



Fig. 2. High-power ($\times 600$) photomicrograph of similar preparation to that shown above. Masses of stainable fat (black) are deposited in all portions of the arterial wall. Lipid in intimal endothelial cells is in the form of fine droplets.

that received a high-fat, high-choline ration containing cholesterol. Lesions developed more rapidly in similar rats that received 1 percent NaCl in their drinking water and were given anti-rat-kidney serum, desoxycorticosterone acetate, and sesame oil injections. The arterial lesions were directly associated with the renal damage that these measures produce. Since choline exerted no protective effect in Wissler's experiments, it is possible that the lesions he has observed and those reported here are not closely related.

The lesions that McCormick and Holman (7) have described in dogs fed a high-fat diet following the production of a "standard" amount of renal damage may be more closely related to those that we have observed in choline-deficient rats. It would be interesting to know whether high levels of dietary choline would exert any protective influence on the vascular lesions seen in Holman's type of experiment.

Subsequent to our reports (1) concerning the production of vascular lesions in choline-deficient rats, Mann *et al.* (8) have produced atherosclerosis in *Cebus* monkeys by giving them large amounts of cholesterol in methionine-deficient diets. Methionine supplements prevented atherosclerosis in control animals. Mann has not specifically investigated the effects of choline deficiency on the vessels of monkeys, and it is possible that he is observing a manifestation of the lipotropic action of methionine in his experiments.

Earlier reports from our laboratory concerning the role of choline in the development of cholesterol-induced atheroma were limited to investigations in rabbits in which choline was added to a basal diet that already contained appreciable amounts of lipotropic factors. The effects of diets very low in lipotropic factors were not studied in that species (9).

We believe that the results of these studies using the rat indicate that an adequate intake of choline favors the maintenance of not only a healthy liver and kidney but also a normal cardiovascular system in young rats. Recent *in vitro* studies by Artom (10) have shown that the rate of fatty-acid oxidation by slices of livers, kidneys, and hearts of choline-deficient rats is enhanced if the animals are given choline just prior

to the time of sacrifice. Morphologic lesions in all these tissues have been found to begin with stainable deposits of fat which precede the irreversible changes associated with cirrhosis, renal damage, and cardiac necrosis.

If it should prove that the vascular lesions reported here are primarily due to a relative lack of lipotropic substances in the diet, another of the factors necessary to maintain the healthy state of the walls of blood vessels will have been recognized. It will obviously be necessary to confirm these findings on other species of laboratory animals before their possible clinical significance is considered.

We suggest that the term *lipotropic* may now be extended to include the prevention of the accumulation of abnormal (stainable) fat not only in the liver and kidneys but also in the heart and blood vessels of animals that exhibit these lesions when choline and its precursors are withdrawn from the diet.

References and Notes

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Retardation of Maturation in Magnolia Flowers by Maleic Hydrazide

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It has been shown that MH (maleic hydrazide) inhibits or retards several processes in plants. Cell division is inhibited in the root tips of *Vicia faba* (1) and onion (2); respiration is retarded in onions through partial inactivation or inhibition of one or more of the dehydrogenases (3); softening of apples is retarded and respiration is reduced, as measured by the quantity of CO_2 given off (4); and in some cases MH appears to act as an antiauxin (5). Evidence is presented in this paper to show that MH retards the aging processes in the flowers of *Magnolia grandiflora* L., the ability of MH to inhibit these processes being correlated with the quantity of starch in the perianth segments at the time of application.

In the Los Angeles area, the flowers of *Magnolia grandiflora* L. last 4 to 5 days. During the first day, they open partially, forming a loose bud (Figs. 1, 4). Anthesis occurs during the morning of the second day, the pollen being shed and the stamens abscised by evening (Fig. 2). During the subsequent 2 or 3 days, the perianth segments turn brown, generally being a deep brown and partially dry by the time of abscission on the fourth or fifth day.

MH-30 and MH-40, commercial preparations of the diethanolamine salt, were used in these experiments. MH-30 was painted on the perianth segments, in concentrations of from 0.1 to 15.0 percent, at various times throughout the principal blooming period. Concentrations of less than 5.0 percent had no noticeable effect. When applied in concentrations greater than 5.0 percent during the first day (loose buds), the areas painted remained white and turgid throughout the life of the flower. These areas were sharply defined, usually being limited to that portion actually touched by the MH-30. The segments were collected at time of abscission and kept under normal atmospheric conditions. Fifteen days after abscission the untreated portions of the segment were deep brown and crumbled, while the treated portion was cream colored and leathery.

Applications made during the second day produced decreasing effects: those made during the early morn-

ing hours produced a reaction almost as pronounced as those of the first day, while applications made at noon produced more localized effects of shorter duration. There was no noticeable effect when MH was applied in the evening of the second day or during the remainder of the life of the flower.

Flowers were sprayed with a 10-percent solution of MH-40 at various times throughout the normal blooming period. The normal maturation processes were inhibited in the flowers that were sprayed during the first day (loose buds) (Figs. 4-6). These flowers did not open further, and the perianth segments remained white except for the edges and some small spots, the processes causing browning of the perianth segments being inhibited (Figs. 5, 6). The normal abscission of the perianth segments was also inhibited. The immature fruits abscised in from 3 to 4 wk, and at that time some segments, although partially brown and dry, were still attached.

Flowers sprayed during the afternoon of the second day showed only slight retardation of the browning processes, generally being lighter brown at the time of abscission. Perianth segment abscission was not retarded or only slightly so. No effects were noted on flowers sprayed during later stages of maturation.

MH-30 in lanolin was applied in concentrations of 8 percent at the base of some perianth segments during the first day. Abscission of these segments was



Figs. 1-3. Untreated flowers of *Magnolia grandiflora* L., showing stages in development: (1) first day, loose bud; (2) second day, anthesis complete and stamens in process of abscising; (3) fifth day, perianth segments abscised the previous evening. Figs. 4-6. Flowers sprayed with 10 percent solution of MH-40: (4) first day, flowers at time of application; (5) second day; (6) fifth day. Both flowers opened the same day. Photographs were taken at noon.

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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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 in-

tussusception 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 hypermotile 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

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