## EFFECT OF SUNLIGHT ON THE STAINING PROPERTIES OF THE LEPROSY BACILLUS

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INTRODUCTION.

At the suggestion of Dr. Ryrie, Medical Secretary of the British Empire Leprosy Relief Association, we investigated the effect of sunlight on the staining properties of the leprosy bacillus. In view of his investigations on the subject he suggested that unstained smears containing the bacilli be exposed to sunlight for varying periods up to 30 minutes, and then stained by Ziehl-Neelsen's method to see if the exposure made any difference to the property of acid-fastness of the bacilli. No difference was however noted after exposure for 30 minutes, but on extending the period of exposure some interesting observations were made, which form the subject of this preliminary note.

Effect of Sunlight.

A number of smears were made from a suspension prepared from leprous tissue and rich in the leprosy bacilli. Some of the smears were stored in the dark to serve as controls, while the others were exposed to bright sunlight for periods varying from 1 to 12 hours. The unexposed and the exposed smears were then stained with the. Ziehl-Neelsen method and examined. The unexposed smears showed large number of bacilli stained red in every field. No appreciable difference was observed in the acidfastness of the bacilli in the smears exposed for less than six hours. However, in the smears exposed for six hours the number of bacilli stained red, was considerably lower, and with further exposure this number gradually became less and less, till at the end of eighteen hours there were hardly any acid-fasts left.

The experiment was repeated with fresh batches of smears made from suspensions prepared from several other patients. Similar results were seen, but it was noted that if the suspension contained excess of tissue matter, the bacilli lying within this tissue matter were protected from the effect of sunlight, and still stained red even at the end of 18 hours.

EFFECT OF COVERING SMEARS BEFORE EXPOSURE.

The question was then investigated as to the particular rays in the sunlight which were responsible for the above noted effect. Smears prepared from a suspension of leprous material were divided into two lots; those in one lot were wrapped in pieces of black paper, and the other lot was left uncovered. Both the covered and the uncovered smears were exposed to sunlight for varying periods from 6 to 18 hours. They were then stained with Ziehl-Neelsen and examined for the acid-fastness of the bacilli. In the covered smears the bacilli remained acid-fast even after 18 hours exposure; in the other series, however, there was a gradual decrease in the number of bacilli stained red, and in the smears exposed for 18 hours there were hardly any acid-fast bacilli to be found. Similar findings were made on repeating the experiment.

It was therefore concluded that it was not the heat rays but the actinic rays at the ultra-violet end of the spectrum that were responsible for the effect noted above.

EFFECT OF ULTRA-VIOLET AND INFRA-RED RAYS.

Smears made from suspensions of leprous materials were then exposed to ultra-violet and infra-red rays, for varying periods and

## Effect of Sunlight

then stained with the Ziehl-Neelsen's method. The smears were placed at a distance of 12" from the ultra-violet and infra-red lamps, and the rise in temperature at the end of exposure was to 33°C and 68°C respectively. It was found that in the smears exposed to ultra-violet there was a gradual reduction in the number of bacilli stained red, till there were hardly any left after 12 hours' exposure. The exposure to infra-red, however, did not make any difference to the acid-fastness of the bacilli.

## THE RAT LEPROSY BACILLI AND THE EFFECT OF LIGHT.

The above experiments were repeated with smears made from suspensions of rat leprosy material obtained from white rats in an advanced stage of experimental rat leprosy. It was found that the property of acid-fastness of these bacilli was not affected by either sunlight or ultra-violet and infra-red rays.

## DISCUSSION.

The above observations have a bearing on certain important matters such as the staining of smears taken from leprosy patients during field investigations, the possible therapeutic value of ultraviolet light in leprosy, and the difference between the organisms of human leprosy and rat leprosy.

In the course of field work in leprosy, it is often necessary to send the skin smears for staining and examination to a laboratory at some distance. If the unstained smears are left lying about exposed to sunlight, either in the field or in the laboratory, it may result in positive smears being reported on as negative, especially in the case of smears with small number of bacilli. However, if the smears are carefully stored, preferably after being wrapped in paper, they can be later stained and examined in the laboratory without vitiating the results.

Regarding the possibility of the use of ultra-violet rays in the treatment of leprosy, it can no doubt be said that previous trials in this connection have not given encouraging results. But in view of the action of the ultra-violet rays on the leprosy bacillus in smears, the matter appears to deserve further investigation. It may be that because of the bacilli being deeper in the skin, or because of some other reason the ultra-violet rays have no effect on the leprosy bacillus in the body of the patient.

The difference in their susceptibility to sunlight is yet another evidence in support of the view that the organisms of leprosy and rat leprosy are two separate organisms. As a matter of fact there is no point in naming the disease in rats as rat leprosy and its causative organism as rat leprosy bacillus. SUMMARY.

- 1. Exposure to sunlight of unstained smears containing leprosy bacilli interferes with the property of acid-fastness of these bacilli.
- 2. If the smears are covered with black paper before being exposed to sunlight, the staining property of the bacilli remains unchanged.
- 3. Exposure to ultra-violet rays has the same effect as exposure of uncovered smears to sunlight, while exposure to infra-red does not affect the staining properties of the bacillus.
- 4. Exposure of the rat leprosy bacillus to sunlight, ultra-violet, or infra-red rays, does not affect its staining properties.
- 5. The above observations have a bearing on certain important matters, such as the staining of smears taken from leprosy patients during field investigations, the possible therapeutic value of ultra-violet light in leprosy, and the difference between the organisms of human leprosy and rat leprosy.