Editorial.

REPORT OF THE LISTER INSTITUTE OF PREVENTIVE MEDICINE.


The first subject dealt with under the heading of Research Work is the study on viruses. During the year studies on various aspects of the virus problem have been continued by workers in the Department of Bacteriology, Serology and Experimental Pathology.

Dr. M. H. Salaman has continued his investigation into the antigenic structure of the vaccinia virus. He has compared the serological properties of the elementary bodies with those of the soluble antigen of the virus which can pass through filters that retain the bodies. It has emerged that the virus neutralizing power of an anti-vaccinal serum can be removed by absorption with a sufficient dose of washed elementary bodies, but not by absorption even with large doses of the soluble antigen. On the other hand, precipitins and agglutinins can be absorbed from such serum in varying degrees by the bodies and by the soluble antigen.

Dr. G. H. Eagles, with the assistance of other workers, has continued the study of a possible virus agent in the causation of acute rheumatism, rheumatoid arthritis and chorea.

The inquiry has been conducted on three main lines: (1) Specific agglutination tests with the sera of patients; (2) infection experiments in monkeys; (3) electrocardiographic investigation of sick animals that appeared to react to experimental inoculations. Suspensions of elementary bodies recovered from rheumatic exudates (pericardial, pleural, and joint), joint fluids and synovial membrane from rheumatoid arthritis and in a few instances from the spinal fluid in chorea have been agglutinated by sera from homologous cases. Evidence of cross agglutination within the rheumatic group suggests that these diseases have in all probability a common aetiological factor. Concomitant infection by streptococci probably plays a part, and the interaction of these with the agglutination reaction has been studied throughout the clinical course of the disease in a series of 200 cases. Infection experiments with monkeys have not so far given unquestioned confirmation of serological findings.

As regards the use of the electrocardiograph in diagnosing the nature of cardiac involvement of monkeys. A complicating factor is that little is known of the simian electrocardiogram either in health or disease. Experiments are under way to discover, if possible, the determining factors in the causation of the prolongation, in monkeys, of the P-R interval,
which is considered to a large extent pathognomonic of early rheumatic carditis in the human subject.

Under the heading of serology, studies on the "Vi" antigen of *S. typhi* are described by Dr. Felix. His earlier conclusion that only strains containing both the "O" and the "Vi" antigens possess the highest degree of virulence of which *S. typhi* is capable, is confirmed by recent investigations. A variant has been described which in all respects behaved like a typical rough variant and was avirulent although it contained "Vi" antigen. From a culture of this strain, which Dr. Petrie had kept for ten months in horse serum containing both "O" and "Vi" antibodies, another variant was isolated, which, though devoid of "O" antigen, otherwise resembled the smooth type. Growth on agar plates and in broth showed the characteristics of smoothness and suspensions of this variant were not agglutinated by salt solutions up to 5 per cent or by heating to 100° C. The virulence for mice was of the same low order as the "O" agglutinable, smooth type, which is devoid of "Vi" antigen.

As regards physico-chemical properties the "Vi" antigen contained in smooth typhoid bacilli appears to differ from that in the "rough" typhoid bacilli. Suspensions of smooth "Vi" strains heated at 100° C. are no longer agglutinable by "Vi" antibody, while similarly treated suspensions of a rough "Vi" strain are still agglutinated. Treatment with 75 per cent alcohol also has different effects on the "V" agglutinability of the two varieties of "Vi" strain—the agglutinability of the smooth variants is much reduced, practically annulled, whereas that of the rough variant is not impaired. On the other hand the two variants do not differ with regard to the effects of heat or alcohol on their capacities of absorbing the "Vi" antibody or stimulating its formation.

In view of these findings the resistance to chemical treatment of the "Vi" antigen contained in "smooth" and "rough" variants was carefully investigated. Suspensions exposed to the action of HCl, NaOH, phenol and formalin did not show any difference in the two variants as regards agglutinability, absorbing power, and capacity of inducing the formation of circulating antibody. It would appear that the physico-chemical behaviour of an antigenic substance may vary as the result of the presence or absence of some other substance, which itself may be either antigenic or non-antigenic in nature. It is considered that the difference noted in earlier experiments between the effects of alcohol on the "Vi" antigens of *S. paratyphi* A and B, and *S. Aertrycke*, on the one hand, and the "Vi" antigen of *S. typhi* on the other hand, is of no greater significance than that now established for the "Vi" antigen contained in "smooth" and "rough" typhoid bacilli.

Experiments with antityphoid vaccine have not yet enabled a vaccine to be prepared in which the "Vi" antigen is preserved in its most effective form. From Dr. Schütze's experiments it appears that active protection
experiments with mice do not disclose those great differences in the antigenic value of various preparations of the “Vi” antigen which are clearly demonstrated by passive protection experiments or by *in vitro* tests.

Dr. Felix, in co-operation with Dr. W. D. Nicol, has tested the antibody response in fifty persons who had been given three doses of an alcohol-killed typhoid vaccine by the subcutaneous route. A marked increase in the “O” antibody was observed in 100 per cent of those inoculated, and a significant increase of the “Vi” antibody in 50 per cent. After storage for half a year, however, it was found that the vaccines had lost the power of stimulating the formation “Vi” antibody in the rabbit.

Dr. Felix has been working on the possible value of “Vi” agglutination in the detection of typhoid carriers. Sera from forty-five typhoid carriers have been examined, and the results obtained suggest that “Vi” agglutination seems to have a strong claim to a definite place in the routine diagnosis of typhoid carriers. It is noted that all the twenty-five strains from chronic typhoid carriers examined were found to contain “Vi” antigen.

The antigens of *B. pestis* have been investigated by Dr. Schütze; his early work showed the importance of the envelope component in prophylactic inoculation of rats. Similar work performed on mice at the Haffkine Institute, Bombay, has given divergent results and Dr. Schütze is now undertaking a comparison of the two types of immunity as developed in the mouse and in the rat. As investigations at Bandoeng, Dutch East Indies, have shown that living avirulent plague inoculations produce excellent immunity, but only when the organism is in what is considered to be the smooth state, Dr. Schütze is including the so-called rough and smooth variants in his analysis.

Modifications in the technique of the Chick-Martiu test for disinfectants have been suggested as a result of experiments carried out for a sub-committee of the British Standards Institution concerned with the standardization of the methods for the testing of disinfectants. The most important modification suggested is the substitution of dried yeast for dried faeces as the organic matter of the test. This was first suggested in 1934, by Dr. Garrod, a member of the committee, and has been found of considerable value.

In order to obtain concordant results special attention has been paid not only to the method of maintaining the stock culture, but also to the medium on which it is grown for the test.

A much criticized point in the technique has been the addition of the disinfectant to the organic matter before the culture. It has been stated that the phenol coefficients so obtained would be lower than if the organic matter and culture were added to the disinfectant together. This criticism has not been supported by experimental data. Tests have shown that the organic matter and the disinfectant may be well mixed together and left
Work on the vitamin-B complex has been carried out by Dr. Macrae and Miss Edgar, who have shown that aqueous yeast extracts contain two factors in addition to B₁ and flavin necessary for the growth of rats. One of these factors is removed with flavin from this yeast extract by absorption in fuller's earth in acid solution, while the other remains in the fuller's earth filtrate and can be eluted with barium hydroxide.

The establishment of these two factors in addition to B₁ and flavin will help to clarify the position of the vitamins in the B group.

Dr. Macrae and Miss Edgar have been engaged during the year on the chemistry and purification of the factor not absorbed by fuller's earth. The results suggest that this vitamin may be distinct from B₆, as described by György.

Miss E. M. Hume and Miss Henderson Smith have been studying a dietary deficiency in rats which affects their breeding capacity, the full-term young dying in utero or failing to survive after they are born. Such young as survived last year showed skin lesions characteristic of the lack of the unsaturated fatty acid (linoleic acid). This year, by the addition of linseed oil the skin lesions have been corrected, but the other features of the syndrome have only been partially improved, and it is thought these symptoms might be due to a partial deficiency of vitamin E which when totally lacking causes resorptive gestations in the female. The effect of the addition to the diet of an unsaponifiable fraction of wheat germ oil is now being tested.

Dr. S. S. Zilva working on the vitamin-C requirements of the guinea-pig has compared the vitamin-C content of selective organs with the intake of ascorbic acid. He found that to attain the maximum concentration in the tissues, ten times the protective dose had to be given. But guinea-pigs which contained only minimal amounts of vitamin C in their tissues lived for a number of years and attained very high weights. Furthermore, the time taken to succumb to scurvy when placed on a scorbutic diet was not appreciably different whether the tissues carried their maximum load of ascorbic acid or only traces of it when the scorbutic diet was commenced. The accumulated vitamin C in the body of the guinea-pig does not act as a store in the true sense of the word. The results of this investigation lend support to the view expressed by Dr. Zilva and Dr. S. W. Johnson, from general observation on human beings, that there is a wide margin of unsaturation with vitamin C which has no obvious detrimental effect on the health of the individual.

A collective investigation has been organized by Miss Hume as Secretary of the vitamin-A sub-committee to compare the results of biological and spectrographic estimations of vitamin A. The aim was to obtain trustworthy figures for the conversion factor required for relation of the results of the biological and spectrographic tests and expression of this in International Units. Ten different laboratories participated in the tests and the
results showed a satisfactory degree of concordance and gave no support to any alteration of the value 1,600 adopted for the conversion factor at the 1934 International League of Nations Vitamin Standardization Conference. Discrepancy observed in the case of some estimations was found to be due to instability of the material during the period of the biological test and this suggested that other vitamin-A concentrates might behave in a similar manner. Further work is contemplated on these lines with a view to explaining other reported discrepancies. If these cannot be satisfactorily resolved there is a possibility that the spectrographic method may have to be abandoned and vitamin-A standardization made to depend on the biological test alone.

Dr. H. Chick has collaborated with Dr. Birch and Sir Charles Martin in an experimental investigation of the nutritive defects of maize in the hope of throwing light on the aetiology of pellagra as it occurs among populations which consume maize as staple cereal. The experimental animals were pigs which were given a diet containing over 80 per cent of ground whole white maize supplemented with peameal and a small amount of purified casein to increase the protein content, extra salts, and cod-liver oil. Dogs on this diet develop a disorder known as nutritional black tongue, which is held by many to be the analogue of human pellagra. After about six weeks on this diet the young pigs ceased to grow, showed loss of weight, anaemia, and severe diarrhoea and died unless the diet was changed. They showed no skin sensitisation to sunlight. The disease was prevented by the addition of 4 per cent yeast in the diet, or if the maize was replaced by a mixture of wheat and barley. The disease was cured in a dramatic manner when yeast or an autoclaved protein-free yeast was added to the diet. These facts pointed to a deficiency in maize of some heat-stable constituent in the yeast extract, e.g. some constituent of vitamin B₂. Preliminary tests with flavin gave negative results.

An investigation of wheat and bread was begun by Dr. Roscoe in 1935, and has been continued by Miss Copping. Samples of whole wheat flour and straight run white flour, as commonly used for bread making in this country, were provided by Messrs. Chitty and from these flours breads of standard composition were baked under controlled conditions in the experimental bakery of Dr. Kent-Jones. The wholemeal flour and the white flour, both unbleached and after bleaching, and the yeast used in the baking of the bread were investigated by means of growth tests on rats for their content of vitamin B₁ and B₂ and by separate tests for that of flavin and other constituents of the vitamin-B complex and the results compared with those obtained from equivalent amounts of baked bread. The vitamin B₁ and B₂ contents of the wholemeal flour and bread were much greater than those of the white flour and bread. These latter, however, proved unexpectedly rich in vitamin B₁ and
the amount in the bread was not to be attributed to the added yeast, seeing that the values for the bread and the flour were equivalent. Control experiments with ground polished rice, in which the rats developed characteristic nervous symptoms of $B_1$ deficiency emphasized the difference between polished rice and white flour in this respect and afforded an explanation of the rare occurrence of beri-beri on one-sided diets which contain wheat as the staple cereal.

The content of flavin was low in both types of flour and breads, the white flour and bread being definitely inferior to the wholemeal. The second constituent of the vitamin-$B_2$ complex (the so-called vitamin $B_6$) was abundantly present in the wholemeal flour and bread and less so in white flour and bread. The nutritive value of wheat flour and bread would appear to be limited by their low content of flavin.