

the most frequent cause is the rupture of an emphysematous bulla. Such bullæ have been demonstrated around scar tissue, such as healed tuberculous foci or collapsed areas. Effusion usually does not follow such a pneumothorax. Complete recovery is usual within a short time, but recurrences may occur. No treatment is ordinarily needed except rest in bed and sedatives during the first few days.

The following case is of interest in this connection :—

The patient was a European, aged 40, who, after a long spell of work in Calcutta without a holiday, had an attack of dengue. He gave a history of tiredness and lack of energy for a few months. He had been given eight injections of emetine, apparently for diarrhoea. A few days before admission he had a sudden pain in his chest over his heart which he attributed to either pleurisy or 'heart'. He said he was unable to rise from his bed at all, and when later he did so, the effort caused him to be drenched with perspiration; the pain disappeared slowly within a few days. The symptoms were more suggestive of indigestion and hypochondriasis, but we admitted him to hospital for more thorough investigation.

Clinical examination of the chest failed to reveal any abnormality, but an orthodiagram was ordered mainly to convince the patient that his heart was sound. There was no evidence of any coronary lesion.

In the absence on leave of the official radiologist, the opinion of another radiologist was obtained. He reported as follows :—

'Apart from slight ventricular predominance the heart appearances are normal.

Extreme cavitation is seen in the right upper lobe. Signs of chronic bronchitis and bronchiectasis to a lesser extent are seen bilaterally.'

Only the first half of the report was communicated to the patient.

The patient was questioned and examined again in view of this report. There was nothing in his history, except that questionable attack of pleurisy, or in the physical examinations to suggest that he had at any time suffered from pulmonary tuberculosis.

A second skiagram was taken by the same radiologist. An almost identical picture was obtained and the radiologist who had now been shown the clinical notes reported: 'See notes on previous skiagram'.

The patient was a nervous individual and we did not feel justified in communicating this report to him. He was discharged from hospital and we have been personally in touch with him for over three years, during which time he has remained in apparently sound health. He has now been in the army for nearly a year.

At our request Col. Shorten, the radiologist to Carmichael Hospital for Tropical Diseases, examined the films and reported as follows :—

'Heavy hilar shadows with calcifications in both sides; in addition, the right lung shows the following: (1) A group of large ring shadows in the right upper zone with thickened broncho-vascular trunks leading to a group of calcifications in the upper part of the hilum. (2) Surrounding the ring shadows there is evidence of a streaky fibrosis but no infiltration. (3) A streaky fibrosis in the periseptal area also extending into the hilum.

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THE RÔLE OF METHÆMOGLOBIN ON THE LEPTOMONAD PHASE OF *LEISHMANIA TROPICA*, WITH SPECIAL REFERENCE TO ITS REVERSION INTO *LEISHMANIA* FORMS IN CULTURE

By R. ROW, O.B.E., M.D. (Lond.), D.Sc. (Lond.)

and

S. S. KULKARNI, L.C.P.S., D.T.M.

(From the Department of Pathology, P. G. Singhanee Hindu Hospital, Bombay)

In a former communication one of us (Row, 1922) had pointed out that the flagellate leptomonad forms of *Leishmania tropica* reverted to the leishmania O-bodies and these made their appearance in the condensation fluid of NNN culture tubes inoculated with the parasite 2 to 3 weeks previously. These were described as the O-bodies, or resistant forms, and it is the presence of these in old cultures of *L. tropica* which was responsible for their infectivity in mice—even to the extent of inducing a generalized infection in them—indistinguishable from experimental kala-azar (Row, 1914). The observation on the infectivity of old cultures containing these resistant forms was confirmed by Southwell and Kirshner (1938)*. It was also pointed out (Row, 1922) that the reversion was due either to the concentration of the fluid or to the alteration of the oxyhæmoglobin into methæmoglobin.

Subsequent work on the culture of the parasite in the hæmoglobin saline medium of Row (1912)

* No original investigation was undertaken by these workers; in their paper they review work already done, including Dr. Row's.

(Continued from previous column)

Conclusions.—These appearances are suggestive of old healed tuberculous disease.

The ring shadows in the upper zone are due to one of two causes: (a) emphysematous bullæ; (b) an irregular pneumothorax with adhesions.

There is no evidence of cavitation.'

Conclusion.—It seems improbable that the patient had a pneumothorax, as there would have been more clinical evidence of this. We are inclined to accept the second radiologist's (Col. Shorten's) alternative opinion that the translucent areas are in this case due to emphysematous bullæ.

There is little to support the first radiologist's interpretation—which he declined to revise—even on the strength of the skiagram, and when the latter was taken in conjunction with the clinical picture, this interpretation was completely ruled out. Had this verdict been communicated to the patient, the result would have been disastrous.

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proved that it was the latter condition more than the former which was responsible for the reversion, but unfortunately it is difficult to demonstrate this feature clearly, even in stained smears, because here the leishmania bodies lie scattered between the large number of flagellates overcrowding and overshadowing them in the form of rosettes which persist in the fluid culture medium even for 100 days, as pointed out elsewhere (Row, 1935), unless one is lucky to hit upon the right day while examining old cultures (*vide* figure 5), and even here the day varies with the strain of the parasite (*vide* figure 6). It was therefore thought interesting to see if any morphological alteration of the flagellates into leishmania bodies could be observed more clearly in the culture fluid containing only methæmoglobin.

Preparation of the culture fluid.—When the parasite of the oriental sore is grown in Row's medium (1912) and the ensuing flagellates allowed to thrive for several weeks at 22°C., the striking character of the culture fluid is its alteration in colour from scarlet lake of the oxyhæmoglobin into a brownish transparent liquid which on spectroscopic examination shows the distinct methæmoglobin absorption band in the red of the spectrum, and the disappearance of the two characteristic absorption bands of the oxyhæmoglobin between the D and E lines. The flagellates flourish in abundance and are found mostly in rosettes of more or less equal size indicating that they continue to grow even when oxyhæmoglobin is altered into methæmoglobin. After a week or two, however, the culture degenerates and consists mostly of granular debris. At this stage the brown culture fluid is centrifuged and the supernatant fluid is distributed in tubes and this constitutes methæmoglobin medium ready for immediate use.

These tubes are inoculated with a drop of young cultures of five to seven days containing only flagellates and incubated at 22°C. and when examined every three or four days reveal the following features:—

During the first week the culture shows that the flagellates have grown in enormous numbers, but is characterized by the presence of rosettes of more or less equal size uniformly distributed in the fluid (*vide* figure 1), and stained smears show that it is from these rosettes that the leishmania bodies are derived and these are found also lying between the rosettes in fair numbers with all transitional forms (*vide* figures 2, 3, and 4).

During the second week the rosettes have a tendency to mass together but slightly degenerate with a large number of oat-shaped bodies and transitional forms, obviously the product of the rosettes (*vide* figure 6). A few days later, all that is left is a degenerate debris of the leishmania bodies with a flagellate here and there.

The transitional forms are exactly like those occurring in the condensation fluid of the old NNN cultures already described elsewhere, except for the absence of the thin capsule-like structure, within which the parasites obviously appear to shrink. These transitional forms, it may be repeated, are briefly as follows:—

(1) The phase where the active flagellates become sluggish with the shortening of the flagellum by its withdrawal inside and its absorption, and with the simultaneous migration of the kinetoplast towards the nucleus.

(2) The shortening phase of the parasite where the shortened stump of the flagellum disappears and where the condensation of the cytoplasm of the parasite takes place side by side with the further approximation of the kinetoplast towards the nucleus, before the final rounding off stage when they become oat-shaped or round bodies (with the differentiation of the nucleus) indistinguishable from those found in the original lesion.

The morphological changes of the flagellates into leishmania bodies when grown in the methæmoglobin medium are illustrated in the accompanying photomicrographs.

Summary

(1) The reversion of the flagellates into leishmania forms can be induced in a few days by growing them in the methæmoglobin medium.

(2) The formation of leishmania forms is always preceded by the appearance of a large number of flagellates mostly in the form of rosettes (distributed uniformly in the medium) from which arise masses of transitional forms and leishman bodies.

(3) The significance of these points may require further investigation.

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DESCRIPTION OF PLATE XVIII

- Fig. 1.—*L. tropica* (Cambay), 12-day culture in methæmoglobin; rosette formation stage.
Fig. 2.—*L. tropica* (Cambay), 12-day culture in methæmoglobin; rosettes going into leishman bodies and transitional forms.
Fig. 3.—*L. tropica* (Delhi), 7-day culture in methæmoglobin; completion of leishman bodies.
Fig. 4.—*L. tropica* (Delhi), 6-day culture in methæmoglobin; transitional forms of leishman bodies.
Fig. 5.—*L. tropica* (Ambala), 39-day culture in Row's medium; oxyhæmoglobin altered into methæmoglobin; most of the rosettes reverting to leishman bodies.
Fig. 6.—*L. tropica* (Cambay), over 4 weeks' old culture in Row's medium; oxyhæmoglobin altered into methæmoglobin; some of the rosettes reverting almost to leishman bodies.

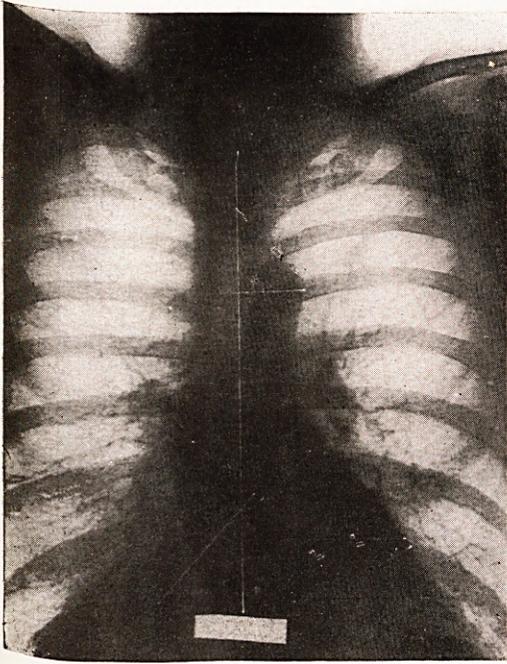


Fig. 1.

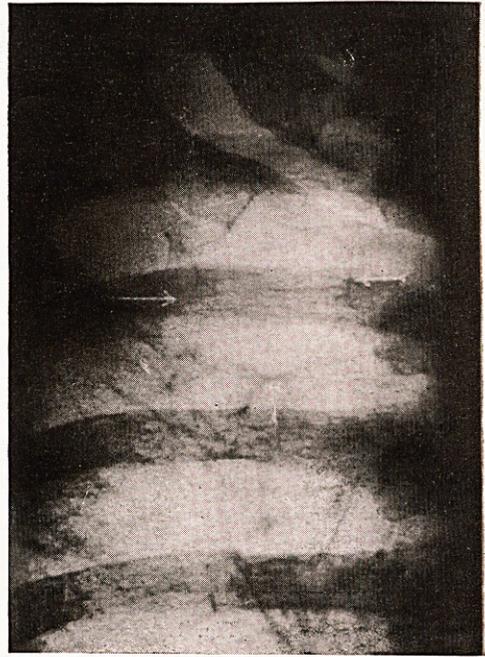


Fig. 2.

THE ROLE OF METHÆMOGLOBIN ON THE LEPTOMONAD PHASE, ETC. : ROW & KULKARNI



Fig. 1.



Fig. 2.



Fig. 3.



Fig. 4.

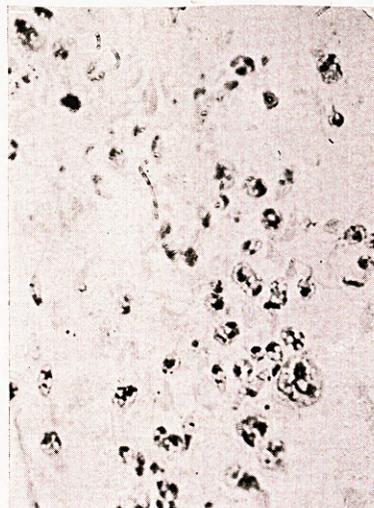


Fig. 5.



Fig. 6.