

tinguishable. The drop toward negativity is faster, and the negativity obtained is greater on the average for the group who had experienced myocardial infarction prior to the age of 40 than for their comparable "normal" control group. The drop toward negative values is slower, and negativity is not obtained on the average for a group known to be mentally defective.

#### References and Notes

1. N. C. Turner and G. E. Crowell, *J. Dental Research* **26**, 99 (1947).
2. M. M. Gertler and P. D. White, *Coronary Heart Disease in Young Adults* (Harvard Univ. Press, Cambridge, 1954).

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4. L. F. Hewitt, *Oxidation-Reduction Potentials in Bacteriology and Biochemistry* (Williams & Wilkins, Baltimore, 1950).

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## Communications

### Preliminary Studies on the Structure of Angiotonin

Angiotonin of a purity 2 to 3 times that of Edman (1) has been obtained by utilizing sodium chloride fractionation, adsorption on and elution from Amberlite IRC-50, and finally partitioning on a Celite column using a butanol-propanol : sodium chloride-acid solvent system (2). By partitioning in a countercurrent apparatus between butanol-propanol and 0.1*N* hydrochloric acid, an angiotonin was obtained of even greater purity. Only one active pressor principle was evident after 100 transfers. Angiotonin prepared in the afore-described manner was used in the following analyses.

A two-dimensional chromatogram (Fig. 1) of a hydrochloric acid hydrolysate shows 13 different amino acids to be present in the angiotonin molecule. The leucine spot was later shown, by a Dowex-50 column (3), to contain both leucine and isoleucine, making a total of 14 different amino acids.

The amino end-group was determined by the 1:2:4-fluorodinitrobenzene (DNP) method of Sanger (4). The ether soluble fraction from the acid hydrolysis of DNP angiotonin was chromatographed by the method of Biserte and Osteux (5) and found to contain only

DNP aspartic and some dinitroaniline. Only  $N^5$  DNP lysine was observed in the water-soluble fraction. The fact that only one amino derivative was found is evidence that the angiotonin used here is mainly one entity.

By hydrazinolysis (6) of angiotonin, the amino acid on the carboxyl end could be isolated as the free amino acid. It was then chromatographed (two-dimension) by the same procedure as is illustrated in Fig. 1 and shown to be either leucine or isoleucine.

Chromatographic analysis of the hydrolysate, using a Dowex-50 column (3), gave the amino acids shown in Fig. 1 in the following molecular ratio: 2 aspartic acid, 1 serine, 1 glutamic acid, 2 proline, 1 glycine, 1 alanine, 1 valine, 1 isoleucine, 1 tyrosine, 1 phenylalanine, 2 leucine, 2 histidine, 1 lysine, 2 arginine.

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#### References

1. Pehr Edman, *Arkiv Kemi, Mineral. Geol.* **22**, 1 (1945).
2. F. M. Bumpus, A. A. Green, and I. H. Page, *J. Biol. Chem.*, in press.
3. S. Moore and W. H. Stein, *J. Biol. Chem.* **192**, 663 (1951).
4. F. Sanger, *Biochem. J.* **39**, 507 (1945).
5. G. Biserte and R. Osteux, *Bull. Ste. Chim. Biol.* **33**, 50 (1951).
6. S. Akabori, K. Ohno, and K. Narita, *Bull. Chem. Soc. Japan* **25**, 214 (1952).

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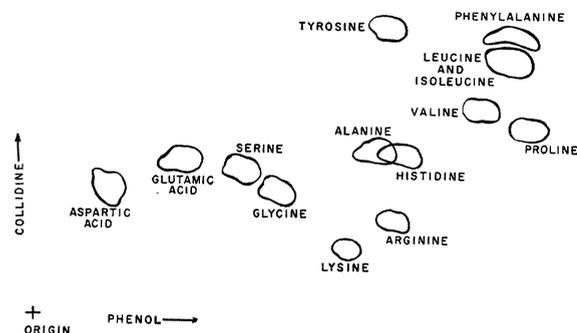


Fig. 1. Two-dimensional chromatogram of an acid hydrolysate of angiotonin, using phenol and collidine as the developing solvents. The amino acids were visualized by spraying with ninhydrin and, after steaming, with diazotized sulfanilic acid.

### The Use of Electrically Conducting Glass for Counting Lesions\*

A device has been constructed that greatly facilitates counting the local lesions on *Nicotiana glutinosa* which appear after viral infection. The novel feature of this counter is the electric conducting glass employed. Because the glass is frosted and translucent, its entire area can be illuminated softly and evenly

\* This work was facilitated by research funds provided by the U.S. Atomic Energy Commission. A sample of EC glass for experimental purposes was supplied by the Corning Glass Works, Corning, N.Y.

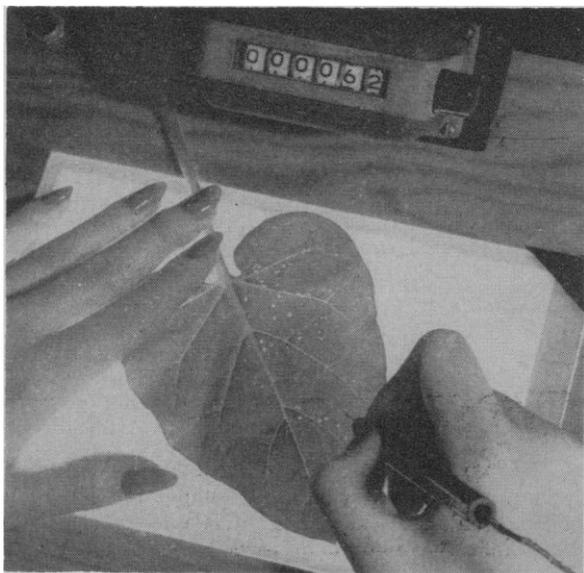


Fig. 1. Counting local lesions on leaf of *Nicotiana glauca* with electrically conducting glass.

by a fluorescent lamp mounted below. As a result, the lesions stand out with great clarity.

The conducting glass is connected to dry cells hooked in series to supply 8 v. Thus, when the delicate leaf tissue next to a lesion is punctured with a metal probe connected to the other pole of the batteries, contact between the probe and glass plate sets up a slight current which trips a relay that calls for 110-v current to activate a reset counter. When the probe is withdrawn, a pinprick of light shines through the puncture showing that the lesion has been counted (Fig. 1). About 500 lesions on a single half-leaf may be counted in approximately 2 min.

The ease of counting that has been achieved with this device has markedly reduced the strain and boredom previously associated with this task. The cost of the batteries, fluorescent light, counter, relay, glass, and so forth, amounts to about \$40. Batteries appear to last about 2 mo when the counter is used twice each week. Further construction details may be obtained by writing to the authors.

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### The Spectrum of Magnesium in Concentrated Sulfuric Acid

Niemann and Ikawa (1, 2) have shown that the type and amount of a carbohydrate in solution can be found from its spectrum in the 210 to 400  $m\mu$  region in strong mineral acid. In some cases, a quantitative determination of one monosaccharide can be made in an admixture of other monosaccharides, and some polysaccharides can be resolved into their component

monosaccharide units. These determinations are based on the production of various furfurals by the monosaccharides on heating in strong mineral acid, each class of monosaccharides condensing into a slightly different furfural.

In crude preparations of carbohydrates, taken from sea water by extraction on charcoal, alumina, and other adsorbents, a discrepancy of 4 to 5 mg/liter in a total of 20 to 25 mg/liter was noted between the amounts of carbohydrates found by examination of the sulfuric acid spectra in the 210 to 400  $m\mu$  region and those found by the N-ethylcarbazole method. Since both methods depend on the formation of a furfural in concentrated sulfuric acid, it was apparent that some compound or compounds other than carbohydrates, could produce ultraviolet spectra similar to those of the furfurals, in hot sulfuric acid.

By a process of elimination, the substance was found to be magnesium. Figures 1 and 2 show the close correspondence between the sulfuric acid spectra of high concentrations of magnesium sulfate and those of a methyl pentose, rhamnose. Certain polysaccharides and some mixtures of monosaccharides duplicate even more closely the spectrum given by magnesium. While the concentrations of magnesium sulfate necessary to give these spectra may seem absurdly high, in working with crude preparations in which the carbohydrate concentration is very low, concentrations of

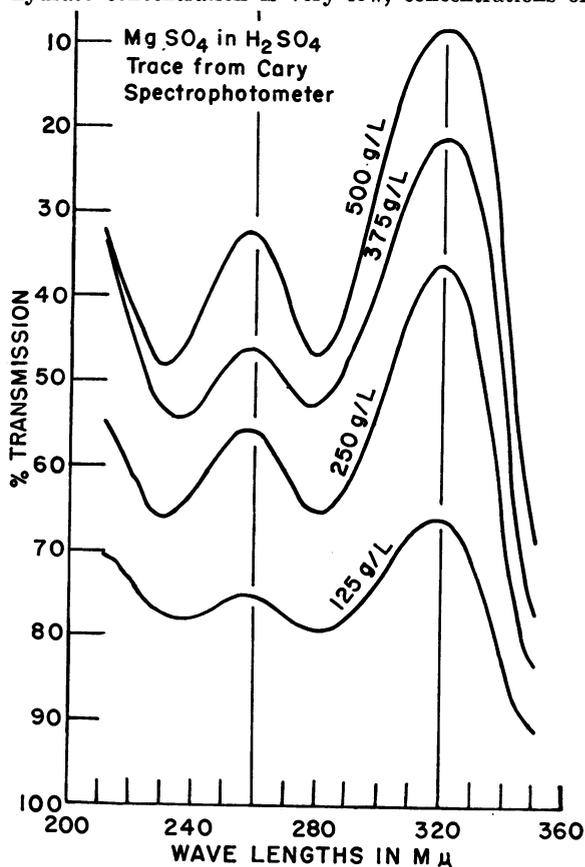


Fig. 1.