Case of malignant leprosy rapidly regressing after treatment by a sulfone associated to physiotherapy. Isolation from a cutaneous lesion of the patient of an acid-alcohol fast bacillus (Non-chromogenic culture) pathogenic to man and to laboratory animals.

by

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1 — Clinical history — A. A. H., a 26 years old, Brazilian of German descent, single, born in Curitiba (Paraná), agronomist at the Department of Agriculture, height 1.73m., weight 71 kg., in very good general health conditions. The patient came to the author's private office, introduced by Doctor Felinto Coimbra, telling that he had a previous laboratory examination by Professor Ruy Noronha Miranda, positive for leprosy. He denies any previous history of leprosy among his ancestors or of contact with lepers.

The patient reports that in the later part of 1947 he worked in the Agricultural Station of Ponta Grossa (Paraná), when he discovered, in the internal part of his right wrist, an anaesthetic patch which in a few months extended from the middle of the palm to about 10 cm. above the wrist (regions innervated by the cutaneous palmar — a branch of the median — and by the cutaneous branch of the cubital; in the wrist, a region corresponding to the anastomosis of the cubital and cutaneous brachial nerves). Very soon, on the anaesthetic patch an erythematous macula appeared and in the center of this mácula, right in the middle of the wrist, a great nodule. The patient has the impression that this should have been the inoculation point. In June 1948, the anaesthetic patch reached about 25 cm. from the middle of the palm to the middle of the forearm, only in the region corresponding to the cubital nerve. At this time, some verrucous lesions appeared on the left knee.

Status praesens — July, 1949. In the middle of the frontal region a moderate erythematous infiltration; an old acne rosacea on the nose and the malar regions; seborrhoeic blepharitis; a slight infiltration of the left auricular lobe. On the right arm lepromatous patches on the
elbow and posterior view of the forearm; a great intradermal nodule on the internal face of the wrist; a *sui-generis* atrophy of the thenar region; marked cyanosis and anaesthesia of the palmar region, anaesthesia of all the cubital region; cyanosis and a little oedema of the dorsal view of the hand. On the left arm: three red, nodular patches in the cubital region; oedema and cyanosis of the same region; two nodules, on the wrist and on the dorsal part of the hand. The buttocks covered by small, newly formed lepromatous patches, of different sizes; a lepromatous seemingly old patch, about 5 by 15 cm., on the left side of the intergluteal furrow; this patch is atrophic and dyschromic in the central part and its external border is crescent-shaped, prominent and hyperchromic. Inguino-crural glands normal. Hair growth of a normal appearance on the thighs and legs, where numerous small, erythematous and prominent patches are found which can be felt on palpation. On the knees two lepromatous patches, in which there are nodules, the one on the left knee being larger; a large nodule on the anterior view of the upper third of the left leg; two smaller lepromatous patches around the malleolus, on the inner side on the left leg and, on the outer side on the right leg; the first toe on the right foot is dark and anaesthetic.

The case was classified as L2-N1, the histopathological examination made at the Instituto Oswaldo Cruz confirming this diagnosis (P.C.15.854, July 13. 1949, corresponding to a fragment of the skin taken in the right buttock). The X-ray examination of the thorax has shown normal lungs (Dr. H. Genú, September 29. 1949).

Diasone, in the dosis of 2 to 6 tables a day at meal time, was prescribed. Galvanocauterization and electrocoagulation were recommended twice a week. The patient decided to follow this treatment for just three months.

September 20. Intradermal tests on the anterior surface of the forearms with Mitsuda-Hayashi's lepromin and the author's leprolin, gave only negative results.

2 — *Routine tests*

*Nasal mucus*, collected by the author and examined by Doctor [Alvaro Lobo](#), gave negative results. This test was repeated by the author on October 8. 1949, three months after the beginning of the treatment, and the result was strongly positive (on the right side: ++○; on the left side: ++ ++. It looks as if the sulfone had activated the infection.

*Cutaneous lymph* — Collected by the author (June 23. 1949) from three lesions, one on the trunk and two on the limbs, after excoriation with a vaccination-needle. Three different smears were made, which have been examined by Doctor [Alvaro Lobo](#) and gave positive results, no information having been given as to the amount of bacilli observed.

July 13. A smear made from a fragment of skin of the left buttock, stained and examined by the author, displayed almost only acid-alcohol
fast bacilli, looking like fragments resulting from a binary division and very similar to the ones found in patients who have been treated for a long time. However, the patient had not been treated as yet.

July 23. Smear of lymph and blood from the left auricular lobe: positive (+). A new examination, on both auricular lobes (October 8, 1949): positive (++)

**Haematological data** — Examination by Doctor Álvaro Lobo (June 24, 1949): red blood corpuscles 4,950,000; haemoglobin 13.0 g.; white blood corpuscles 5,600; eosinophils 2%. On October 10, a new examination shows: red blood corpuscles 4,800,000; haemoglobin 14.0 g.; white blood corpuscles 9,000; eosinophils 2%. Everything else was normal (Doctor Álvaro Lobo). This last examination already been given 500 tablets of Diasone (150.0 g.). Practically, there was no change in the blood situation. The low eosinophil rate contrasted with our experience in other cases treated with sulfones, these last ones being of chronic leprosy.

**Sputum** — In three different instances was the sputum examined (September 26, 28, and 30). The first time, direct smears stained by the Ziehl-Neelsen method gave negative results for acid-alcohol fast bacilli. In the other instances the sputum was treated by Petroff's method, with sodium hydrate, the sediment being stained by the Ziehl-Neelsen method, with the result that groups of acid-alcohol fast bacilli were found. This lead us to submit the patient to X-ray examination which did not show any abnormality. The sputum was inoculated in guinea-pigs in which it did not bring about any tuberculous lesions. It has also been transplanted into Loewenstein's medium with negative results.

**Skin lymph** — Smears were prepared from the skin lymph obtained, on September 17, 1949, from lesions of both arms, by Lleras Acosta's method. Both smears, stained by Ziehl-Neelsen's method were positive (+ +). Skin lymph collected again from a lesion of the left buttock gave also a positive result.

**Lymph glands** — Several times we tried to examine the lymph obtained from the glands of patient but never succeeded, because the inguino-crural glands were not palpable, which should that the case was indeed of acute leprosy.

3 — **Histopathological study** — The first biopsy was made from a florid lesion of the right buttock, on July 13. A skin fragment was studied in the Department of Pathology of the Instituto Oswaldo Cruz, the following statement being furnished:


"Nature and origin of the tissue: Skin fragment from the right buttock.

"Result of the pathological examination: Lepromatous leprosy.

(s) Jorge P. Guimarães."
This result confirmed the author's clinical diagnosis. The second biopsy was made, on August 2., from a lepromatous patch on the left knee, the smears being positive, with large numbers of bacilli most of them appearing as having been broken in fragments. The material then collected has been utilized exclusively for bacteriological work.

The 3rd. biopsy has been made on October 4., from a florid lesion on the left forearm, the histopathological study confirming the former one:

Instituto Oswaldo Cruz — Department of Pathology.


“Nature and origin of the tissue: Skin fragment from the left forearm.

Result of the histo-pathological examination: The sections show an inflammatory granuloma displaying the typical appearance described in lepromata. Occasional occurrence of giant cells of the Langhans type, not associated with epithelioid cells. Search for acidalcohol fast bacilli: positive.

Manguinhos, October 12. 1949 (s) Jorge P. Guimarães.”

Part of the material from the 3rd. biopsy was employed for experimental inoculations.

4 — Bacteriological study — A piece of leprous tissue from the skin, taken by biopsy from the right buttock, on July 13. 1949, was triturated, treated by sodium hydrate, centrifuged and transplanted on the same day, to ten tubes of Loewenstein's medium. Until August 20. when we had to leave the laboratory for a short trip, no growth could be detected. Being back in the laboratory, on September 3., after 50 days in the incubator at 37° C., one of the ten tubes displayed the growth of a yellow-brownish culture, the smears of which, stained by the methods of Ziehl-Neelsen and of Gram, showed that it was a pure culture of an acid-alcohol fast Gram-positive bacillus. The study of this culture will be the matter of a paper to be read at the 5th. International Congress for Microbiology.

Twice did we take blood from this patient, treated it by Petroff's technique and transplanted it to different media, without ever obtaining any growth. Smears from the sediment have been positive for acid-alcohol fast bacilli, displaying the characteristic of Hansen's bacillus. The skin lymph, collected on September 17. and 24. from lesions in the arms and buttocks, and transplanted, did not grow either, as well as the emulsion prepared from the skin of the left forearm transplanted on October 4. Such repeatedly negative results, are disappointing.

5 — Experimental Study — On September 13. we inoculated with a culture obtained from this patient, strain “Hecke,” the first group of black American mice. On September 23., eleven days after the inoculation, we proceeded to make the autopsy of the first mouse of
this group. It displayed a large granulomatous tumour of which the
smears were rich in acid-alcohol fast bacilli, as were also the viscera.
On September 26 and 29 we made the autopsies of two other mice of
the 1st. group, with identical findings in both. The pus taken from
these three mice were transplanted to Loewenstein's medium, where
they gave rise to retrocultures identical to the original one. Those
cultures were utilized for the inoculation of new animals.

On October 4., with the emulsion of skin tissues taken from the
left arm of the patient, we inoculated rats and mice of different
varieties.

During the months of November and December, we inoculated
rhesus monkey, small laboratory animals and also two lepers who
volunteered, these being man and wife. The results obtained in this
wide experiment will be reported by the author in collaboration with
Doctor JORGE P. GUIMARÃES from the Pathological Department of the
Instituto Oswaldo Cruz. This paper will be presented to the 3rd. Pan-
american Conference for Leprology, to be held this year in Buenos
Ayres. Several photographs and drawings illustrate this work.

6 — Treatment — From July 9. to October 10. 1949, the patient
ingested about 500 tablets of Diazone and 250 capsules of B Complex.
By the end of August he complained of indisposition and showed
oedema of the hands, the reason why the use of Diazone was discon-
tinued for a while. During this period, 27 sessions of galvanocauterisa-
tion were made, 10 on the arms and hands, 9 on the buttocks, seven
on the inferior limbs and one on the ears. These treatments were
always followed by the painting of a 33 per cent solution of trichloro-
acetic acid. Four local treatments specially designed to acne rosacea
and blepharitis were also made.

The results have been remarkable and the patient on October 10
went back to Curitiba in order to proceed with the treatment under
the care of Prof. R. N. Miranda, to whom we wrote on December 7.
1949, offering a supply of Leprolin prepared with the "Hecke" strain, so
that some kind of autovaccinotherapy could be tried. The reply came
on January 6. 1950, telling:

"The patient is always improving. I believe he must furnish us
now with negative tests. This I will see with the material I collected
on him to-day. As for the autovaccinotherapy, I think the best opportu-
nity is now over, for the general conditions are excellent and approching
to healing." But, the results I got on September 8. 1949, make it hard
for us to believe in this rapid negative results. We asked Doctor Mi-
randa for further information which we will report later on.

Tuberculosis — The result of the microscopical examination of
the sputum lead me to suspect of an association: leprosy + tuberculosis.
However, the cultures from the sputum gave negative results and
we present here the result of the inoculation to guinea-pigs, of which
one was killed on the 30th. day after the inoculation the following
statement being made by the Section of Pathological Anatomy:
"P.C. 16.043, October 28, 1949. Guinea-pig inoculated on September 27, 1949 and killed on October 28, 1949. Inoculated subcutaneously with sputum of patient Hecke, in the right groin, where a tumour was produced. The examination of a fragment of abdominal muscle gave the following results:

"The microscopical sections show a purulent inflammation associated to the presence of large cells with two and sometimes three nuclei, a highly vacuolized cytoplasm and a dark greenish-yellow pigment. Neither typical tubercular follicles nor acid-alcohol fast bacilli were found.

Manguinhos, November 28, 1949.

(a) Dr. Jorge P. Guimarães".

Since the macroscopical appearance of the "Hecke" culture was similar to that of certain strains of Mycobacterium tuberculosis, S (smooth) type and since Prof. George W. McCoy, when acknowledging receipt of my first cultures from leprosy patients, wrote that he suspected some of them of being of tuberculosis, I inoculated the "Hecke" strain in guinea-pigs. The culture is pathogenic for the guinea-pig, as I could observe several times. However, after three weeks it did not produce tuberculous lesions. The following confirms this statement:

"Section of Pathological Anatomy, Instituto Oswaldo Cruz.


"Anatomo-pathological findings: Skin — The microscopic sections show an acute, purulent inflammatory process, with formation of micro-absceses, around which there is an intensive proliferation of fibrocytes and neoformation of blood-vessels (granulation tissue). No tubercular follicles were found. Acid-alcohol fast bacilli were found. The viscera do not show any changes deserving mention.

Manguinhos, March 1950.

(a) Dr. Jorge P. Guimarães".

On April 18, 1950, in the absence of more recent information about this patient, I close this report, with my sincere thanks to Doctor Joir Monte, for his valuable and constant cooperation for the study and treatment of this patient.

The experimental studies performed with the "Hecke" culture, specially with the rhesus monkey, gave results above all expectation and is not yet finished. Leprolin "Hecke" also is still in experimental study on lepers in several institutions, including the Calcutta School of Tropical Medicine, Department of Leprology, under the able direction of Doctor Dharmendra.

Manguinhos, April 18, 1950.