

# SCIENCE

11 December 1959

AMERICAN ASSOCIATION FOR THE ADVANCEMENT OF SCIENCE



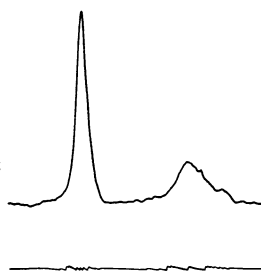
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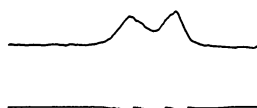
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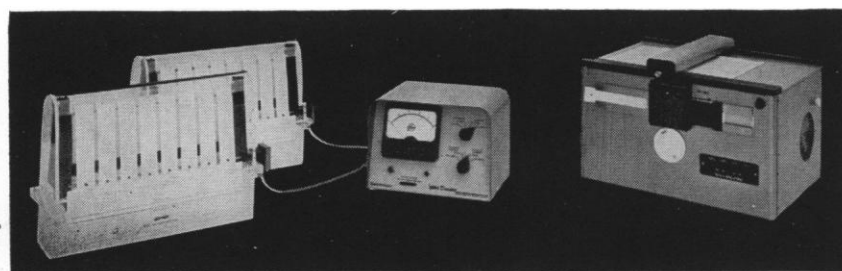
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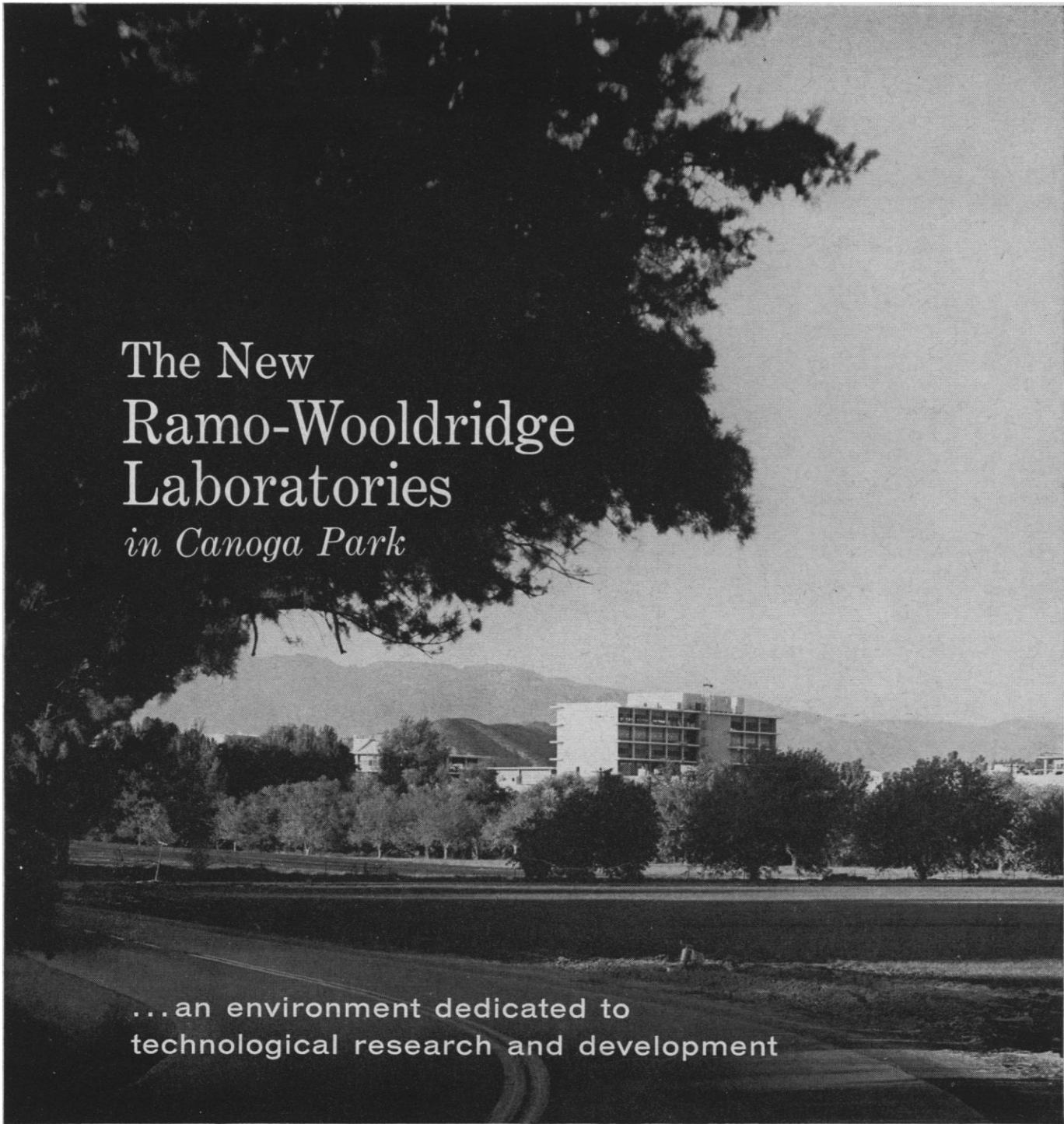
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<b>Cover</b>	Typical midsummer storm over the Gulf of Mexico, from Madeira Beach (St. Petersburg, Fla.), about 10:00 P.M., 23 July 1959. This was one of five separate storm systems parading northward through the Gulf. This time exposure (approximately 15 seconds), made with a Rolleiflex (2¼ inch square), includes four or five separate flashes, only the last of which was "grounded" into the water. [W. R. Stewart]	





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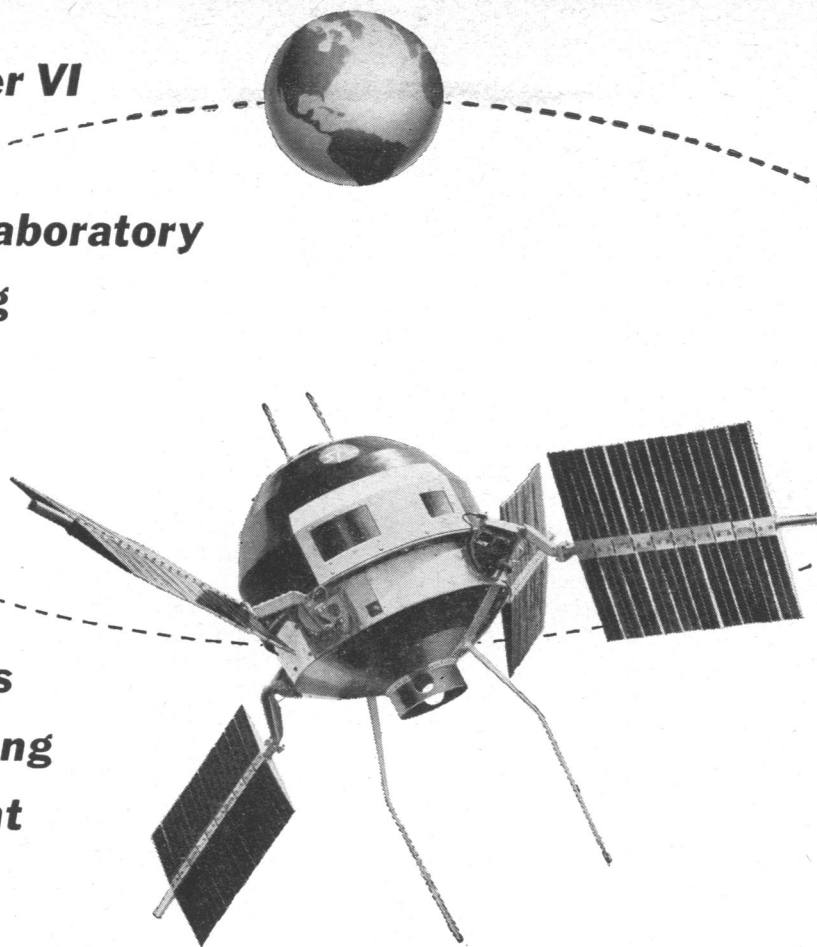
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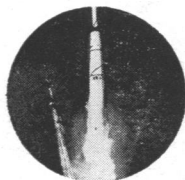
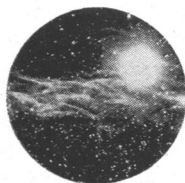
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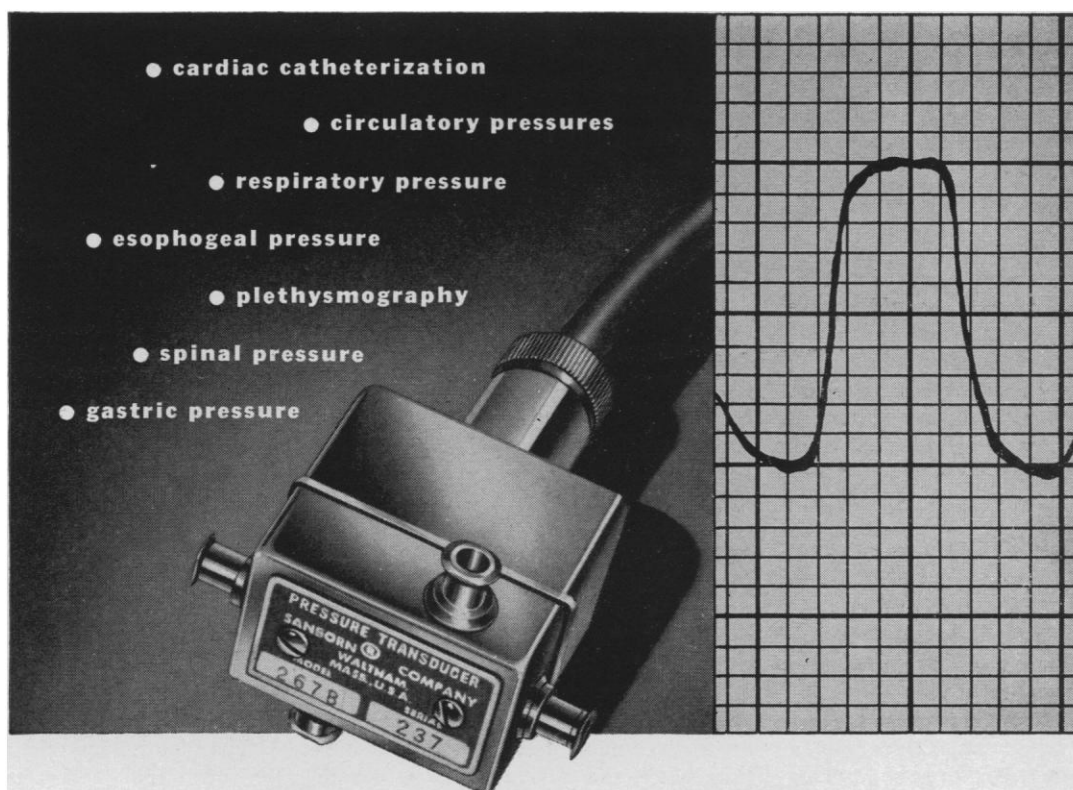
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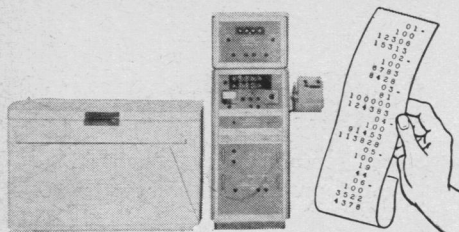


  
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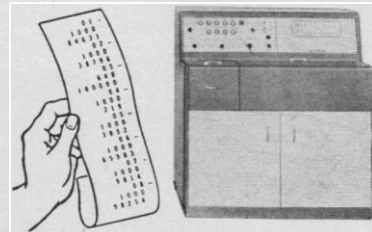
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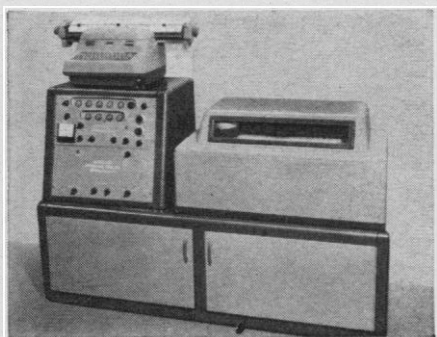


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# Meetings

## **Glucuronic Acid Research**

The study of glucuronic acid in relation to many fields of biology and medicine has been a major continuing activity of Japanese investigators. This interest developed first from the work on the metabolism of drugs in which glucuronic acid conjugation is a frequently encountered feature, and then, with the ready availability of pure glucuronic acid (Chugai Pharmaceutical Company, Tokyo), from a widespread curiosity to examine its biological properties in almost every living situation. A measure of the magnitude of this research effort is the fact that for the last 4 years a research conference on this subject has taken place. I was privileged to be invited to attend the fifth Glucuronic Acid Research Conference which was held 2 and 3 June at Sankei Kaikan, Tokyo. At this conference and on a number of other visits in June to research laboratories in Japan, it was possible to exchange views and to acquire information regarding biochemical studies on glucuronic acid. My present purpose is to transmit this knowledge to American investigators who are working in this area of research.

The conference was opened by M. Ishidate, dean of the faculty of pharmaceutical sciences, University of Tokyo. It was attended by some 250 investigators invited from many research centers of Japan. Two novel features of the meeting were, first, the simultaneous use of two slide projectors and two screens to expedite illustration of the data, and second, the provision of a pocket radio transmitter which not only freed the speaker from the electrical wires which ordinarily chain him to the podium but gave pleasing sound amplification.

Forty-four original papers were presented. Thirteen were concerned with the biochemistry and physiology of glucuronic acid, four with its growth-promoting effect, twelve with detoxication of drugs, viruses, and toxins, and the remainder with the clinical use of glucuronic acid.

### **Biochemistry and Physiology**

The conjugation of amines with glucuronic acid and the significance of this process was reported by Ishidate. Aromatic and aliphatic primary amines easily form N-glucuronides at room temperature in the presence of aqueous solutions of glucuronic acid or its alkaline salts. Crystalline N-glucuronides have been obtained of the following: aniline, toluidine, *p*-chloroaniline, mono-

acetyl-*p*-phenylenediamine, *p*-dimethylaminoaniline, *p*-phenylenediamine, sulfanilamide, sulfapyridine, benzylamine,  $\beta$ -phenylethylamine, ethylamine, isopropylamine, isobutylamine, and cyclohexylamine. A quantitative relation between amine metabolism and glucuronic acid excretion was observed when rabbits were given aniline, sulfanilamide, or sulfapyridine. Also, after the administration of *p*-dimethylaminoazobenzene to dogs, one of the urinary metabolites appeared to be the N-glucuronide of *p*-mono-methylaminoazobenzene. The studies have included the property of amino acids to form N-glucuronides. These can be detected by paper chromatography but, because of their lability, they have not yet been isolated. A reaction of glucuronic acid with diphtheria toxin takes place at pH 7.0 and 38°C with the gradual disappearance of toxicity. The formation of antiserum from the injection of this material was no different than in the case of formalin-inactivated toxin. It was therefore suggested that an amino group is the functional group of toxin and that this reaction depends upon formation of an N-glucuronide.

Ishidate then reviewed the two mechanisms which can explain glucuronic acid conjugation (UDPGA,  $\beta$ -glucuronidase) and found that information regarding the enzymatic mechanism of formation of N-glucuronide is incomplete.

Shimazono *et al.*, (department of biochemistry, University of Tokyo) reported that incorporation of phosphate into rat liver tissues was increased by the injection of D-glucuronate. From an analysis of fractions containing nucleotides and sugar phosphates, it was stated that this incorporation was most marked in the case of glucose-6-phosphate and *d*-glycerophosphate. In a carefully done enzymatic study of glucuronic acid, the following observations were made. D-Glucuronate was reduced to L-gulonate by TPN-gulonic dehydrogenase, and L-gulonate was oxidized following decarboxylation to L-xylulose by DPN-gulonic dehydrogenase. The formation of lactone from D-glucuronate or L-gulonate by soluble lactonase from cell supernatant was observed with the aid of hydroxylamine. The lactonase which acts on D-glucuronolactone was present in liver microsomes of various animals but not in human and monkey liver. L-Gulonolactone was formed easily from D-glucuronolactone by the action of TPN-gulonic dehydrogenase. A dehydrogenase present in microsomes produces ascorbate from L-gulonolactone.

Much discussion centered around in vivo effects of administered glucuronic acid in relation to carbohydrate metabolism, particularly in the liver. Thus,



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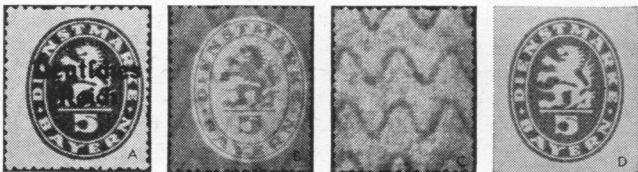
headlined the first column of *The Wall Street Journal* on the penultimate morning of last summer.

"Industry sources expect retail sales of equipment and supplies for industrial photographic use will reach some \$250 million this year, up from about \$100 million five years ago," the story said. (That's a very short time ago. These very discourses have been appearing for six years in this periodical.)

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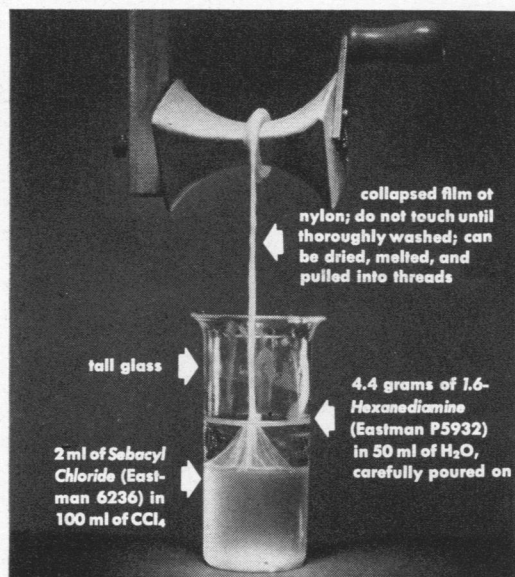
Bavarian stamp of 1920 (Scott No. 052). A. Visible light photograph. The design is green and the "Deutsches Reich" overprint is black. B. Soft x-ray radiograph. Details of both design and paper visible. Design is "negative," indicating absorption of x-rays by the ink. C. Electron radiograph. Only the details of the paper are shown. D. Electron-emission radiograph. The design is "positive," indicating a relatively high

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Obara *et al.* drew attention to effects on the adrenal gland; Oda and Hara observed an increase in liver glycogen following glucuronolactone or xylulose injection; Imanaga stated that the increase in blood ammonia in patients with a portacaval shunt can be controlled with exogenous glucuronic acid; Oji studied urinary pentose in patients with liver disease and in animals with CCl<sub>4</sub> poisoning; Kobayashi *et al.* investigated the effect of glucuronic acid on experimental diabetes mellitus; Kurokawa and Yamagata reported experimental and clinical studies on a beneficial effect of glucuronic acid in

diabetes; Masuda *et al.* investigated phosphorus metabolism in the liver of rabbits given carbon tetrachloride; and Kusuya *et al.* studied the effect of glucuronic acid on sugar assimilation.

With regard to  $\beta$ -glucuronidase, Tsukamoto *et al.* of Kyushu University proposed the use of *p*-nitrophenyl glucuronide as a substrate for both the hydrolytic and transfer activities of the enzyme. The enzyme was stated to catalyze the formation of *m*-aminophenyl glucuronide in a system in which benzoyl glucuronide was the donor substrate, and *m*-aminophenol, the acceptor molecule.

#### Growth-Promoting Effects

Several papers were presented which illustrate the variety of investigations into growth-promoting effects. Thus, Ogawa (National Institute of Genetics) observed that sodium glucuronate and glucuronic acid showed a significant growth-promoting effect on the early development of the embryo (*Triturus pyrrhogaster*) which was most prominent on the 13th day after fertilization (stage 32); Tamura (Tokyo Dental College) noted that the growth of chicks on a vitamin-B-deficient diet was promoted by glucuronolactone; and Wada *et al.* (Sapporo Medical College) reported that the prior administration of glucuronolactone to rats significantly promoted the growth of intraperitoneally transplanted Yoshida sarcoma cells, whereas the opposite effect occurred on the growth of subcutaneously transplanted Yoshida sarcoma.

#### Experimental and Clinical Studies on "Detoxication"

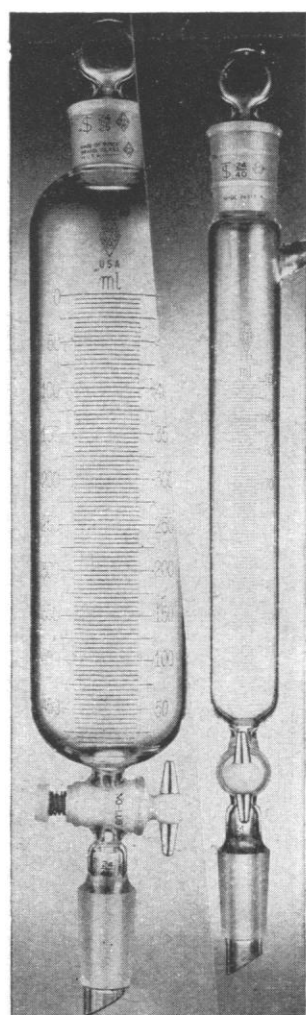
Although the papers on clinical and detoxication studies were the most numerous, yet, because of the great variety of conditions investigated they were not amenable to generalization.

The effect of exogenous glucuronic acid on the excretion of morphine glucuronide was thoroughly investigated by Hosoya and Otobe (Keio University School of Medicine). Thus, during the first 2 hours after administration of morphine, bound morphine appears earlier and in larger amounts in the urine of rabbits receiving glucuronic acid and morphine. Hosoya stated that these results seemed to indicate that exogenous glucuronic acid accelerates conjugation of morphine with glucuronic acid in the living body although it was by no means clear whether the exogenous glucuronic acid did conjugate with endogenous glucuronic acid by some unknown mechanism.

Shirai *et al.* (Kobe Medical College) extended his observations on the ethyl ester of glucuronic acid which augmented the formation of anthranilic acid glucuronide in the rabbit. He observed that the glucuronic acid ester is excreted in the urine more slowly than glucuronolactone.

Sawada (Kyushu University) stated that the direct-reacting bilirubin in cat bile was the glucuronide but that this substance was not produced by cat liver microsomes fortified with UDPGA.

Examples of the experiments and of the toxic compounds studied are Takahashi's (Kyoto) perfusion studies of conjugation by livers damaged with carbon tetrachloride; Tsumoo's (Showa Medical College) study of the effect of 1-phenyl-2-methylaminopropane and ephedrine on blood pressure and respiration in the urethanized rabbit; Tanuora's



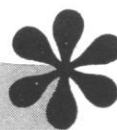
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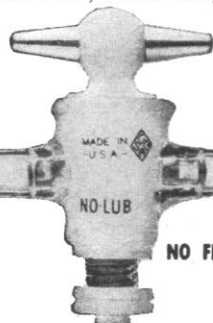
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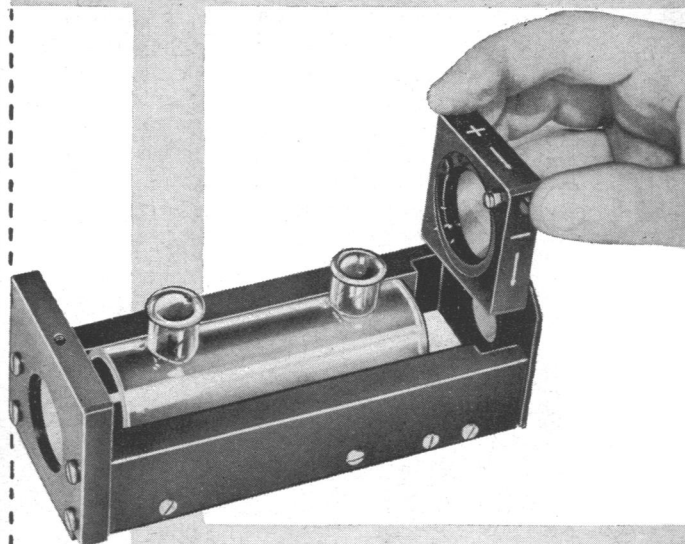


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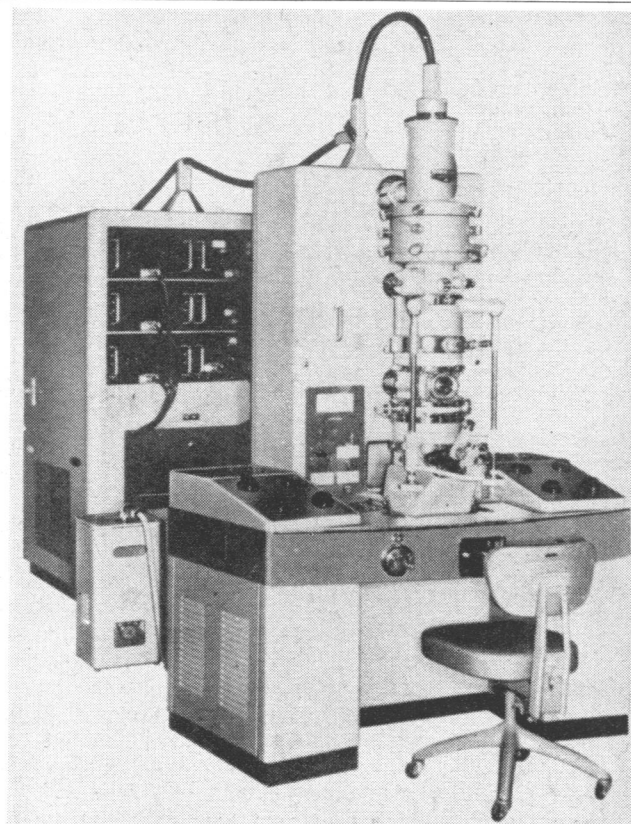
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The General Program of the 126th Meeting of the AAAS in Chicago, 26-31 Dec., 1959, will be available to you within the first week in December—whether you can attend the Meeting or not.

Effective this year, the former General Program-Directory, which had become an unwieldy book of more than 400 pages, has been separated into *two* publications, namely:

- a) The Directory of AAAS Officers and Activities, 96 pp., already published; and
- b) The General Program of the Annual Meeting, c. 200 pp., which will appear early in December

Both of these, sold at cost, may be purchased separately—in advance (see coupon below), or at the meeting. Some of their *respective* contents are:

#### The General Program

1. The two-session general symposium "Moving Frontiers of Science IV," arranged by the Committee on AAAS Meetings.
2. Programs of the 18 AAAS sections (symposia and contributed papers).
3. Programs of the more than 80 participating societies.
4. Sessions of the Conference on Scientific Communication, Conference on Scientific Manpower, and the Academy Conference.
5. The Special Sessions: AAAS Address and Reception, National Geographic Society, Phi Beta Kappa, Sigma Xi, RESA, Tau Beta Pi Association.
6. Details of the Morrison Hotel—center of the Meeting—and of the other session sites.
7. Titles of the latest foreign and domestic scientific films to be shown in the AAAS Science Theatre.
8. Exhibitors in the 1959 Annual Exposition of Science and Industry and descriptions of their exhibits.

#### The Directory

1. AAAS officers, staff, committees, for 1959.
2. Section committees and other AAAS Council members.
3. The 285 affiliated organizations.
4. Historical sketch and organization of the Association.
5. Complete roll of AAAS presidents and their fields.
6. Publications of the Association, including all symposium volumes.
7. AAAS Awards—including all past winners.
8. Future Meetings of the AAAS through 1963.
9. New and current activities of the AAAS.
10. Constitution and Bylaws.

#### Advance Registration

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The following coupon may be used both by advance registrants and by those who wish only the advance copy of the General Program; the Directory may also be ordered.

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(Keio University) work on the isolated surviving frog heart, hypnotic and lethal actions of barbiturate, and changes of oxygen consumption of rat brain tissue homogenate; Harashima's (Keio University) observations on the urinary excretion of glucuronic acid in rabbits experimentally exposed to benzene and carbon disulfide; Ito's study on the detoxication of 2,4-dinitrophenol, and Takatsu's (University of Tokyo) work on the effect of sodium glucuronate on the LD<sub>50</sub> of *Shigella* endotoxin, phenol, pyramine, and noradrenaline in the young mouse.

In the field of virology, Ogasawara *et al.* (Nagoya University) studied the effects of glucuronic acid salts on the activity of influenza virus PR8 and of Newcastle disease virus in producing pyrogenic skin lesions and pulmonary consolidation. On the whole, infectivity, hemagglutinin, and antigenicity of these two viruses were not affected significantly, but the toxicity (skin lesion, pulmonary consolidation) was prevented by preliminary treatment with glucuronolactone. Coto *et al.* (University of Tokyo) observed the in vitro inhibition by glucuronic acid of mouse hepatitis virus, rabies virus, and Japanese encephalitis virus, with regard to their ability to infect the host mice.

Clinical studies included work on the effects of glucuronic acid on steroid hormone excretion during pregnancy (Moriyama *et al.*); studies on glucuronic acid metabolism of newborn infants (Iwanami *et al.*); three separate investigations on glucuronic acid interrelationships with adrenal cortical function (Tokita *et al.*, Oshima *et al.*, and Kawai *et al.*); two reports on a therapeutic effect of glucuronolactone in diabetes mellitus (Katsuki *et al.* and Matsuoka *et al.*); the treatment of epidemic hepatitis with glucuronic acid (Kosaka *et al.*); and the influence of glucuronolactone on experimental liver injury induced by *Penicillium islandicum* Sopp poisons (Suzuki *et al.* and Uraguchi *et al.*).

It appears that with glucuronolactone occupying a central position in the glucuronic acid pathway of glucose metabolism, the basis of interpretation of the results of experimental and clinical work with exogenous glucuronolactone is to be found mainly in the field of carbohydrate metabolism. Nevertheless, "detoxifying" effects of glucuronolactone or glucuronic acid may be explained on a direct basis, such as in N-glucuronide formation, or may result indirectly from alterations in the dynamics of the glucuronic acid pathway. The evidence indicates that glucurono-

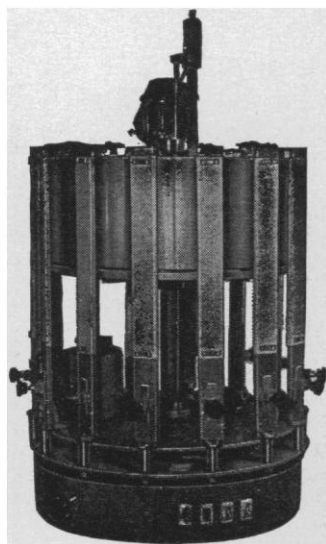
lactone is indeed a physiological nutrient or metabolite and its study provides many indications of rewarding further critical investigation.

The address by W. H. Fishman (Tufts University School of Medicine) was published in monograph form (*Biochemistry of Glucuronic Acid Lectures in Japan*, published by the Glucuronic Acid Research Conference, University of Tokyo, 1959). Worthy of mention was the report of the discovery of a new 3-ketoheptose phosphate by Sie, Nigam, and Fishman, and the presentation of a concept of the role of  $\beta$ -glucuronidase in which the enzyme is believed to be associated with processes of cell and tissue differentiation rather than with cell division or growth per se.

I recall with great pleasure both the many enjoyable events of the conference and the lectures which it was my privilege to deliver at a number of Japanese institutions of higher learning. The kind and sincere hospitality of my hosts are herewith acknowledged with many thanks. My appreciation is expressed also to the National Science Foundation for granting a travel award.

WILLIAM H. FISHMAN

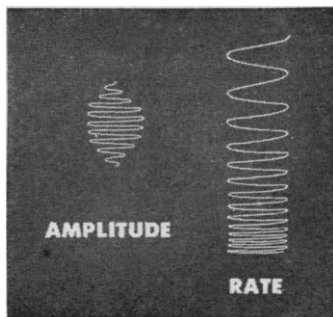
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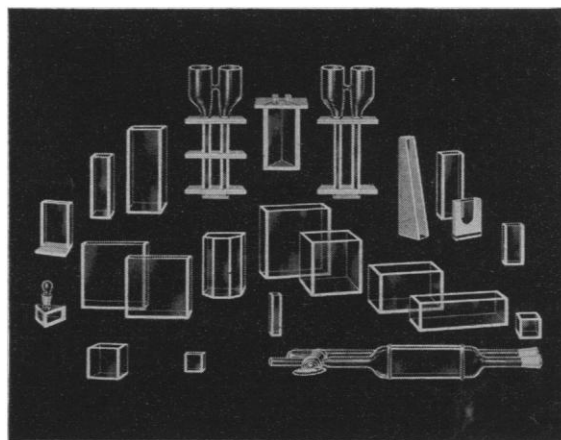
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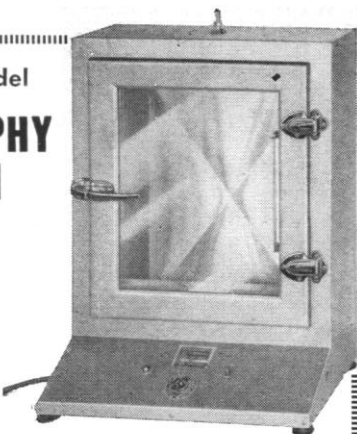
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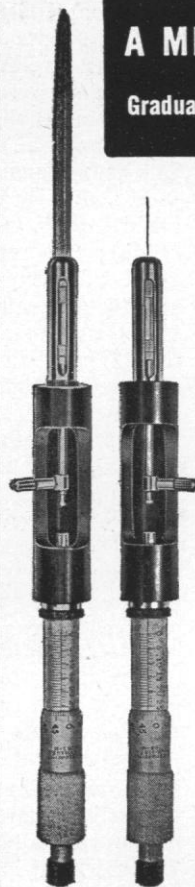
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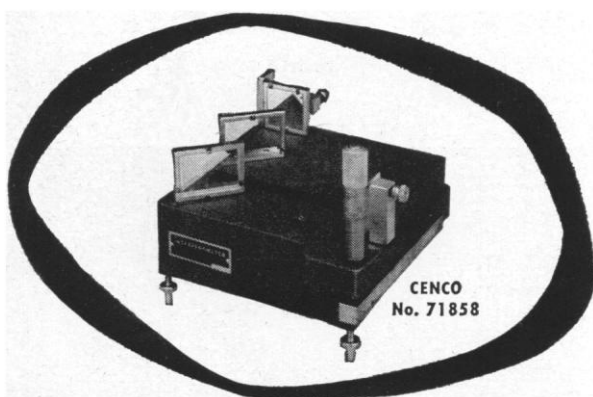
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