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Box for Observation of Living Organs *in situ* and for Abdominal Surgery in the Sculpin.

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The operating box herein described was developed for use in 2 studies upon the sculpin *(Myoxocephalus octodecimspinosus)*, a common marine teleost. Grafflin and Ennis[1] reported experiments upon the blockage of the gastrointestinal tract, and Grafflin and Eisenberg[2] upon the direct observation of the living kidney. There is obviously a wide field for comparative studies in the fishes in which abdominal surgical procedures and direct observation of living organs *in situ* would play an important part. The sculpin is readily available for experimental work; and the operating box which we have developed has been so satisfactory in our experience that it is described here in the belief that it will prove useful to other workers. The box is illustrated in Figures 1 and 2.

With the use of water outlets C and C', the water level in the box is usually maintained at a sufficiently high level. With some larger specimens a higher level is necessary to insure complete covering of the head, and in these cases the rubber tubing is switched from C to D, and the opening at C is plugged. The rather tight fit of the animal between the clamps, and the upward projection of the pectoral fins, suffice to maintain a higher level in the anterior than in the posterior chamber under these conditions.

A constant stream of sea water is supplied through the inlet B. In the experiments previously reported urethane anesthesia was used. Anesthesia was rapidly induced by immersing the fish in a 1% solution of urethane in sea water in a separate chamber. The animal was then transferred to the box, and anesthesia was maintained throughout the experiment by a 0.25% solution of urethane in sea water. This was supplied by gravity from a large container, the flow being sufficiently rapid to effect a constant change of water in the box. The solution becomes somewhat contaminated in passing through the box, and should not be used a second time. If the anesthesia becomes too light, the clamps and lead blocks usually suffice to prevent disturbing movement until the concentration can be adjusted. The angle at which the pectoral fins are held and the

[1] Grafflin, A. L., and Ennis, D., *J. Cell. and Comp. Physiol.*, 1934, **4**, 283.

[2] Grafflin, A. L., and Eisenberg, M. J., *Anat. Rec.*, 1934, **59**, 449.

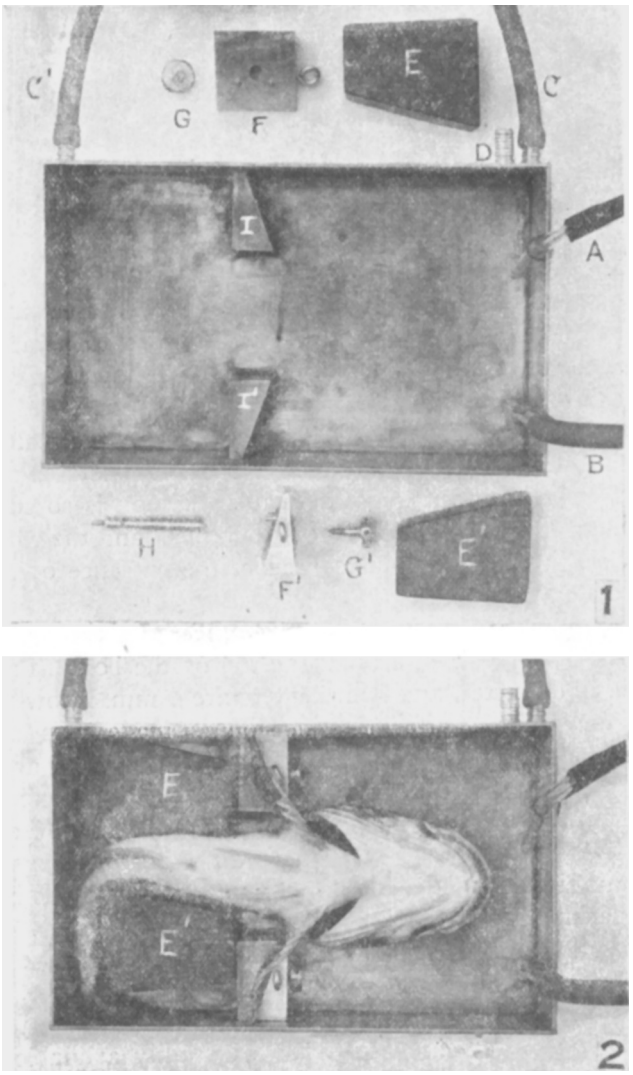

FIG. 1.
Operating box, unassembled, approximately × ⅓.
FIG. 2.
Operating box, assembled, with the sculpin in position for operation.

The over-all dimensions are: length 7⅜″, width 4$\frac{7}{16}$″, height 2$\frac{1}{16}$″. The sides were constructed of 2″ strip brass, the bottom of sheet brass, all $\frac{1}{16}$″ stock, and the joints were soldered. The box and all accessory parts were chromium-plated to prevent corrosion by sea water.

A—air inlet.

B—water inlet; the tube is held in a spring metal clamp.

C, C'—water outlets at the same level, the center of the lumen being 18 mm. below the top of the box.

D—accessory water outlet, with the center of the lumen 12 mm. below the top of the box. C, C' and D are of ¼″ brass tubing.

E, E'—heavy lead blocks for stabilization of posterior portion of fish. Length 5 cm., lesser width 2.6 cm., greater width 4 cm., thickness 2.4 cm.
F, F'—removable portions of pectoral fin clamps. Note the two projecting pins, which fit snugly into holes in the posts I, I'.
G, G'—screws for holding blocks F, F' in position.
H—key for tightening screws G, G'. Key fits into small holes in screw head (see G').
I, I'—permanent portions of pectoral fin clamps, attached by screws to bottom and sides of box. The complete pectoral fin clamp for one side (F and I) is made by sawing at an angle through a block of brass metal 30 mm. long, 19 mm. wide and 32.5 mm. high. The relative dimensions of the two portions are sufficiently indicated.

relatively large size of the anterior chamber allow perfect freedom of respiratory movements. It was found desirable to aerate the anterior chamber, and this procedure should be followed in all experiments.

In ordinary abdominal surgical procedures in the sculpin, it is preferable to have the operative field exposed by suitable retractors. However, if an assistant is not available, the box is so constructed that excellent exposure can be readily obtained, and the walls of the incision held sufficiently high to prevent the entrance of sea water into the abdominal cavity.

On careful examination of Figures 1 and 2 one can see a row of small holes closely spaced around the top of the box. These holes are 2 mm. in diameter, and their centers are 4 mm. below the upper edge of the sides. After the incision is made, ties are taken through the margins of the wound, and the threads are carried through various of these holes depending upon the exposure desired. After the proper tension has been exerted upon the threads, they are held so by inserting tapered, round wooden pegs into the holes. This simple manner of retraction was found to be invaluable in the experiments upon direct observation of the kidney. The box adapted itself readily to such experiments in which the Leitz Ultropak Microscope was used.

The sculpin has been extensively used for investigative work over a period of years, and, in the group of readily available teleosts, seems to be a rather ideal experimental animal. It is a skin fish with a yielding abdominal wall, adapting readily to ordinary surgical procedures. It is relatively resistant to asphyxia and anesthesia, and could be used with little difficulty for experiments involving prolonged microscopic observation of living organs *in situ*.