A CASE OF HISTOPLASMOSIS IN SINGAPORE

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We present a report of the first case of Histoplasmosis, observed and confirmed in Singapore.

Histoplasmosis is a fungal disease which for many years was thought to be both rare and mortal but since 1945 has come to be regarded as a benign malady.

Histoplasmosis was first noted by Strong in 1905. He did not name the infection but likened it to Leishmaniasis. In the same year, Darling discovered Histoplasmosis while carrying out his research on Leishmaniasis in the Panama Canal zone. He reckoned that this parasite was a new pathogenic protozoan and named it Histoplasma capsulatum. In 1912, Da Rocha Lima pointed out that Histoplasma capsulatum was a fungus rather than a protozoan. It was not until 1934 that the mycotic nature of the parasite was confirmed when De Monbreun cultured the organism from the bloodstream and spleen of a 5-month old infant in Tennessee. In 1939, De Monbreun described the first canine case. Since then the organism has been isolated from rat, mouse, cat, cow, sheep, horse and monkey. It has also been isolated repeatedly from soil and dust — the first isolation being done by Emmons in 1908.

Histoplasmosis is endemic in parts of the U.S.A. In the areas of Missouri-Mississippi and Ohio River Valley as many as 80 per cent of the population are shown to have benign Histoplasmosis by the Histoplasmin skin test (Christie and Peterson, 1945; and Palmer, 1945). It has been estimated that 20 to 30 million people in the U.S.A. have benign Histoplasmic infection or pass through a clinically silent infection (Silvermann 1955). It is prevalent in parts of Canada, Panama, Mexico, Argentine, Brazil, Honduras, West Indies, Southern Rhodesia and the Union of South Africa. In Britain about 15 cases have been reported, six of which were confirmed by culture. About 20 cases have been diagnosed among persons from various parts of Africa. Four cases have been reported from Indonesia (Muller 1932 — Bras et al. 1949, Hausman and Hiemstra 1949, Lie Kian Joe 1955). Sporadic cases have been reported from the Philippines (one case), Australia (three cases) and India (three cases — Panja and Sen 1954, Kalna et al. 1957, Sen Gupta et al. 1957). One case was mentioned by Marsden in 1953 from Malaya in an annual report.

Histoplasmosis may be localised or generalised. It may be acute, recurrent or chronic; it may be severe, mild or completely asymptomatic. Lesions are most frequently seen in the lungs, lymph-nodes, spleen, liver, bone-marrow, naso-oral cavity and skin. The disease affects persons of all ages. The incidence is high in infants and in the older age group; it is slightly higher in males. The symptoms are variable. Anaemia wasting, continued fever and sweating are the only constant features. Hepatosplenomegaly, lymphadenopathy, pulmonary symptoms and abdominal symptoms are common.

There are three main clinical manifestations of the disease: (1) Skin manifestations like papular eruptions and ulcers commonly seen in oronasal and anal regions. (2) Visceral involvement including lymph-nodes, spleen, liver, lungs and adrenal cortex. (3) Ulcerations of the gastro-intestinal tract particularly the oropharyngeal site and the terminal portion of the small bowel. In the lung, lesions like interstitial pneumonitis, discrete or confluent areas of lobular pneumonia, miliary and larger nodules, granulomata, caseation necrosis, abscesses or cavities, fibrosis and calcifications are present according to the stage of the disease. Radiologically the lung lesions of Histoplasmosis can mimic any form of pulmonary tuberculosis. The differential diagnosis depends on the bacteriological findings, the skin tests and serological tests. Histopathological studies of glands, if enlarged, may establish the diagnosis. Bone marrow involvement is common in disseminated types of Histoplasmosis. Cases of adrenal cortical deficiency, gastric ulcers, appendicitis, otitis media, and aural canal infection, nasal septal perforation, osteitis, arthritis, endocarditis and meningo-encephalitis have been reported due to Histoplasmosis. Histoplasmosis has been confused with Kala-azar, Leishmaniasis, leukemia, Hodgkin's disease, Banti's disease, atrophic cirrhosis, tuberculosis and carcinoma of the naso-pharynx, oropharynx or larynx. Cutaneous ulcerations often simulate rodent ulcers.

The port of entry of the fungus is now known. More and more the evidence accumulating indicates that the oral route is the most likely way of infection.

CASE REPORT

A male Chinese, aged 59, a taxi-driver by occupation, who had lived in Singapore since 1912, had apparently been well until January 1959. He then noted the onset of progressive loss of weight, associated with cough, extreme nervousness and loss of appetite. There had been no chills, fever, sweats, haemoptysis, jaundice, vomiting, nor pain in the chest, but dyspnoea occurred with extreme exertion.

A diagnosis of pulmonary tuberculosis with cirrhosis of liver was made in the General Hospital in August 1959. He was transferred to Tan Tock Seng Hospital. The sputum cultures grew M. tuberculosum and acid fast bacilli were seen in the direct smears. The patient was given a course of Streptomycin and INH from 29.8.59 till 27.1.60 and then changed to PAS/INH. After December 1959 the sputum findings were repeatedly negative for M. tuberculosum.

In December 1959 he developed sore throat with enlargement of L. upper cervical lymph-nodes. Within the next few days enlargement of the R. upper cervical lymph-nodes ensued. He then developed tinnitus in the left ear with no noticeable loss of hearing and towards the middle of January 1960 he had bilateral nasal obstruction associated with occasional mild attacks of epistaxis. Two weeks later he experienced pain in the throat during swallowing with a sensation of a lump in the throat. He brought up blood stained sputum on a few occasions. The patient was referred to the Ear, Nose and Throat Unit of the General Hospital on 25.1.60 to exclude any malignant condition in the post-nasal space. A provisional diagnosis of tuberculosis ulceration of the naso-pharynx, oro-pharynx and laryngopharynx was made and the patient was admitted to an ENT ward on 1.2.60.

On physical examination the patient was found to be emaciated, poorly developed and he appeared ill and was afebrile. A large granulomatous ulcerative growth was seen in the left lateral wall of the post nasal space extending to the left choana. The right tonsil was enlarged and ulcerated. The left tonsil and the oro-pharynx were congested. Large areas of granulomatous ulcerations were seen in the laryngopharynx, particularly in the valleculae. The patient experienced pain on talking and when the tongue was being depressed. There was tenderness over tonsillar and laryngeal areas. Lymph-nodes in the left side of neck were enlarged to a big lump of 3" x 2" extending from left submandibular region to the left mastoid region. The lump was firm in consistency and not attached to superficial or deep structures. The skin over lump close to auriculo-temporal region was infected and discharging. A few isolated enlarged lymph-nodes were felt in the right upper cervical region which were firm and mobile.

Fine rales were heard over the left upper lung and a soft systolic murmur was heard in the mitral area. The liver was palpable 3 finger-breadths below the right costal margin and the spleen was palpable 1 finger-breadth. Neurologic examination revealed no abnormalities. The ESR was 67 mm/hr., red cell count 4.2 - 4.8/mm^3, haemoglobin 44%-66%, white cell count 4,200 - 7,600/mm^3 with 81% polymorphs, 12% lymphocytes, 2% eosinophils and 5% monocytes. The X-ray chest showed increased infiltration with many radiolucent areas in the left lung suggesting cavitation in left upper lobe. Liver biopsy was done, but was unsatisfactory. Liver function tests revealed no abnormalities. A biopsy was done of the granulating ulceration of the post nasal space and right tonsil (26.1.60).

Biopsy report on tissue from post nasal space: The normal architecture of the tissue was completely replaced by one type of cell — histiocyte. This cell was spindle-shaped, oval or polygonal. In almost every high power field were found myriads of Histoplasma capsulatum. They were rounded or oval bodies which were homogenously basophilic, surrounded by a clear halo. Each body had the size of one-quarter or one-fifth that of a red blood cell. Most of the histiocytes were quite ballooned out. As many as sixty bodies could be counted within one histiocyte (Fig. 1). In one area, surrounded by necrotic tissue, the organisms appeared to conglomerate in cyst-like spaces (Fig. 2). The latter were probably degenerated histiocytes. There was no inflammatory cellular infiltration at all except in the necrotic areas. There were patchy areas of necrosis amidst the tissues. Only in the ulcerated necrotic areas were there a few polymorphs and lymphocytes. There were no granulomatous lesions or giant cells.

Right tonsil: There was an almost wholesale replacement of the lymphoid tissue by histiocytes. The histoplasma bodies were seen

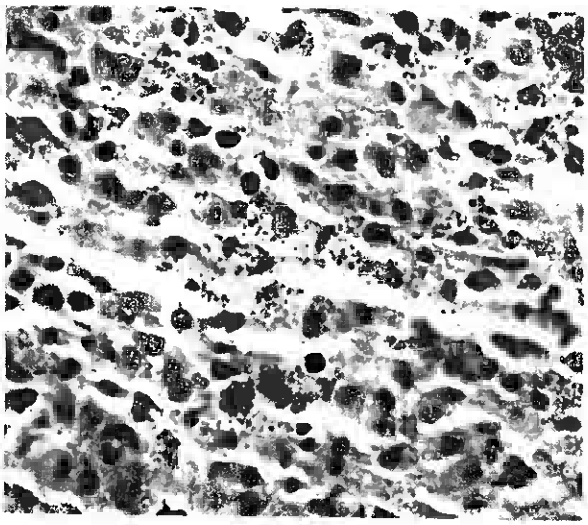

Fig. 1.

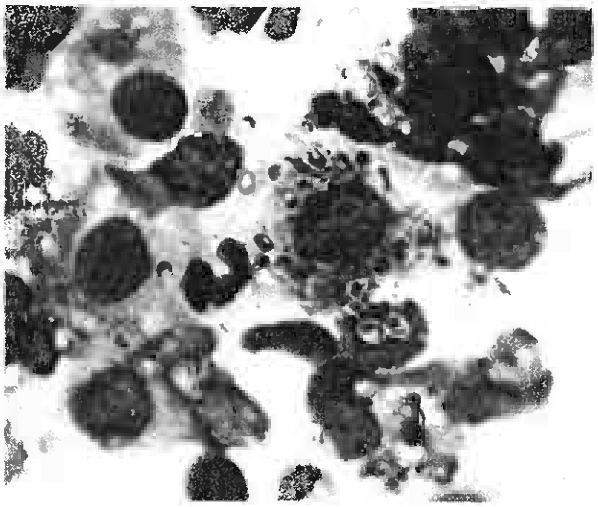

Fig. 4.

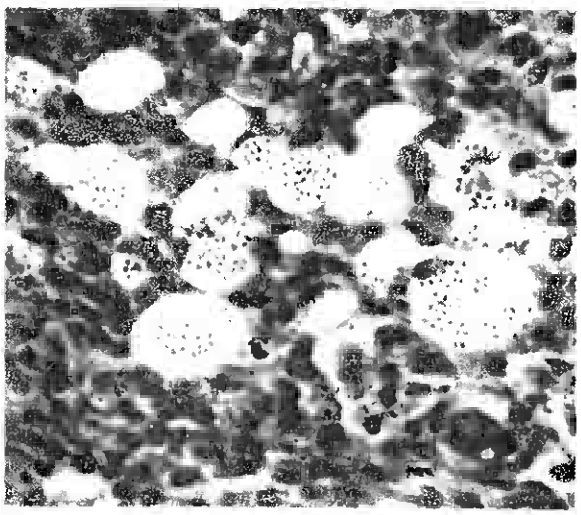

Fig. 2.

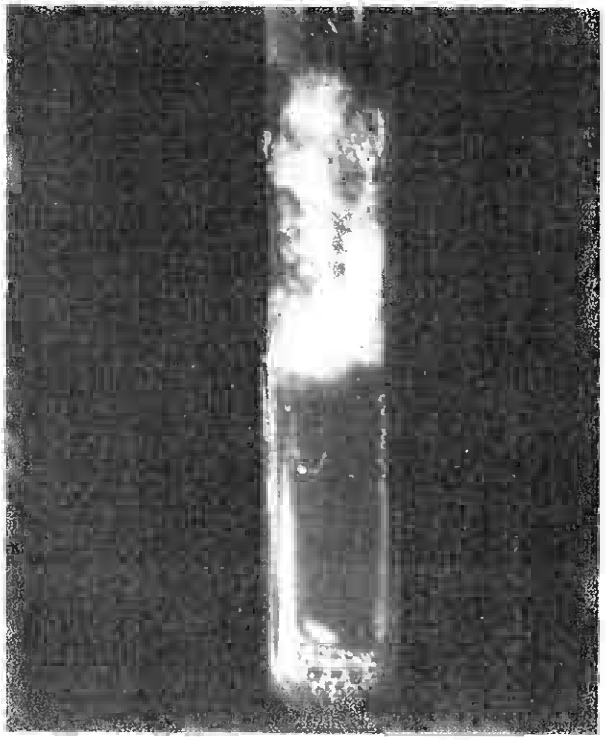

Fig. 5.

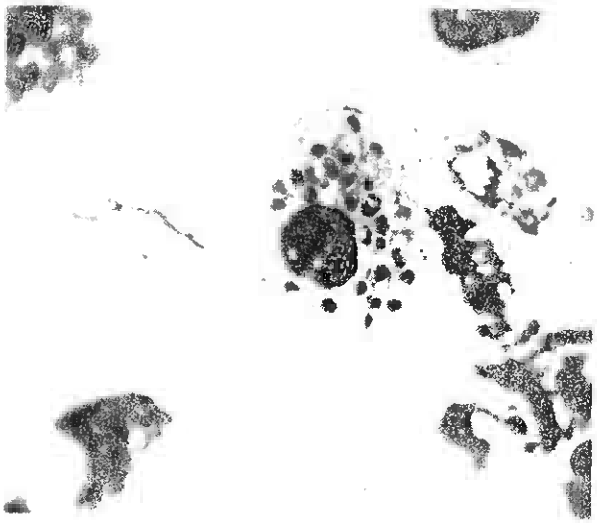

Fig. 3.

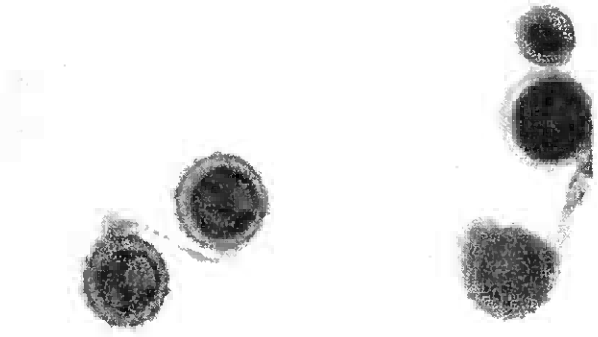

Fig. 6.

infiltrating the intact stratified squamousmucosa, and some were contained within the squamous cells. Not far away histiocytes were following fast behind them. When the squamous epithelium was replaced by the histiocytes ulceration set in.

The post nasal space biopsy and right tonsilar biopsy were repeated again on 16.2.60 along with biopsies from (a) Right cervical lymph-node (b) Left submandibular lymph-node and smears from oro-pharnyx and discharging cervical lymph-node.

Right post-nasal space and right tonsil: The histological pictures here showed more ulceration and many more necrotic areas (including caseation). Otherwise they were similar to the last biopsies.

Right cervical lymph-node: At one end the lymph-node was bordered by a broad stretch of caseation necrosis. The entire lymph-nodal structure was destroyed by a complete replacement of histiocytes. These cells again contained numerous intracellular histoplasma organisms. Once again the singular lack of inflammatory cells was conspicuous.

Left submandibular lymph-node: No lymphoid structure was identifiable. The histopathology was identical to that of the right cervical lymph-node, except that more organisms were seen here, and no necrosis was present.

Smears from Oro-pharynx and discharge from cervical lymph-node: Histoplasma capsulatum was found both intracellularly and extracellulary. The clear halo was quite distinct. Although the rounded or oval "nucleus" was seen, quite often it was pushed to one side to form a crescentic body. The latter phenomenon was seen only in the smears (Fig. 3).

As Histoplasmosis was diagnosed we tried to establish this diagnosis by cultural confirmation.

Bacteriological Report: Pus from cervical gland, swabs from pharyngeal ulcer, sternal marrow, peripheral blood and specimens of sputum were sent for culture. Leishman stained smears from all these specimens, except the bone-marrow and the peripheral blood showed numerous small oval yeast-like cells (See Fig. 4). All specimens were cultured directly on to sealed blood-agar slants and on Sabouraud glucose agar slants, screened and unscreened: the screening consisted of Penicillin 250 u/ml and Streptomycin 2500 u/ml. The sealed slants were incubated at 37°C and a duplicate set was held at room temperature. After 7-13 days incubation, a fine cottony growth appeared on some of the blood and Sabouraud slants from the pus of the cervical gland incubated at room temperature (Fig. 5). A needlemount in lactophenol-cotton blue revealed fine septate mycelia with smooth pyriform spores on long lateral branches, indistinguishable from Blastomyces dermatitidis. Subcultures on cornmeal agar slants, however, yielded the typical tuberculate chlamydospores of Histoplasma capsulatum after 7 days incubation at room temperature (Fig. 6). On the sealed blood slants incubated at 37°C, smooth whitish cerebriform pin-point colonies, perched on the medium, appeared between the 15-20th day, which on subsequent incubation turned yellowish brown (Fig. 7a and 7b). Some of the colonies grew a few cottony threads. This growth consisted of small oval budding cells, 1-3 micron in diameter with a few abortive fine mycelia (Fig. 8). This yeast like growth was easily converted to the mycelial phase by subculturing on to Sabouraud agar at room temperature. The conversion of the mycelial phase into the yeast phase proved difficult, but was finally accomplished on congealed potato-starch-egg medium (Kurung and Yegian 1954).

One specimen of sputum was injected intraperitoneally into 3 white mice after treatment with Penicillin and Streptomycin (Moffat et at. 1956). The first mouse was killed after 2 weeks. Neither the lungs, spleen nor liver showed any sign of Histoplasma infection either histologically or culturally. The lung, spleen, and liver of the second mouse, killed after 4 weeks, showed no evidence of Histoplasmosis in sections, but cultures of an emulsion of these organs resulted in a heavy growth of Histoplasma capsulatum.

The cultural and microscopic characteristics as reported above are diagnostic of Histoplasma capsulatum (Lewis and Hopper 1958, Schaub et al. 1958).

Histoplasma capsulatum was cultured from the pus of the cervical gland, from the throat swabs, from the bone-marrow and from several specimens of sputum. The throat swabs and sputum showed a heavy growth of Candida albicans as well. Intra dermal old tuberculin 0.1 ml of 1:1000 dilution produced a skin response. Intra dermal histoplasmin 0.1 ml of a 1:1000 dilution produced no response.

The results of the complement-fixation tests were:

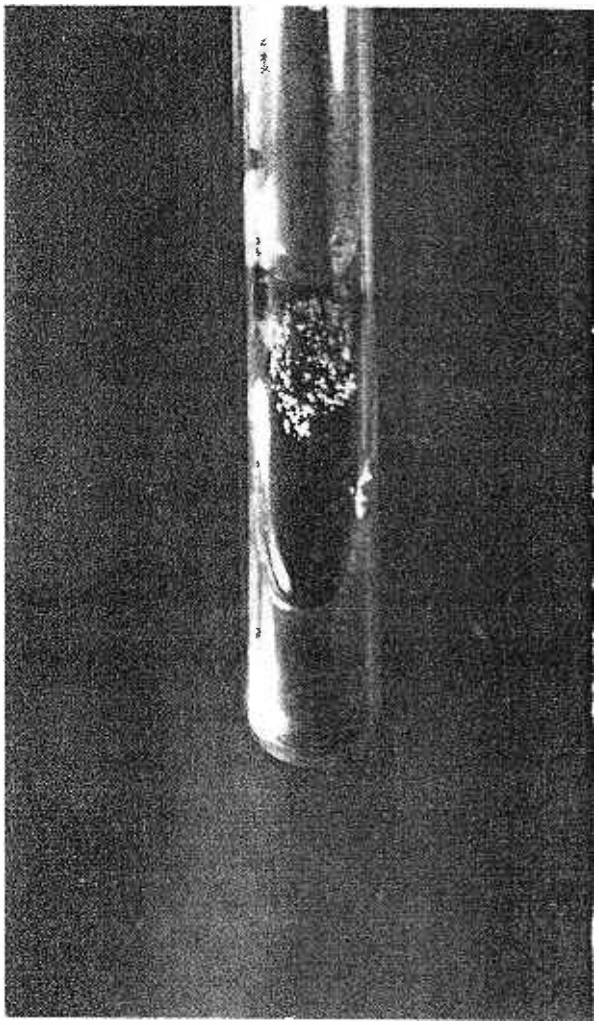

Fig. 7 (a).

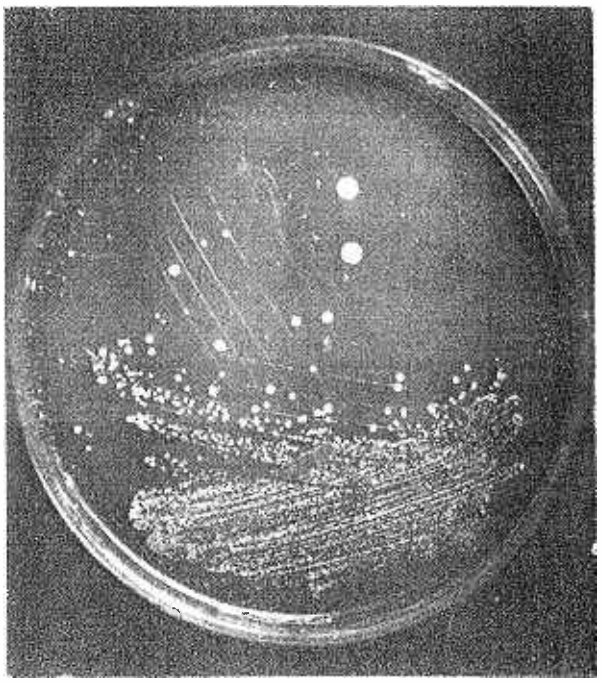

Fig. 7 (b)

Fig. 8.

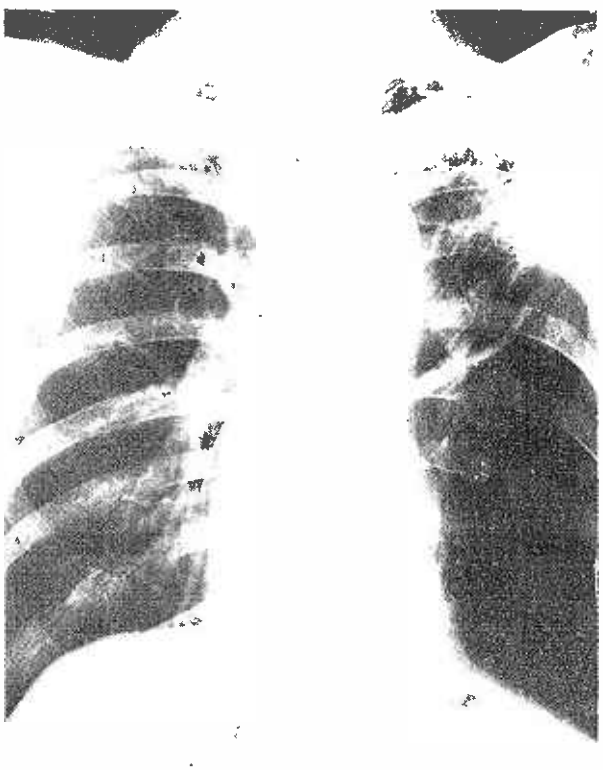

Fig. 9.

Histoplasmosis:

Whole yeast phase antigen — Negative
Histoplasmin — Negative

Blastomycosis:

Yeast phase antigen — 1 : 8

Treatment: Despite the relatively benign course of chronic pulmonary histoplasmosis the effects of therapy were disappointing until quite recently. Amphotericin B, an antifungal agent derived from a species of Streptomyces, is at present widely used. This drug claims to be effective against a wider variety of deep seated fungi and yeasts than any other antifungal agent now available. It is administered by slow intravenous infusion over a period of approximately six hours. Amphotericin B was given in 2 courses between February and March 1960, daily for 25 days with a break of 5 days as the patient developed toxic symptoms — nausea, loss of appetite, headache, febrile reactions and anaemia — and again in May 1960 for 7 days. The treatment was started with a dosage of 50 mg. daily for 4 days and then increased to 75 mg. for 4 days and to 100 mg. for 12 days. On the second occasion the treatment was initiated with 100 mg. and this dose was maintained throughout. This treatment was combined with a course of oral Sulphatriad. Haematological investigations, serum proteins, liver function tests and blood urea estimations were done weekly during the course of the treatment. The treatment altered the clinical course of this patient. The ulcerations in the nasopharynx, oropharnyx and laryngopharnyx disappeared, the cervical glandular enlargement subsided, the liver attained its normal size and the sputum remained persistently negative for Histoplasma capsulatum.

Discussion: This case presented is a typical Histoplasmosis. The organism was proved culturally and histologically. It is suggested that the Penicillin and Streptomycin in the media acted as definite growth promoting substances. The mouse proved the best medium for isolating the organism from contaminated material (sputum). The organism was isolated by culture in cases where the direct smear showed no organism (bone-marrow, mouse organs). The fact that the patient did not react to intradermal histoplasmin could be attributed to the state of acute generalised infection in which case the skin test often proves negative. The complement fixation test of his serum was negative. Dr. Charlotte C. Campbell who did the test adds the following comment, "Your histologically confirmed case of histoplasmosis was negative with our Histoplasma capsulatum antigens. The minimal titer of 1:8 with Blastomyces antigen is not significant. We frequently encounter cases of Histoplasmosis in which the Blastomyces antigen is the only one to react. The essentially negative serologic pattern is suggestive of a limited localised active infection or a more extensive one which has healed." In our case the infection was generalised and not healed.

It is difficult, if not impossible, to date accurately the onset of this illness. The mode of infection in this case is not known. Since leaving China 47 years ago, he had never left Singapore except for Johore in Malaya. He lived in the rural areas of Singapore (Katong), but had no interest in gardening, farming, raising pets or live-stock. He was not exposed to any environment likely to be the source of infection. He had been a taxi-driver for more than 30 years and his work brought him into frequent contact with American visitors; this is probably without significance, as there is no evidence that the infection is transmitted from person to person. He handled baggage and other belongings of tourists.

It is possible that being in close proximity with an infected visitor contamination of his clothes could have introduced the organism into the soil in his garden and subsequent infection has taken place or it may be that this fungus was already to be found in Singapore and Malaya undiscovered. The possibility of future cases of Histoplasmosis in Singapore and neighbouring countries should be borne in mind.

ACKNOWLEDGEMENTS

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