A Chronic Subcutaneous Gastric Cannula In Adult Rats¹

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ALTAR, A. A chronic subcutaneous gastric cannula in adult rats. PHARMAC. BIOCHEM. BEHAV. 12(4) 629-631, 1980.—The design and method of implantation for a chronic gastric cannula in adult rats is described which obviates the necessity of externalizing the peritoneal cavity or its contents. This procedure minimizes tissue damage, post-operative recovery time, and permits intragastric infusions without the oropharyngeal sensations present with nasopharyngeal gastric catheters.

Subcutaneous gastric cannula

Chronic preparation

INTRAGASTRIC infusions of experimental animals have been accomplished with rubber [1] or plastic [11] gavage tubes, nasopharyngeal catheters [2, 3, 5, 6, 9, 13] and, in the adult rat [2, 8, 10] or neonatal rat [7], via chronically implanted gastric cannulas. Gastric loading by gavage requires anesthetization or immobilization and either of these procedures are stressful to the animal. Anesthetization introduces pharmacological agents into the animal (e.g., ether) that may confound interpretations of experimental results. Use of the nasopharnygeal catheter, while relatively stress-free during stomach loading of freely moving rats, has several drawbacks. Intragastric injections of food [8] or water [2] via the catheter produce thermal and/or mechanical cues in the nasopharynx which appear to be crucial for the reinforcement of self-injection behavior. Implantation of the nasopharyngeal catheter is particularly difficult in young animals, due in part to the small diameter of the neonatal nasopharynx. Even in older rats, tubing of relatively narrow gauge must be used in order to prevent respiratory blockade. The narrow tubing lumen limits intubations to solutions of low viscosity and may become blocked with small digestive particles. Finally, gastric cannula implantation of adult rats has until now required relatively large peritoneal and gastric incisions and the use of purse string sutures [10,12] which may complicate post-operative recovery with infection and internal bleeding. The present report describes the design and method of implantation for a chronic subcutaneous gastric cannula in adult rats which allows the intragastric injection of a variety of materials without the stress of gavage or the necessity of intraperitoneal surgery. Hall's gastric cannula technique for neonatal rats [7] has been modified here for use with adult rats and these modifications differ significantly from Hall's technique in design and most procedural steps.

METHOD

Male albino rats of the Sprague-Dawley strain (338-495 g; Simonsen Laboratories, Gilroy, CA) are housed individually in an animal colony room maintained at $22\pm2^{\circ}$ C and 50% relative humidity. Animals are usually 90-120 days of age at the time of surgery, although animals as young as 40 days of age can be used with the technique described here.

Surgery

Animals

Animals are injected intraperitoneally with sodium pentobarbital (Nembutol, 50 mg/cc, 0.9 cc/kg; Abbott Laboratories, Chicago, IL) and with atropine sulfate (0.4 mg/cc, 0.5 cc/kg; Eli Lilly and Co., Indianapolis, IN) to prevent respiratory blockage by mucous. The abdominal and scalp regions are closely shaved.

A 16 cm guide tube (Fig. 1A) of PE 205 ga tubing (i.d. = 1.4mm; o.d.=1.9 mm) is pushed down the esophagus, passes the esophagogastric sphincter and enters the stomach. The guide tube is slightly curved so that it can be directed towards the left inside wall of the stomach. The guide tube is gently pushed and slowly rotated until the end makes contact with the stomach wall and can be felt by touching the skin over the abdomen. The location of a contact is marked on the skin with an ink pen and the guide tube removed. After a 5 mm incision has been made at the ink mark, the guide tube is reinserted in the mouth and pushed down the esophagus until it makes contact with the stomach wall underneath the incision. The guide tube is held in place while a 25 cm section of 27 ga stainless steel wire (Fig. 1B), sharpened to a coneshaped tip, is inserted down the guide tube. The wire tip penetrates the stomach wall and is passed through the muscle and fascia to appear externally at the incision. A 45 cm

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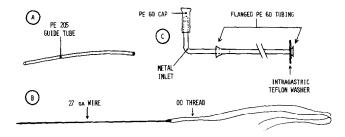


FIG. 1. Specially prepared parts necessary for chronic gastric cannula surgery. See text for physical dimensions, materials used, and methods of fabrication.

loop of 00 surgical thread, run through a closed loop (5 mm long; 2.5 mm wide) at the unsharpened end of the wire, is pulled down the guide tube and through the abdominal incision as the wire is pulled out of the abdominal incision. The protective guide tube is then removed via the mouth. This leaves the double strand of surgical thread running from the mouth, through the esophagus and stomach, and exiting at the abdominal incision. One loop of the thread is pulled out of the animal via the incision. The oral end of the remaining strand is tied securely to the end of the cannula, a 25 cm section of PE 60 tubing (i.d. = 0.76 mm; o.d. = 1.2 mm). Previously, the other end of the cannula has been heated to produce a 3 mm diameter flange and a flexible teflon washer has been cut with cork boring tools to the following dimensions: 0.20-0.25 mm thick; inside diameter=2 mm; outside diameter=3.75 mm. The teflon washer is placed in front of the flange which prevents the washer from slipping off the tube (Fig. 1C). The cannula is lightly coated with mineral oil and is drawn down the esophagus by pulling the surgical strand exiting at the incision. This causes the washer to slide past the epiglottis and cardiac pylorus and enter the stomach. The cannula is pulled out the abdominal wound until the washer and cannula terminal can be felt to lodge on the inside of the gastric mucosa

A 2.5 cm midline incision is made in the scalp. The connective tissue is removed with a bond scraper and the area allowed to dry. The exposed cannula tubing is washed in 80% ethanol and then pulled subcutaneously from the abdominal incision to the scalp would with the aid of a stiff wire pressed tightly into the cannula lumen. It is important that an additional 2.5 cm of cannula tubing be included between the abdominal and scalp incisions in order to provide slack for animal movements and body growth following the operation. The cannula terminal is fitted to the arm of a right-angle, blunted 2.5 cm length of 20 ga stainless steel tubing. The metal outlet is cemented to the skull with acrylic dental cement anchored to three stainless steel skull screws (Fig. 2). The scalp wound is sutured with 00 surgical thread. The abdominal wound is closed with two wound clips or with surgical steel suturing wire and the animal is allowed to recover for one week before experimental use.

Post-operative Care

Animals typically feed and drink normally within two days after surgery and body weights return to pre-operative levels within one week after the operation. Cannulas should

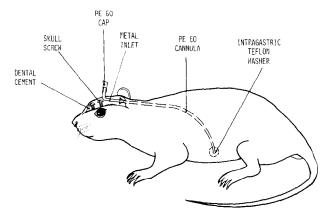


FIG. 2. Side view of complete gastric cannula assembly *in situ*. Head mount is shown as if uncovered by skin and as if clear dental acrylic had been used to cover skull screws and metal inlet.

be flushed with distilled water once a week and after the injection of any solution. When the cannula is not in use, the exposed metal inlet should be capped by a heat sealed 1 cm section of PE 60 tubing (Figs. 1C and 2).

DISCUSSION

The chronic gastric cannula described here has been used with eight animals receiving intragastric injections of distilled water during operant responding [2] and is presently being used with 30 animals receiving intragastric injections of carbohydrates or their metabolites and radioactively labelled tryptophan. A 1, 3, or 10 cc syringe is connected to the animal via a 10–100 cm length of PE 60 tubing and injections are always made while the animal moves freely on a laboratory counter or cart.

Successful gastric cannula implantation requires experience with small animal surgery and at least several practice implantations. Experience has shown that following extensive surgical practice, about 85% of all operated animals survive with intact cannula assemblies for at least one month. Cannula system failure is almost always due to either loosening of the head mount from the skull or from infection along the subcutaneous cannula path. Dermal ruptures resulting from infection may expose the cannula tube which may be chewed by the animal. The frequency of infection can be attenuated with antibiotic treatment of the animal immediately after and two weeks following surgery. Head mount stability is promoted by tightly inserting all three skull screws and thorough scrapping of the skull and connective tissues before dental cement application.

In summary, the present technique is sufficient for the implantation of a chronic subcutaneous gastric cannula in adult rats that is well suited to the injection of a large variety of solutions. This technique is an improvement over previous adult gastric cannulation techniques by virtue of the ease of cannula insertion, the small amount of tissue damage, and the opportunity it affords the investigator to intragastrically infuse solutions without concomitant nasopharyngeal stimulation, the stress of gavage, or long-term surgical recovery.

REFERENCES

- 1. Adolph, E. F. Thirst and its inhibition in the stomach. American J. Physiol. 161: 374-386, 1950.
- 2. Altar, A. and H. J. Carlisle. Intragastric drinking in the rat: Evidence for a role of oropharyngeal stimulation. *Physiol. Behav.* 22: 1221-1225, 1979.
- 3. Borer, K. T. Disappearance of preferences and aversions for sapid solutions in rats ingesting untasted fluids. J. comp. physiol. Psychol. 65: 213-221, 1968.
- Deutsch, J. A. and W. T. Hardy. Ethanol tolerance in the rat measured by the untasted intake of alcohol. *Behav. Biol.* 17: 379-389, 1976.
- 5. Epstein, A. N. Water intake without the act of drinking. *Science* 131: 121-124, 1960.
- Epstein, A. N. Oropharyngeal factors in feeding and drinking. In: Handbook of Physiology, Vol. 1, Alimentary Canal. Section 6, edited by Charles F. Code. Washington, DC: American Physiological Society, 1967.
- Hall, W. G. Weaning and growth of artificially reared rats. Science 190: 1313-1315, 1975.

- 8. Holman, G. L. Intragastric reinforcement effect. J. comp. physiol. Psychol. 69: 432-441, 1968.
- 9. Kissileff, H. R. Oropharyngeal control of prandial drinking. J. comp. physiol. Psychol. 67: 309-313, 1969.
- 10. Miller, N. E. and M. L. Kessen. Reward effects of food via stomach fistula compared with those of food via mouth. J. comp. physiol. Psychol. 45: 555-564, 1952.
- 11. Myers, R. D. General laboratory procedures. In: Methods in Psychobiology. Vol. 1., Laboratory Techniques in Neuropsychology and Neurobiology, edited by R. D. Myers, New York: Academic Press, 1971, pp. 27-65.
- Nicolaidis, S., N. Rowland, M. Meile, P. Marfaing-Jallat and A. Pesez. A flexible technique for long term infusions in unrestrained rats. *Pharmac. Biochem. Behav.* 2: 131-136, 1974.
- 13. Teitelbaum, P. and A. N. Epstein. The role of taste and smell in the regulation of food and water intake. In: *Olfaction and Taste*, edited by Y. Zotternam. London: Pergamon Press, 1963, pp. 347–360.