neuronal activities of areas MI and SMA.

No increased processing time for bimanual as compared to unimanual movements

First, we found that both on the behavioral and on the neurophysiological level, bimanual movements did not require longer processing time than unimanual ones. The reaction times of bimanual movements never exceeded those of unimanual movements of the slower arm. In a
sessions, bimanual movement times were significantly related to the amount of training in the task. In our human subjects, who experienced the task for only three to six months before data collection began, and their bimanual movements were even faster than unimanual ones. This suggests that bimanual movements only need more time than unimanual ones when they are not well trained. One possible explanation for this would be that prolongation of movement times is caused by independent motor errors in the two arms, which have to be corrected by additional small compensatory movements. With increased training, the subjects may learn to make more accurate movements, resulting in smaller deviations of the desired trajectory.

It is noteworthy that we did not find any difference in RTs and MTs of different bimanual movement types, although it is known that symmetric movements are more easily performed than non-symmetric movements. One explanation for this finding could be the fact that the data collection (in both humans and monkeys) was started after differences between the easier, symmetric and more difficult, non-symmetric movements had been reduced by the training processes.

Progressive decorrelation of movements is accompanied by neuronal decorrelation

As a second point, this paper investigates a possible neuronal substrate for the observed dynamics of bimanual coordination. We found that temporal coupling between movements of the arms underwent a continuous decrease during movement execution, with RTs of the two arms being more strongly correlated and less variable than successive temporal landmarks along the velocity profile of each arm. This finding of progressive bimanual decorrelation is consistent with several previous studies employing similar simple bimanual movements (Boesen-kool et al. 1999; Fowler et al. 1991). In more complex bimanual activities, other patterns of temporal coupling have been observed. For example, the task of opening a drawer with one hand, and retrieving a food morsel out of it with the other hand, has revealed the strongest temporal coupling in the middle of the sequence of movements, at the point when the drawer is fully opened and the retrieving hand reaches into it (Kazennikov et al. 1994). Variable patterns of temporal synchronization between the arms seem to account for the specific behavioral demands of the task at hand. In the drawer-opening task, the most critical phase for the success of the operation is the retrieval of the food. In our task, the most critical point for successful performance was to complete both reaching movements quickly and accurately, while the exact timing at the point of target acquisition was not of crucial importance. But why did bilateral movements in our task start in a coupled way and become decoupled over time? Again, the answer to this question seems to be related to the task’s design. For successful performance, both cursors had to be brought simultaneously into the small targets (0.8 cm in diameter). To perform the movements accurately, the subjects and monkeys had to
correct small deviations from the instructed trajectories by corrective movements. This is demonstrated by the asymmetric velocity profiles in our task (Fig. 5). The acceleration phase was fast and more stereotypic, while the following deceleration phase was slower and more variable, caused by the corrective movements. Such asymmetric velocity profiles have been shown to be typical for movements in which the final location must be precisely controlled (MacKenzie et al. 1987; Milner et al. 1990). Thus, the behavioral decorrelation observed may be explained by the fact that bimanual coupling had to be overcome in order to be able to produce independent corrective movements for the two arms.

Assuming that each arm is controlled mainly by the activity in the contralateral cortical hemisphere, we hypothesized that temporal coupling between the arms may be accomplished by correlated activity in the two hemispheres. Confirming this hypothesis, the interhemispheric correlations, like the behavioral correlations between the arms, decreased during movements. Monkey F, who showed almost no behavioral decorrelation, also did not show a significant neuronal decorrelation (Figs. 5, 7). This individual correlation of the time course of neuronal correlations with the time course of bimanual correlations strengthens the argument that the two phenomena are tightly related.

Interestingly, we found progressive decorrelation also within the same hemisphere. This finding can be explained taking into account that there is also a considerable ipsilateral representation in primary motor cortex (Wasserman et al. 1994), and many single cells are active during both ipsilateral and contralateral movements (Tanji et al. 1988; Donchin et al. 1998; Kermadi et al. 1998). Thus, it is possible that neuronal correlations, even within the same hemisphere, can control or reflect the level of coupling between the two arms.

The fact that decorrelation also occurred during unimanual movements seems more difficult to explain. It may be plausible, however, under the above-mentioned hypothesis that behavioral decorrelation is the result of corrective movements. Clearly, corrective movements occur also in unimanual movements, and may cause decorrelations when only one arm moves while the other is stationary.

SMA and MI activities correlate equally well with motor behavior

The neuronal data of both MI and SMA supported our major finding that bimanual movements did not require more neuronal processing time than unimanual movements. This result is in agreement with previous studies showing that MI and SMA seem to process bimanual movements in a similar way (Donchin et al. 1998; Kermadi et al. 1998). The second similarity between MI and SMA was a comparable movement-related decorrelation in both cortical areas. This shows that interhemispheric interactions of both cortical areas may be engaged in controlling temporal coupling of the two arms. It extends previous studies finding a correlation between spatial bimanual coupling and interhemispheric correlations of MI (Cardoso de Oliveira et al. 2001).

In spite of these general similarities, we cannot rule out the possibility that there was a slight difference between the two areas. The onsets of neuronal activity occurred slightly earlier in SMA than in MI (5 ms in monkey F, and 25 ms in monkey G). These differences are well within the range suggested by the conduction delays measured between SMA and MI neurons (around 5 ms, Aizawa and Tanji 1994). However, because of the high variability in the onset values, these differences failed to reach significance. There was a large overlap of onset distributions in MI and SMA. This indicates that although some pairs of neurons in the two areas may be activated consecutively, a large number of MI units are activated before many of the SMA units. This finding is in agreement with previous studies reporting overlapping response onsets of SMA and MI neurons (Chen et al. 1991) and no difference between the time courses of movement-related potentials in MI and SMA (Ikeda et al. 1992; Rektor et al. 1994). While our results as well as these studies are consistent with the notion of a distributed processing in which SMA and MI work in parallel, we cannot rule out the alternative hypothesis of serial processing where SMA is considered a “supramotor” area (Deecke et al. 1985; Goldberg 1985).

In any case, our findings suggest that MI and SMA are equally involved in temporal aspects of the planning and execution of bimanual movements. However, this does not mean that these are the only areas involved in this function. Indeed, bimanual related neuronal activity was also found in other cortical (dorsal premotor, posterior parietal and cingulate cortex, Kermadi et al. 2000) and subcortical (striatum and pallidum of the basal ganglia, Wannier et al. 2002) areas, indicating that the functional network participating in bimanual coordination may be highly distributed.

Conclusion

The fact that bimanual movements did not require more processing time than unimanual ones supports the view that dedicated representations of complex movements exist in the brain. This notion is also consistent with our previous accounts of bimanual related activity in MI and SMA (Donchin et al. 1998, 2001). Furthermore, we extend our previous findings of neuronal dynamics and spatial bimanual coordination (Cardoso de Oliveira et al. 2001) by showing that neuronal correlations are also related to temporal aspects of bimanual coordination. The fact that we found the same temporal characteristics of bimanual coordination in monkeys and humans suggests that the same mechanisms operate in human and non-human primates.
Acknowledgements We thank G. Goelman for obtaining the MRI pictures. Histological evaluation was performed in collaboration with S. Haber (University of Rochester, USA).

References


Abstract We recorded local field potentials (LFP) in primary (MI) and supplementary (SMA) motor areas of rhesus monkey cortex in order to compare movement-evoked potentials (mEP) in bimanual and unimanual movements with single-unit activity recorded concurrently. The mEP was often different during bimanual and unimanual movements (a "bimanual-related" effect), but, unlike the single units, the size of the mEP in both MI and SMA was always greater during bimanual movements than during unimanual movements. This increase primarily reflected an increase in the late positive peak of the mEP, a result that may reflect greater overall cortical activation during bimanual movements. In addition, analysis of the mEP revealed differences between MI and SMA not seen in the single-unit activity. mEP in MI had greater contralateral preference than in SMA. Also, SMA mEP was more correlated to the single-unit activity than in MI. This greater correlation was also more apparent in the late peaks of the mEP than in the early peaks and may reflect a greater influence of recurrent activation in SMA than in MI. Our results further reinforce the idea that unimanual and bimanual movements are represented differently both in MI and in SMA and also show that a complex relationship between spikes of individual neurons and LFP may reflect the different input-output relations of different cortical areas.

Keywords Motor cortex · Supplementary motor area · Frontal cortex · Movement physiology · Bimanual coordination · Single-unit recording · Evoked potentials · Rhesus monkey

Introduction

Single neurons in the proximal arm area of primary motor cortex (MI) and the arm area of supplementary motor cortex (SMA) behave differently during bimanual and unimanual movements (Donchin et al. 1998). This paper analyzes local field potential (LFP) during bimanual movements. LFP is a signal that arises largely as a result of synaptic activity in the area of the recording electrode (Mitzdorf 1994). The relationship between LFP and the activity of individual neurons remains unclear: there is evidence that they are highly correlated (Laas 1968; Kenmochi and Eggermont 1997), but other evidence shows that this correlation can vary over time (Murthy and Fetz 1996a) or depend on context (Eggermont and Mossop 1998), and that the response properties of the LFP and single units may differ (Mitzdorf et al. 1994). In human motor cortex, studies have addressed the complex sequence of evoked EEG potentials preceding movement (Shibasaki 1975; Lang et al. 1990; Cui and Deecke 1999). However, animal research on field potentials in motor cortex has focused on the relationship of synchronous oscillations to movement and to single-unit activity (Eckhorn and Obermueller 1993; Sanes and Donoghue 1993; Murthy and Fetz 1996b; Donoghue et al. 1998; Baker et al. 1999). The character of the evoked potential in this area and its relationship to movement has not been fully explored.

The interpretation of the LFP has been hindered because its source is poorly understood. It is widely accepted that strong negative deflections reflect excitatory, spike-causing input to neurons in the neighborhood of the electrode (Arieli et al. 1995). Current source density analyses of LFP can be used to determine the cortical layers in which synaptic currents are generated, and, in
primary sensory cortices, such analyses have provided an interesting picture of the spatiotemporal events underlying sensory processing. These results allow interpretation of the LFP evoked by sensory stimulation (Mitzdorf 1985, 1987), but it is not clear whether such studies would have relevance for other cortical areas, particularly agranular cortex, or in animals which are actively performing a task. In this study, we present an analysis of the activity evoked in the LFP by movement, analyze the relationship of different components of the LFP signal to a motor task, and compare the activity in the LFP with activity in single units.

Methods

Behavioral paradigm and data acquisition

The task is identical to that described by Donchin et al. (1998). Two female rhesus monkeys (Macaca mulatta; monkey F, 4 kg, and monkey G, 3.5 kg) were trained to operate two separate low-weight, low-friction manipulanda. Each manipulandum was controlled with one arm and restricted to move in the horizontal plane; its motion controlled the motion of a corresponding cursor on a vertically oriented video screen placed in front of the monkey. The monkey was trained to use the manipulanda to perform unimanual movements (of either the right or the left arm) and bimanual movements (using both arms). During unimanual movements, the monkeys were required to keep the non-moving arm still, and, during bimanual movements, the monkeys were required to begin and end movements of both arms simultaneously. All movements were made from central “origin” locations located in front of each of the monkeys shoulders and ended on circles of radius 3 cm around these origins.

Two recording chambers (27×27 mm) were surgically implanted above the left and right hemispheres of the monkeys while they were under general anesthesia, in aseptic conditions. The animals' care and surgery procedures were in accordance with The NIH Guide for the Care and Use of Laboratory Animals (revised 1996) and the Hebrew University regulations. Neural activity was recorded by eight glass-coated tungsten microelectrodes (impedence 0.2–0.8 MΩ, at 1 kHz) from homologous sites in the two hemispheres (four electrodes in each hemisphere). Location of MI and SMA was determined using microstimulation and neural response during passive manipulation of the joints, as well as from the surgical pattern seen during surgery. Interelectrode distance was approximately 500 µm at the dura. However, since the electrodes were individually driven, this distance only reflected the perpendicular dimension, and the interelectrode difference in depth varied from recording session to recording session.

The neural signal recorded on each electrode was amplified and filtered (MCP, Alpha-Omega, Nazareth, Israel) in two different ways to generate two different signals. One bandpass filter (300–8,000 Hz) was used to generate the signal from which we isolated the action potentials of individual neurons, and the analysis of that signal will be reported separately. A second bandpass filter from 1 to 140 Hz was used to generate an LFP signal. This signal was sampled continuously at a rate of 400 Hz using in-house software built around data acquisition boards (DAP 3200e; Microstar Laboratories, Bellevue, Wash.) on a personal computer. Fifty-hertz noise caused by the A/C power supply was removed using a notch filter applied digitally after data collection (48- to 52-Hz, 4-pole Butterworth applied forward and backward to prevent phase shift). There were two different types of recording sessions: those involving two directions of movement and those involving eight directions of movement.

In order to allow pooling of the data, data analysis in this paper is restricted to movements in two directions. That is, for each of the eight-directions sessions, we restricted our analysis to data from two directions. This was done by determining the mean pre-
zero crossing before this maximum to the zero crossing after this maximum was area P1.
3. Similarly, area P2 enclosed the maximum in the range from the end of area N1 to 500 ms after the end of area N1.
4. Area N2 enclosed the minimum found between the end of area P2 and 500 ms after the end of area P2.

The algorithm depends on the fact that there is no DC offset in the LFP signal, so that zero volts is the overall mean of the LFP signal over time. This is indeed the case. Occasionally, any one of these peaks might not be significantly different from the noise; however, the algorithm did not treat such cases differently. For each of these areas we took the square root of the integral of the square (rms) of the mEP enclosed in that area as a measure of the strength of the peak (Eq. 1). We estimated the standard deviation of this value by projecting the signal in each trial onto the mean signal (in the window that defined the peak; Eq. 2) and then taking the standard deviation of these values (Eq. 3):

$$\text{rms} = \sqrt{\sum_{t \text{-window end}} \text{LFP}^2(t)}$$

$$\text{projection}_t = \sqrt{\sum_{t \text{-window start}} (\text{LFP}_t - \text{LFP}(t)) / \text{RMS}}$$

$$\sigma_{\text{rms}} = \sqrt{\frac{1}{N} \sum_{t} (\text{projection}_t - \text{projection}_t)^2}$$

(A bar over a value indicates the averaged quantity of that value across trials.) The overall rms was also calculated in a window extending from 250 ms before movement onset to 700 ms after movement onset, and the standard deviation of projections onto the mean was calculated as with the other areas. Significant differences between two mEPs were detected by t-tests, and the nominal threshold for significance was \(\alpha = 0.001\). We also repeated the analyses using the maximum and minimum values of each area and of the whole signal but, since the results were the same as with the rms values, we do not present them here.

The contralateral preference of the mEP at a recording site was calculated using the formula:

$$\text{Contralateral preference} = \frac{\text{contralateral mEP} - \text{ipsilateral mEP}}{\sigma_{\text{mEP}}}$$

Out of the two unimanual contralateral mEPs performed during the recording of each LFP, we selected the one where the mEP was greatest. Similarly, out of the two unimanual ipsilateral movements, we selected the one which evoked the greater mEP. Thus, this is a comparison of the maximal mEP during a contralateral movement with the maximal mEP during an ipsilateral movement. \(\sigma_{\text{mEP}}\) is the standard deviation combined from the mEP in the two movements. The standard deviations were combined using the standard weighted average:

$$\sigma_{12} = \sqrt{(N_1 - 1) \sigma_1^2 + (N_2 - 1) \sigma_2^2} / (N_1 + N_2 - 1),$$

where \(N_1\) and \(N_2\) are the number of trials over which each standard deviation is calculated. The strength of the “bimanual-related” effect was generated using a very similar formula:

$$\text{Effect Strength} = \frac{\text{bimanual mEP} - \text{unimanual mEP}}{\sigma_{\text{mEP}}}$$

where \(\sigma_{\text{mEP}}\) now calculated using the standard weighted average to combine the unimanual and bimanual standard deviations. This measure was calculated four times (once for each type of bimanual movement), and in each case the mEP was compared with the stronger of the two associated unimanual mEPs. The most significant of these differences, as determined by a t-test on the two responses, was taken to be the strength of the bimanual-related effect. Because four t-tests were performed in generating the final significance value, the actual significance is overestimated. One simple correction that can be used is to multiply the final significance achieved by the number of tests. While this is not an exact correction, it is generally conservative (that is, it underestimates the statistical significance). In our case, the probability was multiplied by 4 to account for the repeated tests. Selection of the maximally significant effect produced bimodal distributions of effect strength. As a result, nonparametric statistical tests were used when analyzing these distributions. We used the binomial test on the signs to test whether the bimanual-related effect was significantly skewed in the positive or negative direction and the Mann-Whitney U-test to compare the distributions. The bimanual-related effect is also presented when calculated separately for each bimanual movement type.

In addition, we calculated the degree of correlation between the mEP recorded by an electrode and the single units recorded by the same electrode. Correlations in this paper are calculated on the averaged activity correlated across recording sites and not, as is more common, on the trial-by-trial activation correlated within a given recording site. This reflects an interest in the task-related characteristics of the LFP and its relation to the task-related characteristics of single-unit activity. We address trial-by-trial correlations in a separate paper (S. Cardoso de Oliveira, O. Donchin, A. Green, and H. Bergman, E. Vaadia, unpublished work).

For the purposes of calculating correlations, the single-unit firing rate was averaged over all trials from activation onset (as determined by a CUSUM algorithm; Davey et al. 1986) through 500 ms after activation onset. Baseline firing rate for a neuron was averaged from 300 ms before activation onset to 50 ms before activation onset. The measure of neural activation used in the correlations was:

$$\text{neural activation} = \frac{\text{abs(activated firing rate} - \text{baseline firing rate)}}{\sigma_{\text{response}}}$$

Similarly, for the LFP, the measure of response was:

$$\text{LFP activation} = \frac{\text{rms}_{\text{peak}}}{\sigma_{\text{peak}}}$$

where peak indicates one of N1, P1, N2, P2, or the overall rms as already described. For correlation of the activation of a neuron with the LFP, we chose either those movements for which the neuron was most responsive or those movements in which the mEP was greatest and performed the analysis on these two possibilities separately. For correlation of the contralateral preference or the bimanual-related effect, we calculated the measures for the single-unit activity in the same manner as they were calculated for the mEP. The strength of these correlations was assessed using Spearman’s r, a nonparametric measure of correlation, and the significance of this statistic was tested using the standard transformation to Student’s t-distribution.

Results

Recording sites

The database included a total of 96 penetrations (usually paired simultaneous recordings at four recording sites in both the left and the right hemisphere), which included 347 recording sites. Of all the recording sites, 117 passed our criteria for continued analysis: 45 recording sites in MI (35 from monkey F and 10 from monkey G) and 72 recording sites in SMA (44 from monkey F and 28 from monkey G).
Shape of the mEP

mEPs were recorded in both MI and SMA in nearly every recording site with a characteristic shape. This characteristic shape can be demonstrated by averaging the mEPs recorded at all different recording sites (Fig. 2). The peaks we characterized in the Methods section can be clearly seen in the means from each recording area in both monkeys. Table 1 shows that peaks N1 and P2 were significant in 80% or more of the mEPs in both SMA and MI, and that peak P1 was less often significant than the others. Table 1 also shows a weak tendency for the mEP to include several (or all) peaks more often than expected by chance. This is an additional indication that the shape of the evoked mEP is preserved across different recording sites.

Evoked potentials during unimanual movements

Figure 3 shows the contralateral preference (Eq. 4) of recording sites in MI and SMA for the overall rms. A dotted line separates those recording sites for which a t-test indicated a significant difference in contralateral and ipsilateral activation of the peak (below the line) from those in which there was no significant difference (above the line). While for many of the recording sites there is not a significant difference between bimanual and unimanual activation, testing the distributions showed that the mean contralateral preference of the overall rms in MI was significantly greater than 0 at \( P < 0.001 \). Figure 4 extends this analysis to the separate peaks in the mEP. The contralateral preference of peak P2 in MI was significantly greater than 0 at \( P < 0.001 \). The mean of the contralateral preference of peaks P1 and N2 in MI was significantly greater than 0 at \( P < 0.01 \). In contrast, in

Table 1  Percentages of significant peaks in movement-evoked local field potential (mEP). The numbers (and percentages) of mEPs in which each peak was significant, and the number of mEPs (and percentage of mEPs) in which two peaks were simultaneously significant. Where the simultaneous occurrence of peaks is shown, the first percentage is the actual value, and the second is the expected percentage under an assumption of independence [expected(peak1, peak2)=actual(peak1) \times actual(peak2)/100]. Note that the actual percentage is always slightly greater than the expectation in the combinations of the individual peaks. The two percentages are equal in combinations of peaks with the overall rms because it was always significant.
SMA, the means of the contralateral preference of peaks N1 and N2 were both less than zero ($P<0.01$ and $P<0.05$, respectively). Thus, MI shows a strong contralateral preference not shared by SMA, which shows a slight preference for the ipsilateral arm.

Evoked potentials during bimanual movements

Figure 5 shows the mEP during unimanual and bimanual movements at a recording site where the difference between bimanual mEP and contralateral mEP is quite small. However, even in this example, the small effect (bimanual-related effect of 0.50; Eq. 5) is significant at $P<0.001$; this is because there is a significant difference between the bimanual activity in row 2 (bimanual parallel movement to $270^\circ$) and the unimanual activity. The value of the bimanual-related effect for each peak is as follows: $P_1$, 0.27; $N_1$, –0.24; $P_1$, 0.82; $N_2$, –0.68. Of these, peak $P_2$ and peak $N_2$ have significant bimanual-related effects at $P<0.001$. Figure 6 shows the LFP recorded at a different site in MI. Here the difference between the mEP during bimanual movements and unimanual movements is more striking. This is particularly evident in bimanual parallel movements to $315^\circ$, where the strength of the bimanual-related effect in the overall rms is 2.60 (broken down by peak: $P_1$, –0.06; $N_1$, 2.08; $P_2$, 2.56; $N_2$, –1.08. All peaks except $P_1$ are significantly bimanual-related at $P<0.001$). A final example taken from SMA is shown in Fig. 7. Here, the strength of the
bimanual-related effect for the overall rms is 1.33 as measured in bimanual parallel movements to 270° (P1, 0.35; N1, 0.90; P2, 1.49; N2, −1.17. All peaks except P1 are significant at \( P < 0.001 \)).

Figure 8 demonstrates that positive bimanual-related effects in the overall rms characterize the population. The figure shows the bimanual-related effect for all recording sites in both MI and SMA. Below the dotted line are recording sites for which the bimanual-related effect was significant. (***\( P < 0.001 \))

Figure 9 shows the bimanual-related effect in the different peaks. A binomial test on the signs shows that peak P1 in the SMA is significantly stronger during unimanual movements (\( P < 0.001 \)), and that peak P2 in both MI (\( P < 0.001 \)) and SMA (\( P < 0.01 \)) is significantly stronger during bimanual movements. Mann-Whitney tests comparing the distribution of the bimanual-related effect in MI and SMA showed significant differences (\( P < 0.01 \)) in peaks P1 and P2.

Figures 8 and 9 do not rule out the possibility that the overall rms of the mEP was larger during some bimanual movements but smaller during others. Figure 10 repeats the analysis of Fig. 8, but includes all four bimanual movements in the analysis rather than selecting the largest effect. The figure clearly indicates that most bimanual mEPs in both MI and SMA were larger than the associated unimanual mEPs (\( P < 0.001 \)). A comparison of the distributions failed to find any significant difference between them.

In sum, for nearly all recording sites, bimanual mEPs are greater than unimanual mEPs, and this increase is caused by an increase in the positive components of the mEP, particularly P2. This result is different from the single-unit result (Donchin et al. 1998; O. Donchin, A.
Gribova, O. Steinberg, H. Bergman, E. Vaadia, unpublished work), where the bimanual-related effect can be either an increase or a decrease in activity during bimanual movements.

Correlation of single-unit activity with LFP

In order to compare the functional relationship between the LFP and single-unit activity, we looked for correlations between the firing rate of single units recorded by an electrode with the size of the mEP recorded by the same electrode. In the Max Cell columns, the firing rate during movements in which the neuron was most responsive was paired with the mEP during that movement. In the Max LFP columns, the movement selected was that in which the LFP was maximal, and the neural activity was taken from that movement also.

<table>
<thead>
<tr>
<th>Peak</th>
<th>MI Max Cell w/LFP</th>
<th>MI Max LFP w/Cell</th>
<th>SMA Max Cell w/LFP</th>
<th>SMA Max LFP w/Cell</th>
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<tbody>
<tr>
<td>P1</td>
<td>0.09</td>
<td>-0.05</td>
<td>0.01</td>
<td>-0.17</td>
</tr>
<tr>
<td>N1</td>
<td>0.14</td>
<td>0.04</td>
<td>0.29*</td>
<td>0.17</td>
</tr>
<tr>
<td>P2</td>
<td>-0.02</td>
<td>0.02</td>
<td>0.24*</td>
<td>0.38***</td>
</tr>
<tr>
<td>N2</td>
<td>-0.01</td>
<td>0.16</td>
<td>0.20</td>
<td>0.33***</td>
</tr>
<tr>
<td>Overall rms</td>
<td>-0.03</td>
<td>0.24*</td>
<td>0.30*</td>
<td>0.37***</td>
</tr>
</tbody>
</table>

Numbers shown are Spearman’s r.
* Significance at P<0.05;
** significance at P<0.001

Table 2 Correlation between maximal single-unit response and maximal mEP. The correlation of the response of single units recorded by an electrode with the size of the mEP recorded by the same electrode. In the Max Cell columns, the firing rate during movements in which the neuron was most responsive was paired with the mEP during that movement. In the Max LFP columns, the movement selected was that in which the LFP was maximal, and the neural activity was taken from that movement also.

Fig. 11 Correlation of contralateral preference in LFP and single units. This figure demonstrates that contralateral preference in LFP and units in SMA are more strongly related than in MI. Only those peaks which were significant in SMA (P<0.05) and the overall rms are shown. Numbers in each plot are Spearman’s r. (*P<0.05)

Fig. 12 Correlation of bimanual-related effect in single-unit activity and LFP. Format is similar to Fig. 11, but here the absolute value of the bimanual-related effect is compared rather than contralateral preference. Only those peaks with a significant correlation in either MI or SMA are shown. (*P<0.05)

SMA are less clear. For peaks P1 and N2, Spearman’s r is weakly significant in SMA (P<0.05). In fact, all of the mEP peaks in SMA showed a (positive or negative) correlation of contralateral preference with the single units with |r|>0.1, while for MI all of the peaks had |r|<0.1.

We also compared the strength of bimanual-related activation in the mEP and simultaneously recorded single units. Bimanual-related effects can be positive or negative (in single units they are often both, while in the mEP they are always positive) so we repeated the analysis both on the signed and on the absolute values of the effect. The analysis of the signed effect produced lower correlations (not shown) than the analysis of the absolute values (Fig. 12). The correlation of the bimanual-related effect...
effect in the N2 peak of the mEP with the bimanual-related effect of neurons recorded by the same electrode is highly significant in SMA and weakly significant in MI. The bimanual-related effect in the overall rms in SMA is also weakly correlated with the bimanual-related effect in the neurons. Here, as in the other correlation analyses, the late mEP in SMA is more strongly correlated with the single-unit activity than it is in MI.

Discussion

Bimanual-related effect always positive in the mEP

This paper analyzes the LFP, a mean of electrical fields from the vicinity of the electrode. We find that a bimanual-related effect exists in the LFP, as it does in the single-unit activity (Donchin et al. 1998). The bimanual-related effect in the LFP is different from the effect in the single units. In the LFP, activity during bimanual movements (as measured by the overall rms of the mEP) is always greater than activity during unimanual movements; whereas, in the single units, the bimanual-related effect was often a decrease in bimanual activation as it was an increase (O. Donchin, A. Gribova, O. Steinberg, H. Bergman, E. Vaadia, unpublished work). The unidirectional nature of the bimanual-related effect in the LFP supports the hypothesis that the motor cortices represent bimanual movements differently from unimanual movement (Donchin et al. 1998). Apparently, bimanual movements require neuronal control beyond simultaneous production of two unimanual control signals. However, while providing support to the hypothesis above, the result raises its own questions. Is there any physiological explanation for the increased LFP activation during bimanual movements? Is there any functional significance for the result?

There are four (not mutually exclusive) possibilities that offer an immediate explanation for the increased mEP during bimanual movements:

1. Neurons are more active in the area of the electrode.
2. Those neurons active in the area of the electrode are better aligned.
3. More neurons are active far from the electrode with efferent connection to the area of the electrode.
4. The synaptic activity in the area of the electrode is more synchronized.

The first possibility can be rejected because the number of active single units and the mean firing rate do not increase in either MI and SMA during bimanual movements (O. Donchin, A. Gribova, O. Steinberg, H. Bergman, E. Vaadia, unpublished work). The second possibility would be difficult to prove or disprove, and may warrant further investigation. Specifically, if pyramidal cells tend to show increased activity during bimanual movements, while interneurons tend to show decreased activity, then the greater anatomical alignment of pyramidal dendrites might lead to an overall positive effect on the mEP.

### Table 3

<table>
<thead>
<tr>
<th></th>
<th>Unimanual left</th>
<th>Unimanual right</th>
<th>Bimanual</th>
</tr>
</thead>
<tbody>
<tr>
<td>Left hemisphere</td>
<td>7.3</td>
<td>8.2</td>
<td>8.2</td>
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<td>Right hemisphere</td>
<td>8.0</td>
<td>7.7</td>
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</tbody>
</table>

The fourth possibility is particularly intriguing in light of recent disagreements about the functional significance of neural synchronization. Work on synchronization of LFP oscillations has shown a relationship between synchronized oscillations in the LFP and synchrony in single-unit activity (Murthy and Fetz 1996b), but research which specifically studied bimanual movements did not find increased LFP synchrony during bimanual movements (Murthy and Fetz 1996a). However, this negative finding is inconclusive because these studies analyzed periods of LFP synchrony rather than evoked potentials, and it is still possible that increased neuronal synchrony would correlate well with increases in mEP. Studies of unimanual movements suggest that synchronized activity in the oscillatory components of the LFP is decreased during movements (Sanes and Donoghue 1993; Donoghue et al. 1998) and that LFP oscillation is phase-locked to the single-unit activity (Baker et al. 1999), results which suggest that there may be no functional role for synchrony during movements. However, it is feasible that, specifically during bimanual movements, synchrony does have such a functional role.

The second of the four possibilities listed above is not implausible. While for any particular neuron maximal bimanual activation may be less than maximal unimanual activation, it is still possible that the sum of bimanual activation across both hemispheres is more than the sum of unimanual activation. For instance, neurons in left cortex may be more active during movements of the right arm, while neurons in right cortex are more active during movements of the left arm, but during bimanual movements both sets of neurons are active (see Table 3). Since MI and SMA receive input from both contralateral and ipsilateral cortex, the amount of input each cortical area receives may be greater during bimanual movements than during unimanual movements. A group investigating the neuronal response as a function of stimulus size in visual cortex found a similar result: induced oscillations in LFP increase with increased stimulus size while single-unit discharge rates may increase or decrease (Bauer et al. 1995). Further information regarding this
of recurrent local collaterals. Hypothesis is that SMA, often thought to be involved in the area is to events outside that area. Thus, an appealing nonlocal input to an area might reflect how responsive understood, it seems reasonable that the ratio of local to tomical differences between cortical areas is not well un-

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be stronger in SMA than in MI is a question open to

mEP at that site (Table 2). Why this correlation should at a recording site and the magnitude of the late peaks in the correlation between the magnitude of neural response activation of the neurons receives additional support from the correlation between the magnitude of neural response at a recording site and the magnitude of the late peaks in mEP at that site (Table 2). Why this correlation should be stronger in SMA than in MI is a question open to speculation, although one simple hypothesis is that a larger percentage of neurons have recurrent local collaterals in SMA. While the functional significance of anatomical differences between cortical areas is not well understood, it seems reasonable that the ratio of local to nonlocal input to an area might reflect how responsive the area is to events outside that area. Thus, an appealing hypothesis is that SMA, often thought to be involved in self-generated movements, would have a high proportion of recurrent local collaterals.

Strong contralateral preference of the mEP in MI

We found that the contralateral preference of LFP from recording sites in MI was much greater than the con-

lateral preference of single units recorded in the same loc-

ations and in the same task. In the single-unit results, contralateral preference in MI was only slightly greater than the contralateral preference in SMA (O. Donchin, A. Gribova, O. Steinberg, H. Bergman, E. Vaadia, unpublished work). In contrast, in the LFP, contralateral preference in MI was quite strong, while in SMA a bilateral activation with slight ipsilateral preference was found (Fig. 3). This difference between single-unit activity and the mEP was highlighted by a relative lack of correlation between the degree of contralateral preference of the mEP and the contralateral preference of the neurons recorded at that site (Fig. 11), particularly in MI.

The greater contralateral preference of the LFP in MI relative to the single units is consistent with findings in auditory cortex showing greater contralateral preference in LFP than in single-unit activity (Eggermont and Mossop 1998). In our study, this increased contralateral preference is more evident in the late peaks of the LFP than in the early peaks (Fig. 4). In sensory cortices, the wider, late peaks in evoked responses have been seen to result from recurrent collaterals within a cortical area (Mitzdorf 1985). This suggests that those neurons forming significant local connections may have greater contralateral preference than other neurons.

Magnitude of single-unit response correlated with mEP magnitude in SMA

The suggestion that the late peaks in the mEP reflect re-
current local activity following the movement-related ac-
tivation of the neurons receives additional support from the correlation between the magnitude of neural response at a recording site and the magnitude of the late peaks in mEP at that site (Table 2). Why this correlation should be stronger in SMA than in MI is a question open to speculation, although one simple hypothesis is that a larger percentage of neurons have recurrent local collaterals in SMA. While the functional significance of anatomical differences between cortical areas is not well understood, it seems reasonable that the ratio of local to nonlocal input to an area might reflect how responsive the area is to events outside that area. Thus, an appealing hypothesis is that SMA, often thought to be involved in self-generated movements, would have a high proportion of recurrent local collaterals.

The relatively weak correlations between the neuronal response and mEP contrast with reports showing that single-unit activity can be quite tightly related to the LFP signal. Spike-triggered averaging has shown that spikes tend to occur preferentially during negative deflections of the LFP (Eckhorn and Obermueller 1993). On the other hand, other work has shown that the relationship between LFP and single-unit activity can be quite complex (Mitzdorf 1994; Murthy and Fetz 1996b; Eggermont and Mossop 1998). In our analysis, it was the late peaks (P2 and N2), and not the sharp negative deflection of peak N1, which were correlated with the neuronal activity. This probably reflects a difference in the correlation being measured. Usually, correlations are measured in the trial-by-trial variations in single-unit activity and LFP. This study focuses instead on correlations between average evoked potentials. This correlation had the advantage here of addressing directly the results on mean evoked potentials reported in this paper and in our previous work (Donchin et al. 1998). Trial-by-trial correlations will be addressed extensively elsewhere (S. Cardoso de Oliveira, O. Donchin, A. Gribova, H. Bergman, E. Vaadia, unpublished work).

Correlation of bimanual-related effect in neurons and mEP in SMA

A strong correlation was seen between the bimanual-related effect in the single-unit activity and the bimanual-related effect in peak N2 of the LFP, and a similar, but weaker, correlation was seen in the overall rms in SMA. As already discussed, the overall rms of the mEP was always greater during bimanual movements. To a large degree, the increase in overall rms during bimanual movements was the result of an increase in the rms of peak P2 (Fig. 9). The difference between the functional significance of peak P2 and peak N2 is difficult to guess, because no current source density analysis of the mEP in motor cortices exists in the literature. However, peak N2 seems to reflect the bimanual character of the neural activity more directly, while peak P2 may represent a different aspect of cortical processing of bimanual control.

Conclusions

Many questions remain regarding interpretation of the mEP, but it seems clear that understanding of cortical processing can be aided by examining this signal in addition to the single-unit activity and the oscillatory components of the LFP. Specifically, our results are consistent with an interpretation that understands early components of the LFP to reflect the input to a cortical area, and late components of the mEP to reflect recurrent synaptic activation. They show differences in the role of MI and SMA in controlling movement, and differences in the way that bimanual and unimanual movements are controlled. In this last sense, the results support the hypothe-
sis (Donchin et al. 1998; O. Donchin, A. Gribova, O. Steinberg, H. Bergman, E. Vaadia, unpublished work) which holds that bimanual movements have specific neuronal representations and are not generated by simple combination of two unimanual movements, and this fact is reflected in the activity of the cortical networks which produce the movements.

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References


Neuronal populations in primary motor cortex encode bimanual arm movements

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Keywords: bimanual coordination, electrophysiology, Macaca mulatta, population vector, single-cell recording

Abstract
Previous studies have shown that activity of neuronal populations in the primary motor cortex (MI), processed by the population vector method, faithfully predicts upcoming movements. In our previous studies we found that single neurons responded differently during movements of one arm vs. combined movements of the two arms. It was, therefore, not clear whether the population vector approach could produce reliable movement predictions also for bimanual movements. This study tests this question by comparing the predictive quality of population vectors for unimanual and bimanual arm movements. We designed a bimanual motor task that requires coordinated movements of the two arms, in which each arm may move in eight directions, and recorded single unit activity in the MI of two rhesus (Macaca mulatta) monkeys during the performance of unimanual and bimanual arm movements. We analysed the activity of 212 MI cells from both hemispheres and found that, despite bimanual related activity, the directional tuning and preferred directions of most cells were preserved in unimanual and bimanual movements. We demonstrate that population vectors, constructed from the activity of MI cells, predict accurately the direction of movement both for unimanual and for bimanual movements even when the two arms move simultaneously in different directions.

Introduction
Natural voluntary movements require coordination among limbs, joints and muscles. A prevailing approach in experimental motor research was isolation of a single movement parameter, by studying simplified movements (for example – movements around a single joint). While this approach proved fruitful for studying the gross organization of motor cortex, it does not provide insight into the emergence of more complex movements.

Simultaneous movements of the two arms constitute a relatively simple example of complex movements and may serve to test whether and how the brain generates unique representations of complex movements from their constituent elements, as suggested by Leyton & Sherrington, (1917) ‘... the motor cortex may be regarded as a synthetic organ for compounding [...] movements [...] from fractional movements’.

Very little is known about cortical involvement in bimanual coordination. Studies by Tanji and coworkers suggested that, except for a small zone, the MI is not involved substantially in bilateral, distal movements (Tanji et al., 1988; Aizawa et al., 1990). Another study also failed to find bimanual specific activity in MI when more proximal movements were tested (Kazennikov et al., 1999). In contrast, two groups, using different tasks (Kermadi et al., 1998; Donchin et al., 1998) reported strong bimanual related effects for MI neurons. In the latter, we demonstrated that for many of MI cells, ‘bimanual related activity’ was relatively insensitive to small differences in muscular activity and arm kinematics between bimanual movements and unimanual movements and concluded that neural activity in the MI, as well as SMA, can reflect specialized cortical processing associated with bimanual arm movements (Donchin, 1998).

The finding that the activity of MI cells does not depend only on the limb’s movement per se but also on the larger context (such as whether the arm moves together with the other arm) is consistent with other recent findings suggesting that the MI is not only responsible for muscle activation but may be involved in ‘higher’ aspects of motor control (Georgopoulos et al., 1989; Porro et al., 1996; Zhang et al., 1997; Carpenter et al., 1999; Kakei et al., 1999).

However, bimanual related activity in the MI seems inconsistent with the notion that single cells in the MI are characterized by symmetric directional tuning around a ‘preferred direction’ and, thus, that a population of MI cells can represent the direction of upcoming movements (Schwartz et al., 1988; Georgopoulos et al., 1986; Kalaska et al., 1989; Caminiti et al., 1990a; Caminiti et al., 1990b; Schwartz, 1993). If the preferred direction of a neuron is not consistent in different contexts, the whole concept of a ‘population vector’ (PV) may lose its validity. Additionally, one may ask whether the PV method is appropriate for representing multiple directions simultaneously, as is required for execution of bimanual movements. In this study we show that the preferred directions of single cells are relatively well preserved across different movement types, and thus, PV analysis is a valid tool for describing bimanual movements, including simultaneous representation of the directions of the two arms. Preliminary reports of this work have been published previously (Steinberg et al., 1998; Donchin et al., 1999).
Methods

Monkeys

Two female rhesus (Macaca mulatta) monkeys, G and P (weighing 4–4.5 kg each), were used in the experiments. The monkeys were kept in the animal facilities of the faculty of medicine. The animals’ care and surgical procedures used were in accordance with The NIH Guide for the Care & Use of Laboratory Animals and the Hebrew University regulations. Unless mentioned specifically, all details of the behavioural task, surgical procedures, and recordings are identical to and can be found in Donchin et al., (1998).

Behavioural task and training

The two monkeys were trained to move two separate manipulanda, one with each arm. A trial began when the monkey aligned both cursors on ‘origins’ and held them still for 500 ms. For each arm, one of eight peripheral target circles (0.8 cm diameter) could appear at a distance of 3 cm from the origin. The movement of each cursor was mapped to its corresponding manipulandum movement such that each millimeter of manipulandum movement caused one millimeter of movement of the cursor on the video display. The direction of movements from the origin to the right was defined as 0°, and movements away from the monkey (upward motion of the cursor) as 90°. There were four main types of trials (Fig. 1). In unimanual trials (Fig. 1A and B), only one target appeared and the monkey moved the appropriate arm and had to keep the other still. In bimanual trials (Fig. 1C and D), two targets appeared and the monkey moved each arm to the corresponding targets. As in Donchin et al. (1998), there were only two classes of bimanual movements that were tested: bimanual parallel (Fig. 1C, where the arms moved in the same direction) and bimanual opposite (Fig. 1D, where the directions of movement of the two arms differed by 180°). In contrast to Donchin et al. (1998), we recorded activity during performance of unimanual and bimanual movements in all eight directions, in all sessions.

Surgery

A head holder and two 27 × 27 mm recording chambers were fixed onto the skull under general anaesthesia [induced by ketamine (10-15 mg/kg) and sustained with isoflurane] to allow recordings from the primary motor areas of both hemispheres. In Monkey P, the implants were made of plastic to allow magnetic resonance imaging.

Neural recordings

Neural activity was recorded simultaneously by eight glass coated tungsten microelectrodes. Spike detection and online sorting was aided by MSD® (Alpha-Omega, Nazareth, Israel) spike sorters. All data, including spike shapes, were stored for off-line analysis.

Experimental procedures

Recording sessions started after full recovery from surgery (2–5 days later). In each session, two sets of four electrodes were inserted into the primary motor areas (MI) of the two hemispheres (one set into each hemisphere). The depth of each electrode was individually controlled and monitored by EPS® (Alpha-Omega). The recording area was selected on the basis of mapping sessions where we examined the effects of intracortical microstimulation (ICMS) and the neuronal activity evoked by passive manipulations of the monkeys’ limbs. In addition, at the end of each recording session, we also tested ICMS effects and responses to passive limb manipulation from each of the eight electrodes. After completion of data collection, monkey G was killed by injection of ketamine (13 mg/kg) followed by nembutal (100 mg/kg) and its brain was removed, dissected, and analysed histologically. Monkey P is still participating in experiments, and anatomical confirmation of the recording sites was only possible using MRI imaging.

Data analysis

Tuning and preferred direction

For most cells, the directional tuning curve can be approximated by a cosine function, although the method probably overestimates tuning width (Amirikian & Georgopoulos, 2000). We continued to use the cosine approximation in order to allow comparison of our results with the previous studies using population vectors to predict the direction of unimanual movements. The method we used to quantify the cells directional tuning and their ‘preferred directions’ (PD, the direction of movement to which the cell has the strongest response) was similar to the one used by Georgopoulos et al., (1982). We used the coefficient of determination, $R^2$, to quantify the fit of the neurons’ directional tuning to a cosine function, and defined all cells with an $R^2$ above 0.7 as ‘directionally tuned’ and the others as ‘nontuned’. This $R^2$ threshold was selected to facilitate comparison to previous PD studies (e.g., Georgopoulos et al., 1982; Schwartz, 1992).

We were interested in the degree of similarity of PDs in different types of movement, therefore, we compared PDs in ipsilateral unimanual, bimanual parallel and bimanual opposite movements to the PDs in contralateral unimanual movements. We examined whether the distributions of the differences in PDs were spaced equally using the Rao test (Mardia, 1972).

Measuring directional tuning and PDs under different behavioural conditions

A few possible methods could be used to quantify a single PD for each cell in different types of movement. In principle, we could have calculated four different preferred directions from each type of movement and calculate a simple average of the four measures to get...
one measure for each cell. However, such a method wouldn’t have
taken into account the magnitude of response in the different
movement conditions. Therefore, we used the following technique to
calculate a single estimate of a PD for each cell: we calculated five
different PDs for each cell. The first four were taken from the
neurons’ activity during execution of the four different types of
movement (unimanual right, unimanual left, bimanual parallel, and
bimanual opposite). The fifth PD was computed by fitting one PD for
each cell, based on its activity during performance of all types of
movement. It was called the ‘best-fit PD’ (BFPD). To compute it, we
used a least-squares fit that finds the best parameter set to fit the actual
data recorded, with the restriction that the PD ($\theta_0$) is constant for all
types of movement (constr function, optimization toolbox,
MATLAB, Mathworks Inc.). The tuning function to which we fitted
the data is thus:

$$y_T(\theta) = a_T + c_T + \cos(\theta - \theta_0)$$

(1)

where $y_T(\theta)$ is the firing rate of the cell in a certain type of
movement ($T$) in direction $\theta$, $a_T$ and $c_T$ are the parameters of
the cosine function (specific for the movement type $T$) and $\theta_0$ is the
preferred direction, which is assumed to be the same in all four
types of movement. The movement direction $\theta$ in bimanual
opposite trials was assigned according to the arm yielding
stronger activation for that cell. This was determined according to
the amplitude of the fitted cosine. For example, if a cell
responded more strongly to unimanual movements of the left arm,
then the movement direction was determined according to the
direction of movement of the left arm regardless of the
hemisphere the cell was recorded from. However, in some
analyses (specified below), the movement direction in bimanual
opposite was assigned according to the contralateral arm. This
decision was made to ensure that all cells were included in the
analysis of bimanual opposite movements.

In order to check that the tuning characteristics do not result
from random variation in the firing rates, we tested whether the
distribution of $R^2$ for the cosines around BFPDs, could be
obtained by chance. Therefore, the distribution of the original $R^2$
values was compared to a distribution of $R^2$ values that were
created by calculating four cosines, based on the recorded data,
with noise equal to the noise in the original data, but with
random preferred directions. To do that we performed the
following calculations: (i) the values of the cosine function fitted
to the data were subtracted from the eight points of data, for each
type of movement. (ii) These residuals were randomly ordered.
(iii) A PD was randomly chosen, and a cosine function was
calculated from this random PD and from the amplitude and
offset of the original cosine. (iv) The randomly ordered residuals
were added to this new cosine. This procedure was repeated for
data from the four types of movement. Then, four cosine
functions, with the same preferred direction in all four of them,
were fitted to the new 32 points. An $R^2$ value was calculated to
provide an estimate for the goodness of fit of all points (total 32)
to the four cosine curves. The Kolmogorov–Smirnov two-sample
test was used to determine whether the distributions of the real
and randomized $R^2$ values could have been drawn from the same
population.

Bimanual related effect

The definition of the ‘bimanual related effect’ is shown in equation 2,
where ‘bimanual’ and ‘unimanual’ represent the cell’s firing rates in a
selected bimanual or unimanual movement, respectively.

$$Bimanual related effect = \frac{\text{bimanual} - \text{unimanual}}{\text{bimanual} + \text{unimanual}}$$

(2)

To select the movements for comparison, we first chose the
direction of movement closest to the BFPD of the cell and the
direction 180° away from it. We compared each bimanual movement,
each of those directions (four comparisons), to the stronger
response in the two unimanual movements composing it (as in
Donchin et al., 1998). Then, we used the comparison that gave the
largest difference to calculate the bimanual related effect. To evaluate
the correlation between the bimanual related effect and the $R^2$ values
we used the Spearman rank correlation coefficient.

Population vectors

Population vectors have been calculated in the customary fashion
(Georgopoulos et al., 1986). In this study, we extracted three different
types of PV values from the activity of the neurons: (i) for each
movement type, the PV was constructed using the PD determined in
that specific movement type. (ii) For all kinds of movements, the PD
of contralateral unimanual movements was used, and (iii) a common
PD was determined by the BFPD and used for all types of
movements.

The error between the PV prediction and the actual movement
direction was calculated as the mean of errors in all eight directions.
A bootstrap analysis was used to estimate the confidence interval of
differences in the average error for the different methods of
calculating population vectors. In this analysis we calculated PV
values using random weighting functions. Each such weighting
function was a number randomly chosen from a distribution with a
mean and standard deviation equal to those of the real weighting
function. We repeated this 200 times, and each time we calculated the
mean error of the direction of the resulting population vectors across
the eight directions. This gave us a population of 200 averaged errors,
from which we estimated the standard error of the mean.

Calculating population vectors for simultaneous movements of two arms

In order to arrive at two PV values, one for each arm, we divided the
cells into two subpopulations using two approaches. First, we
followed the classical hypothesis that each hemisphere controls the
contralateral arm, and divided the cells by the hemisphere in which
they resided. Second, cells were assigned to a subpopulation
controlling the arm for which the modulation depth of the unimanual
tuning-curve was larger (preferred arm, PA) regardless of its
anatomical location. The population vectors were calculated in both
cases using the BFPD. In calculating population vectors from
subpopulations selected according to hemisphere, we used BFPDs in
which the direction of bimanual opposite movements was assigned
according to the contralateral arm. For population vectors based on
subpopulations divided according to modulation depth in unimanual
movement, we used BFPDs in which the direction of bimanual
opposite movements was assigned according to the stronger activity
in unimanual movements (the PA).

Length of the population vectors

The length of the population vector is always equal to or larger than
zero. Small PV values may reflect a slow or small movement in a
given direction, or they may be the outcome of ‘random’ directionless
population activity. Therefore, it is important to estimate if a given
vector’s length is significantly different from one resulting from
TABLE 1. The number and percentage of cells tuned to different types of movement

<table>
<thead>
<tr>
<th>Movement type</th>
<th>Cells (n)</th>
<th>Cells (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unimanual, ipsilateral</td>
<td>54</td>
<td>34</td>
</tr>
<tr>
<td>Unimanual, contralateral</td>
<td>86</td>
<td>55</td>
</tr>
<tr>
<td>Bimanual, parallel</td>
<td>94</td>
<td>60</td>
</tr>
<tr>
<td>Bimanual, opposite</td>
<td>76</td>
<td>48</td>
</tr>
</tbody>
</table>

The groups are not mutually exclusive. The total number of cells tuned for at least one movement type is 156. The \( R^2 \) threshold for significance is 0.7.

TABLE 2. The numbers and percentages of cells tuned in different combinations of types of movement.

<table>
<thead>
<tr>
<th>Number of types of movement</th>
<th>Cells (n)</th>
<th>Cells (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 (nontuned)</td>
<td>56</td>
<td>26</td>
</tr>
<tr>
<td>1 type</td>
<td>64</td>
<td>30</td>
</tr>
<tr>
<td>2 types</td>
<td>44</td>
<td>21</td>
</tr>
<tr>
<td>3 types</td>
<td>34</td>
<td>16</td>
</tr>
<tr>
<td>4 types</td>
<td>14</td>
<td>7</td>
</tr>
<tr>
<td>Total cells</td>
<td>212</td>
<td>100</td>
</tr>
</tbody>
</table>

Note that only 26% of the cells are not tuned to any type of movement. Forty-four percent of all cells (which constitute 59% of the 156 tuned cells) are tuned to more than one type of movement.

![Fig. 2. An example of a cell with tuned responses in different types of movement. The figure depicts activity of one cell from the left MI during performance of four different types of movement. Each quadrant of the figure shows the activity of the cell in one type of movement, in eight directions. The rasters are aligned around movement onset (cyan line, time 0) in a time window of 750 ms before movement onset till 1000 ms after it. The red arrows indicate the preferred direction. Their lengths are proportional to the \( R^2 \) of the cosine fit. The cosine fit of this cell with its \( R^2 \) values and directional indices are shown in Fig. 4.](image-url)
random activity. To do this, we calculated the distribution of PV values that would have resulted from a population for which the weighting functions and the PDs are independent, and checked whether the real PV length was significantly different from the expected from this distribution.

Results

Database

Neural activity was recorded from the MI in 13 sessions in monkey G, and 12 sessions in monkey P. Overall, we recorded the activity of 415 cells (207 in monkey G and 208 in monkey P). The activity of each cell was evaluated from the recording period in which its firing rate was stationary. A number of additional criteria were used to include cells in the database: the anatomical location of the cell (see Methods), the quality of isolation, the number of successful trials performed during its recording, and the total number of spikes fired. Two hundred and twelve cells fulfilled all these criteria (135 in monkey G, 66 in the left hemisphere and 69 in the right hemisphere, and 77 in monkey P, 37 in the left hemisphere and 40 in the right hemisphere).

Directional tuning in unimanual and bimanual movements

Cells are directionally tuned in different types of movement

As is expected from repeated reports of the arm area in the MI, most of the cells in our sample (156/212) exhibited broad symmetrical directional tuning around a preferred direction for at least one movement type. Table 1 shows the percentages of tuned cells in different types of movement, and indicates that there are groups of cells in the MI, which contain directional information not only about the contralateral movements, but also about the direction of bimanual movements and unimanual movements of the ipsilateral arm. Few cells (7% of the sample, Table 2) were tuned significantly to all four types of movement. An example is shown in Fig. 2. However, a significant portion of the cells was tuned to more than one type of movement (Table 2).

Preferred direction in different types of movements are similar

The finding that a single neuron may be tuned to more than one type of movement calls for comparisons of PDs in different movement conditions being compared were included in the analyses. N, number of cells included in each analysis.

FIG. 3. Histograms of differences in PD values: (A) bimanual parallel vs. contralateral; (B) bimanual opposite vs. contralateral; (C) ipsilateral vs. contralateral. Only cells in which the $R^2$ was above 0.7 in both movement conditions being compared were included in the analyses. N, number of cells included in each analysis.

FIG. 4. (A) An example of best-fit preferred direction (BFPD) showing four cosines that fit to the activity of one cell (same cell as in Fig. 2) for four movement types, with the restriction that the preferred directions (peaks of all cosines) are the same. The BFPD for this cell was $4.5^\circ$ (marked by a vertical arrow below the horizontal axis). The polar plot at the top-left corner shows the four preferred directions calculated separately for each type of movement (coloured arrows) and the BFPD (black arrow). For this particular cell all five measures are close to $0^\circ$, namely the cell is best tuned to arm movements to the right side. The $R^2$ values and directional index are also given in the figure. The directional index ($DI = A/b_0$, where $b_0$ is the average firing rate across all movement types and $A$ is the cosine’s amplitude, given by $A = \sqrt{b_1^2 + b_2^2}$ (for details, see Georgopoulos et al., 1982). (B) $R^2$ value distribution validates BFPD. The figure shows histograms of $R^2$ values of the fit of BFPD for all cells (filled blue bars), and $R^2$ values calculated from randomly ordered data (red line).
types. One possibility is that the cell’s PD is fixed. If so, it could be represented either by the value measured during contralateral movements, or on the basis of the cell’s activation during all types of movement. Another possibility is that the PD is not fixed and for each movement type the cell may ‘prefer’ a different direction. To begin studying these possibilities we tested whether the PDs measured during performance of different types are similar. The results are summarized in Fig. 3, where the distributions of differences of PDs (contralateral vs. bimanual parallel, contralateral vs. bimanual opposite, and contralateral vs. ipsilateral) are shown. In this analysis, we included only cells for which the $R^2$ values for the two compared types of movement were above 0.7. All distributions were proven statistically to be nonuniform ($P < 0.01$, Rao test). The figure demonstrates that PDs in bimanual parallel movements had the greatest similarity to the PDs in the contralateral unimanual movements, while PDs in ipsilateral movements showed larger deviations.

We also tested the hypothesis that the PDs for the ipsilateral arm movement match the mirror-symmetric PD for movements of the contra-lateral arm. For example, if the PD for the contralateral arm is 0°, the expected matched PD for the ipsilateral arm is 180°, while for contralateral PD of 90° the expected ipsilateral PD is 90° as well. To perform that, we calculated the difference between the PD in ipsilateral movements and the ‘mirror’ of the PD of contralateral movements. The PD differences were much larger than those seen in Fig. 3C, therefore, we could not find support for the ‘mirror’ hypothesis in our data.

Can a cell be represented by one preferred direction?

As the PD of a single-cell in different types of movement were similar, but not identical, the next step was to estimate one PD for each cell based on its activity in different movement types. The algorithm for generating a unique PD (the BFPD) is described in the Methods section. Figure 4A shows the estimated BFPD of one cell. As the figure shows, this cell was tuned to all four types of movement, with quite similar PDs, and its BFPD was 4.5° (black arrow). Note that the BFPD is indeed close to each of the four different PDs of this cell. Also note that the tuning curve for the contralateral arm (in red) seems to show better directionality tuning as compared to the tuning for the ipsilateral arm (in purple). However, computing a directionality index (see Georgopoulos et al., 1982) to each of the curves indicate that the modulation depth is similar in all conditions (for details see figure legend). This result is explained by the fact that the average firing rate across movements in all directions is highest for unimanual movements of the contralateral arm and lowest for movements of ipsilateral arm.

Figure 4B shows the distribution of $R^2$ values of the BFPDs for the whole population of cells (blue filled histogram) and the distribution of $R^2$ values calculated from the fit of a cosine model to randomized data (red line histogram). Comparing the two histograms shows that the distribution of $R^2$ of the cells is markedly shifted to the right (higher values) relative to the $R^2$ of bootstrapped data. The Kolmogorov–Smirnov two-sample test revealed that it is improbable that the distributions of the $R^2$ values calculated from the data and the distribution of the randomized $R^2$ values were drawn from the same population ($P < 0.05$). Half of the cells had BFPD $R^2$ values above 0.7 (107/212).

Bimanual related cells are also directionally tuned

To examine the relation between bimanual related activity and directional tuning of MI cells, we first calculated the bimanual effect (see Methods). As expected from our previous work, we found that many cells exhibit a strong bimanual effect (in our sample, 52% of the cells shown in Fig. 5 had a bimanual related effect higher than 0.5). We then tested the relation between the strength of the bimanual related effect of each cell and its cosine tuning, as estimated by the maximal $R^2$ value across the four types of movements. The results, as shown in Fig. 5, indicate that the correlation between the $R^2$ and the bimanual effect is very weak even if it is statistically significant (Spearman rank-based correlation coefficient, $r = -0.21$, $P < 0.01$). The weak correlation suggests that many cells may encode both ‘bimanuality’ and movement direction independently.

Population vector analysis

In analysing population activity we first tested that the three conditions, which guarantee a good prediction of direction by population vectors (PV values), are met in our sample. The first – that the cells have symmetric tuning around the PD – was proven true by receiving high $R^2$ values ($> 0.7$) for about 70% of the cells in at least one type of movement. The second – that the PDs of the cells we sampled were uniformly distributed in all directions – was tested by a Rao test. Here, we failed to show nonuniformity for $P > 0.2$. The third is that the distributions of the amplitude and offset of the cosine functions are independent of the PDs. We ruled out correlation between those two parameters and the PDs (Spearman rank correlation coefficient, $r < 0.1$, $P > 0.3$).

Then, we calculated PV values for each direction and each movement type in three ways, generating three sets of PV values for each movement. First, for each type of movement, we used the preferred direction taken from the cells’ activity during execution of that same type of movement. This generated the first set, called the four-PD set. This set must provide the most accurate prediction, as our data meets the conditions of Georgopoulos et al. (1982). Indeed, the resulting PV values predicted movement directions quite accurately, in all movement types, as shown in Fig. 6A.

In the second and third set, the PV values were calculated using one preferred direction for each cell. As the classical approach for the motor cortex is that each hemisphere represents movements of the contralateral side of the body, we first chose the PD of the contralateral unimanual movements to generate a second set of PV values (the contralateral-based PV). As shown in Fig. 6B, these PV values were only accurate for the contralateral unimanual movements, and particularly inaccurate for predicting the movement of the ipsilateral arm.

Finally, we produced a third set of PV values, using the BFPD (BFPD-based PV values). The results of this analysis are shown in Fig. 6C. The first impression is that these PV values are almost as accurate as the best possible PV values (four-PD set, compare Fig. 6C and A). To validate this impression, we calculated the average deviation of the different PV values from the actual direction of movement. Indeed, Fig. 7 shows that the prediction errors of BFPD-based PV values are not much larger than the four-PD based PV values. Thus, the results indicate that BFPDs represent the cells’ tuning better than the preferred directions of the contralateral unimanual movements, and almost optimally.

Population vectors represent simultaneous movements of the two arms

To construct a separate population vector for each arm, we divided the population of sampled cells into two subpopulations, hypothesizing that bimanual movements are generated by two separate (although possibly coordinated) neuronal networks. The division into two subpopulations was performed in two different approaches.
better than selection by the hemispheric bimanual movements. Also note that the PV values for the nonmoving arm, are a little smaller in B as compared to A. For unimanual movements, this is an inevitable result of the reselection, but the improvement in the bimanual movements is not a trivial result. Yet, examining the PV values for all movement directions, we could not validate that the accuracy of PA-based PV values is higher then the hemisphere-based PV values.

Discussion

Summary of results

This paper investigates the activity of MI cells during performance of unimanual and bimanual arm movements, and demonstrates that directional tuning coexists with ‘bimanual related activity’ and that PDs of single cells in different types of movement are correlated. Further, we show that populations of cells can accurately predict the direction of movement even when the two arms move simultaneously in different directions.

Directional tuning

The previous finding of ‘bimanual related activity’ raised a simple question. Do single cells maintain directional tuning properties despite differences in activation intensity in unimanual and bimanual arm movements, or does the tuning change along with the activation intensity? The results of the current study show that the classical directional tuning properties that were described for unimanual movements (Georgopoulos et al., 1982) remain valid also for bimanual arm movements. Namely, firing rates of most cells are well described by a symmetric tuning curve around a preferred direction. Many cells showed directional tuning in more than one type of movement, and their preferred directions tended to remain similar in these different types. This finding is in agreement with a previous study where ipsilateral and contralateral responses were compared (Perepelkin & Schwartz, 1996). For these cells, when a ‘bimanual related effect’ existed, it reflected significant changes of evoked activity not associated with a major change in the preferred direction of the cell. For example, Fig. 4A demonstrates a case where the ‘bimanual related effect’ reflected an overall change in firing rate without a shift of PD or change in the modulation depth.

Cells code for more than one parameter of movement

Our results are also in agreement with our own previous studies showing that many cells in the MI show a bimanual related effect (Donchin et al., 1998). Here, we extend these findings by showing that neuronal activities can depend both on the direction of movement and on the context of the arm movement, i.e., whether it is a unimanual movement or a bimanual arm movement in which the other arm is also moving (see Fig. 5). This also corroborates other studies showing that the activity of single-cells in the MI may be related to more than one parameter of the movement (Fu et al., 1993; Moran & Schwartz, 1999).

We also looked at how these two parameters combine at the level of a single-cell. We demonstrated a way of describing the cell’s activity as a function of a number of parameters of movement (see equation 1). The equation could be easily adapted into a general approach when more than one parameter of behaviour or stimulus may influence the cell’s activity.

Population vectors

There is still debate over the physiological significance of the population vector. Some researchers argue that downstream struc-
tures decode the signal from the MI by calculating a population vector. Others have argued that no real evidence exists that the PV is actually used to determine movement direction and that it is nothing more than a data-reduction technique. It has been further argued that the neurons giving rise to the PV may actually be muscle-related and not related to the arm kinematics (Mussa, 1988; Scott & Kalaska, 1995; Todorov, 2000). Todorov, for example, presented a model, which ‘… reinterprets the neural population vector to afford unified control of posture, movement and force production’.

Taken together, our previous accounts, that bimanual related activity was not sensitive to small variation of the movements of each arm or to muscular activity in bimanual vs. unimanual movements (Donchin, 1998), and the present result, that population activity codes for bimanual as well as unimanual movement direction, suggests that the motor cortex may also code for abstract movement parameters, independent of movement dynamics. This does not mean that motor cortex does not encode dynamic parameters of the movement. To conclude, while our experiment demonstrates that the MI contains information of abstract parameters of movements, it was not designed to differentiate between coding for intrinsic vs. extrinsic parameters of movements. It merely demonstrates that information about kinematics and context of the movement is present in motor cortex activity.

### Hemispheric control

A more emphatic interpretation can be made on the issue of hemispheric control. The assumption that each hemisphere is related only to movements of the contralateral side of the body no longer seems accurate. There is ample evidence that the MI is active during ipsilateral movements (Evarts, 1966; Tanji et al., 1988; Kermadi et al., 1998). In addition, results obtained from human subjects using various techniques (Hoshiyama et al., 1997; Shibasaki, 1975; Kim et al., 1993; Chen et al., 1997) also demonstrate activation of the motor cortex during ipsilateral movements. Our findings are in

![Fig. 6. Population vectors (PV values) calculated with different preferred directions. (A) PV values calculated using a different preferred direction for each type of movement. (B) PV values calculated using for each cell its preferred direction in contralateral movements. (C) PV values calculated using the BFPD, for all types of movement. The different colours represent the PV values of the different types of movements, as noted in the figure. Dark colours indicate PV values with lengths greater than the threshold of significance; Light colours indicate PV values with lengths smaller than the threshold of significance. Note, that the movement direction in bimanual opposite trials was assigned according to the preferred arm.](image)

![Fig. 7. Mean deviation of the population vectors (PV values) from the actual direction of movement. The deviations are shown for the three different methods of PV values calculation: (i) Four different preferred directions (black). (ii) The preferred direction of the contralateral movement (grey). (iii) The best-fit PD (BFPD – white). The error bars represent the standard error of the mean. Note that the BFPD method gives much smaller deviations in comparison to the method using the PDs from contralateral movements.](image)
agreement with the notion that MI is active both during ipsilateral and contralateral movements.

Here we examined the ‘natural’ hypothesis that the two cerebral hemispheres collaborate in encoding the direction of movement of the two arms using two neuronal populations, each coding for one arm.

We divided the cells into two subpopulations either by hemisphere or by their arm preference. This type of division ‘replaces’ approximately a quarter of the cells in the contralateral hemisphere by cells from the ipsilateral one, and yet, the PV values calculated in bimanual movements from this division are not less accurate than PV values calculated when dividing by hemisphere. This result further supports the notion that both hemispheres are active and contribute to execution of both unimanual and bimanual movements. The way the two hemispheres interact and collaborate with each other, have been further examined, and is the subject of a separate study (Cardoso de Oliveira et al., 2001).

Conclusions

The results presented in this paper show that even though single cells have very different activities in unimanual and bimanual arm movements, they still maintain directional tuning. Thus, MI cells can represent both the direction of movement and its context. Reasonable population vectors are obtained under the assumption that the PDs of neurons in MI are independent of the movement type. The accuracies of PV values are equally good if populations are formed...
by hemisphere or by response, indicating that the cortical networks related to movements of both arms may be distributed over both hemispheres.

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Abbreviations

BFPD, best-fit preferred direction; MI, motor cortex; PA, preferred arm; PD, preferred direction; PV, population vector.

References


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