social hygiene, 9 were architectural students and 90 were graduate school teachers. Three diplomas in tropical medicine and two in public health were granted. The course in tropical medicine occupies a period of three and a half months, and corresponds closely to those of London and Liverpool. The faculty of medicine in the university has recognized the importance of the school by appointing its director, Dr. Harvey Sutton, professor of preventive medicine. This is the first chair in Australia in this branch of medicine. A hookworm and filariasis survey of Norfolk Island was undertaken during the month of January, 1931, completing the hookworm survey of the Australian Commonwealth, Papua and the Mandated Territory of New Guinea. This work, which was conducted for a period by the Rockefeller Institute in collaboration with the Commonwealth Government, has been carried on by the Commonwealth Health Department.

THE German Hygiene Museum at Dresden, which sponsored the International Hygiene Exhibition last year on the occasion of the opening of its new building, reports, according to *Museum News*, that 7,200,000 people visited the exhibition from May 15 to October 1, 1930. In view of the world-wide interest, the directors have decided to repeat the exhibition during the same period in 1931. Many of the foreign governments are enlarging their exhibits and it is said that the participation of the United States Government seems to be assured. In the museum itself the exhibits dealing with the health of the mother and child have been amplified and the exhibits on biology and comparative anatomy have been completely revised and enlarged. In anticipation of the exhibition, the museum has opened offices in several cities throughout the world. The American office is in charge of Dr. R. Woerner, at 1880 Broadway, New York City.

DISCUSSION

CRYSTALLINE PEPSIN OF NORTHROP

THE comprehensive investigation of Dr. Northrop on the crystalline pepsin prepared by him justifies the conclusion of the protein nature of this enzyme and makes probable the individuality of the crystalline substance. Its low isoelectric point stands out as the most characteristic property of the substance from the physicochemical view-point. It seemed to us of importance to detect some purely chemical characteristics of the crystalline pepsin, and with the consent of Dr. Northrop such an investigation was undertaken in this laboratory.

In course of the study of the products of hydrolysis of the crystalline pepsin which is now in progress, a peculiarity was found in the composition of this protein which differentiates it from any other protein, namely, the extremely small content of the basic components. Thus, the twice crystallized material contained only 4.65 per cent. of its total nitrogen in the form of basic substances. The significant feature, however, is that the material crystallized five times contained only 3.3 per cent. of its total nitrogen in the form of basic nitrogen.

The conclusion to be drawn, then, is that the crystalline pepsin, in the main, is an individual protein but it probably contains a small admixture of an extraneous protein. We are endeavoring to prepare the crystalline material free from the impurity.

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VITAMIN D IN WHOLE CORN

IN SCIENCE of January 23, 1931, Harris and Bunker[1] reported irregularity in the development of rickets in rats fed on diet No. 2,965 of Steenbock and Black.[2] The authors suggested that this may have been due to the presence of antirachitic factor in the corn contained in the diet.

It is true that of a group of rats fed on the same batch of diet No. 2,965 containing freshly ground whole corn, one or more animals occasionally fail to develop rickets, at least in the usual period of observation. This also holds true for diet No. 3,143 of McCollum,[3] if administered in the form in which it is usually prepared. But most investigators of experimental rickets have attributed this rather unusual occurrence either to a large store of antirachitic factor in such animals or to partial or complete inanition which interferes with the development of the disease. Another possibility that must always be ruled out is the contamination of the cage by a minute amount of very potent antirachitic substance (irradiated ergosterol) that may have been used in a previous experiment. Most investigators of the subject have now had this untoward experience. I am of the opinion that a fourth very likely explanation is the settling out of the calcium carbonate of the diet so that, as consumed by the animal, it does not have the high ratio of Ca to P which is a necessary condition for the production of rickets by this diet. Even if the diet is thoroughly

[1] R. S. Harris and J. W. M. Bunker, SCIENCE, 73: 95, 1931.

[2] H. Steenbock and A. Black, *J. Biol. Chem.*, 64: 263, 1925.

[3] E. V. McCollum, N. Simmonds, P. G. Shipley and E. A. Park, *Am. J. Hyg.* 1: 492, 1921.

mixed before serving, and is served fresh daily, the settling out of the calcium carbonate is still a very likely occurrence.

To avoid the settling out of the $CaCo_3$ and NaCl from diet No. 3,143 I have been in the habit of melting the gelatine it contains, in warm water (60 cc to every 15 grams of gelatine), incorporating the remaining constituents of the diet in this solution and stirring thoroughly until the mixture is a solid jelly. This insures the maintenance of the even distribution of the soluble and insoluble ingredients of the diet and keeps the ratio of Ca: P quite constant. A weighed portion of the jelly is served to the animal, and the residue is weighed after an interval of 24 hours to determine the amount consumed. It is easier to do this when the food is served in this form rather than in the form of a dry granular mixture, because the residue is in the form of one or a few dry masses which do not fall through the wire floor of the cage and can be easily separated from the feces. This form of diet No. 3,143 is eaten greedily by young rats and they develop a remarkably uniform degree of severe rickets in the usual period of four weeks.

Recently Shohl and Brown,[4] working with diet No. 2,965, modified by the addition of calcium or phosphorus compounds, so that the ratio of Ca to P was varied in a number of ways, obtained rather unexpectedly irregular results. Upon the assumption that the cause may have been the inconstancy of the various ratios of Ca: P, they have adopted the above method of maintaining them constant. They have effected this by replacing 6 per cent. of the corn in diet No. 2,965 by gelatine, and by incorporating the other ingredients in the jelly, as outlined above for diet No. 3,143.

It is not claimed that the results of Harris and Bunker[1] are accounted for by any of the explanations given above, but it seems timely to draw attention to some of the other possible explanations of the occasional irregular results obtained with rickets-producing diets served in the usual form of dry, granular mixtures, and to indicate a way of avoiding one of the causes.

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VIABILITY AND RETENTION OF VIRULENCE OF A CULTURE OF CORYNEBACTERIUM DIPHTHERIAE

IN December, 1928, a pure culture of *C. diphtheriae* was isolated from the throat of a patient. It fermented glucose and gave a typical virulence test in guinea pigs. The culture was grown on chocolate agar, placed in the refrigerator at the time of maximum growth, and transplanted every two weeks until June, 1929. At that time generous loopfuls of culture, each mixed with a drop of sterile rabbit's blood, were placed on each of a number of small bits of sterile filter paper, transferred to sterile Wassermann tubes, and kept *in vacuo* in anaerobic jars (Smillie) for a period of three months. The anaerobic jars were kept in an electric refrigerator.

In October, 1929, the culture was removed from the anaerobic jar, grown in meat infusion broth for twenty-four hours, and then planted on chocolate agar. It grew well, fermented glucose, and again gave a positive virulence test in guinea pigs. The culture was kept *in vacuo* for a second period of three months in the summer of 1930 and, when used for class work in December last, appeared to have undergone no loss of virulence.

It is known that *C. diphtheriae* survives preservation *in vacuo* after the method of Brown,[1,2] but it may be of interest to teachers to know that the culture will remain virulent under such simple conditions as those described.

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FORMALIN POISONING

FEW laboratory workers in the biological sciences have escaped some disagreeable experience with formalin. I know one pathologist who always wears gloves to hide the condition of his skin and another who should if he valued appearances. His hands are swollen, discolored and cracked. (Since I first wrote this he has gone into the hospital with a severe infection that started in these sores.) When I last saw my former anatomy professor he had both hands bandaged for formalin sores. I never heard that sinus or lung trouble had been traced directly to inhaling the fumes, but in some cases they are very irritating to the mucous membrane.

One of the worst features of formalin is the insidious and cumulative nature of the trouble it causes. A person may use it for a year or more before any symptoms appear; then the skin of the hands begins to dry, harden and form painful cracks that heal very slowly. Sometimes suppuration starts under the nails. After the victim has once become susceptible he generally has the trouble with him as long as he has anything to do with formalin. Rubber gloves give only partial protection, for a very slight exposure is enough to start trouble.

Formalin is such a useful laboratory reagent that it is almost impossible to avoid some contact with it. Aside from alcohol and water it probably has more uses than any other fluid.

[4] A. T. Shohl and H. B. Brown (unpublished).

[1] J. H. Brown, *Abs. Bact.*, 9, No. 1, 1925.
[2] *Ibid.*, SCIENCE, 64: 429, 1926.