

**Bacteriology of Leprosy. IV. Bacteremia.\***

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Cultures were made of the blood of 462 patients suffering with advanced leprosy in its various manifestations. Only one sample of 10 cc. was taken from each patient and a separate sterile syringe was used in each instance. The skin over the arm vein was thoroughly scrubbed with alcohol and tincture of iodine was applied and allowed to dry 2 min. before the insertion of the needle. The contents of the syringe were transferred directly into the culture or centrifuge tubes.

Two hundred specimens were cultured by the technique of Löwenstein.<sup>1</sup> The blood was centrifugated at once, the serum withdrawn with Pasteur bulb pipettes, the red corpuscles dissolved by the addition of sterile distilled water until the hemoglobin was entirely removed. The precipitate was then transferred to the surface of Löwenstein's medium which had been distributed and sterilized in Blake bottles of 250 cc. capacity. After inoculation, the bottles were sealed with wax and placed in the incubator at 37° for 6-8 weeks. At the end of this interval the flasks were removed from the incubator and the surface of the medium carefully examined macroscopically for the presence of colonies and later microscopic studies were made of stained smears of the water of condensation. No growth was noted.

Two hundred samples of blood were transferred directly into 30x250 mm. tubes, each containing 50 cc. of cooked meat beef heart broth prepared by the addition of heart infusion broth to pieces of cardiac muscle saved from the infusion. The tubes were placed in the incubator and at weekly intervals for a period of 8 weeks a loopful of the broth suspension was streaked over the surface of NNN medium in test tubes. No growth appeared on this medium after incubation.

Sixty-two specimens of blood were allowed to clot in 50 cc. sterile plugged centrifuge tubes. The clots were separated from the walls of the tubes with a sterile wire and subsequently centrifugated

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<sup>1</sup> Löwenstein, E., *Inter. J. Leprosy*, 1933, 1, 39.

at 500 R.P.M. for 10 minutes. The serum was removed with Pasteur bulb pipettes and 50 cc. of sterile glycerol infusion broth were added to each clot. The centrifuge tubes containing the clot and medium were then placed in the incubator. The tubes were removed and the medium examined daily for evidence of growth. At weekly intervals, for 6 weeks, a loopful of the broth was streaked over the surface of NNN medium and subsequently incubated. No growth was obtained.

The negative results obtained as a result of this rather large series of experiments are rather hard to interpret in the light of many recent studies<sup>2</sup> on the isolation of organisms from the blood stream in health and disease. It is possible that the therapeutic measures instituted against the Hansen infection, namely, the weekly injection of 4-5 cc. of the iodized ethyl esters of *Hydnocarpus wightiana* oil, may have had a direct influence on the incidence of blood stream invasion. It is also true that these studies were made during the rainy season so that the possibility of air borne contamination was at a minimum.

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### Effect of Testicle Extract on Primary Tuberculous Infection and Reinfection in Guinea Pigs.

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1. *Effect on Primary Infection.* Testicle extract greatly increased the extent of the local lesions resulting from the intradermal or subcutaneous injection of virulent human tubercle bacilli into guinea pigs. When compared with lesions resulting from the injection of an equal number of tubercle bacilli suspended in salt solution, the increase was estimated at from 10 to 15 fold. The testicle extract was prepared by mincing bull testicle in 2 volumes of physiological salt solution, straining the material through cloth and finally filtering through a Berkefeld V candle. Kept at ice-box temperature, the extract maintains its activity for a long period of time. Two groups of 6 guinea pigs were used in the first experiment, in which the injection was made intradermally. One group

<sup>2</sup> Callow, Bessie R., *J. Inf. Dis.*, 1933, **52**, 779.