sizing system. It is also significant that the 2 principal odd-numbered carbon fatty acids found, pentadecanoic acid and heptadecenoic acid, are one carbon less than palmitic acid and oleic acid, respectively, the 2 major acids of animal fats. This is indicative that the odd-numbered fatty acids are synthesized by the same systems that produce the normal fatty acids having an even number of carbon atoms.

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Effect of Flavins on Cramp caused by Phenol Husemann, Salkowski, Morita and others<sup>2</sup> have reported that phenol provokes a characteristic convulsion in animals; so the effect of flavins on cramp caused by phenol was studied.

The flavin mononucleotide was supplied by Hoffmann-La Roche Co. Flavin adenine dinucleotide was prepared by the method of Yagi et al.<sup>2</sup> The phenol used was a commercial sample. Male albino rats weighing about 130 gm. were used for the experiment.

Forty-five animals, 10 days after feeding under the same conditions were divided into three groups, each group consisting of 15 animals. The animals of the first group were injected intra-abdominally with the mixture of 1.0 ml. of 0.1 mole/l. phenol and 2.0 ml. of water; those of the second group were injected with the mixture of 1.0 ml. of 0.1 mole/l. phenol, 1.0 ml. of 0.04 mole/l. flavin mononucleotide and 1.0 ml. of water ; those of the third group were first injected with 2.0 ml. of 0.02 mole/l. flavin mononucleotide, and 10 min. after the injection, 1.0 ml. of 0.1 mole/l. phenol was further injected.

Cramp was observed by electromyogram of the muscle biceps femoris. A needle electrode was placed in the muscle, and the electromyogram was recorded by a Grass model III-D electroencephalograph.

The time from the injection of phenol to the beginning of the convulsion (1), and that of the duration of the convulsion (2) were measured. The arithmetic mean of the results of each experiment and the mean of the population (with the probability of 99 per cent) are listed in Table 1.

The results were statistically examined, and the differences between the first group and the second or the third group were significant (with 99 per cent These results indicated that flavin possibility). mononucleotide could inhibit the provocation of phenol convulsion and shorten the duration of the convulsion. On the other hand, statistical examination of the difference between the second and the third groups showed that the difference was significant between the second group (1) and the third group (1), but not between the second group (2) and the third group (2) (with 99 per cent possibility). This result 
 Table 1.
 Effect of Flavin on the Provocation and Duration of Convulsions caused by the Injection of Phenol

		Time from the injection of phenol to the beginning of cramp	Time of duration of cramp
First	$\bar{X}$ $(n=15)$	1 min. 4 sec.	50 min. 6 sec.
group	$M\left(a\!=\!0\!\cdot\!01\right)$	1 min. 15 sec 53 sec.	53 min. 53 sec 46 min. 19 sec.
Second	$\bar{X}$ $(n=15)$	1 min. 17 sec.	33 min. 58 sec.
group	$M(a\!=\!0\!\cdot\!01)$	1 min. 30 sec.– 1 min. 4 sec.	37 min. 59 sec.– 29 min. 57 sec.
Third	$\bar{X}$ (n=15)	1 min. 34 sec.	33 min. 27 sec.
group	$M\left(a\!=\!0\!\cdot\!01\right)$	1 min. 49 sec.– 1 min. 19 sec.	36 min. 30 sec.– 30 min. 24 sec.

 $\bar{X}$ , Arithmetic mean. M, Mean of the population.

showed that flavin mononucleotide previously injected is more effective than that injected at the same time with phenol for the inhibition of the provocation of phenol convulsion, and is equally effective with that injected at the same time with phenol for the duration of the convulsion.

A similar experiment was also carried out with flavin adenine dinucleotide, and the results showed the same tendency. These results suggest that the riboflavin part of flavin nucleotides may be an active component in the above-mentioned effect.

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## RADIOBIOLOGY

## Assay of Plasma Insulin in Human Subjects by Immunological Methods

WE have previously reported on the immuno-assay of beef insulin and certain other animal insulins, employing antiserums from human subjects treated with commercial mixtures of beef and pork insulin<sup>1</sup>. The insulin-binding antibodies present in these antiserums do not form precipitable complexes with insulin, but with the use of insulin labelled with iodine-131 the complexes are readily demonstrable by paper chromatography and electrophoresis<sup>2</sup>. Beef, pork, sheep and horse insulins can be assayed quantitatively by measurement of the degree of competitive inhibition of binding of any insulin labelled with iodine-1311-3. As might have been anticipated, however, human insulin competes too weakly in systems employing human antiserum to be measurable at concentrations which obtain in vivo. Furthermore, the lack of availability of significant quantities of pure human insulin precludes its use as an antigen for animal immunization. However, in the present work, it has been found that human insulin cross-reacts strongly with insulin-binding antibodies in guinea pigs immunized with crystalline beef insulin, and that guinea pig anti-beef insulin serum has characteristics suitable for the detection and measurement of human insulin at concentrations which exist in the plasma of normal fasting subjects.

The methods employed and results obtained in this