with others, also in cases where there is neither histological nor embryological relationship, *e. g.*, trachea immune serum on brain. Only the sperm immune serum proved to be entirely organ specific. The immune sera were Wassermann negative and had moderate (kidney), little (trachea), or practically no (thymus, sperm) hemolytic activity for ox blood.

[1] Halpern, J., *Z. f. Immunitätsf.*, 1911, xi, 609.

[2] Fleisher, M. S., Hall, T. G., and Arnstein, N., *J. Immunol.*, 1920, v, 437. Fleisher, M. S., and Arnstein, N., *J. Immunol.*, 1921, vi, 223. Fleisher, M. S., *J. Immunol.*, 1922, vii, 51.

[3] Witebsky, E., and Steinfeld, *Centralblatt f. Bakteriologie*, 1927, civ, 144.

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Composition of Bone. II. Analytical Results.

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The micro-technique which we[1] have devised for analyzing small specimens of calcified tissue has been subjected to further test and study. The crushed bones are extracted with ether and alcohol immediately after removal from the body; they are then dried and pulverized. About 10 mg. of bone powder are digested with HCl, trichloracetic acid is added, the solution is made up to 10 cc. and then filtered. Calcium is determined on aliquots of this filtrate. To another aliquot molybdic acid reagent (Briggs-Bell-Doisy) is added, the volume is made up to 10 cc. and then filtered. Inorganic phosphorus is determined on aliquots of this second filtrate. Digestion with HCl of different samples of the same specimen of bone powder gives solutions whose content of calcium and inorganic phosphorus agree as well as duplicate determinations on aliquots of the same solution.

CO_2 is determined on about 20 mg. of bone powder in Van Slyke's manometric gas apparatus.

A specimen of crushed bone was divided into two portions. One portion was extracted with ether and alcohol immediately, the other was allowed to stand unextracted during the summer. After standing for 5 months it was extracted and analyzed. The results for calcium, inorganic phosphorus and CO_2 were the same for both portions. Similar results were obtained with a second specimen of nor-

mal bone. The second precipitation with the molybdic acid reagent does not interfere with the accuracy of the method; the values for the ratio residual Ca/P so obtained are normal within the experimental error. When phosphorus values obtained from the first filtrate are employed, low ratios result.

In studying the method 11 specimens of normal calcification and 7 specimens of pathological calcification were analyzed. In all cases the value of the ratio was 1.96 within the experimental error; the theoretical value from $Ca_3(PO_4)_2$ is 1.94. One type of exception was noted. Three specimens of calcified fibroid of the uterus gave ratios of 2.23, 2.23, and 2.18 respectively.

The normal adult rats analyzed contained about 15% carbonate calcium, *i. e.*, carbonate Ca/total Ca × 100 was about 15. In the young rats analyzed the carbonate calcium was only from 8 to 10% of the total calcium. This indicates that the composition of bone in rats is not constant, but changes with age.

[1] Shear, M. J., and Kramer, B., *J. Biol. Chem.*, 1927, lxxiv, 9.

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Immunization Against Pneumococcus by Feeding Desiccated or Milk Suspended Organisms.

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The author previously demonstrated that white rats can be protected against many lethal doses of pneumococci, intraperitoneally injected, if these animals are first fed either the tissues of animals killed by this germ,[1] living pneumococci,[2] or HCL killed germs.[3] It was next decided to determine whether the acid killed germ when dried and mixed with cracker meal and the acid killed germ when still moist and suspended in milk could also be used. The object was to simplify the method of administering the germs as far as possible, consistent with successful immunization. At the same time success or failure accompanying a given modification gradually helps toward an explanation of oral immunization with the pneumococcus.

The organisms were grown, killed and centrifuged as described elsewhere.[3] A thin layer of germs was spread on a watch crystal and warmed on a water bath, so that they were at no time heated above 37° C. Desiccation was complete in 15 minutes. For 20