After more prolonged centrifugation (4 hours at 20,000 rev/min), the nuclei are more sharply banded away from the "cell fragments" or membrane fraction. The results of enzymatic studies on these fractions will be reported elsewhere. While much theoretical and experimental work remains to be done before optimal fractionation conditions are achieved, these results suggest that quantitative separations of gram quantities of tissue can now be achieved.

In addition to liver cell fractions, monkey heart cells (*8*) have been fractionated in studies preliminary to attempts to isolate viruses directly from homogenates. Partial separation of the albumin and globulin peaks in rat serum was obtained in 9 hours, suggesting that molecular separations can be easily effected by this very gentle method when higher speeds become available.

It should be emphasized that rotor II used in these experiments was designed to study (i) rotor and gradient stability, (ii) performance of fluid-line seals which allow liquids to be pumped into and out of the centrifuge during rotation, (iii) the fractionation of cell populations into different cell types, (iv) the separation of subcellular organelles and viruses, and (v) the fractionation of protein and nucleic-acid mixtures on the basis of sedimentation rate. The rotor is necessarily of a compromise design that is not ideally suited to doing any single one of the above. These results therefore do not demonstrate the resolution ultimately attainable.

It appears that zonal or density gradient centrifugation in hollow rotors may prove to be a general separation method for particles ranging from whole cells, cell particulates, and viruses to small proteins. The theoretical aspects of large, tubeless-rotor density gradient centrifugation will be discussed in detail elsewhere.

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Positive and Negative Reinforcement from Intracranial Stimulation of a Teleost

Abstract. Tests in which an animal receives or avoids direct electrical stimulation of its brain according to its position in the tank as well as tests of free operant behavior demonstrate the existence of both positively and negatively rewarding areas in the brain of the goldfish.

Since Delgado *et al.* first obtained negative reinforcement by stimulation of certain areas in the brain of the cat (*1*) and Olds and Milner showed that stimulation of certain areas of the brain of the rat is positively reinforcing (*2*), similar areas have been found in the brains of several mammalian species. Areas in which stimulation is positively reinforcing have been found in cat (*3*), dog (*4*), and primate (*5*), and areas in which stimulation induces aversion have been found in rat (*6*) and primate (*7*). If these areas are related to the normally functioning reward systems of animals, they would be expected to exist in any species with a well-developed nervous system. An attempt was made to demonstrate that such areas exist in the brain of the goldfish, *Carassius auratus*, a more primitive animal than those previously studied.

Monopolar electrodes, about 10 inches of 10-mil Formvar wire exposed only at the cut end, were implanted in the brains of goldfish anesthetized with tricaine methane sulfonate (1:10,000). The electrode, in a stereotaxic instrument, was lowered into the brain through a hole made by a dental drill and fixed to the head with dental cement and small anchoring wires. The stereotaxic instrument served only to regulate the depth at which the electrode was implanted; the placement in all coordinates was variable and not reproducible. The electrode was attached by a brass swivel to a suspending wire centered over the fish tank. Stimuli of 0.5-second pulses of 60-cycle current were delivered between the implanted electrode and an indifferent electrode in the fish tank.

After testing, 2 ma of direct current were passed through the electrode for 15 seconds, the fish was killed by decapitation, and the head was fixed in formalin containing ferrocyanide to stain the iron deposit. Frozen brain sections (40 to 140 μ) were photographed by the method of Guzman *et al.* (*8*) to record the electrode placement.

Each fish was subjected to two test procedures. The first procedure was a series of side preference tests at currents between 5 and 150 μa. Each test consisted of ten consecutive 5-minute intervals at a set current intensity. During each interval, the animal received a stimulus every 1 to 2 seconds when it was on one side of the tank, and no stimulus when it was on the other side. The side associated with the stimulus was reversed between each of the intervals, and the presence or absence of a light gave the fish a cue as to which side was associated with the stimulus

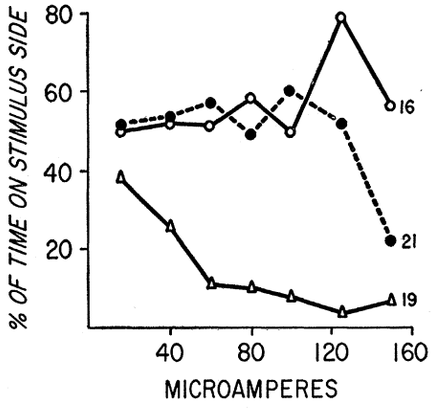

Fig. 1. The performances of fish 16, 19, and 21 in side preference tests (see text) at various current intensities. A percentage above 75 indicates positive reinforcement, below 25, negative reinforcement.

Fig. 2. Sagittal section of the goldfish brain 1 mm from the midline, showing the location of the tips of the electrodes in fish 16, 19, and 21.

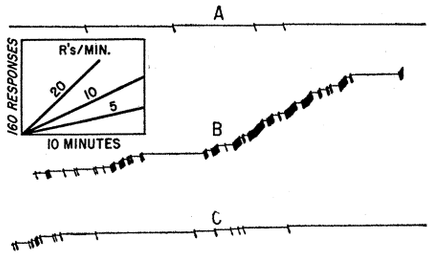

Fig. 3. Continuous records of self-stimulation of fish 16 at 125 μa. *A*, Light off and target inoperative. *B*, Light on and target operative. *C*, Light off and target inoperative. Each response is marked both by an upward motion of the pen and a reinforcement pip. The inset gives the scale of responses versus time.

at any particular time. The animal was thus free to receive or avoid the stimulus. The data are expressed as the percentage of the total time (50 minutes) which the animal spent on the side associated with the stimulus. A side preference could be associated with factors other than the stimulus. For instance, there may have been a marked preference for one side of the tank which was manifest only during the alternate 5-minute intervals when the cue light was off. This and other possible factors could conceivably result in percentages between 25 and 75, and therefore a percentage less than 25 was taken as a definite indication of negative reinforcement, and a percentage greater than 75 was taken as a definite indication of positive reinforcement. The results with three fish are shown in Fig. 1. For fish 19 the stimulus became progressively more aversive with increasing current intensity. In fish 21 the stimulus was neutral up to 150 μa, when it became aversive, and in fish 16 it was neutral except at 125 μa when it was a positive reinforcement. The placements of these three electrodes are shown diagrammatically in Fig. 2.

After the side preference tests, each animal was tested in a free operant situation with a Lucite target in an arrangement similar to that used by Aronson and Herberman (*9*). Responses and stimuli were recorded with a Gerbrands cumulative recorder. If the animal had shown negative reinforcement in the previous tests, it was exposed to a Sidman type (*10*) avoidance schedule (stimulus-stimulus interval, 1 or 2 seconds; response-stimulus interval, 20, 30, or 60 seconds). The fish would strike the target to escape the stimulus, but not to avoid it. If the animal had shown positive reinforcement, it was allowed to strike the target to receive a stimulus when a cue light was on. When the light was off, the target was inoperative. Figure 3 shows a typical result with fish 16 at 125 μa. In the upper record, the target was inoperative and the animal responded five times in 35 minutes. This was followed immediately by a test series, shown on the center record, when the target was operative. There were 174 responses in 31 minutes. This series was followed by another control period, the lower record, with 28 responses in 35 minutes.

Of the 13 fish tested so far nine placements have been negative and three positive at one or more current intensities, and one has been neutral between 5 and 150 μa (*11*).

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Preservation of Old, Waterlogged Wood by Treatment with Polyethylene Glycol

Abstract. The excessive cracking and distortion that old waterlogged wood undergoes when it is dried can be substantially reduced by treating the wood with polyethylene glycol. The process was used to dry 200-year-old waterlogged wood boats recently raised from Lake George, New York.

The preservation of waterlogged wood from archeological sites has long been a problem for the archeologist and museum conservator. Wood that has been removed from these sites generally develops cracks and flakes while it is being dried, and it undergoes severe changes in dimension and shape.

Various techniques have been developed to counteract this degradation (*1*), but many of them are cumbersome and too complex to use in the field. In the United States the problem of preserving large objects developed suddenly when remains of Colonial bateaux were found under the waters of Lake George, New York, in the summer of 1960. A few of these 200-year-old boats, from the French and Indian Wars and perhaps the Revolutionary War period, were raised by skindivers in the fall of 1960. Experimental work to determine whether such wood could be successfully treated was carried out jointly by the Adirondack Museum, Blue Mountain Lake, N.Y., and the U.S. Forest Products Laboratory, Madison, Wis.

The results of this work showed that the severe degrading of old waterlogged wood, which normally occurs during drying, can be eliminated or greatly reduced by treatment with polyethylene glycol.

The wood used in this study was from the boats that had been raised from Lake George. The material consisted of ½- and 1-inch-thick sections of planking identified as white and yellow pine and short lengths of white oak that were used in the ribbed structure. The wood was kept water-soaked until it was treated. The white oak was quite sound except for surface decay. The white pine, however, was badly decayed; boards 2 and 3 feet long were difficult to handle without breakage. Yellow pine was intermediate in degree of deterioration.

Polyethylene glycol (mol. wt. 1000) (*2*) in 50-percent aqueous solution was used to treat the wood. The waterlogged sections (12-inch lengths) were soaked in the solution at room temperature for periods ranging from 4 hours to 1 week, and by multiple dip treatments. The treated wood was then air dried at 80°F and 30 percent relative humidity. Control specimens without previous treatment were dried in a similar manner.

Shrinkage in width of the untreated specimens was of the order of 5 to 7 percent, but shrinkage of the specimens treated for 2 to 7 days was approximately 0.5 percent. When the treatment period was shorter (4 to 24 hours), the subsequent shrinkage on drying was 2 to 4 percent, because the diffusion of the chemical was incomplete. However, the degree of surface