## L. A. Nazarov and A. G. Komlyagin

A stand for holding fish of various sizes during electrophysiological investigations is suggested. The stand is mounted in an aquarium in such a way that the fish can be placed in any desired position in space.

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Interest in electrophysiological investigation of individual analyzer systems of fishes has grown appreciably in recent years. This interest is attributable partly to the specific features of the aquatic life of these animals and partly to the comparatively simple techniques required to approach various parts of the brain.

Most lectrophysiological investigations on fishes have been carried out usually in acute experiments. To hold he fish during the operation and in the course of the experiment, various types of stand are used. The best of them is a universal stand with screw-operated head holder and with curved supports for the body of the fish made of thick brass wire [1]. However, this stand, like all fixing devices, when used for electrophysiological investigations, especially in connection with microelectrode techniques, requires the fish to be under general anesthesia or completely immobilized by relaxants. Administration of anesthetics (Nembutal, urethane) has a considerable effect on bioelectrical activity of the fish brain, and may even suppress it completely, while relaxants, if producing complete immobilization, as a rule abolish respiration through the gills also.

The suggested apparatus (Fig. 1) for holding fish of different sizes is an improved version of the stand described previously [1], and consists of a head holder and two supports for the body of the fish, mounted on two rods (1). The head holder consists of two jaws, the lower one of which is inserted into the fish's mouth while the upper presses on the fish's upper jaw. The clamps for the body consist of rubber bags (3)

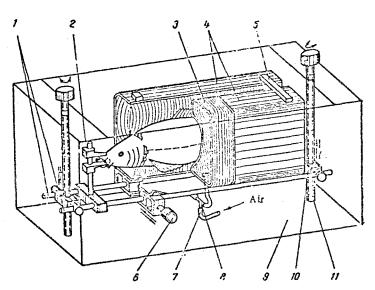


Fig. 1. Diagram of stand for holding fish. Explanation in text.

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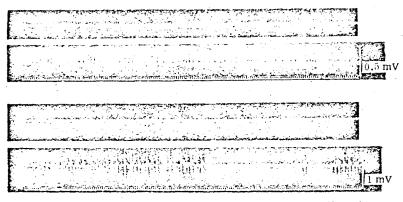


Fig. 2. Response of mitral neurons of olfactory bulbs of a burbot (I) and pike-perch (II) to application of vanillin solution to olfactory epithelium of fish. 1, 3) Initial background; 2, 4) action of 0.5% vanillin solution for 1 min, time marker 20 sec.

filled with air and placed in rigid metal boxes (4) preventing expansion of the bags in an outward direction. The supports can rotate around the rods and are held in the closed position by two struts (5). When fixing fish of different sizes or volumes, the supports can be moved transversely by means of screws (6). Air enters the bags through a tube (7). The interiors of the bags are connected by a T-tube (8) to equalize the pressures. The air pressure in the bags is controlled by a manometer. The whole stand is placed in the aquarium and fixed to it by two carriages (10) and screws (11). By turning the screws (11), the stand can be raised or lowered to any height in the aquarium, and by means of the carriages (10) it can be inclined in a longitudinal direction.

Before the operation begins the aquarium is filled with water. The supports are opened and the fish placed between them so that its head remains free. The supports are then closed and fixed in this position by nuts, after which air is introduced into the bags. Under the action of excess pressure the rubber bags are expanded and fit snugly against the body of the fish, holding it tight. The head is fixed by the jaw (2) introduced into its mouth, holding the upper jaw of the fish by the lip, allowing free access to the olfactory sacs, olfactory tracts, and then to the brain as a whole. The lower jaw of the fish remains free, allowing it to perform the full range of respiratory movements. By turning the screws (11) the stand can be raised into a position so that the upper part of the skull lies above the surface of the water but the gill slits are in the water, after which the experimenter begins the operation.

More than 100 acute experiments have been carried out with the aid of this type of stand on pike-perch and burbot. During the experiments microelectrode extracellular recordings were made of spontaneous and evoked activity of the mitral neurons of the olfactory bulbs during application of stimulants to the olfactory epithelium of the fish (Fig. 2).

The gentle and elastic fixation of the fish between the two rubber chambers prevented all movements except the full range of respiratory movements of the branchial arches. As the experimental results showed, no additional immobilization by general anesthesia or muscle relaxants was required. The reasons for the artefacts recorded in approximately 10% of cases when microelectrodes were used to record the biopotentials were either improper fixation of the fish in the stand (inadequate air pressure in the rubber bags, inadequate fixation of the upper jaw, incomplete fixation of the pectoral fins, and so on) or to entry of air into the gill slits, when the water level in the aquarium was too low and the fish drew in air along with the water. Artefacts arising from these causes are easily eliminated and their occurrence is directly dependent on the attentiveness of the experimenter.

It may be concluded from these experiments that the principle adopted in the design of this stand enables fish of any size to be quickly and securely immobilized, yet at the same time it provides conditions which are closest to physiological. It is to be hoped that the stand will allow many physiological problems involving the use of modern electrophysiological methods of investigation to be solved.

## LITERATURE CITED

1. Yu. G. Kratin, N. P. Bekhtereva, V. I. Gusel'nikov, et al., Techniques and Methods of Electroencephalography [in Russian], Moscow-Leningrad (1963), p. 246.