REPORT ON FURTHER OBSERVATIONS ON BACILLUS VIRIDANS.

[From the Pathological Laboratory of the University of Aberdeen and the
Institut Pasteur, Paris.]

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Pleomorphism.—At the date of the first communication¹ I had observed the microbe under various guises—bacterial, bacillar, filamentous—and since that time I have seen it assume almost every known form. There is a short rod form whose diameter is half its length; there is a bacillus whose length is $2\frac{1}{2}$ to 4 times its diameter; there are short threads whose length varies from 8 to 20 times the diameter. There are filaments, wavy as a rule, which stretch across the whole field of the microscope. These long solid threads often entangle themselves into beautiful spirulina forms, like ropes twisted of two strands. Filaments are seen which taper at both ends, running away to scarcely perceptible points; others are of equal diameter throughout. Vibrio forms, wavy and snake-like, have been seen. In various cultures a beautiful spiral of 8 to 10 turns has been observed. Comma forms and spirals of one turn were numerous, especially when cultivated in Koch's gelatine plus $\frac{1}{2}$ per cent. of boric acid. Chains of bacilli occur when a little gentian violet is added to the nutrient medium. In a twelve days' culture, to which boric acid had been added, beautiful cocci were developed. Lastly, $\frac{1}{2}$ per cent. sodium formate added to nutrient gelatine furnished an enormous number of spore-bearing filaments. These observations were checked by fractional cultivations. And no more convincing example of pleomorphism could be demanded.

Abeysance of Pigment Formation.—While the microbe is restrained by various antiseptics from secreting pigment, it still retains pigment-forming properties and needs only a suitable concurrence of circumstances to render these apparent. Although boric acid inhibits its pigment-producing faculty, the culture removed from its influence and transferred to bouillon at once displays its green colour as beautifully as before. If, however, the microbe is passed through the system of the rabbit, it loses permanently its power of producing pigment in bouillon. If now this "race" or altered form is inoculated on Koch's gelatine it reassumes its chromogenic power. These two peculiarities are the same as those which M. Gessard has so beautifully shown as appertaining to *B. pyocyaneus*.

Relation to Aniline Dyes and Coloured Media.—The microbe is immediately decolorised by the alcohol in Gram's method. Gentian violet stains it deeply, methylene blue very faintly. The affinity for aniline dyes, however, varies according to the medium upon which the microbe has been nourished. Thus the organism grown on cocoa-nut stains very faintly by Kühne's methylene blue, whereas the same solution stains most intensely those microbes which were grown on carrot. The same holds good of Kühne's solution of gentian violet in regard to the growth on beetroot and parsnip. Such instances could be multiplied, and seem to indicate that the microbial protoplasm varies according to the nutritive media employed. The bacillus grows well on media coloured by aniline dyes, and never under these conditions takes on the dye. The tinted media become decolorised, or altered in colour by the microbe. Thus aniline green completely loses its colour. The action of the organism on methyl blue is very remarkable. Tubes of Koch's gelatine deeply tinted by this dye completely lose their colour except at the surface, where there remains a mere film of dusky green. On agitation, the whole medium becomes of this green hue, but when left at rest the fluid once more becomes colourless with the exception of the surface layer. This phenomenon may be recalled as often as desired, and evidently depends on the action of oxygen on the liquefied mass.

Effect on Animals.—Rabbits, guinea-pigs, mice, pigeons, etc., fed upon enormous masses of the microbe remained unaffected. Subcutaneous injections in large quantity into

white mice, white rats, guinea-pigs, and pigeons were harmless. Guinea-pigs after injections into the peritoneal cavity invariably suffered from a severe malady from three to five days. They became much depressed, remained in one place, ate nothing, were evidently in pain if touched on the abdomen, and their temperature was somewhat elevated. It is noteworthy that, after recovery, no amount of intraperitoneal injection produced even the slightest *malaise*, even when daily repeated in large quantity. By far the most interesting result was obtained with rabbits. Subcutaneous injections on the trunk were harmless; intraperitoneal injections were followed by slight and transitory *malaise*, while intravenous injections (40 m distilled water made milky by the microbe) occasionally caused death in from thirty-six to forty-eight hours, but left no organic changes recognisable after death.

The pathological effect *par excellence* was, when the microbe was injected subcutaneously over the ears, the production of baldness. This was easily elicited in all the rabbits experimented upon—twenty-five. In about ten days, as a rule, the hairs commence to fall out, and gradually the spot of baldness widens until the area comes to present exactly the appearance of a patch of alopecia areata in man. After several weeks the hairs reappear, and ultimately the baldness, as is the case in human alopecia, completely vanishes. I have produced this alopecia two and three times in the same ear of the same rabbit, so that one attack does not confer immunity against a second. This baldness is produced by small quantities of the microbe by 5 m of a milky suspension of the organism in distilled water. If 20 m or more are injected there arises a severe inflammation, the whole ear is much swollen, and hangs heavily against the side of the animal's head. In a few days the tissues slough away in such a manner as to leave an oval perforation from 1 to 2 inches long. In time the inflammation subsides, the hairs grow to the margin of the perforation, which remains permanently open, and looks as if it had been clearly punched out by some sharp instrument. These effects could not be produced with cultures of the bacillus which had been sterilised by heat or by passage through Chamberland's filter.

I desire, in conclusion, to return my warmest thanks to Professor D. J. Hamilton, and to express my admiration for the unrivalled facilities of the Institut Pasteur.

PRELIMINARY REPORT ON THE GLYCOLYTIC FERMENT OF THE PANCREAS.

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UPON consideration of the remarkable experiments of Lépine, Minkowski, and others, on the presence of a glycolytic ferment in the blood, and the apparent dependence of this ferment in great measure on the presence or absence of the pancreas, as well as the well-known pathological connection between disease of that organ and diabetes mellitus, it seemed reasonable to hope that a substance of the nature of a glycolytic ferment could be discovered in the pancreas itself. Another function of the pancreas appeared to be necessary, the production of a ferment which would bring about the glycolytic changes in the blood. Obviously, it would not be poured out, in all probability, into the common duct, as the ordinary amylolytic ferment would neutralise its action, but it would pass away from the pancreas by the portal vein. In other words, as Dr. Osler has neatly put it:—

The pancreas on this view has, like the liver, a double secretion, an external, which is poured into the intestines, and an internal which passes into the blood. This latter is supposed to be of the nature of a ferment, in the presence of which alone the normal assimilative processes can take place with the glycogen. Disease of the pancreas causes diabetes by preventing the formation of the glycolytic ferment.¹ This is rendered still more probable by Minkowski's² later experiments in the prevention of diabetes after extirpation of the pancreas by transplanting a piece of the organ to another part of the abdominal cavity, thus showing clearly that as long as even a small portion of the gland retained its vascular

¹ BRITISH MEDICAL JOURNAL, December 12th, 1891.

² BRITISH MEDICAL JOURNAL EPILOGUE, December 10th, 1892.