Stereotaxic presentation of subcortical aspiration¹

GEORGE H. GOLDEN, LAWRENCE D. MIDDAUGH, and JOEL F. LUBAR, UNIVERSITY OF TENNESSEE, Knoxville, Tennessee 37916

A method of producing subcortical brain lesions by the stereotaxic presentation of aspiration is described. Advantages of the method are noted and a description of the instrumentation is given.

The production of brain lesions of exacting size and reproducibility has been an impediment to research dealing with the localization of function. The traditional methods of lesion production are plagued with problems of scar formation, deposition of metallic particles (Reynolds, 1963), and the production of peripheral damage by heat dissipation in the case of radio-frequency lesions.

In 1967, Lubar, Wolfe, and Ison described a technique for producing septal lesions by subcortical aspiration. This procedure offered the advantages of being free from the deposition of ions and damage to surrounding structures. The resulting lesion is symmetrical with minimal scar formation.

A further refinement of the aspiration technique described here involves adapting it to a stereotaxic instrument, thus allowing the production of precise, reproducible lesions anywhere in the brain. Lesions involving large areas of the cortex as well as tract and nuclear lesions are equally facilitated by the use of stereotaxically placed aspiration.

METHOD

Apparatus

Any stereotaxic instrument can be modified quickly so that it can be used to implement the aspiration technique. In this study, we used the Kopf Model 1404 stereotaxic instrument modified for rats with the Model 1220 rat adapter and rat ear bars. All that is necessary for stereotaxic aspiration is to replace the standard electrode holder with a universal clamp (David Kopf, 1272 Universal Holder) fitted to a common tuberculin syringe barrel. See Figs. 1 and 2 for the syringe placement within the stereotaxic framework and its valve connection. The top of the syringe barrel is then connected, via surgical rubber tubing (1/4 in. o.d., 1/8 in. i.d.) to a vacuum pump capable of producing from 20-25 in. of vacuum (Gomco Surgical Manufacturing Corporation). Variable control over the amount of suction can be attained by inserting a valve somewhere along the length of rubber tubing. We have found that a very effective valve can be made from the barrel of a 5/4-in. disposable pipette (Medi-pak, General Medical Corporation). If the tip of the pipette is occluded in a Bunsen flame and then heat is applied to a spot in the middle of the pipette shaft, a hole can easily be blown in the tube wall. This hole should be from 1/4 in. to 3/8 in. in diam. The tapered end of the pipette can then be cut off and the end of the resulting hollow glass tube fire-polished. The resulting tube should be about 3 in. long, with a valve opening in the side about 1 1/2 in. from the ends. This control valve is then inserted into the surgical rubber tubing so as to connect the tube from the syringe to the tube from the vacuum pump. With this valve arrangement, one can control easily the amount of applied suction by partially or totally occluding the hole with a finger.

The actual aspiration pipette is fashioned from stock hypodermic needles by grinding off the point and finishing the tip in a 45-deg bevel. One can then regulate the magnitude of the lesion by merely varying the gauge of the hypodermic needle

Fig. 1. Instrumentation for producing stereotaxically placed aspirated lesions.

Fig. 2. Close-up of valve and pipette arrangement.

used. A 20-ga needle has been found to be quite satisfactory for general ablation work in the rat. In the case of cat surgery, a 16- or 18-ga needle is more appropriate.

The use of the glass syringe barrel has the advantage of allowing rapid changes of pipette size as well as presenting the E with a clear view of the aspirated material. It is also readily apparent when the flow of suction from the surgical pipette is in any way impeded so that the advance of the pipette can be slowed, stopped, or reversed as dictated by the situation. The technique is also potentially useful for cortical lesions, especially in animals with smooth brains, such as the rat.

For cortical lesions, a most effective pipette can be fashioned by (after grinding and beveling) compressing the tip of an 18-ga hypodermic needle into an elliptical opening. This pipette can then be swept over the area of the cortex to be ablated.

Fig. 3. Quadrant, unilateral, and bilateral lesions as represented in coronal sections. The section in the lower right-hand corner was prepared from fresh, unstained material.

Maintaining a distance between the pipette tip and the cortical surface of about 0.5 mm produces very clean lesions.

APPLICATION OF THE TECHNIQUE FOR THE PRODUCTION OF SEPTAL LESIONS IN THE RAT

Subjects

Twenty male Long-Evans rats, approximately 150 g in weight, were obtained from Blue Spruce Farms, New York. These animals were assigned randomly to three groups in which they received, respectively, anterior, middle, and posterior septal lesions.

Procedure

Surgery was done aseptically under sodium pentobarbital (50 mg/kg). The animals were prepared for surgery and placed in the Kopf stereotaxic instrument with the tooth bar set at zero. Both bilateral and unilateral lesions were made. All lesions were placed at an angle of 25 deg toward the midline, 2.5 mm lateral to the superior sagittal sinus, and 6.5 mm below dura. The anterior-posterior coordinates for the respective groups were: anterior, 2.8 mm anterior to midbregma; middle, 1.8 mm anterior to midbregma; posterior, 0.8 mm anterior to midbregma.

The degree of vacuum was set at 11 in. while the pipette was being lowered to the lesion site. When the lesion site was reached, the negative pressure was increased to 20 in. and was applied for 4 sec. The pipette was then withdrawn from the brain under 11 in. of suction. Three months after surgery, the animals were perfused with saline and 10% formalin in saline, and the brains were removed and prepared for frozen sectioning. They were sectioned at 40 microns and stained with Cresyl-Violet. Figure 3 shows representative sections.

Results

It can be seen that the lesions resulting from the stereotactically applied aspiration technique are quite clean and relatively free from scar formation. The pathways created by the lesion-producing pipettes in advancing to the brain area to be removed are minimal. Examples of puncture lesions through the cortex are shown in Fig. 4. These are even smaller than those described by Lubar, Schaefer, and Wells (1969), which were produced by hand-applied aspiration.

Fig. 4. Dorsal view of cortical puncture lesions.

DISCUSSION

With a minimum of experience, an investigator can produce a wide variety of lesions accurately and quickly. One person working alone can complete an entire bilateral septal ablation in 15 min. Compared with lesions produced by other techniques, the stereotaxically placed aspirated lesion is relatively free from scar formation and damage to peripheral structures.

REFERENCES

LUBAR, J. F., WOLFE, J. W., & ISON, J. R. Effects of medial cortical lesions on appetitive instrumental conditioning. *Physiological Behavior*, 1967, 2, 239-244.

LUBAR, J. F., SCHAEFER, C. S., & WELLS, D. G. The role of the septal area in the regulation of water intake and associated motivational behavior. *Annals of the New York Academy of Sciences*, "Conference on the Neural Regulation of Food and Water Intake": 1969, 157, 875-893.

REYNOLDS, R. W. Ventromedial hypothalamic lesions without hyperphagia. *American Journal of Physiology*, 1963, 204, 60.

NOTE

1. This investigation was supported by a United States Public Health Service grant, MH-14182-03, to Joel F. Lubar.