Section 4. Intro to Neurophysiology

4.1 Action potentials at work (Cockroach Receptive Fields)

Overview

The goals of this unit are to: 1) introduce you to the basic concepts, equipment, and methodology of neurophysiology research, and 2) to familiarize you with the fundamental unit of neural activity – the action potential.

The wet lab session will allow you to see how electronic equipment can be used to learn about action potentials. In this session (Cockroach receptive fields), you will consider how action potentials can be used to infer how the nervous systems of animals communicate information from the sensory periphery to the central nervous system.

4.1 Action Potentials at Work (Cockroach Receptive Fields)

Equipment Connections

- 1. Connect the electrode pins to the differential inputs of the amplifier.
- 2. Connect a BNC from the amplifier output to the audio monitor and to the oscilloscope CH 1 simultaneously using a T junction.
- 3. Set the filter settings on the amplifier to 300 Hz and 3 kHz, respectively. Set the amplification to 1000x.
- 4. When these connections are in place, have an instructor check that the connections are correct, and then proceed to obtain a cockroach leg.

Cockroach Leg Preparation

**The teaching staff will assist you will all aspects of this procedure. It is critical that NO cockroaches escape during this process!

1. Obtain a cockroach from the teaching staff. It will be in a vial with the top covered in parafilm.

2. Using the handheld gas trigger attached to the CO2 tank, poke a hole in the parafilm and inject CO2 into the vial until the cockroach is no longer moving.

- Be careful not to over expose the cockroach to CO2 and kill it.

3. Remove the cockroach from the vial, and lay it on its back.

4. Using dissection scissors, disconnect a large posterior leg from the body at the upper coxa.

-Two groups should use a single cockroach, so the other limb should also be used.

5. Using forceps (gently, don't crush it), carry the leg back to your recording station.

6. Place the hind leg on the molding clay on the steel plate such that the coxa and femur are securely on the clay but the tibia and claw are freely extending off of the edge of the block.

7. Place one electrode pin in the femur ("active electrode"), connected to Input 1 of the amplifier. Place two electrode pins in the coxa, connected to Input 2 ("reference") and Common ("ground"). Try to place the electrodes so that the tip is inside the leg; you can still record if the tip passes all the way through and into the clay (these electrodes are not insulated), but you'll get better signals from the tip.

General "Hand Mapping" of Receptive Fields

1. Turn up the volume the audio monitor in order to hear action potential activity.

-Be mindful that other groups are listening to their audio monitors at the same time, so try not to turn the volume up too high.

2. Look at your oscilloscope to determine if you can see action potential activity.

3. Using your forceps, gently deflect the claw.

-You should hear a large increase in the degree of neural activity.

4. Gently deflect individual spines on the leg, apply vibrations to the desktop, etc. to get a general sense of the evoked neural activity.

-Be careful not to move the preparation itself.

Qualitative "Hand Mapping" of Receptive Fields

1. Systematically deflect each spine on the tibia on both sides of the leg, in all directions.

2. Do all of your action potentials look the same, in terms of amplitude and waveform, or do you appear to have more than one population of spikes? If you're able to distinguish spikes from more than one cell, you can describe the receptive field for each, or just focus on the cell that produces the largest spikes.

3. Determine qualitative responsiveness to deflection of each spine, for both the direction of deflection and for spine position.

-Was the firing rate stronger when you deflect the spine toward the claw or toward the knee? Was the firing rate equal across all spines? Did spines on the back of the leg have stronger responses than those on the front of the leg? Did spines near the knee have stronger responses than those near the claw?

4. Reposition the recording electrode near the dorsal (nonspiny) side of the femur. Repeat steps #1-3.

5. Reposition the recording electrode in the ventral portion of the femur. Repeat steps #1-3.

-Do you notice any change in responsiveness when you change the electrode position?

6. Other characteristics of the response you should consider: What is the maximal firing rate? Are the spike trains regular or irregular? Is the response maintained as long as the spine is deflected or does it adapt; if it adapts, how long does adaptation take? Does the response to repeated stimulation differ from the response to a single stimulation? (By this point, you should start to appreciate the value of better stimulus control when investigating the subtleties of neural response properties.) Are there other aspects of the neural response you can investigate?

7. Position both the recording and indifferent electrodes in the femur, with the recording electrode more distal by \sim 4 mm. Can you observe biphasic action potentials? If so, estimate the time delay between the two sites; with a ruler, estimate the distance between the sites; estimate the conduction velocity.

Illustration of the basic anatomy of a cockroach leg removed due to copyright restrictions. See Figure 1A. Ramos, Raddy L., Andrew Moiseff, et al. "Utility and Versatility of Extracellular Recordings from the Cockroach for Neurophysiological Instruction and Demonstration." *Journal of Undergraduate Neuroscience Education* 5, no. 2 (2007): A28–A34.

Basic anatomy of a cockroach leg.

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