PHOTOGRAPHIC ANALYSIS OF RELATION BETWEEN UNIT ACTIVITY AND MOVEMENT

JEROME M. SIEGEL, STEPHEN M. BREEDLOVE and DENNIS J. McGINTY

Neurophysiology Research, V.A. Medical Center, Sepulueda, Calif. 91343; Department of Psychiatry and Brain Research Institute, UCLA School of Medicine, and Department of Psychology, University of California, Los Angeles, Calif. 90024 (U.S.A.)

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A technique for the cinematographic analysis of relationships between unit activity and movement is described. The method permits mapping of the topographic and temporal relationships between unit activity and movement in the unrestrained animal. We have used this technique in the cat to document relationships between unit activity in the pontine reticular formation and movement.

INTRODUCTION

Observation of the behavioral correlates of unit discharge provides a powerful insight into the functional roles of brain areas. As the application of new techniques that allow the recording of unit activity in behaving animals becomes more widespread, a more quantitative description of observed relationships will be needed to document observations and differentiate between the unit activity—behavior relations of different brain areas.

The analysis of relationships between neuronal unit activity and movement patterns has generally been restricted to animals trained to make specific movements such as bar presses (e.g. Burton and Onoda, 1977; DeLong, 1973) or to angular movements of eye and head (e.g. Keller and Robinson, 1972; Collewijn, 1977) and locomotor (Orlovsky, 1972) movements, which can be readily quantified. Since only one component of behavior is measured in such procedures, results cannot provide a complete picture of the relationship of unit discharge to natural movements and may therefore be misleading.

The understanding of the functioning of neural units in sensory systems has been greatly facilitated by the ability to map receptive fields. However, a similar 'mapping' technique is not available for studies of unit activity in motor systems. Photography has long been used in the analysis of the temporal interrelations of movement patterns in unrestrained animals. The present report describes the adaptation of cinematographic techniques to the study of relations between unit activity and behavior. We have used these techniques in freely moving cats to map the movement relations of neurons in the gigantocellular nucleus of the pons, an area in which the activity of most units is related to specific movements (Siegel and McGinty, 1977).

METHODS AND RESULTS

Procedures for the long-term recording of single units in the brain stem of the unrestrained cat have been developed in our laboratory and reported in detail elsewhere (Harper and McGinty, 1973). Briefly, bundles of six 32 /um insulated microwires protruding 5 mm from a 24-gauge support cannula are attached to a mechanical microdrive consisting of concentric cannulae controlled by a miniature machine screw. Each microdrive can hold one or two bundles of microwires. Two microdrives are normally implanted in each cat. After recovery from surgery the cat is placed in a recording chamber and microwires are scanned for discriminable unit discharge. Microwire bundles are advanced in 50 /um steps until one or more isolated units with signal-to-noise ratios exceeding 3/1 and initially negative spikes are found. Signals are amplified with high impedance preamplifiers and conditioned with high pass 300 Hz filters to prevent interference from cable movement artifact. Unit signals are led to a window discriminator (Neurofeedback Instruments) which produces a pulse output for each unit spike whose amplitude falls between two experimenter set voltage levels. The pulse output can be used to drive polygraph pens, an audio monitor, and an electronic counter. With this technique, individual cells may be studied for periods ranging from several hours to many days, depending on the region studied.

Cats are placed in a shielded, 58 X 60 X 85 cm chamber with a glass front door (see Fig. 1). A mirror, mounted at 45° off vertical at the top of the chamber, allows simultaneous photographing of two views of the cat. Two digital LED counters are placed at the side of the chamber. The top counter is incremented by each unit discharge via the pulse output of a window discriminator. The bottom counter displays the current reading on a binary coded decimal time code (Biotronic Designs CGD200) with intercount intervals of 1 sec. Since this same time code is recorded on the polygraph and on magnetic tape, it serves as a reference point that allows one to correlate unit discharge—movement relations with EEG variables. It also permits the experimenter to compare changes in the reading of the unit counter with the magnetic tape of the unit recording, and to determine retrospectively if noise has triggered the counter. The chamber has a 'discriminative stimulus' light visible to both the cat and the camera. This light can be used to label behavioral contingencies, such as the operant reinforcement of increased rates of unit discharge with hypothalamic stimulation. This reinforcement procedure was used to increase discharge rates during the filming sessions used in the present study and will be reported on in detail elsewhere. The chamber has an LED indicator light which can be used to visually label short

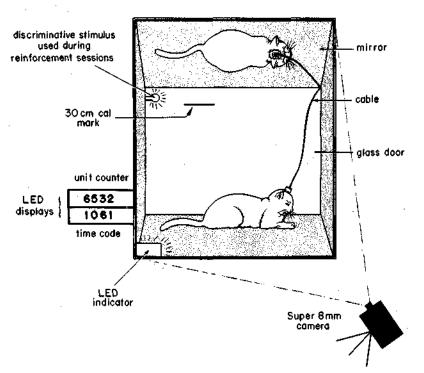


Fig. 1. Arrangement of equipment used for filming unit discharge—movement relations.

duration events such as discrete auditory stimuli or pulsed brain stimulation. Thirty cm rulers placed on the rear wall, floor and door of the cage are used to calibrate movement measurements. The cat, unit counter, time code and indicator lights are simultaneously filmed by a Super 8 mm 'existing light' camera placed 1 m from the cage door and run at 18 or 36 frames/sec.

Films are analyzed on a Bolex V180 Duo editor. This editor has a flat, wide screen and bright image, which facilitate data analysis. For each analysis, we locate a minimum of 10 instances in which an isolated unit discharge has occurred, by observing the unit counter. To be considered isolated there must be at least 5 frames without unit activity preceding the discharge.

Measurements of movement are made as follows. (1) Each frame is centered on the screen. The relative location of a selected body landmark is measured and plotted on graph paper. In the cells presented here the cats' nares served as a landmark for mapping head movements. (2) The next frame is then viewed and the relative location of the body part again plotted. (3) A vector between the two plotted points indicates the movement. (4) By plotting the vectors from the same starting point for each pair of frames, a figure that describes movement in the interframe interval before the first unit discharge in an isolated burst, and a second one to describe movement

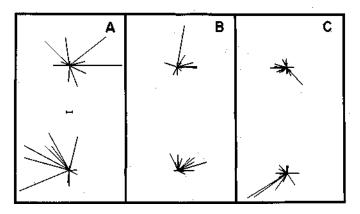


Fig. 2. A. Upper portion: vectors describing the movement of the cat's head during the film frames spanning the 56 msec period before the Onset of unit activity. Calibration mark for A, B and C is 10 mm. Lower portion: vectors describing the movement of the cat's head during the film frames spanning 56 msec period in which the unit began to discharge. Note the nearly random distribution of vectors in the upper portion of the figure, and the preponderance of movements toward the upper left-hand quadrant accompanying the onset of unit activity in the lower portion of the figure. B: same as A. Note the shift in movement to upper right-hand quadrant during onset of unit activity. C: same as A and B. Note the shift in movement in the left-hand quadrants during the onset of unit activity.

when the unit begins discharging can be constructed. (5) Separate sets of vector diagrams for each level of rate increment can be plotted, i.e. one set of plots for cases in which the unit discharges once between adjacent frames, another set for cases in which it discharges twice, etc. (6) Movements in each interframe interval following or preceding isolated unit discharge can be plotted. In this way one can determine at what time the maximum movement excursions occur relative to unit discharge. (7) A continuous plot of the movements before, during and after individual unit discharges can be drawn by putting the beginning of each interframe vector at the end of the previous vector. This allows one to determine the relationship of unit discharge to complex ongoing movement patterns. Plotting may be done in two or three dimensions. The present report used two dimensional plots. Unlike magnetic techniques of recording head and eye movement, our photographic procedure can record both angular and linear translations of any body part. The complete vector analysis procedure, including filming and vector plotting, takes approximately 12 h for each unit. Significance of changes in vector distribution can be assessed by appropriate statistical tests. Fig. 2 presents plots of head movements related to unit discharge in three units recorded in the pontine nucleus gigantocellularis. These cells were histologi-cally localized betweeen P 1.5 and P 5.5 and L 1.5 and L 2.0 (Berman, 1968). Note the random arrangement of movement vectors during the interframe intervals prior to the start of unit discharge. During the film frames

spanning the interval in which unit discharge begins, the distribution of vectors shifts.

Our previous work, which employed visual and EMG observations to describe the behavioral correlates of pontine gigantocellularis unit discharge, found that activity in these cells was related to specific movements (Siegel and McGinty, 1977; Siegel et al., 1977). The most common relation was to head movements. The preliminary data presented here indicate that these movements begin within 56 msec of unit discharge and that the movements have a high degree of directional consistency. Within a 56 msec interval more than one movement can be executed. Therefore the scatter observed in Fig. 2 could result either from other movements within the interval obscuring a very tight coupling of unit discharge to a highly specific movement, or from the units' relation to a wide range of movements concentrated in one direction. Higher speed filming could resolve such questions.

DISCUSSION

This procedure provides an objective method of mapping a unit's motor correlates. The temporal relation of unit firing to behavior, and the consistency of this relation, can be documented with great precision without using restraint or movement restriction. This technique can be applied to describe unit activity—movement relations in unrestrained animals during a wide range of natural behaviors.

Correlative data cannot itself prove causal relationships. Thus a unit discharge that precedes motion of an identified body part need not be directly in the pathway mediating that movement. Rather, the unit activity and movement may both be temporally related to other aspects of the complex sequence of sensory and motor events surrounding the observed movement. For example, unit discharge may relate to increased tone in axial musculature, alterations in reflex excitability, activity in cutaneous muscles, etc. The photographic technique is able to detect unit discharge relations with a wide variety of movements and is therefore less likely to miss underlying relationships than restricted movement paradigms that measure only the movement of a single body part. However, results from both of these techniques must be correlated with behavioral, physiological and anatomical findings in order to determine the mechanisms underlying observed relationships.

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REFERENCES

- Herman, A.L. (1968) The Brain Stem of the Cat, University of Wisconsin Press, Madison, p. 175.
- Burton, J.E. and Onoda, N. (1977) Interpositus neuron discharge in relation to a voluntary movement, Brain Res., 121: 167–172.
- Collewijn, H. (1977) Gaze in freely moving subjects. In R. Baker and A. Berthoz (Eds.), Control of Gaze by Brain Stem Neurons, Developments in Neuroscience, Vol. 1, Elsevier/North-Holland Biomedical Press, Amsterdam, pp. 13–22.
- DeLong, M.R. (1973) Putamen: activity, of single units during slow and rapid arm movements, Science, 179: 1240—1242.
- Harper, R.M. and McGinty, D.J. (1973) A technique for recording single neurons from unrestrained animals. In M.I. Phillips (Ed.), Brain Unit Activity during Behavior, Thomas, Springfield, 111., pp. 80—104.
- Keller, E.L. and Robinson, D.A. (1972) Abducens unit behavior in the monkey during vergence movements, Vision Res., 12: 369—382.
- Orlovsky, G.N. (1972) Activity of vestibulospinal neurons during locomotion, Brain Res., 46: 85–98.
- Siegel, J.M. and McGinty, D.J. (1977) Pontine reticular formation neurons: relationship of discharge to motor activity, Science, 196: 678—680.
- Siegel, J.M., McGinty, D.J. and Breedlove, S.M. (1977) Sleep and waking activity of pontine gigantocellular field neurons, Exp. Neurol., 56: 553—573.