THE PIORKOWSKI-FRIEDMANN TURTLE TUBERCULIN.¹

BY DR. PIORKOWSKI.

I SHALL first describe the historical development of the origin of my turtle tuberculin preparations. In 1901 Friedmann came to me and brought two large turtles which had become spontaneously tubercular. Their lungs contained a large number of tubercular nodules, and he asked me to prepare pure cultures from these organs. I did this, and I succeeded in obtaining pure cultures on suitable nutrient media, as glycerine agar and bouillon. Friedmann remained with me nearly a whole year in order to do further work on the cultures. Meanwhile I made a large number of experiments on my own account. Some turtles I fed, others I inoculated, and of the large number of the latter only two manifested the symptoms shown by Friedmann's turtles. It was therefore evident that we were dealing with a special strain of bacillus, which, when developing on a specially prepared nutrient medium, exhibited the symptoms of tuberculosis. It is very noteworthy that the turtle tubercle bacillus in its further behaviour, both culturally and morphologically, displayed an extraordinary resemblance to the human tubercle bacillus. Its growth at 37° F. is remarkably characteristic. The main point about this strain is that it can be used without the risk of any manifestations—a circumstance which may be ascribed to the fact that for the last ten years it has been reinoculated afresh daily, and thus has acquired generally an extraordinary innocuousness, becoming both avirulent and atoxic.

The differentiation of the various tubercle bacilli is especially well carried out by Knoll's modifications of Unch's method, consisting of a double stain by means of fuchsin, methyl violet and resorcin. This method reveals long slender structures with regular nuclei in human tubercle bacilli, but in this variety the nuclei are often seen to be irregularly divided and of a variable size. In avian tubercle bacilli they are somewhat shorter, and in turtle bacilli they are very small—almost like dots. In my opinion the extraordinary resemblance of the various tubercle bacilli is to be attributed to the zoological grouping, and such resemblances only occur in lung-breathing animals.

We must differentiate between mammals which produce their offspring alive, the class to which human beings and oxen belong; and birds—*i.e.*, that is, animals which lay eggs; and, thirdly, reptiles, which possess horny or long integument and also lay eggs. Lizards, croco

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diles, and turtles belong to the last class. Finally, we have to think of fishes which breathe as long as they are young through gills or by their lungs, and also lay eggs. We thus see very clearly that resemblances are to be found only among lung-breathing animals, and it is for this reason, probably, that the results described are obtained on the injection of tubercle bacilli of similar kind. It became very evident that turtles were especially adapted for our purpose.

The method of working with living bacilli is not entirely new, for the same principle was used by Behring in his experiments on the extermination of bovine tuberculosis. He employed human tubercle bacilli, which are atoxic to oxen, with good results; and therefore the attempt to immunize human beings with turtle tuberculin is quite rational. I can explain the process very satisfactorily by the side-chain theory of Ehrlich. According to this theory, the nutriment of an animal is taken up by means of cells. Each cell possesses a central active nucleus which carries out the function of the cell, and a so-called side-chain—a receptor. The cell takes up nutriment by means of this receptor, and then a very remarkable proceeding occurs. The portion of the cell which has absorbed the nutriment breaks away from the rest, becomes metabolized, and a new receptor is developed in the original cell; but not merely one receptor—an actual overgrowth of receptors takes place (Weigert's law of over-compensation). The new receptors have charged with nutritive material, break away, and carry the nutriment to different parts of the organism, thus effecting nutrition. This theory not only accounts for the method of nutrition, but it also explains how toxic action occurs; because cells not only possess receptors for food-molecules, but also for other substances, and these receptors are as numerous as they are varied. Some of them take up particles of nutriment, others take up poisons, absorb ferments, others the products of bacterial fermentation or similar processes.

Let us, for example, consider a toxic action a little more closely. When a poison enters the body—*e.g.*, tubercle toxin—the first point concerns the existence of receptors which can take up the tubercular poison. If these do not exist, no infection by tubercle bacilli can occur, for the organism possesses *congenital immunity* towards the action of these bacilli. But if there be some arrangement for receiving them, and appropriate receptors for the tubercle bacilli exist, these take up the poison, and if absorbed in sufficient quantity, a general toxæmia ensues, which may lead to the destruction of the organism. If, however, the receptors have the opportunity of loading themselves with the poison, and if they drop off the cell after they have become useless and harmless, then an excessive production of these receptors occurs, as stated previously. A constant succession of new receptors becomes formed; but there is not room enough on one cell for the attachment

of so many receptors, and the result is that the newly-formed elements become discharged into the serum before they have been able to take up any of the toxin. The receptors thus discharged come into contact with poison in regions far distant from the site of the sensitive cells, and are there seized upon, whereby a ventilization of the poison takes place and it is rendered injurious. In this manner an acquired specific immunity is developed, and the newly-formed bodies are termed "anti-toxins."

The harmless turtle tuberculous toxin combines with the receptors, and the combination is thrown off into the blood as antitoxin. Now receptors are formed in large quantity, but they are capable of seizing not only the turtle tubercle bacilli, with which they have been hitherto dealing, but also human bacilli, and thus render them harmless. If there is a profuse formation of new receptors, and if the human tubercle bacilli have increased unduly, complete recovery may be effected, and the *rationale* of the cure is along these lines. There is also the additional advantage that turtle tubercle bacilli are innocuous and harmless, and therefore this method is especially well adapted for protecting inoculation. But even if the body is already affected with tuberculosis the disease can be suppressed, because its course is so protracted. The process is very much the same as that of Jenner's vaccination against small-pox, except that in the latter case the agent is calf-lymph, whereas here it is the turtle tubercle bacillus. All the endeavours of therapeutics in tuberculosis based upon Koch's work, which consist of injections of living tubercle bacilli or the virus, attain a certain result, but, if accurately judged, fail to reach their ultimate aim. In order to attain this end it is necessary that the antigen which is to invoke the protective and curative powers of the body should be alive, but deprived of the poisonous properties—avirulent and atoxic. It is, further, necessary that the tubercle bacillus which is concerned in this matter should be treated as carefully as possible, and should be both active and specific. To a very considerable extent these desiderata have been arrived at.

I have made two preparations. One is the suspension of living turtle tubercle bacilli in physiological salt solution, and the other is turtle tuberculin prepared like Koch's, but no temperature higher than 35° C. *in vacuo* is employed in its preparation. The administration should be intramuscular, but it may also be given subcutaneously or into a vein. The suspension should first be given in doses of 0.5, 1, 2, 3 c.c.; later on, the turtle tuberculin in doses of 0.1, 0.3, and 1 c.c., according to the stage of the disease and temperature—in other words, according to the clinical condition. It is best to begin with 1 c.c. of the suspension, to double the dose in eight to ten days, and then to inject the tuberculin after another eight days, repeating the later

injection at frequent intervals. It has appeared, as a rule, that the doses of living turtle tubercle bacilli do not require to be used more than three times. Numerous experiments on animals and the treatment of a large number of cases have confirmed the absence of any toxic effects. Owing to the careful methods of preparation of this remedy, there have hitherto been no instances of induration or abscess; there have only been slight infiltrations, which subsided very satisfactorily. The most striking clinical results have been the rapid subsidence of fever, night sweats, pain in the chest, and other pains. Very soon there is an increase in weight, appetite returns, and the feeling of fatigue disappears.

Professor Meyer has obtained very good results with turtle tuberculin, and in his opinion it is in every way superior to Koch's tuberculin. Bandolier refers to a case which could not tolerate even the smallest doses of sensitized bacilli and albumose-free tuberculin, but which tolerated large doses of turtle tuberculin easily and satisfactorily. Klemperer is able already to point to a large number of cases wherein the remedy was well borne. Karfunkle of Berlin has also injected a large number of patients. Heid of Pittsburg has demonstrated good results in 80 per cent. of the cases.

Detailed records will be published in due course. It is obviously necessary to treat a very much larger number of cases, and the remedy requires further trial. It is best to inject into the muscles of the gluteal region. Cases of tuberculosis of the larynx and skin, and also surgical tuberculosis, have also been benefited, and it is to be hoped that the trials which are being made by many reliable investigators will be followed by good results.