BCG, by mouth weekly, most of them (35) receiving 5 doses of 100 mgm. each, the others 4 doses. Five or six weeks after the last dose they were all tested with lepromin. Of the 47 recorded after 48 hours, only 9 were positive, and in only 3 of them did the early reaction reach 10 mm. in diameter. After 25 days, 47 (96%) were Mitsuda positive, 1 being ± and 1 negative; of the positives, 20 were ++ and of them 9 had ulcers. These very high positive results cannot be ascribed to reinoculation of the antigen, as Tissieu would have it, for only the one test was made.

The other table shows the results obtained in 27 children of leprous parents in a preventorium here in Goiania, of whom 22 were tested with lepromin for from 4 to 9 times (average 5.2 times), within periods ranging from 11 to 64 months (average 26.8 months), without becoming reactive to that antigen. Each was then given 3 doses of BCG, but in unusual sequence and dosage. The first dose, 200 mgm., was given in November or December 1952; the second dose, also 300 mgm., was given in March 1953; the third dose, this one of 400 or 500 mgm., was given 40 days later. The lepromin tests were made after another month. This group showed very little early reactivity—only in 2 cases, their reactions measuring 5 and 8 mm. The late reaction was now positive in 24 cases (89%), 3 remaining negative; of the positives, 7 were ++ and of them 3 with ulceration.

---

**EARLY EXPERIENCE WITH BCG**

To The Editor:

This is in reply to your inquiry about what I have seen in the contact children whom I vaccinated with BCG.

I started this experiment in 1940 with a small group of about 20 cases, which I followed up for several years. Later, some of them moved from Rosario but the others are still under observation.

While under observation none of those children developed lepromatous leprosy. One developed typical tuberculoid lesions, and another had achromic macules of the indeterminate type. Both recovered spontaneously, without treatment.

All of these children were living with open cases. All were vaccinated once, with 0.15 cc. of BCG given intradermally at birth.

25 de Diciembre, 811
Rosario, Argentina

**SKIN TESTING WITH LEPROSY BACILLUS SUSPENSIONS**

To The Editor:

As requested, I am supplying a free translation of the summary of a report which I made at the April 1918 session of the Japanese Bacteriological Association on attempts which I had made to cultivate the leprosy bacillus, and of the results of skin tests made on leprosy patients with the supposed cultures which
had been obtained. This report appeared, under the title, “On a Pure Culture of Leprosy Bacilli, and a Skin Reaction by Means of the Pure Culture Suspension,” in the Saikinigaku Zasshi (Journal of Bacteriology) No. 272 (1918) 51-53, published by the Kitasato Institute. The entire summary is given, because the part dealing with the cultivation work explains the material used in the skin tests.

On a pure culture of leprosy bacilli.—Since 1914 I have been investigating methods of obtaining pure cultures of the leprosy bacillus, but have not yet found a perfect method. However, I have obtained relatively marked reproduction of the bacilli in vitro by means of the following method:

(a) Ringer's solution is placed in test tubes, 10 cc. per tube, and sterilized. This is used as the culture medium.

(b) A new nodule is extirpated entire, and many tiny slices are made of it. One such slice is placed in a tube of the culture medium, and the tubes are incubated at 34° to 37°C. The entire procedure is carried out with care to avoid contamination with other bacteria.

In this way, by 3 to 5 weeks we can observe a slightly yellowish and viscous material manifesting gradual multiplication around the slice at the bottom of the test tube. The upper part of the medium is almost transparent. If the tube is shaken the whole medium becomes turbid, of a thin milky appearance. A smear of this suspension reveals large numbers of leprosy bacilli in aggregates which are much larger than what can be found in the original nodule. Furthermore, the numbers of bacilli observed in such smears are far greater than those in a preparation made from an equivalent quantity of a suspension of a nodule slice of the same kind and same size. No colony formation can be seen in transplants of the culture upon normal agar or glycerine agar media.

From these facts it is believed that the numerous bacilli in the Ringer's solution signify reproduction and isolation from the original nodule slice. It is therefore concluded that leprosy bacilli are able to reproduce in vitro.

2. On a skin reaction in leprosy.—I tried a skin-reaction test by means of an antigen prepared in the following manner: The culture suspension of leprosy bacilli described above was heated at 60°C. for 30 minutes, an equal quantity of sterile Ringer's solution was added, and finally 0.5 per cent of carbolic. For a control, sterile Ringer's solution was used.

In the first step, 0.1 cc. of the antigen was inoculated into the abdominal skin of five healthy guinea-pigs. This resulted in absolutely negative reactions.

In the next step, 60 leprosy patients, of which 44 were of the lepromatous type and 16 were neural-type cases, were tested. The antigen was inoculated, 0.1 cc., into the skin of the flexor surface of the upper arm. The results were as follows: Twelve of the 44 lepromatous-type cases (27.3%) were positive, and 32 (72.7%) were negative. Twelve of the 16 neural type cases (75%) were positive, and 4 (25%) were negative.

According to these results the neural type gives a high rate of positive reactions, while the lepromatous type gives few positive and many negative reactions. This is a very interesting result which calls for attention.

Practically the same results were obtained by means of a suspension
of nodule, except that the reactions were always stronger than those caused by the culture suspension, especially in the case of the neural-type patients.

I had not tried this skin-reaction test on normal healthy people, and therefore cannot discuss the diagnostic value of the test in leprosy. My idea when the work with the skin test was undertaken was that lepromatous cases should give strongly positive reactions and neural-type cases weakly positive reactions, and that nonleprosus people should be negative. The results were completely opposite to that assumption, and I was much interested in that fact.

Higashimurayama
YOSHINORU HAYASHI
Tokyo, Japan
Director
Tama Zenkho-en

VIRCHOW’S DESCRIPTION OF THE LEPIRA CELL

To THE EDITOR:

The question has been asked, precisely what was Virchow’s original description of the cells of the leproma to which his name is so commonly given. That description appeared in his treatise, Die krankhaften Geschwulste (The Morbid Tumors), which was published in Berlin in 1864-1865 (Bd. 2, S. 497). If any translation of the pertinent passage has ever been published in the leprosy literature, we have not learned of it. It appears that the treatise itself was never translated into English, although a French edition, entitled Pathologie des Tumeurs, was made by one Dr. Paul Aronssohn and was published in 1869 (Paris, Germer Bailliere, Libraire-Editeur, Rue de l’Ecole-de-Medecine, 17).

Introducing his description of the lesions as seen in the fresh state and otherwise, he described the low-magnification picture and went on to say:

Seen with high magnification the mass of new tissue is composed mainly of cells which present great variations in shape and size. It is doubtful if I have ever before seen, as well as here, the progressive development of connective tissue cells, first simply fusiform or stellate and going through the phase of nuclear and cellular division. The immediate products of cellular division appear here in a striking fashion. The more frequently the division is repeated, the smaller and more globular are the new cells. The old intercellular stroma becomes thinner and thinner, to the extent that between the cells, which are arranged in layers or groups, one can no longer clearly distinguish the narrow strands of interstitial tissue, which become granular and turbid when acetic acid is added. Often in this tissue only nuclei are seen; in preparing the specimen for examination the cells are greatly disrupted, so that the freed nuclei (cytoblasts) are seen in large numbers...

With regard to the cells, I will again comment that when fully developed they consist of round structures, pale, slightly granular and delicate