# THE VIRULENCE OF LEPRA BACILLI.

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Does the virulence of M. leprae vary to any appreciable extent in nature? Is it stepped up when leprosy is first introduced into a region where it has not previously existed, and where it spreads rapidly among the inhabitants as is reported to have occurred, for instance, in Hawaii and in the island of Nauru? Our inability to culture M. leprae *in vitro*, or to reproduce signs corresponding to human disease in experimental animals, makes it difficult to give a definite answer.

The whole question has been raised in a paper by J. W. Fielding (1945) on Rat Leprosy. The fact that rat leprosy is, like the human disease, confined as far as is known to one genus, and that M. leprae mur. like M. leprae has not been successfully cultured *in vitro*, has encouraged the study of the former disease with the hope of shedding light on the latter. Fielding in his paper assumes that the virulence of rat leprosy varies considerably. He says, for instance :—

" Experimental evidence, however, indicates that the virulence of the bacilli of fresh fæces is greater than that of the same fæces stored dry at room temperature for twelve to fifteen or more months.'

and again :—

" For the production of superficial lesions two things appear to be necessary. The first is the breaking down of local or general resistance accomplished by repeated inunction, or by subcutaneous inoculations with any of the various types of organisms. The second is the use of organisms of a naturally high order of virulence, Such as those from the fæces or urine, or the use of organisms of long-standing ulcers or granulomate whose virulence has been built up by passage through other animals, as the guinea-pig or rabbit by subcutaneous inoculation . . . The first evidence of infection by superficial lesions in my series was obtained by breaking down resistance by combining inoculations of small doses with inunction with large numbers of organisms. Virulent organisms for the final inunction were produced by passage through a guinea-pig, which showed a pyrimidal-shaped lesion with organisms, reaching a maximum diameter of 17 millimetres in seven days. Subinoculated into a rabbit, bacilli from this lesion resulted in a non-vascular nodule of

maximum measurements of 44 millimetres in about thirty days. Subsequently the lesion broke down after an inflammatory reaction, and the organisms being injected into a rat, produced a nine-millimetre nodule, the washings of which were injected into the same rat on the fourth day and resulted in a lesion measuring twelve millimetres. Both these rat lesions ulcerated three days later. Breed counts of bacilli from washings of these ulcers resulted respectively in one to ten fields and two organisms per field. Inunction on three rats with the second washing diluted to contain 60,000 organisms resulted in a permanent lesion in one rat. In all rats inflammatory reactions occurred with thickening of the skin; these reactions subsided in two, but the other was granulomatous, covered the whole treated area by the sixth day and contained loose bacilli; these were obviously increasing in number. By the eighth day a breed count of skin serum yielded sixty bacilli per field, which was equal to 18,000,000 per cubic centimetre; on the twelfth day many organisms had invaded polymorphonuclear leucocytes, which had now been thoroughly mobilized. Owing to interference six weeks later the lesion broke down, but gradually healed, breaking down permanentlly some eleven weeks later. The animal died nine months after primary inoculation, from an extremely heavy generalized infection which was reflected in the uro-genital and alimentary systems. The two rats in which skin thickening subsided were killed at 17 weeks and at 22 weeks respectively; in neither were internal abnormalities detected. The last mentioned rat showed no evidence of infection in lesions or organisms; in the former, however, bacilli were found in kidney, capsules, spleen, heart, liver and inguinal glands."

The evidence here that the organisms used for inunction were more virulent than ordinary rat leprosy bacilli is rather vague. The writer has frequently found that subcutaneous inoculation of a rat with a bacillary emulsion taken from a cutaneous nodule of an infected rat produced a marked local nodule, often followed by ulceration, and intraperitoneal inoculation produced a large tumour (Muir, Henderson, Landeman, 1927); whereas an emulsion prepared from infected liver or spleen seldom produced a marked local reaction, but in practically cent per cent of inoculations caused progressive disease which showed definite clinical signs in a maximum of 4 to 5 months. This tends to show that emulsion prepared from the skin lesions often contains some contaminant which when injected is chiefly responsible for the local reaction. It is not unlikely that the reactions described in Fielding's inoculated guinea-pig, rabbit and rat were due to a contaminant carried over; also that the inflammatory reaction produced in the rats treated by inunction was due to the same contaminant.

When a suspension of rat leprosy bacilli killed by heat is inoculated intracutaneously in man it produces in a majority of cases a local nodule (Muir, 1933); so that the production of a local nodule, however large, in a similarly injected experimental animal is no proof that the bacilli are alive, and still less that the animal is susceptible to rat leprosy, or that the bacilli are multiplying in its tissues. And if no multiplication is taking place then obviously the virulence could not be stepped up by even a series of such passages.

Much stronger evidence is required before it can be proved that the actual virulence of the individual rat leprosy bacillus varies. Disease-producing power of an emulsion containing a hundred per cent of live bacilli would be much greater than that of an emulsion with only ten per cent of the bacilli alive. Also an irritating contaminant such as that mentioned above, or the Kieselgur used in Ota and Nitto's experiments (1941), may possibly produce more active results. But in neither case would it be true to say that the virulence of the bacilli had been increased.

Proof has been obtained that different strains of tubercle bacilli vary in virulence, by injecting pure fresh cultures in laboratory animals, and noting the time required to produce death. Here the bacilli are presumably all, or almost all, alive and no irritating contaminent (dead or alive) is present in the suspension injected. Also a standardized number of bacilli is injected, and the speed of death is increased by injecting subdurally so that extraneous factors associated with a long period of illness are excluded as much as possible.

In the case of rat leprosy an approximately pure and standardized suspension of bacilli might be obtained by Dharmendra's (1941) chloroform method, but it would be impossible to tell how many, if any, of the bacilli were alive after treatment with chloroform.

If it is difficult to estimate the virulence of the rat leprosy bacillus for which we have at least one experimental animal available, how much more difficult is it to estimate the virulence of the human leprosy bacillus, for which we have no practicable experimental animal.

Clinical evidence of varying virulence of M. leprae is no less difficult to find. As has been remarked above, the more prolonged the course of a disease the more opportunity is there for complicating factors to obscure the evidence of the disease-causing power of the specific organism. If the history of a family widely infected with leprosy is studied, severe lepromatous cases are found alongside of cases with only slight neural leprides. The difference is obviously not due to infection with strains of varying virulence, but to varying resistance and degree of infection. As Dharmendra and Santra (1945) have shown :

"A factor which appears to have a bearing on the observed variations [in lepromatous-rate and child-rate] is the attitude of the people towards leprosy, and the presence of a custom of isolation of leprosy patients in a community. In areas in which there exists ostracism of the leprosy cases, and where some sort of isolation is practised, a high lepromatous rate is associated with a low gross incidence, and a low child-rate. On the other hand, in the areas where no isolation is practised, a low lepromatous rate is associated with a higher gross incidence and a higher child-rate."

Lowe (1938) has shown that in Burma the disease is more severe in Burmans than in Indians living in the same climatic conditions, but here again the difference cannot be conceived as being due to varying strains.

Leprosy has been shown to pursue an epidemic curve, increasing for a time and then diminishing. This is best shown in an isolated community such as that on the island of Nauru in the Southern Pacific. But here from the beginning severe and mild cases were found side by side, and the rapid increase and elimination of the disease was obviously not dependent on changing virulence of the infection, but on initial absence of precautions followed later by well-planned preventive regulations.

Leprosy is in a very real sense a sociological disease. It spreads in areas where the sanitary standard is low; it is controlled and stamped out when society reacts against it either by taking special precautions or by raising the general sanitary level.

If the virulence of M. leprae is fixed and, unlike other pathogenic organisms, does not vary appreciably in different strains, can this be accounted for by other peculiarities of the disease? Virulence of organisms can be modified in the case of anthrax by cultivation at a raised temperature, in tuberculosis by growing in the presence of an antagonistic substance like bile, and in smallpox by passage through other species. But such methods are not so far available with the leprosy bacilli. The virulence of influenza or the common cold may be raised by rapid human passage, but one passage of the leprosy bacillus takes at least a year, generally far longer. It may thus be that the slow growth of M. leprae and the practical impossibility of its culture outside the human body may account for its fixed virulence. Even in advanced lepromatous cases with the whole of the skin surface deeply involved, clinical signs of toxicity may be absent and the general health of the patient good, while death is generally the result not of leprosy but of complications. It would thus appear that the toxicity and lethal effect of M. leprae is of a low order, and perhaps because of this its virulence as regards aggressiveness also
at floor level.

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