partial immunity in 26 per cent. A second series of toxin injections successfully immunized most of the refractory cases.

2. Subsequent exposure to scarlet fever showed that the induced active immunity protected the children from infection.

3. From 10 to 20 per cent of individuals who gave a negative Dick reaction after toxin injections showed a slightly positive reaction at later re-tests.

4. Groups of children who were re-tested from 9 to 14 months after the toxin injections show by a negative Dick retest that the immunity can last for at least that period of time.

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Immunity results with diphtheria toxoid (modified toxin anatoxin) and 1/10 L + mixtures of toxin antitoxin.

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The 1/10 L+ mixture of diphtheria toxin-antitoxin, when prepared with a slight excess of free toxin so that 5 cc. will kill a guinea pig in 5 to 7 days, is an efficient immunizing agent. Three doses, each 1 cc., will immunize from 90 to 98 per cent of injected persons. The fact, however, that an underneutralized mixture has to be used which produces the local effects of free toxin at the site of injection; that the mixture has to be very carefully prepared so that the amount of free or partly bound toxin is not in excess; and finally that the mixture on standing loses some of its immunizing value, indicates that such a mixture is not yet the ideal preparation for the universal immunization of all children of school age and pre-school age. In diphtheria toxoid (anatoxin) we have such a preparation. It is easily made by adding 0.25 to 0.4 per cent formalin to the toxin, and leaving the formalinized toxin in the incubator for 30 days. The preparation is stable, *non-toxic* and highly efficient as an immunizing agent. With a preparation sent us by Ramon, we injected one-half of the children in three large schools, the other half being injected with 1/10 L+ toxin-antitoxin. The dose of the anatoxin

was as follows: 1st dose, 0.5 cc.; 2nd dose, 0.5 cc.; and the 3rd dose, 1.0 cc. The dose of the toxin-antitoxin was 1 cc., repeated three times. The injections were given every two weeks. The Schick re-test was made in one of the schools at the end of 6 weeks, and the following results were noted:

(A) Three doses anatoxin 98.0% Negative Schick re-test.
(B) Two doses anatoxin 86.4% Negative Schick re-test.
(C) One dose anatoxin 60.0% Negative Schick re-test.

With the toxin-antitoxin, 91 per cent of all the injected children gave a negative Schick re-test. The local reactions to the toxoid (anatoxin) were slight. In fact the toxin-antitoxin, being slightly toxic, gave more pronounced local reactions. In pseudo-reactors the toxoid (anatoxin), being undiluted formalinized toxin, gave more marked local reactions.

The diphtheria toxoid prepared at the Research Laboratory was slightly toxic, following the suggestions of Glenny, Hopkins and Pope. On account of the excess of free toxin we[1, 2] used only small doses of the toxoid at first (0.1 to 0.05 cc.). From 52.0 to 94.0 per cent of over 3,000 school children injected with these doses in different schools gave a negative Schick re-test. By increasing the dose with later preparations, the results were better and more uniform.

With one preparation of toxoid after three doses, each 1 cc., 96 per cent of the children showed a negative Schick re-test; with another toxoid after three doses, each 0.5 cc., 79 to 95 per cent became immune.

The smaller the amount of formalin used, the better were the immunity results. From one toxin, three preparations of toxoid were made—one with 0.25 per cent formalin (Toxoid 7), one with 0.5 per cent (Toxoid 7 A), and one with 0.75 per cent (Toxoid 7B). Three doses of 0.25 cc. each were given of the 0.25 (Toxoid 7) and of the 0.5 per cent formalinized toxin (Toxoid 7 A), and three doses of 0.5 cc. each of 0.75 per cent formalinized toxin (Toxoid 7 B). Of 359 children receiving Toxoid 7 (0.25 per cent formalin) 66 per cent gave a negative Schick re-test after 3.5 months; of 336 children receiving Toxoid 7 A (0.5 per cent formalin) 45.8 per cent were negative at the re-test,

[1] Zingher, A., and Park, W. H., PROC. SOC. EXP. BIOL. AND MED., 1924, xxi, 383.

[2] Park, W. H., and Zingher, A., *Am. J. Dis. Child.*, 1924, xxviii, 464.

and of 100 children receiving Toxoid 7 B (0.75 per cent formalin) 36 per cent were negative.

CONCLUSION.

Diphtheria toxoid (anatoxin) is an ideal preparation for active immunization against diphtheria. This is especially true for children of school age and of pre-school age who are relatively free of protein sensitiveness and in whom the toxoid produces very slight local or constitutional reactions.

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Studies upon the biological reactions of growing tissues to radiant energy. I. Effect of radiumized media upon tissue cultures in vitro.

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The *in vitro* cultivation of embryonic tissues seemed to us the most promising method of gaining important information of the biological reaction of growing tissues to radiant energy. Up to the present, experimentation along physical lines has claimed major interest, and very scant attention has been paid to the biological importance of radiant energy. Consequently very little is known concerning the biological phenomena accompanying radiation.

It was highly desirable therefore to study the immediate and latent behavior of tissues growing *in vitro,* when placed in immediate contact with radium salts and radium emanations.

The experiments to be reported were conducted upon chick embryo hearts, embryo spleens, and upon Flexner-Jobling rat carcinoma. Along with these, numerous experiments were made upon the effects of radium salts, radium emanations, as well as X-rays, upon different media, such as plasma, serum, embryonic tissue extracts, etc.

In the present studies chick embryo hearts from 7 to 11 day embryos, and spleen from 18 to 20 day embryos were used for *in vitro* studies. Small fragments were planted in a medium of chicken plasma and embryonic tissue juice. In the cultures where