inhibited, while adjacent segments, either without application or with pure lanolin applied, abscised normally. Only the portion of the segment to which the MH was applied remained white and turgid; the rest browned and dried in the normal length of time.

Cytological examinations of untreated perianth segments were made at intervals during the life of the flower to ascertain the amount of starch normally present. There is an abundance of starch in the parenchyma cells of the segments during the first day. During the second day, starch decreases rapidly; in the early morning, there is as much present as on the first day; at noon, there is about one-half as much; and in the evening, starch is absent or nearly so. Examination of flowers in later stages showed starch to be absent. In treated material, however, considerable starch remains for a longer period of time.

The foregoing observations indicate that the ability of the MH to retard maturation processes is related to the amount of starch present at the time of application. There is no noticeable effect when starch grains are absent or nearly so. The retardation of starch digestion in the presence of MH may account in part for the lowered respiration observed by others. Further experimental work is in progress.

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Received February 26, 1954.

Intestinal Intussusception in Chronic Nephrectomized Dogs Maintained by Peritoneal Dialysis*

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Despite widespread interest in clinical aspects of intestinal intussusception, few experimental studies have been made. These have consisted of the deliberate production of intussusceptions by direct stimulation of the animal intestine by faradic current, acetylcholine, salt crystals, and actual manual manipulation (*1*), and by faradic stimulation of the premotor area of the monkey cerebral cortex (*2*). Intussusceptions have been cited as a frequent complication in very young dogs following a constricting ligature of one renal artery (*3*) but have never been encountered by Goldblatt in some 1500 renal hypertensive dog preparations (*4*).

This paper presents an incidence of intestinal intussusception as a complication in 11 of 36 (30.6 percent) chronic bilaterally nephrectomized dogs maintained for from 6 to 111 days on a low-salt diet by intermittent peritoneal dialysis as described elsewhere (*5*). Either a 17-gage 3½-in. needle or P.E. 320 polyethylene tubing was used in the abdominal cavity for the dialysis (*6*). This incidence is believed to be the highest for intestinal intussusception so far reported as an experimental complication. Muirhead *et al.* (*7*) found intussusceptions in 11 percent of 71 nephrectomized dogs untreated or subjected to the artificial kidney.

Nine of the 11 intussusceptions were single, and two were dual; 10 were forward, and one was retrograde. According to the classification reported by Finkelstein (*8*), four were enteric, four ileocolic, one colic, and two entericileocolic (dual). They occurred on post-nephrectomy days 2, 4, 5, 6, 7, 8, 8, 10, 11, 15, and 35. Six intussuscepta were necrotic and presumably not recent; five were moderately to marked congested and presumably were recent. Five of the dogs were sacrificed when obstruction appeared imminent; six were found dead. In three of the ileocolic cases, the dark red gangrenous ileal intussusceptum protruded through the anus as far as 6 in. An intussusception was, for the most part, manifest by failure to retain food, by watery bile-stained vomitus and yellow-tan colored watery diarrhea, gradually becoming tarry and frankly bloody when the lower tract was involved. The intussusceptum was palpated rectally on four occasions and was diagnosed fluoroscopically on another (*9*).

Ten normal dogs were maintained on the same diet and dialysis schedule as the nephrectomized series for from 9 to 12 days, then were kept under observation for up to 18 days. No intussusceptions occurred. Consequently, such technique of maintenance alone was probably not the cause of the intussusceptions.

Although the cause of the intussusceptions was not readily apparent in this study, it could have been related to the irritation of the gastrointestinal tract, manifest in some of the dogs by hyperemia, congestion, and petechial hemorrhages in the mucosa, presumably due to some toxic substance incident to the absence of the kidneys. The combination of such irritation with visceral peritoneal irritation from the indwelling dialysis fluid might have created certain hypermotal regions of the intestine which acted as a nidus for an intussusception.

The possibility of bacterial contamination from the dialysis fluid as a cause of intussusception has been considered but insufficiently studied to warrant any definite conclusions. A preliminary investigation of this point was recently carried out on only a few of the aforementioned animals, and the data are reported elsewhere (*10*). Counts of bacterial colonies in cultured dialysis fluid before administration into, and after retention in, the abdominal cavity for 1½ to 2 hr were determined in 7 control dogs. All solutions put into the abdomen contained per liter the usual 25 mg of streptomycin and 25,000 units of crystalline

* Supported in part by grants from the American Heart Association, the National Heart Institute, U.S. Public Health Service, and Ciba Pharmaceutical Company.

penicillin used throughout the study and were sterile. In six animals, all withdrawn samples were sterile; in one, severely contaminated. Four of the foregoing six animals with sterile dialysis fluids were subsequently nephrectomized. One developed an intussusception. All withdrawn dialysis fluids initially were contaminated but subsequently could be made sterile by the intramuscular injection of penicillin. Nephrectomy appears to lower the resistance of the animal to bacterial contamination (*10*). Since the nature of the contaminant was not investigated, no definite statement concerning the source of such contamination can be made at this time.

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6. This work would have been impossible without the tireless efforts of the following, who carried out the dialyzing and feeding of the dogs: Dr. Max Wyatt, Dr. James B. Reddick, Robert Ford, Lowry Kirby, Milton Davis, George Bryan, Irving Fleming, Royal Lea, and Frank Giglio.
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Received March 5, 1954.

A Method of Making Prefilled Microelectrodes

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In making microelectrodes for intracellular recordings, the greatest difficulty encountered is in the step of filling the tip with electrolyte. The method described by Ling and Gerard (*1*) uses fairly vigorous boiling to drive out the air, but this process destroys a large percentage of the microtips. Nastuk (*2*) has recently described another method in which boiling is avoided, but it requires several delicate manipulations.

The difficulty of filling the microelectrode can be circumvented by drawing the tips from capillary tubes that have been previously filled and maintained in a closed electrolyte system during the pulling. A specific application in connection with the Gamma pipette puller will be described. In this instrument, which is operated by gravitational force, the capillary is in a horizontal position and lengthens as heat is applied to it by a hot wire. For drawing prefilled microelectrodes, lucite blocks carrying deep wells filled with the electrolyte solution in use are attached to the jaw elements of the puller (Fig. 1). Capillary tubing of

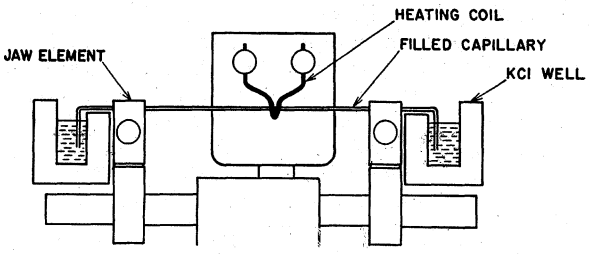

Fig. 1. Diagrammatic representation of the prefilled glass capillary in the Gamma pipette puller modified by adding a lucite well at each of the jaw elements.

appropriate lengths is bent at both ends. The bent tubing is first filled with the electrolyte solution and then is inserted into the jaw elements of the puller with the ends dipped in the reservoirs.

When heat is applied to the middle of the filled capillary, thermal expansion expels the fluid from the heated region into the wells. As the capillary is pulled apart to form the microtips, these are again filled by the electrolyte drawn from the reservoir. The filled electrodes are then immediately removed from the clamps and are hung vertically with the tips downward. This allows any large air pockets to rise toward the coarse end of the capillary where they can be dealt with easily.

In about 40 percent of the microelectrodes made in this manner, small air bubbles remain in the shaftlet or in the tip. However, when these electrodes are stored for a few days on a rack in a container filled with the electrolyte, most of the small air bubbles disappear. Thus most of the microelectrodes become usable without further manipulation.

Several other variants of this technique have been used successfully with the Gamma puller and other instruments. Microelectrodes with 1.0 to 2 μ tips are made with the greatest of ease from capillary tubes of 0.7 mm outside diameter. For tips smaller than 1 μ,

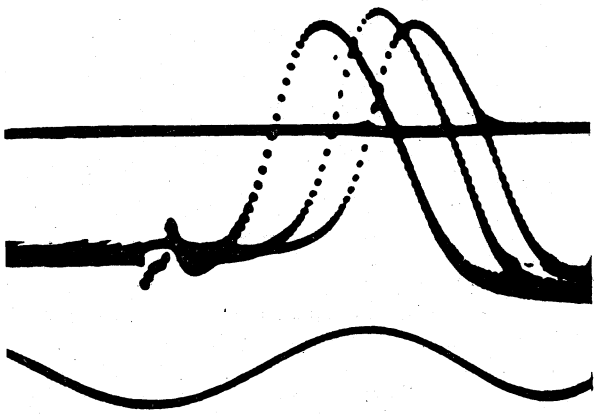

Fig. 2. Simultaneous records of activity at three microelectrodes inserted into a squid giant axon of diameter 480 μ. The distances between the electrodes (from left to right) were 3.7 and 2.5 mm. The resting potential (50 mv) is indicated by the displacement of the three electrode traces from the base line. Spike heights, 94 to 100 mv. Time curve, 1000 cy/sec.