生物物理学 I Handout No.4



(A) Channels activated by physical changes in the cell membrane



Voltage-activated

(B) Channels activated by ligands



Stretch-activated



Figure Micropipettes used far patch-clamp recording.

A rod cell from the eye of a salamander is shown held by a suction pipette while a fine-tipped glass pipette, pressed against the cell so that the glass is sealed tightly to the plasma membrane, serves as a microelectrode. The term "clamp" is used because an electronic device is generally utilized to "clamp" the voltage across the patch so that the voltage is maintained at a fixed value. (from T.D. Lamb, H.R. Matthews, and V. Torre, J. Physiol. 37:315-349, 1986.)



FIGURE 2.2 Modes of Channel Activation.

The probability of channel opening is influenced by a variety of stimuli. (A) Some channels respond to changes in the physical state of the membrane, specifically changes in membrane potential (voltage-activated) and mechanical distortion (stretch-activated). (B) Ligand-activated channels respond to chemical agonists, which attach to binding sites on the channel protein. Neurotransmitters, such as glycine and acetylcholine, act on extracellular binding sites. Included among a wide variety of intracellular ligands are calcium ions, subunits of G proteins, and cyclic nucleotides.



FIGURE 2.4 Examples of Patch Clamp Recordings.

(A) Glutamate-activated channel currents recorded in a cell-attached patch from locust muscle occur irregularly, with a single amplitude and varied open times. Downward deflections indicate current flowing into the cell. (B) Acetylcholine-activated currents from single channels in an outside-out patch from cultured embryonic rat muscle reach a maximum amplitude of about 3 pA and relax to a substate current of about 1.5 pA. Downward deflections indicate inward current. (C) Pulses of outward current through glycine-activated chloride channels in an outside-out patch from cultured chick spinal cord cells are interrupted by fast closing and reopening transitions to produce bursts. (A after Cull-Candy, Miledi, and Parker, 1980; B after Hamill and Sakmann, 1981; C from A. I. McNiven and A. R. Martin, unpublished.)



FIGURE 2.3 Patch Clamp Recording.

(A-E) Patch configurations, represented schematically. The electrode forms a seal on contact with the cell membrane (A}, which is converted to a gigaohm seal by gentle suction (B}. Records may then be made from the patch of membrane within the electrode tip (cell-attached patch). Pulling away from the cell results in the formation of a cell-free vesicle, whose outer membrane can then be ruptured to form an inside-out patch (C). Alternatively, the membrane within the electrode tip may be ruptured by further suction to obtain a whole-cell recording (D) or, by pulling, to obtain an outside-out patch (E). (F) Recording arrangement. The patch electrode is connected to an amplifier that converts channel currents to voltage signals. The signals are then displayed on an oscilloscope trace or computer screen so that amplitudes and durations of single-channel currents can be measured. (A-E after Hamill et al., 1981.)