

## METHODS

### A REVERSIBLE METHOD OF BLOCKING THE NEOCORTEX BY COLD FOR USE IN LONG-TERM EXPERIMENTS

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A large part of the neocortex or individual regions of it can be reversibly blocked under chronic experimental conditions by means of capsules fixed to a cat's head. Cold liquid is passed through the capsules, reducing the cortical temperature beneath them to 23-20°, at which temperature the cortical electrical activity is blocked. The body temperature and the temperature of the subcortical structures remain virtually unchanged.

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The method of extirpation of the cerebral cortex which is widely used to study brain functions has many disadvantages, the most important of which is the development of retrograde degenerative changes in subcortical structures closely connected morphologically with the extirpated part of the neocortex. In addition, investigations on decorticated animals usually start sometime after the operation, when the defect resulting from cortical extirpation has been largely compensated. This explains the urgency for the introduction of new methods of cortical blocking which are purely functional and reversible.

Cold as a method of blocking the cortex has been used previously [1, 3, 4, 6, 7], but the methods adopted on these occasions did not prove popular.

The method now developed for complete and partial cold blocking of the neocortex consists essentially of the implantation of specially made capsules, which can be fixed above the region of the cortex to be blocked. Cold liquid is passed through the capsule, lowering the temperature of the brain in its vicinity to 22-17°.

**Preparation of capsules.** So that as wide an area of the cortex as possible could be blocked, it was found that capsules made of organic glass with the addition of a small quantity of plasticizer gives best results. To prepare the capsule, casts of the cat's brain were used, made by filling skulls of different sizes with plaster of Paris. An elastomer (composition: emulsified polymethyl methacrylate 43 g, acetone 57 ml, dibutyl phthalate 3 ml) was stretched over the cast. This element of the future capsule is its inner part, which is applied to the brain. The cast was next covered with organic glass, and over it a specially prepared lead mold, 3-4 mm in thickness, was fitted to its surface. This mold had a T-shaped projection on its convex surface from which inlet and outlet channels could be formed. Another similar elastomer was stretched over the lead mold. After 2-3 days to allow maturing of the polymer, the upper and lower elastomers were cut off the edge of the cast and the lead mold was removed. These layers were then glued together with dichloroethane glue. An organic glass block was then glued to the upper part of the capsule, and the holes in it were connected with the channels of the capsule (Fig. 1). To cool the cortex, the block was connected by rubber tubes (Fig. 2A) with a refrigeration unit, pumping liquid through the capsule.

When blocking separate parts of the cortex it was found that the best was to use metal capsules (Fig. 1A). These are made from brass foil 0.1 mm in thickness. To form the inner surface of the capsule a metal die was used, made from a plaster of Paris cast of this part of the brain. The outer surface of the capsule was made from another die. Inlet and outlet tubes were mounted in the cover. The outer and inner parts were joined together to form a space between them. The capsule was assembled by soldering and then chromium plated.

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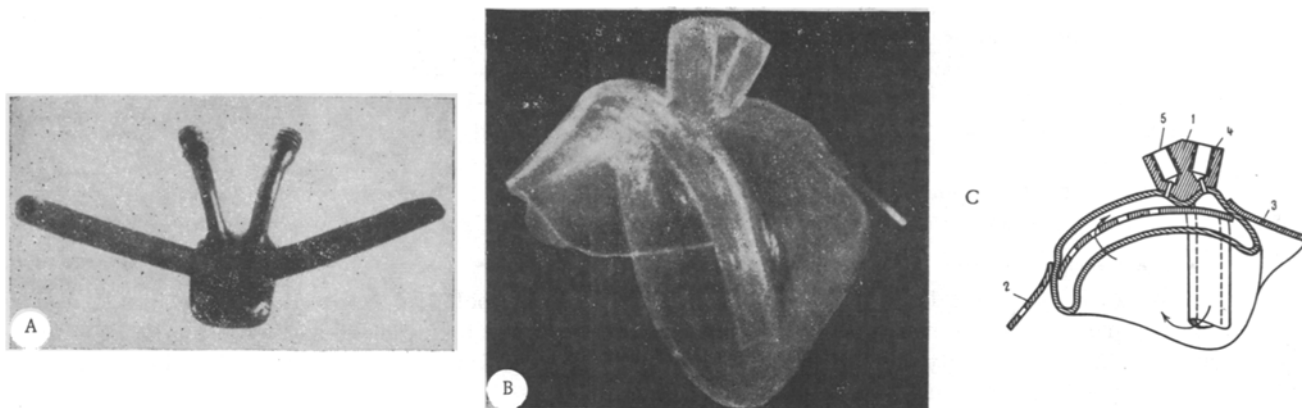


Fig. 1. Metal capsule for blocking temporal region of cortex (A) with plates soldered at the sides for fixation to the skull; plexiglas capsule for blocking the greater part of the neocortex (B) and diagram of this capsule (C). 1) Block; 2 and 3) organic glass plates for fixing capsule to cranial bones; 4) inlet orifice; 5) outlet; arrows indicate path of circulation of fluid.

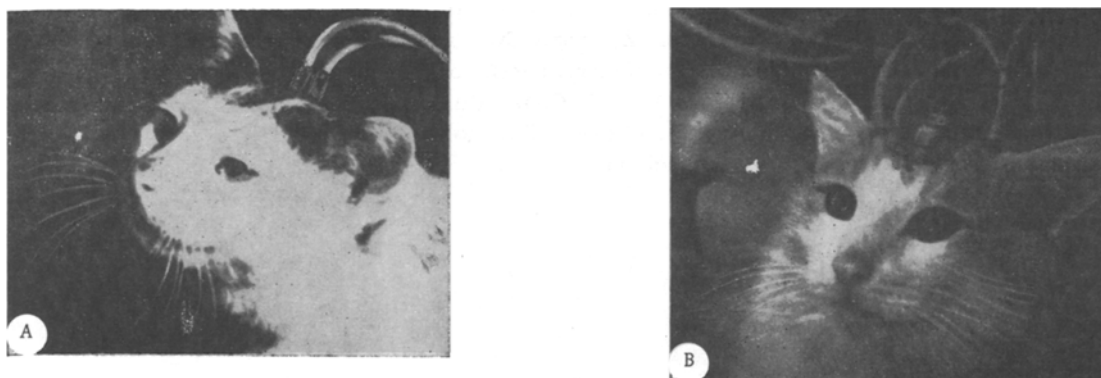


Fig. 2. Cat's head with implanted Plexiglas capsule (A) and with metal capsules implanted bilaterally over temporal cortex (B).

The plexiglas capsules were sterilized with ammonia vapor and ultraviolet irradiation. The metal capsules were sterilized by boiling.

**Operation.** The operation to fix the capsules was carried out under general anesthesia. To insert the "total" plexiglas capsule, the parietal bones and as much as possible of the occipital, temporal, and frontal bones were removed, leaving the dura intact. To insert the "local" metal capsules, smaller areas of bone above the region of cortex to be investigated were either removed or were thinned considerably by means of a dental drill. The "total" capsule was fixed with screws to the remnants of the frontal and occipital bones, and the "local" capsules by means of quick-hardening plastic (styracryl, butacryl, etc.) and screws. During the operation thermocouple pick-ups were introduced into the relevant parts of the cortex and sub-cortex. The leads must run without touching the capsule to the plexiglas contact block fixed to the remnants of the frontal bones, or otherwise inaccurate measurements of the brain temperature will result. The wound was sutured. Only the inlet and outlet tubes of the capsules and the contact block of the thermocouples were visible on the cat's head (Fig. 2B). The animal could be used for experimentation 1-1.5 weeks after the operation.

**Experimental results.** Experiments with cooling of the greater parts of the neocortex and of individual regions were carried out on 23 cats. The animals were placed in a chamber where they could move about freely. The capsule was connected to the cooling system and the thermocouple pick-ups to an electrothermometer for recording the temperature. Cooling was carried out with liquid at a temperature of between 1 and 3°. During the first 20-30 min of cooling the temperature of the whole thickness of the cortex fell to 22-19°, and during the next 40-50 min it was cooled by not more than 1-2°. The rectal temperature remained essentially unchanged.

During measurement of the brain surface temperature next to the capsule, and also at different depths from it, it was found that if the cortical temperature under the capsule fell during 40-60 min to 22-19°, at a depth of 4-5 mm (in the white matter) it fell to 28-25°, and at a depth of 8-10 mm, i.e., in the subcortex, it was the same as the animal's body temperature. The cortex was blocked only in the region where it was in contact with the capsule, for at a distance of 2-3 mm away from the edge of the capsule the surface temperature of the brain was reduced only to 28-27°, and at a distance of 10 mm it corresponded to the body temperature. After the end of cooling the temperature of the cortex returned to its initial level within 15-18 min.

According to data in the literature [3, 5], the function of the cooled cortex could be expected to be completely suppressed at a temperature of 21-20°. That this in fact took place was shown by the results of investigation of the cell activity of the neocortex (under acute experimental conditions), when the spontaneous unit activity stopped as the brain temperature fell to 23-22°.

The suggested method thus affords ample possibilities for physiological (reversible) blocking of the cortex as a whole and of various parts of it.

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