

A Microdrive With a Movable Microelectrode for Single Unit Recording in Freely Behaving Rats

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In neurophysiological studies of behaviors, investigation of unit activity of the brain in freely behaving animals is important to clarification of the neural mechanisms involved in behaviors.

Many devices have previously been proposed for recording extracellular unit activity of the brain in behaving animals. Although in these devices, unit activity recording has been performed with the fixed or movable electrodes, which were either fine wires (Ainsworth & O'Keef, 1977; Bland et al., 1980; Chorover & Deluca, 1972; Fontani, 1981; Harper & McGinty, 1973; Palmer, 1978; Rank, 1973; Rosseto & VanDercar, 1972; Sainsbury et al., 1983; Sasaki et al., 1983; Yamamoto, 1987; Vertes, 1975; Zhang & Harper, 1984) or standard metal microelectrodes (Deadwyler et al., 1979; Sinnamon & Woodward, 1977; Winson, 1973), fine wire bundle electrodes have been utilized more often than standard microelectrodes, since the former make possible long-lasting unit activity recording from the same neurons.

However, fine wire electrodes with large tips are probably biased toward larger neurons than are microelectrodes with small tips (Towe & Harding, 1970). Taking account of the properties of these electrodes, the microelectrode may be capable of recording more unit activities from large and small neurons than is the fine wire electrode.

Moreover, the movable microelectrode seems to have an advantage over the fine wire electrode in that it can record single unit activity with higher signal-to-noise ratio from several different neurons in a given region of the brain, whereas the fine wire electrode tends to record multiple unit activity, either fixed or movable.

For the clarification of neural mechanisms involved in behaviors, both thorough analysis of single unit activity and long-term recording of unit activity should be useful. Furthermore, in case of use of a movable electrode, estimation of the depth of the recording site in the brain through unit activity recording must be performed, but the previous microdrive did not offer such a function.

The present microdrive is equipped with both a movable microelectrode and a ruler to estimate the depth of unit activity recording site, which has been successfully used in our laboratory for long-term recording of single unit activity from the dorsal hippocampus in freely behaving rats (Yamaguchi, 1991).

CONSTRUCTION

All the materials for making the microdrive are readily available on the market. Fig.1A, 1B, and 1C show the construction process.

In Fig. 1A, the body of the microdrive is a support (a; 8 mm in diameter, 10 mm high) with a screw hole, 3 mm in diameter. First of all, after two shallow, vertical grooves were carved in parallel on the support's surface, two stainless pipes with a diameter of 1 mm (b; 13 mm, c; 12 mm long) were settled into these grooves and fixed with an instant glue. The distance between the two pipes was about 1.5 mm. The upper and lower parts of the pipe (b) were adjusted to protrude 2 or 1 mm from the edge of the support. As for the pipe (c), only the upper part protruded 2 mm from the support. Then, the stainless pipe (d; 1 mm in diameter, 7 mm long) was attached to the pipe (b) with an instant glue, which is necessary when the microelectrode is implanted into the brain stereotaxically. All three pipes were then covered with an epoxylite adhesive.

In Fig. 1B, the stainless spring (e; 5 mm in diameter, 13 mm long) was soldered to the washer (f; 8 mm in diameter), and after the screw (g; 3 mm in diameter, 15 mm long) was inserted into the spring from the side of the washer, it was turned into a screw hole of the support until its tip reached the bottom of the support. At this time, the base of the support was covered with a thin plastic plate in order to protect the screw hole.

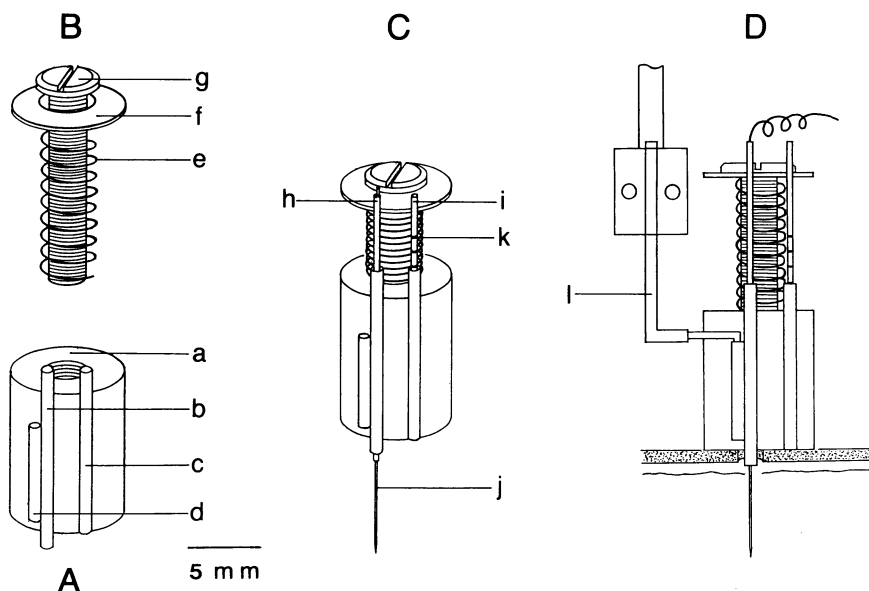


Fig. 1 A, B, and C show stages in construction of the microdrive assembly. D shows a stage of the stereotaxical implantation of the microelectrode. See text for details.

In Fig. 1C, the fine stainless pipe with a diameter of 0.5 mm (h; 18 mm, i; 17 mm long) was inserted into the pipe (b, c) and the upper part of the pipe (h, i) was soldered to the edge of the washer. The lower tip of the pipe (h) protruded 0.5 mm from the pipe (b). Then, the microelectrode (j) was inserted into the pipe (h) from beneath and fixed with an instant glue. The total length of the microelectrode was settled by adding the length of the pipe (h) to the depth from the skull surface of a given region in the brain. The material of the microelectrode was a tungsten wire, 100 μm in diameter. Its tip was sharpened at 5 μm by electrolytic etching.

The pipe (i) serves two functions. The first is to estimate the depth from the skull surface to the microelectrode's tip through unit activity recording

with the inscription of marks (k) on its length (pipe i) at 1 mm intervals. The lowest mark on the pipe (i) should be inscribed so as to hold the depth of the microelectrode's tip more than 1 mm from a given region of the brain, in order to keep brain tissue intact until unit activity recording begins. The second is to protect the pipe (h) against a load arising by turning the screw.

In short, by turning or loosening the screw, the stainless-spring contracts or expands, and in accordance with this movement, the microelectrode also moves up and down in a vertical line. However, the microelectrode itself does not rotate at all. Consequently the tip of the microelectrode should be held at the arbitrary depth in the brain so that unit activity may be investigated at will. If revolution of the screw is tight, movement may be improved by adding oil between the washer and the thread.

The movable range of the microelectrode is 8 mm and one full turn of the screw moves the electrode 500 μm . In addition, the minimal movable range of the electrode is about 20 μm . Further, this microdrive assembly is so compact and light-weight (3 gm.) that it cannot completely interrupt all kinds of behaviors in freely behaving animals.

IMPLANTATION

Fig. 1D shows a state of the stereotaxical implantation of the microelectrode. The subjects were male Wistar strain albino rats. Under anesthesia, standard surgical procedure was performed. Ground and indifferent electrodes (stainless steel screw, 1 mm in diameter) were placed over the frontal sinus.

Then, the stainless pipe with a L-shaped end (l) was inserted into the pipe (d) and soldered. Moreover, the pipe (l) was installed on the electrode holder of a stereotaxic instrument. In this way, the tip of the microelectrode may be adjusted stereotaxically on the skull over a given region of the brain. After the coordinates for implanting the microelectrode were decided according to the atlas (Paxinos & Watson, 1986), a small craniotomy was made in the skull. The diameter of this hole for inserting the electrode must be larger than that of the pipe (b).

Subsequently, the microelectrode was inserted into the brain slowly

until the bottom of the microdrive contact the skull. Here the tip of the microelectrode should be arranged to remain at least 1 mm over a given region of the brain, in order to keep brain tissue in the recording site intact. If need be, it is possible to monitor unit activity through implantation. After the microdrive assembly was fixed on the skull by dental resin, the pipe (I) was removed. Finally, the microdrive assembly was covered by a transparent plastic cylinder (15 mm in diameter, 20 mm high) in order to protect it against behaviors such as scratching or self-grooming in behaving animals. Fig. 2 shows the microdrive assembly mounted on the head of a rat.



Fig. 2 Photograph of a rat with the microdrive assembly mounted ready for use.

RESULTS AND DISCUSSION

The basic recording method of unit activity in the brain was similar to that reported by Rossetto and VanDercar (1972). But the Field Effect Transistor (FET) circuit for minimizing movement artifacts was not mounted directly on the head connector of the animals. In the present recording, an input box with a built-in buffer amplifier was used, and had the same functions as an FET circuit. Accordingly, the slip-ring connector was linked between the recording cable from the head connector of the animal and this input box. Unit activity recording was performed by turning the screw until the tip of the microelectrode reached a given region of the brain. The mark on the pipe (i) was a guide to the recording site. This simple ruler was very efficient for recording unit activity within a given region of the brain.

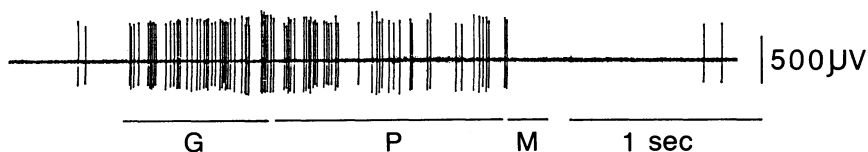


Fig. 3 Example of single unit activity recorded from the dorsal hippocampus during copulatory behavior in the male rat. This unit activity showed a conspicuous increase of impulse discharges while the male rat gazed (G) and pursued (P) the female rat but disappeared as soon as the male rat mounted (M).

Fig. 3 shows an example of single unit activity recording with the present microdrive, recorded from the dorsal hippocampus during copulatory behavior in male rats. Although a chain of behaviors such as pursuit-mount in male rats was very active, movement artifacts were eliminated completely in this unit activity recording. Consequently, the present recording system seems to protect unit activity against artifacts due to twisting or shaking of the recording cable caused by animal's movements. Therefore, single unit activity with higher signal-to-noise ratio may be easily and stably recorded from freely behaving animals. In some cases, the same unit activity was recorded over a period of one week.

If the amplitude of unit activity lessens gradually during recording, there is a possibility that its amplitude can be improved by turning or loosening the screw slightly. Therefore, the same unit activity may be maintained for a relatively long time. Even if unit activity disappear completely during recording, other unit activity should be detected by turning the screw. Consequently, the present microdrive seems suitable for investigating the relation between brain unit activity and behaviors in behaving animals.

On the other hand, Sainsbury et al. (1983) have used the microdrive in order to record the representative hippocampal slow waves within the fascia dentata of the hippocampus in the behaving guinea pig. Likewise, the present microdrive seems to be useful in the studies such as EEG, evoked potential or electrical stimulation of the brain in behaving animals by substituting a macroelectrode for the microelectrode, because this helps adjust the tip of the electrode to the most adequate site of the brain for provoking good responses while monitoring behavioral or electrical responses. Therefore, the present microdrive shows wide applicability in various fields of behavioral physiology.

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