In the present patient extreme debility and prolonged antibiotic therapy were probably the most important factors in the development of systemic candidiasis. Latterly the progress of the disease may have been accelerated by cortisone therapy and by intravenous administration of 20% glucose saline. Indeed, Duhig and Mead (1951) and Schaberg and his colleagues regarded the prolonged use of intravenous glucose saline as an important factor in establishing the infection in their cases. Some form of injection appears to be common to all cases of systemic candidiasis irrespective of the use of antibiotics. Here, however, infection from the alimentary tract may have occurred at operation, since local peritoneal exudate infected with *C. tropicalis* was present between the stomach and liver at necropsy. Systemic candidiasis has been reported after duodenal perforation or abdominal operations on a number of occasions (Brown *et al.*; Schaberg *et al.*; Caplan, 1955; Matthias and Rees; Barrett *et al.*).

This case illustrates the various clinical factors predisposing to the development of systemic candidiasis. The history suggests that *Candida* may proliferate in the blood of a patient without any clinical signs of systemic moniliasis appearing other than pyrexia. Unexplained pyrexia occurring in prolonged debilitating diseases, especially where antibiotics and intravenous alimentation have been used, is an indication for culturing the blood in media suitable for the growth of fungi.

Summary

A case of systemic candidiasis due to *C. tropicalis* infection is recorded, and the clinical features, bacteriological investigations, and pathological findings are described. *C. tropicalis* endocarditis affected the right atrium. Other cases of monilial endocarditis are reviewed, and the aetiology and pathogenesis of the condition are discussed.

We thank Dr. W. Sircus for permission to publish this case, and are indebted to Dr. W. Blyth and Mr. D. W. R. Mackenzie, of the Experimental Mycosis Unit, Department of Botany, Edinburgh University, for confirmation of the *Candida* species.

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INDUCTION OF SARCOMA IN THE RAT BY IRON-DEXTRAN COMPLEX

BY

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[WITH SPECIAL PLATE]

Pathological amounts of iron may accumulate in the body in a variety of conditions—for example, haemochromatosis and transfusional, malnutritional, and occupational siderosis. In all these conditions arguments for and against the cytotoxic effects of iron have been advanced, but the possibility that this metal plays any part in carcinogenesis has hardly been entertained. Therefore, in the belief that iron was non-carcinogenic if not altogether bland, an iron–dextran complex ("imferon," Benger Laboratories Ltd.) was deliberately chosen in order to establish a progressive mobilization or "hyperplasia" of histiocytes. This need arose when it had been discovered (Richmond, unpublished observation) that repeated subcutaneous injections of trypan blue commonly induced a pleomorphic histiocytic sarcoma in the rat at the site of injection. By a collateral series of experiments with a non-carcinogenic substance it was hoped to detect some critical difference between a "hyperplasia" and a neoplasia of histiocytes. This hope was not realized because another crop of pleomorphic histiocytic sarcomas histologically identical with those induced by trypan blue was obtained as detailed below.

All the rats belonged to a home-bred hooded strain in which no spontaneous tumours of this kind have previously been recognized in this laboratory. They were fed on Diet No. 1 (Thomson cube) supplemented with greens.

Experimental Procedures

Experiment 1.—The animals were all adult males within the weight range of 250–300 g. at the beginning of the experiment. (*a*) 40 received a weekly intramuscular injection of 0.4 ml. iron–dextran complex into the right upper thigh. Each dose contained 20 mg. of iron as ferric hydroxide in complex with low-molecular-weight dextran. (*b*) 12 received weekly intramuscular injections of 0.5 ml. "ferrivenin" (Benger) under ether anaesthesia, with occasional interruptions owing to the development of ulceration. Each dose contained 10 mg. of iron as saccharated oxide of iron. (*c*) 12 received weekly injections of 0.5 ml. low-molecular-weight dextran. (*d*) 12 received weekly injections of 0.5 ml. normal saline solution.

Experiment 2.—The animals were weanling rats of both sexes averaging 48 g. in weight. (*a*) 20 males and 20 females were given twice-weekly intramuscular injections of iron–dextran complex into the right upper thigh in graduated dosage according to weight—namely, 0.1 ml. up to 100 g., 0.2 ml. up to 150 g., 0.3 ml. up to 200 g., and 0.4 ml. thereafter. This regimen was stopped after three months, each animal

having received 9.5 ml. iron–dextran complex. (*b*) 6 males and 6 females received corresponding volumes of low-molecular-weight dextran solution.

Results

In Experiment 1, 17 of the 40 animals receiving iron-dextran complex were killed during the early months, but 16 of the remaining 23 developed tumours at the site of injection between the 11th and 16th months of the experiment. Rats given low-molecular-weight dextran and saline did not develop tumours, nor did those animals receiving saccharated oxide of iron over a period of 17 months. In the last group the injections commonly gave rise to necrosis which might be associated with ulceration of the overlying skin and secondary infection; collections of siderocytes and an abundant granulation-tissue reaction were demonstrated on histological examination, but no evidence of malignancy was seen.

In Experiment 2, tumours developed in 10 female and 12 male rats at the site of injection from six to eight months after cessation of iron treatment, while those receiving dextran alone showed no tumour formation.

Histological Observations.—The essential change at the site of injection of the iron–dextran complex in the early stages is a progressive accumulation of histiocytes laden with iron pigment. Previous to the inception of tumours it has been established from biopsy material that occasional histiocytes develop enlargement and hyperchromatism of the nucleus associated with mitotic activity and other aberrant changes (Special Plate, Fig. 1). Therefore it is deduced that the neoplastic process originates in such histiocytes, which gradually lose their avidity for iron as the neoplasia gathers momentum. The characteristics of established tumours vary from spindle-cell sarcoma to a highly pleomorphic growth including many giant cells and exhibiting numerous mitoses (Special Plate, Fig. 2). Iron-laden histiocytes are distributed throughout the tumour, and, while some neoplastic cells contain traces of the metal, the great majority are free from it.

Transplantation.—Metastases were not observed in the tumour-bearing animals, but transplantation of the tumour was successful in three of four examples in which this was attempted. In the first generation the transplants grew slowly after a lag period of four, six, and eight months respectively, but more rapidly in subsequent generations. One line is now in the 27th generation, requiring transplantation every four weeks. Histologically, some degree of the initial pleomorphism is still seen in this transplanted tumour, but the growth is essentially a spindle-cell sarcoma.

Discussion

From these observations it is clear that intramuscular injection of iron–dextran complex is carcinogenic in the rat. It is also clear that the dextran fraction alone is free from carcinogenic activity under the conditions of the experiments, so that iron is likely to be the responsible agent. The possibility that this effect is exerted only when it is in complex with dextran has been considered, but this is regarded as unlikely owing to the fact that the dextran is rapidly split off after injection and the iron chelated with protein to form haemosiderin (Golberg, 1957). The attempt to decide the issue by the repeated injection of saccharated iron was upset by the occurrence of necrosis and ulceration, necessitating interruptions in the sequence of the injections. These findings in the rat following the injection of iron–dextran complex have been confirmed in a small series of mice at the Chester Beatty Research Institute (Haddow, 1958).

The clinical significance of these observations cannot be assessed pending an elaborate study of the many factors involved. The iron–dextran complex has been widely used in the treatment of iron-deficiency anaemias in adults (Scott and Govan, 1954; Cappell *et al.*, 1954; Baird and Podmore, 1954; Jennison and Ellis, 1954; Scott, 1956) and in infants (Gaisford and Jennison, 1955). The amount administered in the adult human subject will vary according to the need of the patient, up to 54 ml. being recommended by the manufacturer for a patient weighing 180 lb. (81.64 kg.) with an observed haemoglobin value of 50%. Against this may be contrasted the volume administered to the rat—namely, 17.6–25.6 ml. in Experiment 1 (*a*) and 9.5 ml. in the younger rats of Experiment 2 (*a*). Therefore it will be evident that the dosage used in these experiments is relatively massive—some 200–300-fold—when compared with the therapeutic dose—for example, 20–40 ml. for a 70-kg. man.

Hitherto iron has not been proved to be a carcinogenic agent, and this finding may have some wider implications. For example, frequent reports appear in the literature concerning the high incidence of hepatoma supervening on the cirrhosis of the liver which is associated with haemochromatosis. The usual incidence of liver cancer in relation to cirrhosis of Laennec type has been placed at 3 to 7% (Anderson, 1957). In contrast, Warren and Drake (1951) reported the development of hepatoma in 18% of their cases of haemochromatosis, Stewart (1931) found an incidence of 10% in 151 collected necropsy cases of haemochromatosis, and Willis (1953) found three cases of hepatoma in seven necropsies on patients suffering from haemochromatosis. These figures suggest that there is a special tendency for the cirrhosis of haemochromatosis to undergo malignant change as compared with cirrhosis in general. While Edmondson and Steiner (1954) believed that the difference might be due to patients with haemochromatosis living longer than patients with portal cirrhosis of Laennec type, the present experiments suggest that gross iron deposition may well be the factor responsible. Incidentally, Branwood (1958) has investigated 100 examples of portal cirrhosis, of which slight to gross deposition of haemosiderin in the liver cells was observed in 33 cases. Hepatoma supervened in six of the 100 cases of portal cirrhosis, and all six were included in the group showing haemosiderosis.

The evidence submitted also suggests that iron may be the important causal agent in the development of pulmonary carcinoma in haematite miners. The rising incidence of pulmonary carcinoma in haematite miners was pointed out by Faulds and Stewart (1956), who found 17 cases of carcinoma of the lung in 180 necropsies (9.4%). These lungs contained silica as well as iron, but there was no similar incidence of pulmonary carcinoma in coal-miners dying of silicosis in the same area of West Cumberland, nor had this been reported from other coal-mining areas. Faulds added more information in 1957 with regard to the incidence of pulmonary carcinoma in haematite miners, making it clear that the only other silicious trades associated with

an increased incidence of carcinoma of the lung were asbestos workers, moulders and foundrymen, and chromate workers, and pointing out that asbestos and chromate contain varying amounts of iron in their chemical composition. In this context there is further support from the work of Campbell (1940, 1943), who exposed mice of a strain susceptible to lung cancer to an atmosphere laden with ferric oxide. He found that 32.7% of the mice exposed to the dust developed tumours of the lung compared with 9.6% of the controls, while mice exposed to a mixture of silica and ferric oxide showed a tumour incidence halfway between these figures (19.4%).

Mode of Action of Iron.—It has been observed that the histiocytes at the site of injection and elsewhere in the body (for example, Kupffer cells) contain, in addition to iron pigment, globules of a lipofuscin pigment of ceroid nature. The development of ceroid throughout the tissues of the body is one of the characteristics of vitamin-E deficiency (Mason, 1944), and in recent experiments Golberg and Smith (1958) have shown that the ceroid developing through iron overload can be largely prevented by supplementing the diet with α-tocopherol. It may therefore be inferred that one way in which iron influences intracellular metabolism is by blocking the antioxidant activity of vitamin E and possibly other antioxidants in the cell. The destruction or interference with these natural antioxidants allows oxidation of unsaturated fats to form the yellow pigment which is termed ceroid (Casselman, 1951). It remains to be shown whether this action of iron in the cell has any relation to the development of malignant change.

Summary

In the adult rat weekly-repeated intramuscular injections of iron–dextran complex induced sarcoma at the site of injection. A bi-weekly series of similar injections, begun in weanling rats and stopped after 12 weeks, also induced sarcoma at the site of injection some seven months later.

I wish to thank Professor J. S. Young for continuous encouragement and advice during this work, and Professor A. Haddow for a helpful discussion and the personal communication of his observations on the mouse. My thanks are also due to Mr. A. Bodie, senior technician, for his co-operation, and to Miss E. M. Gillies, who carried out much of the technical work. Mr. W. Topp kindly supplied the photographs.

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MELIOIDOSIS

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[WITH SPECIAL PLATE]

Melioidosis is an uncommon yet highly lethal disease caused by *Pfeiferella whitmori*, which closely resembles the organism responsible for glanders in animals. The disease was first reported by Whitmore and Krishnaswami in 1912 from necropsies on vagrants in Rangoon. In 1932 Stanton and Fletcher, of the Institute for Medical Research, Kuala Lumpur, Malaya, collected 83 cases of human infection from the literature—six of them in Europeans, and only nine of which were diagnosed before death. In this series there were only two survivors. Couture in 1935 reported 35 cases with a 95% mortality, and since then single instances or small series of cases have been recorded by a number of authors.

In the past the disease has been restricted to that part of Asia east and south-east of India, but in recent years isolated cases have been reported in Great Britain, the U.S.A., and South Africa among nationals who have returned from periods of residence in these endemic regions.

We have recently seen three patients with this disease in Malaya. We report them because they demonstrate the successful outcome that may be expected in cases suitably treated, and also because, owing to the large numbers of men and women of various nationalities who visit Malaya, isolated cases may be expected almost anywhere in the world.

Case 1

A 50-year-old male Chinese vegetable seller was admitted to the General Hospital, Kuala Lumpur, on March 21, 1955, complaining of cough and fever of two weeks' duration. There was much purulent sputum, at times blood-stained, and he was wasted and looked toxic. Temperature 100.6° F. (38.1° C.), pulse 120, respirations 30. There were signs of consolidation in the right infraclavicular region, and a pleural rub in the left mid-axillary line associated with a few moist sounds. Liver and spleen were not enlarged, and no lymph nodes were palpable. Hb 7.7 g. per 100 ml.; W.B.C. 11,800 per c.mm.; E.S.R. 83 mm. in one hour (Westergren). Sputum examination repeatedly negative for acid-fast bacilli. Radiography showed ill-defined patchy opacities in the upper zones of both lungs, with a larger homogeneous opacity in the left mid-zone peripherally (Special Plate, Fig. 1). There was a large soft-tissue swelling in the root of the neck on the left side.

A provisional diagnosis of pulmonary tuberculosis was made, and streptomycin, 1 g. daily, and isoniazid, 100 mg. thrice daily, were administered. By the eleventh day the fever had subsided, but treatment was continued up to a total of 31 g. of streptomycin. Despite this an abscess developed in the right supraclavicular fossa from which dark, offensive material was aspirated. No organism was demonstrated on smear, but some haemolytic streptococci

H. G. RICHMOND: SARCOMA INDUCTION BY IRON-DEXTRAN COMPLEX

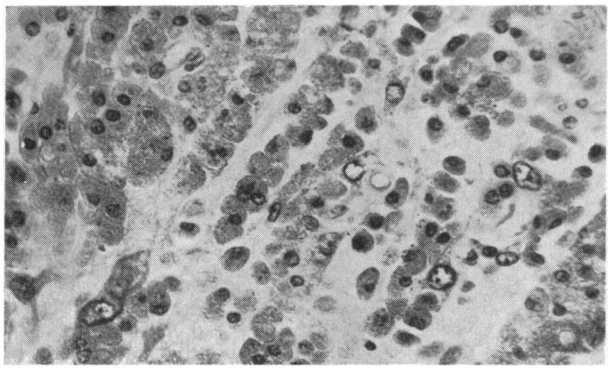

FIG. 1.—Biopsy of site of injection at 16 months. Iron-laden histiocytes mingled with aberrant cells. (H. and E. ×300.)

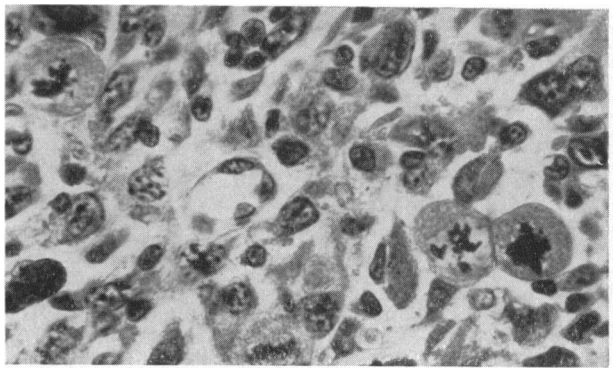

FIG. 2.—Biopsy from same rat as Fig. 1 five weeks later. Pleomorphic tumour showing numerous mitotic figures. (H. and E. ×450.)

B. S. KHAIRA, W. B. YOUNG, AND P. de V. HART: MELIOIDOSIS

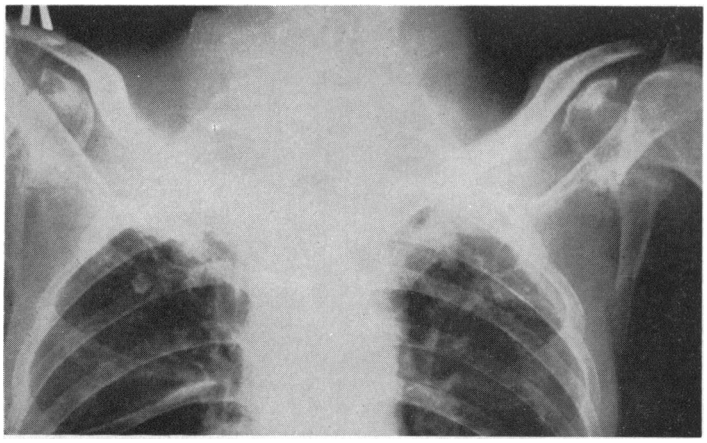

FIG. 1

FIG. 1.—Case 1. Ill-defined patchy opacities in upper zones of both lungs, with larger homogeneous opacity in left mid-zone peripherally.

FIG. 2.—Case 1. Porosis of lower cervical and upper dorsal spine and upper ribs.

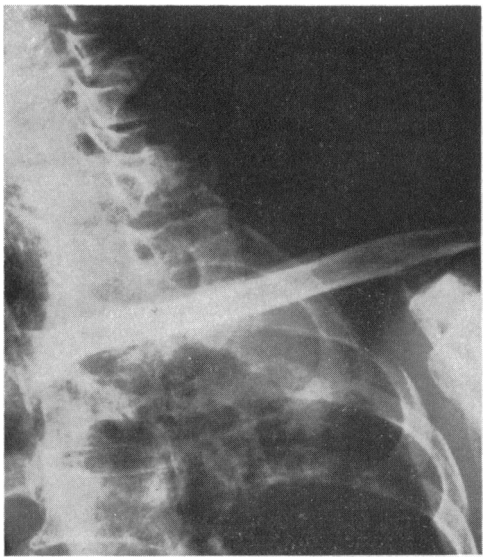

FIG. 2

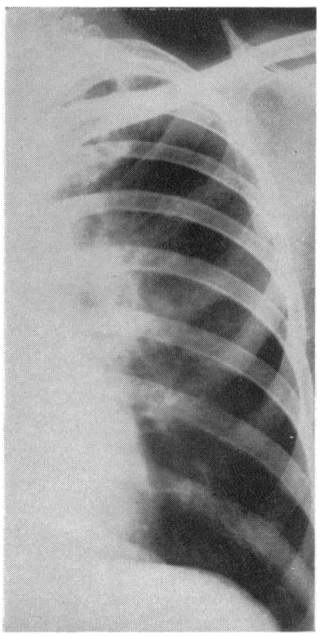

FIG. 3.—Case 2. Infiltration in left upper and middle zones of left lung.

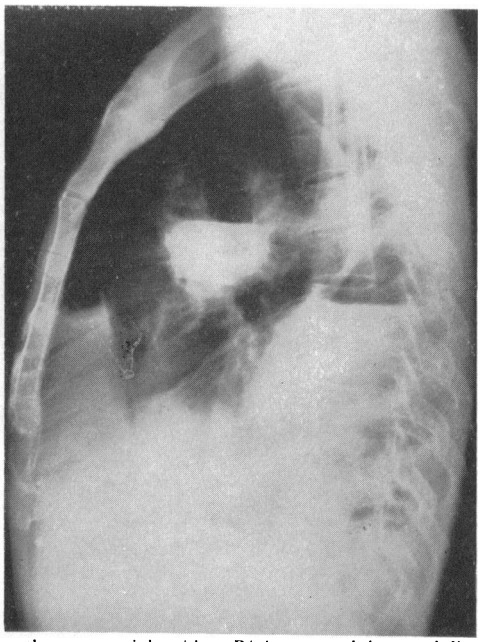

FIG. 4.—Case 3. Loculated hydropneumothorax on right side. Right upper lobe partially collapsed and containing medium-sized abscess cavity surrounded by irregular area of consolidation.