

must be kept in mind in any evaluation of the factors involved in the pathogenesis of human hypercholesteremia.

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Growth of the Scutellum of Maize in Culture

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The discovery (1) that maize embryos of small size upon excision gave rise to small plants, and that those of larger size at time of excision gave larger ultimate sizes of embryos, and Merry's (2) evidence of the same situation in barley embryos suggest that if embryos could be held in an embryonic condition until they had grown beyond the normal size of embryos in mature seeds, and then planted, they should give rise to seedlings and mature plants of abnormally large size. Kent and Brink (3) had considerable success in preventing germination of barley embryos and in continuing their embryonic growth by the use of tomato juice in culture.

In trials with maize, tomato juice had little or no effect on embryos, although it did serve to expedite the growth of corn endosperm in culture (4). Any check in germination of the embryos was so slight as to be of no importance in producing embryos of abnormal size.

Since the root and the shoot could not be kept from growing out, thus producing germination and setting up seedling growth, rather than the desired embryonic growth, it seemed that the scutellum at least might be led to continue embryonic growth by the removal of the root-stem axis. Following this idea, numbers of embryos of the variety Evergreen, which were nearing mature size, were excised and the root-stem axis was removed from each. At the same time embryos of Golden Bantam variety were treated in the same way, but these were in an earlier stage of growth than the Evergreen. One set of each variety was grown on White's medium and one on White's medium plus tomato juice, as used by Kent and Brink.

A set of Evergreen embryos was weighed and meas-

TABLE 1
WEIGHTS OF CULTURED, CONTROL, AND RIPE
SCUTELLI OF MAIZE

| Variety | Treatment | Av wt (mg) | |
|---------------|-----------|------------|-------|
| | | Wet | Dry |
| Evergreen | Cultured | 977.3 | 224.2 |
| " | Control | 171.0 | 126.8 |
| " | Ripe | 393.9 | 154.3 |
| Golden Bantam | Cultured | 90.0 | 27.3 |
| " | Control | 18.9 | 13.3 |
| " | Ripe | 252.6 | 150.0 |

TABLE 2
LENGTHS AND WIDTHS OF CULTURED, CONTROL, AND
RIPE SCUTELLI OF MAIZE

| Variety | Treatment | Av length (mm) | Av width (mm) |
|---------------|-----------|----------------|---------------|
| | | | |
| Evergreen | Cultured | 12 | 9.0 |
| " | Control | 5 | 4.5 |
| " | Ripe | 6.5 | 6.0 |
| Golden Bantam | Cultured | 5.5 | 3.0 |
| " | Control | 3.0 | 2.0 |
| " | Ripe | 6.0 | 5.0 |

ured to serve as controls, then dried and weighed to give the dry weight. Golden Bantam embryos were treated in the same way. It is obvious from Table 1 that the Golden Bantam embryos were a great deal younger than those of the Evergreen. The cultures were continued until growth ceased. Wet and dry weights and measurements of length and width of the scutelli were obtained.

For comparison with the cultures and controls, ripe grains of the two varieties were soaked and germinated. When the embryos were fully expanded the root-stem axes were removed from them, and wet and dry weights and length and width measurements were taken.

In the cultures a number of scutelli formed outgrowths that were partial roots or partial stems. These growths were apparently due to incomplete excision of the root-stem axis rather than to regeneration. All such scutelli were removed from the experiment. No true regeneration of scutelli was seen, but some scutelli did develop papillate or even calluslike outgrowths on their surfaces. More outgrowths were seen on the medium with tomato juice than on the plain White's medium, but the differences were not great and both sets were thrown together in the tables.

The scutelli of Golden Bantam corn grew fairly well and showed nearly a fivefold increase in net weight and more than a doubling of the dry weight. They never, however, approached the size or weight of the scutelli from ripe seeds—a result, probably, of excision at a stage definitely too young.

The Evergreen scutelli made a much better showing and at the end were nearly twice as large and as heavy as those from normally ripened seeds.

Tables 1 and 2 show that one can take scutelli from

embryos near mature size and, by removing the meristems, thus making them incapable of germination, carry the scutelli on in growth of an embryonic type until maximum size. Since these cultured entities lack any organized meristems, they do not give positive evidence that if whole embryos could be held back in development and induced to continue embryonic growth they would eventually be able to produce mature plants of abnormally large size, but they do at least suggest this possibility.

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Perchloric Acid Extraction of Ribose Nucleic Acid from Cytological Preparations

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Ogur and Rosen (1,2) have reported a method for differential extraction of ribose and desoxyribose nucleic acids from tissues. They find that treatment of tissues with 10% perchloric acid at 4° C for about 18 hr will selectively extract ribose nucleic acid. Desoxyribose nucleic acid may then be removed by treating with 5% perchloric acid at 70° C for 20 min. Cytochemical adaptations of this technique have also been reported by Erickson, Sax, and Ogur (3) in plant material and by Seschachar and Flick (4) in protozoan material. These authors found that cold perchloric acid treatment does in effect remove the basophilia due to ribose nucleic acid from the cytoplasm and from nucleoli. Koenig (5), however, found that a somewhat higher temperature is necessary to remove basophilia from cytoplasm and nucleoli of formalin-fixed mammalian tissues, which was also found to be true by the author (unpublished) for Carnoy-fixed mammalian material. Sulkin and Kuntz (6), working with Zenker-fixed mammalian tissues, reported that cold perchloric acid treatment has no effect on deparaffinized sections. They did find that when the cold perchloric acid is allowed to act on the fixed tissue before it is embedded, or if it is allowed to act on frozen sections of the fixed material, this method can be used as a substitute for ribonuclease digestion in the removal of ribose nucleic acid from cytological preparations. In this laboratory perchloric acid extraction of ribose nucleic acid from cytological preparations and its effect on desoxyribose nucleic acid during this time were studied photometrically. Results of ultraviolet absorption studies and of absorption studies of Feulgen-stained sections are herein presented.

For these studies, rat pancreas was fixed in Carnoy's acetic alcohol (1:3), sectioned at 3 μ , and all slides were prepared from the same ribbon.

The method of photometric analysis employed throughout the experiment has been described in detail by several authors (7-11). The apparatus used is essentially the same as the one described by Pollister and Moses (12).

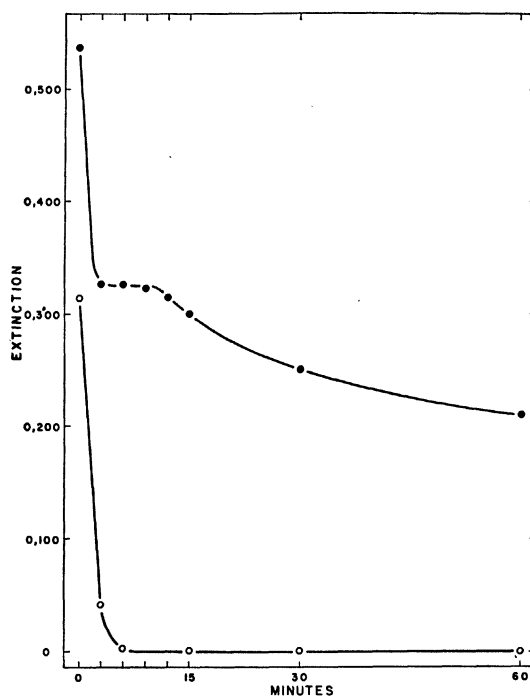


FIG. 1. Curves showing the relationship between ultraviolet extinction and duration of perchloric acid treatment: ●—●, curve for nuclei; ○—○, curve for cytoplasmic cores. Each point represents a mean extinction value of 15-25 measurements.

The natural ultraviolet absorption of nuclei having a diameter of about 6 μ , and of cores of cytoplasm 4 μ in diameter, was measured in sections treated for varying periods of time with 10% perchloric acid at 25° C. In order to study the effect of the perchloric acid treatment on the desoxyribose nucleic acid, after the perchloric acid treatment, the sections were stained by means of the Feulgen reaction. Sections were hydrolyzed for 12 min at 60° C in 1 N hydrochloric acid (8,9), stained for 1 hr in Schiff reagent, prepared according to the directions published by Stowell (13), and measurements were then made of the intensity of the Feulgen dye bound by the nuclei. In addition, photometric measurements were also made of Feulgen control slides which were placed in Schiff reagent together with the test slides. Except for hydrochloric acid hydrolysis, these were treated in the same manner as were the test slides.

A Photovolt photometer (Model 512) with an electron multiplier tube was used for measuring the ultraviolet absorption of nuclei and cytoplasmic cores. The light source for these measurements was the 2537 A