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CERTAIN PRACTICAL ASPECTS OF IMMUNITY.*

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In reviewing the great amount of work being carried out in immunology and its vast literature, the general practitioner, medical or veterinary, has a right from time to time to ask of the workers in these fields what advances in medicine have resulted—advances so generally accepted as to be reasonably considered permanent, and which he must therefore be prepared to use in his practice. It was arranged that, as Professor Browning is dealing with the more attractive explanation of the phenomena underlying certain aspects of immunity, I should deal with what we may call the practical achievements of immunology in the recent past, and also survey briefly the tasks just ahead; since comparative medicine is represented at this Section, a review of these achievements in human and veterinary medicine side by side may not be out of place.

In endeavouring to estimate future tasks and possible lines of advance, what lessons can we learn from the immediate past? During the past century the death rate from many human diseases has been much reduced. Those that spring at once to the mind in this connexion are the intestinal diseases—typhoid, cholera, and dysentery, and typhus and plague. I do not know that we can draw from veterinary epidemiology a close parallel.

If we endeavour to draw up an order of merit for those who have achieved this reduction, we shall have to award the greater honour to the hygienist and sanitarian, who insists on general measures of common-sense cleanliness, fresh air, a decent allowance of space for the individual—that is, the avoidance of overcrowding—and the guarding of food and water supplies. Next in order would come the immunologist, who presents himself mostly with a syringe and needle. Last of all the clinician, when disease surmounts the barriers set up by the hygienist and vaccinator, to-day treats his patients with more knowledge than of old, and reduces the death rate either with "specific" remedies, such as antitoxin, as in diphtheria, or with greater clinical skill, until he to-day expects a mortality not more than half that of an earlier date.

* A paper read in a discussion on immunity at a joint meeting of the Sections of Pathology and Bacteriology and of Comparative Medicine at the Annual Meeting of the British Medical Association, Edinburgh, July, 1927. Professor T. J. Mackie was in the chair.

But will the contribution of these three groups of medical workers bear the same relative proportion in the advances of the future? In considering these lines of advance the teachings of general epidemiology, and particularly those of experimental epidemiology, and of the influence of avitaminosis on susceptibility to infection, must be borne in mind. We are beginning to have a clearer idea of many points, thanks to the work of Topley and others, particularly of the exact way in which isolation of infected groups of people and the division of these groups into small units may control infection.

The condition of nutrition of the animal exposed to infection may be of at least as great importance as the "virulence" of the threatening organism. To judge from our own experience over some years, it is almost capable of direct proof that a stock of guinea-pigs, for instance, with certain potentially pathogenic bacilli present in their intestinal flora, and therefore in their surroundings, may remain healthy and not show an outbreak of the disease in question until the content in vitamins, salts, etc., of their diet be reduced beyond a certain point, or their living conditions be made unsuitable by exposure to extremes of temperature, uncleanliness, etc.[1] Present evidence suggests that the outstanding advances of the future will come more from the immunologist than from the sanitarian or hygienist.

We are perhaps apt to forget how big a part the man with the syringe and needle already plays. Some striking figures can be gleaned from recent veterinary literature. Thus we find that probably upward of two to three hundred thousand pigs are injected yearly on the Continent with swine fever serum, the number in the United States of America being apparently greater; in Holland and California alone probably half a million fowls are vaccinated against avian diphtheria in a year; of rinderpest serum East Africa and India last year used between four and five million doses; in one year about two million dogs in America and Japan were vaccinated against rabies; eight million cattle were tested last year in the United States with tuberculin. The only large figures in human medicine that may bear comparison are the millions vaccinated with the various typhoid vaccines during the war and those protected against tetanus with serum, and the 500,000 children in the United States who have been tested by the Schick method or immunized with diphtheria prophylactic.

If, then, we agree that the future is largely with the immunologist or vaccinator armed with syringe and rustless steel needle, we may well consider what he is to inject and how he may best inject it.

"Bacterial Vaccines."

In human medicine we may expect universal acceptance of the statement that immunity by thorough vaccination can be conferred on those brought into contact with typhoid and the paratyphoid fevers, bubonic plague, probably

cholera and dysentery, and possibly, under certain circumstances, pneumonia; in veterinary medicine—anthrax, blackleg, chicken cholera, leptospiral jaundice, and haemorrhagic septicaemia, and probably swine erysipelas and avian diphtheria or " roup."

How best can we immunize in these diseases—with live or dead vaccines, at what intervals? For human work *living* or " attenuated " living vaccines have been used, but have gone almost wholly, and probably permanently, out of use. Presumably, as recently ably argued by Ledingham, the living bacillus will produce a higher immunity than a dead one, and perhaps the ideal vaccine would be a certainly non-pathogenic strain. But in human work the tide has set strongly against living vaccines, because of the obvious danger of the non-pathogenic agent becoming pathogenic.

In veterinary work this same danger is well recognized. The only living bacilli widely used are those for swine erysipelas, contagious abortion of cattle, and " blackleg " of cattle and sheep. We must freely admit that we do not know what is the best method of using these agents. An examination of many of the commercial swine erysipelas vaccines available reveals a wide difference in bacterial content. The wisdom of vaccinating against abortion is much debated. Some authorities maintain that dead vaccines do not give sufficient protection to be of use; others condemn the living vaccine because it may introduce the infection and abortion into a herd previously free. The recent use in the United States of a bacillus apparently entirely free of pathogenicity may help in solving the problem. But at present it must be freely confessed that authorities vary so much that we must wait until further experience gives us riper judgement. The plan favoured in the United States of trying to build up " certified " herds by rigorous exclusion of " reactors " will be watched with interest. For many years " blackleg vaccines " containing live spores of *B. chauvoei* were used. Of late bacteria-free " vaccines," the filtered muscle juice of artificially infected cattle, or the simple filtrate from a culture of the organism, have come widely into use.

Various living " non-pathogenic " cultures of *B. tuberculosis* have been used during the past two decades. The latest is B.C.G., an " attenuated " strain used by Calmette and Guérin; it is believed by these workers to be completely non-pathogenic and to possess considerable immunizing power. Although much evidence in its favour is being brought forward on the Continent, the efficiency and safety of this vaccine have not been widely accepted in Britain or America. Kraus and Gerlach in Vienna, and various speakers at the recent Russian congress, sound a note of caution. Until we begin to have some understanding of the immunological relation of tuberculin to tuberculosis comparable to our present conception of the relations between toxin and antitoxin in diphtheria, one avenue to progress in our attack on this disease remains closed. It is curious that ordinary " exotoxic toxins " of

diphtheria, tetanus, etc., are toxic for the normal as well as the infected animal, whereas tuberculin, of which 0.1 c.cm. will readily kill a tuberculous guinea-pig, is innocuous to healthy animals in doses fifty times as large. A clear understanding of the "tuberculin" reaction would probably give us the key to much.

We have examples of the successful use of *filtrates of bacterial culture* in Dick toxin for immunization in scarlet fever in human medicine, and of filtrate ("artificial aggressin") for immunization against blackleg in cattle. Filtrate is also used for protection against braxy, the winter disease of sheep caused by *Vibrion septique*. Whether the braxy filtrate can be made of such antigenic potency as to yield an immunity equal to that given by properly balanced toxin-antitoxin mixtures is not yet certain.

Toxin deprived of all toxicity has attracted much attention of recent years. With formalinized diphtheria toxin, rendered completely atoxic to animals, good immunity to diphtheria can be secured. Whether this "toxoid" or "anatoxin" will eventually replace toxin-antitoxin (U.S.A.) or toxoid-antitoxin (England) prophylactic mixtures for immunization against diphtheria in man and tetanus in animals must be left unsettled until further experience has been gained. Toxin "detoxicated" with ricinoleate of sodium (Larson) has been introduced during the recent past. It is doubtful whether this will replace the ordinary mixtures and formalinized toxin, but here also we must await the verdict of further experience.

Mixtures of the toxin-antitoxin (or, better, *antigen-antibody*) type have given satisfactory results on the large scale clinically or in fully controlled laboratory experiments in protecting against diphtheria, tetanus, and "gas gangrene." Most of this practice has been founded on our knowledge of the use of mixtures of diphtheria toxin and antitoxin. It is therefore interesting to follow the course of clinical and experimental diphtheria prophylaxis, through the toxin-antitoxin mixture and toxoid-antitoxin mixture, formalinized toxoid (that is, "anatoxin"), and "floccules." In the Ramon method of titration when toxin and antitoxin are mixed in certain "neutralizing" proportions a flocculant precipitate is formed. A suspension of these insoluble toxin-antitoxin floccules when injected into animals produces good immunity. Similarly formalinized atoxic toxin or "toxoid" when mixed with antitoxin causes "floccules" to form, and my colleague Mr. Glenny has recently shown that these floccules also have high immunizing power. It is hoped that the "floccule" antigens may prove less liable to cause reactions in sensitive adults than the ordinary prophylactics and may possibly shorten the course of immunization. Recent work by Glenny (unpublished) suggests that by physical agents the toxoid can be dissociated from the toxoid-antitoxin floccules, and thus yield a much "purer" immunizing agent than has hitherto been available. An interesting special use of toxin-

antitoxin mixture has been made by Dalling in his successful work on the prevention of lamb dysentery. Here the ewe is given two widely spaced injections of the prophylactic, the second as near to the date of lambing as possible, in order that the concentration of antitoxin produced in the mother's blood may be high when the lamb is born, and the lamb may thus have enough antitoxin derived from the mother to carry it through the dangerous period of the first few weeks of its life. So far as I know, this is the only instance of the intentional and satisfactory use in human or veterinary medicine of the well known transmission of maternal immunity.

Antigen-antibody mixtures are met with, as live swine erysipelas bacilli and antiserum, and are given either as a mixture made immediately before injecting or injected simultaneously but separately into the animal's body. In rinderpest and swine fever the mixture used consists of the blood of an infected animal containing the live filterable virus and the blood of an immunized animal containing antibody. Both methods are used on a very large scale and are successful when properly made materials are used. The *serum of convalescent animals* is not used in veterinary medicine. In the prophylaxis and treatment of human measles the serum of human convalescents where available continues to be used with most satisfactory results, to bestow, according to the method adopted, either complete temporary protection to the very young or ill when given very shortly after infection, or active immunity to those receiving the serum at a late stage, when it will allow only a mild attack of measles to occur. This use of the antitoxin-containing serum at different periods after exposure to infection closely resembles that common in England in dealing with swine fever. Here the serum of immunized pigs is used; it is given to the pigs in contact, who may have accidentally received from their fellows doses of the virus of unknown size at various periods before the serum was given. A degree of immunity found in practice to be satisfactory is given by this method.

How to Use Injections of Vaccines, etc.

Some materials may give a reasonable immunity after one injection—for example, vaccinia and blackleg vaccine. Here one uses a large amount of material consistent with safety and no question of spacing of injections arises. But when giving a "course" of vaccine or prophylactic, the question of the best spacing and dosage arises. In immunizing horses against tetanus during the war Glenny[2] obtained a remarkable result when he found that horses receiving an initial injection and allowed to rest completely for three months produced within six months of entering a serum stable a higher concentration of antitoxin than horses immunized continuously over the same six months. This result suggests that a revision of methods of spacing ordinary "courses" of prophylactic vaccines might be wise.

Amongst promising and interesting lines of inquiry in the

recent past we may recall that on the "mosaic structure" of antigens, particularly in the group of pathogenic intestinal organisms, on the chemical aspects of immunity, the relation of undulant fever to contagious abortion, and the continued speculation in connexion with "non-specific therapy."

Antigens.—Much work has yet to be done before we understand clearly the import of the recent work on "rough" and "smooth" colonies, flagellar (ectoplasmic) and bacillary (endoplasmic) antigens, heat-stable and heat-sensitive agglutinogens, and "type and group specificity," but already the man concerned with the diagnosis of the intestinal pathogens, particularly of the "food poisoning group," in men or pigs and other animals, must bear all this knowledge in mind, and he who makes vaccines must inquire whether he is to make his most efficient vaccines from "smooth" cultures only. Apparently most typhoid vaccines in use are thus made.

The stimulating work on the chemistry of the proteins, carbohydrates, and lipoids in immunity continues to grow, and the astonishing changes in specificity that can be produced by introducing chemical groupings promise to lead far towards a rational conception of specificity, one of the greatest puzzles of immunity, but up to the present there is not much result from this field which concerns the daily work of the practitioner. So-called "non-specific therapy" continues to attract a considerable amount of notice, and some bacteriologists, proceeding from the assumption that clinical benefit may in some cases be definitely due to this treatment, seek an explanation in the production of fever, stimulation of the leucocytes, etc. Since there are few if any instances in which the curing of an infection can be obtained by the use of these substances in controlled animal experiments, the laboratory worker cannot have a first-hand opinion on any part played in clinical medicine by these methods of treatment.

With regard to vaccines themselves, there is nothing of new significance in recent work. The attractive hypothesis of Heist and Solis-Cohen, if confirmed, may be helpful in diagnosis and the making of autogenous vaccines. These authors hold that a patient becomes infected with a certain bacillus because the blood has an unusually low power of resisting growth of that organism; the patient's blood in a culture tube will therefore allow the infected bacillus to grow faster than any other pathogenic organism. Thus the patient's blood used in the culture tubes or plates is a favourable selective medium for the infecting organism which one wishes to obtain.

Recent inquiries suggest that some attacks of epidemic mild jaundice may be due to leptospira, and if severe can be treated with the corresponding serum. But even if they are not due to this organism the general feeling is that the practitioner would be prudent to treat all patients as potentially highly infective, and to take the same precautions with regard to infection of food by urine, faeces,

etc., as in an attack of typhoid fever. The interesting relationship between the *B. abortus* and the organisms of undulant fever and tularaemia does not as yet much affect the general practitioner.

From the Pasteur Institute have come three methods—vaccination per os, the use of B.C.G., and of " antivirus " for local immunization, which are attracting great interest, probably more on the Continent than in England or America. Apparently under carefully controlled conditions it is possible to show that by giving dead vaccine (without bile) by the mouth to rabbits, they can be immunized against otherwise lethal doses of the Shiga bacillus, as my colleague Miss Runge and others have shown. In the earlier work the margin between the effective vaccinating dose and a dose dangerous to the health or life of the rabbit was not very large, and the immunizing results were capricious. Bacteriologists are not all agreed that these difficulties have yet been entirely overcome. So far as the typhoid vaccines are concerned, the oral method, though it is used in France and elsewhere, has not established itself in England or America.

Foot-and-Mouth disease, which three years ago cost us about £4,000,000, has become less of a menace at the moment. The immunologist can hardly claim any share of the credit due to the methods of slaughter used. We may hope that by a combination of applications of the lessons of experimental epidemiology, and the promising research work going on in England and on the Continent, directed to a study of the conditions of survival and propagation of the virus and of active immunization, we may eventually be able to face outbreaks of this disease with a less heavy heart than in the past.

In canine distemper the confirmation in England of Carré's work on the filterable virus, and the highly promising results obtained by Laidlaw and Dunkin, lead us to hope that a practical method of immunization, if not already here, is almost at our disposal.

Of actual achievements in the recent past veterinary medicine can count to its credit the successful protection against braxy. The two methods in use—vaccination with filtrate alone or with toxin-antitoxin mixture—apparently both give good results.

Among pig diseases a most interesting development in the recent past has been the big outbreak in the United States and the frequent failures of vaccination in the later part of the epizootic, probably due to the use of serums of low activity that had to be employed in the emergency.

In the poultry world there are signs that the destructive disease bacillary white diarrhoea of chicks is to be seriously attacked in England and the results of research applied in the field. In one instance I knew a hatch of 2,000 fine chickens was reduced to 27 in about two weeks. It is reasonable to expect that by the application of the agglutination test, and perhaps the intradermic test, the dangerous infected hens may be so dealt with as to free

infected farms from the disease, financial loss being kept low by wise disposal of flocks during the process of "cleaning up" farms. Poultry breeders whom I have met are agreed that it will "pay" to build up "B.W.D.-free" poultry farms.

With much trepidation I venture to say that I think veterinary practitioners would here play a useful and proper part if they would take deeper interest in this work and train the poultry breeder to look to them constantly for aid in their troubles. In "B.W.D.," by testing and administration, and in roup by vaccination, the veterinary surgeon can promise practical aid of high value to the breeder. Veterinary surgeons on the Continent and in America find it well worth while to pay constant attention to these diseases.

International and State control of immunological materials tends to increase. The new Therapeutic Substances Act will control all such materials used in human medicine. In passing I may remark that examination for some years of various veterinary "biological products" leads me to say that at least as strong a case can be made out for the need of control of such materials used in veterinary as in human medicine.

In reviewing recent work in practical immunology to me the two greatest puzzles seem to concern tuberculin and local infection in the presence of circulating antibody. I have spoken earlier of the baffling relation between tuberculin, the tuberculin reaction, and infection with tuberculosis. The second problem arises when we remember that it is possible to immunize a horse or rabbit against the pneumococcus until a small quantity of its blood will protect a mouse against many lethal doses of live pneumococcus, and yet the horse or rabbit may have a damaging local progressive infection in the heart valves or elsewhere.

Parish and Okell have shown that we can give scarlet fever antitoxin to a rabbit and so protect it against a dose of live streptococci that will kill a control rabbit in twenty-four hours, and yet the rabbit injected with the serum, though it remains quite well for perhaps a fortnight, will most probably later show an inflammation in the joints, which are then found to be infected heavily with apparently the streptococcus originally injected. The obvious explanation that the local nidus is shut off from the general circulating antibodies may be sufficient, but it does not meet all our doubts. The problem has an obvious bearing on the inefficacy of scarlet fever antitoxin for the relief of late septic complications despite its clear curative effect in the early toxaemia of scarlet fever.

In conclusion, the immunologist has reasonable ground for pride in the achievements of the immediate past and a confident hope that continued work will give further advances in the control of disease.

REFERENCES.

[1] Glenny and Allen, *Lancet*, 1921, ii, 1109; Trevan and Boock, *Biochemical Journal*, 1922, xvi, 780. [2] Glenny, *Journal of Pathology and Bacteriology*, 1925, xxviii, 481.