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An Imported Case of Kala-azar in Korea

Je G. Chi, Young Kee Shong, Sung Tae Hong,* Soon Hyung Lee*
Byong Seol Seo,* Kwang Won Choe**
Departments of Pathology, Parasitology, and Internal Medicine,***
College of Medicine, Seoul National University

INTRODUCTION

Kala-azar is visceral type leishmaniasis which is caused by *Leishmania donovani*. *Leishmania* is a genus of hemoflagellates and requires both vertebrate and invertebrate hosts in its life cycle. Human and some mammals become the vertebrate hosts, and sandflies are the invertebrate and vector host. Three species in genus *Leishmania* are known to affect human beings. They are *L. donovani*, *L. tropica* and *L. brasiliensis*. Since their morphological characteristics are identical they can only be distinguished by clinical manifestations, biology, serology and geographical distribution.

Kala-azar is not known to occur indigenously in Korea. It is an endemic disease of the Mediterranean countries, Middle Asia, Africa, India, China, and Central and South America. All the cases of Kala-azar reported in Korea were the patients who returned home after being infected from mainland China or Manchuria.

This communication is a case report of Kala-azar that we experienced recently, which is thought to be imported from Mideast Asia.

CASE RECORD

A 26-year-old Korean male living in Seoul visited Seoul National University Hospital (SNUH) on September 17, 1982, complaining of prostration and abdominal distention. Since May 1982, he had suffered from easy fatigue, and felt a hard mass in the abdomen. Also there had been a daily spiking fever which reached its peak in the early afternoon for the last two months. He lost 12 kg during 4 months. Several episodes of epistaxis were presented during this period. The patient had been well before the onset of the symptoms.

On admission the patient was emaciated and chronically ill-looking with huge liver and spleen. His body weight was 54 kg, and his height 177 cm. A firm, but not tender liver was palpable 14 cm below the right costal margin. The spleen was 14 cm palpable (Fig. 1). Ascites was not present. Both inguinal lymph nodes were palpable and firm. Under the impression of hematologic malignancy, he was admitted to the Department of Internal Medicine on September 28, 1982.

At the time of admission, Hb 9.6 gm%, Hct 30.8%, WBC 3,200/mm^3, platelet 54,000/mm^3 and reticulocyte 0.9%. Bleeding time was 2min. and 50sec., prothrombin time 100%, activated partial thromboplastin time 56sec. (control 38 sec) and fibrinogen 320mg%. Serologic test for syphilis was negative. Serum calcium 9.4mg%, phosphorus 3.3mg%, blood urea nitrogen 14 mg%, and serum uric acid was 6.0mg%. Total serum protein 9.9g% and albumin 2.9g%. Serum electrophoresis revealed polyclonal gammopathy with increased IgG and increased IgM, kappa and lambda chains. Total serum bilirubin 0.7 mg%, serum alkaline phosphatase over 350 units, SGOT 67 IU/L and SGPT 37 IU/L. Urinalysis revealed one positive albumin. Chest X-ray was normal. Supine simple abdomen showed hepatosplenomegaly. Esophagogastroscopy

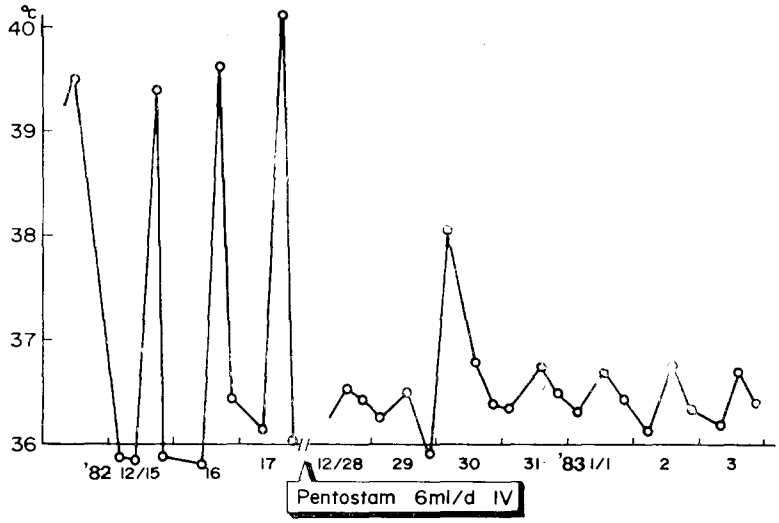

Fig. Temperature curve before and after Pentostam treatment.

revealed no abnormality. Blood culture for pathogenic bacteria was negative.

Daily spiking fever of up to 39°C continued after admission along with dyspnea and palpitation. For his anemia packed cell transfusion was given several times. Bone marrow aspiration revealed normocellular marrow with monohistiocytic hyperplasia and increased megakaryocytes. Inguinal lymph node biopsy showed atypical lymphohistiocytic proliferation suggestive of virus-associated hemophagocytic syndrome. On October 14, 1982, peritoneoscopic biopsy of the liver was performed, which showed intra- and extracellular amastigotes.

After the diagnosis of Kala-azar was made the patient disclosed the fact that he had been in Gizan, Saudi Arabia from September, 1980 to September, 1981 as a construction worker. He remembered that there were numerous flies and mosquitoes in and around the living quarters when in Saudi Arabia. He also lately informed the doctors that he had been previously admitted to National Seoul Hospital from July 9, 1982 to July 22, 1982, where he was suspected to have Kala-azar. No specific treatment was given there. After discharge he was on herb medicine until he finally decided to visit SNUH.

The specific chemotherapeutic agent, pentavalent antimonial, was unfortunately not available at that time in Korea. And while waiting for the arrival of the chemotherapeutic, the patient was under general supportive measurements, such as packed cell transfusions, albumin infusions and vitamin K injections. As his general condition became worse, with the development of facial edema and bleeding tendency Amphotericin B was tried as an alternative on December 19, 1982. The dose was raised gradually up to 25mg daily with serial checking of serum creatinine and potassium. No apparent clinical improvement was noted in that period. Finally sodium antimony gluconate (Pentostam®) was obtained through the Welcome Foundation. The patient was given daily 6 ml (0.6μg) intravenously from December 28, 1982. Three cycles of 10 day treatment was repeated with intervals of 10 days. Laboratory data at the beginning of Pentostam® treatment was Hb 8.6 g%, Hct 25.7%, WBC 9,000/mm^3, platelet 61,000/mm^3, ESR 58 mm/hr, protein 9.1 g% and albumin 2.5g%. Following the drug administration the spiking fever subsided promptly (Fig. in the text), and five days later his appetite improved, abdominal discomfort lessened, and he started to gain weight. After the first cycle of chemotherapy ended on January 7, 1983, his liver size shrank to 12cm below costal margin. The spleen decreased also in size to 9.5cm. At that time laboratory data were Hb 7.0g%, Hct 20.8%, WBC 4,500/mm^3, platelet

788,000/mm³, ESR 62mm/hr, protein 8.7g% and albumin 2.7g%, The second course continued from January 17 to 26, 1983. At completion of the second cycle, laboratory data were Hb 10.5g%, Hct 22.1%, WBC 4.300/mm³, platelet 114,000/mm³, ESR 45mm/hr, serum protein 10.8g% and albumin 3.8 g%. The liver size decreased to 10 cm and the spleen 7cm. The third course was given from February 6 to 15, 1983.

After completion of chemotherapy, follow-up liver biopsy was taken on February 21, 1983, to show focal scar and otherwise intact liver parenchyme with no amastigotes demonstrable. The liver was palpable 8cm and the spleen 3cm, at the time of his discharge on February 28, 1983. On April, 1983 the liver size decreased to 6cm and was soft. The spleen became no longer palpable. Body weight returned to 68 kg. The laboratory findings were in normal ranges except for an increased serum alkaline phosphatase. On June, 1983 the latest checkup so far, the liver was soft and palpable 2cm below the right costal margin. And no subjective symptoms were complained about.

PATHOLOGICAL AND PARASITOLOGICAL FINDINGS

The liver biopsy specimen (S82-11578) was reddish purple tissue speckled with gray white patches. It was granular without notable necrosis under the stereoscope. A small fragment was prepared for transmission electron microscopy (TEM), and the rest was fixed in 10% formalin and stained by various methods.

The light microscopic examination showed lobular architecture of the liver, which was distorted due to multifocal areas of cell necrosis and fibroblastic proliferation (Fig. 2). Amastigotes were present prominently in necrotic areas. The organisms were easily found by hematoxylin and eosin staining as round bodies of approximately 2-7 μm in diameter, in which dark spots were seen. They were clustered or dispersed in necrotic areas. The Kupffer cells near the junction of necrotic area and relatively normal tissue were often filled with the small intracellular parasites (Fig. 3). These were observed also by Giemsa staining. Most of the inflammatory cells were mononuclear cells mixed with plasma cells. Pyknotic nuclei were frequently seen in the necrotic areas where numerous amastigotes were seen.

An inguinal lymph node biopsy (82-11193) showed a considerable follicular hyperplasia as well as atypical proliferation of reticular cells in the medullary sinuses. There was erythrophagocytosis of the activated histiocytes. However, no parasites were seen.

Follow-up liver biopsy after treatment revealed normal liver tissue mixed with focal fibrosis (Fig. 4). The areas of necrosis were replaced by fibrosis.

TEM of the liver tissue demonstrated numerous round to ovoid organisms which were amastigotes. Although the organisms were seen intracellularly, it was sometimes difficult to be sure of intracellularity in necrotic centers where the organisms were not particularly related with cytoplasmic organelles. The cytoplasm of the leishmania body was packed with numerous ribosome granuless. And occasionally unidentifiable osmiophilic inclusions were observed in the cytoplasm. The subpellicular microtubules were observed beneath the cell membrane. The nucleus was large and eccentric. Central karyosome was slightly eccentric in the nucleus and composed of fine granules. And peripheral chromatin was located along the nuclear membrane with irregular density of granules. The organism had a flagellum which merged in an invaginated flagellar pocket (Fig. 5). Cross section of the flagellum showed 9+2 pattern of axonemal microtubules (Fig. 6). The length of longitudinally sectioned flagellum varied by the organism, occasionally reaching whole dimension of cytoplasm. Flagellum may be extruded from the cytoplasm but always covered with cell membrane (Fig. 7). Basal body was identified at the beginning of the flagellum. Close to the basal body, kinetoplast was observed in the double layered

membrane of mitochondrion. It consisted of highly extended and convoluted DNA fibrils. And the cristae structure of mitochondria was also sectioned in separate membrane from kinetoplast.

DISCUSSION

Recently there are increasing cases of tropical diseases in Korea although they are not necessarily published. Since cutaneous leishmaniasis (Yoo *et al.*, 1978) was reported in Korean workers returned from Saudi-Arabia, it was naturally expected that visceral leishmaniasis would be encountered sooner or later.

It was quite natural that the diagnosis and treatment were unnecessarily delayed in this patient because any kind of leishmaniasis is not indigenously endemic in Korea, and therefore no experience of kala-azar could be obtained among Korean physicians. In this context, hematologic malignancy has the diagnostic priority over infection origin for such cases. Furthermore at first the patient failed to report the important history of his travel to Saudi Arabia. It took another two months to procure the chemotherapeutic agent Pentostam® even after the correct diagnosis was made. It was tried through 3 routes; the Korean Embassy in Saudi Arabia, the Center for Disease Control (CDC), Atlanta, Georia, USA, and the Welcome Foundation. Thanks for the Welcome Foundation. The drug, Pentostam® proved to be very effective in this case. The improvement was quite impressive, and it was morphologically confirmed by the follow-up liver biopsy of this patient, that showed fibrous scar formation and no residual organism.

This case is not the first case of Kala-azar ever reported in Korea. Professor Heu (1952) of Seoul National University Hospital reported three cases of Kala-azar in 1952. All those cases were returnees from Northern China. Therefore, they represent imported cases. The diagnosis was based on the finding of Leishman donovan bodies from splenic aspirates. We are not aware of any other reported cases of Kala-azar in Korea. This case therefore seems to be the first proven case of Kala-azar imported from Saudi Arabia. Despite the fact that Cochrans (1914) described distribution of Kala-azar along the midwestern coast in Korea it is believed by many parasitologists in Korea the Kala-azar is not indiginous disease in Korea, and Cochrans' statement might have been based on imported cases. Kala-azar is a systemic disease and can jeopardize human life. Very recently schistosomiasis cases were reported by Min *et al.* (1983). These along with tropical malaria raise a very important issue that we should be ready for it on a government scale.

Lastly we like to stress the point that despite the meticulous search the amastigote organisms were not detected in the bone marrow and lymph node by light microscopy. The lymph node interestingly showed the features that were suggestive of "virus-associated hemophagocytic syndrome." Atypical lymphohistiocytic proliferation with increased phagocytic activity was the prominent feature. Histological findings of the liver biopsy were also different from what are described in standard textbooks in showing a significant parenchymal destruction and necrosis. There were definite foci of necrosis, and in these areas numerous fibroblasts were demonstrable under electron microscope. The Kupffer cell was not the only cell involved. The hepatocytes were actually involved and destroyed by the disease, although we have not demonstrated organism inside the hepatocytes. Many organisms were found extracellularly in these zones. In other words the liver lesion in this case is not simply diffuse but diffusely patchy instead. The ultrastructural demonstration of *Leishmania donovani* in this case was very helpful in confirming the diagnosis. And the findings were identical to those described by Aikawa and Sterling (1974).

SUMMARY

An imported case of Kala-azar in a 26-year-old Korean man is reported. The diagnosis was made by liver needle biopsy. Amastigotes were

seen in Kupffer cells under light microscope, and their characteristic ultrastructural features were recognized under the electron microscope.

This case represents an imported disease from Saudi Arabia where the patient spent one year as a construction worker, 8 months before the onset of the disease. This report also signifies the second description of Kala-azar in Korea, and the first case of Kala-azar imported from Saudi Arabia.

This patient was successfully treated with sodium antimony gluconate (Pentostam®), and follow up liver biopsy showed focal fibrous scar and otherwise normal liver without demonstrable organism.

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=국문초록=

中東에서 유입된 Kala-azar 1例

서울대학교 의과대학 병리학교실, 기생충학교실* 및 내과학교실**

지제근 · 송영기 · 홍성태* · 이순형* · 서병설* · 최광원**

Kala-azar는 우리나라에는 분포하지 않는 것으로 인정되고 있으나 과거 중국에서 감염되어 국내에서 관찰된 보고가 있는 질병이다. 최근에 中東 지방에서 감염되어 귀국하고 발병하였다고 생각되는 例를 보고한다.

서울에 주소를 둔 26세의 한국인 남자가 1년간 사우디 아라비아에서 건설기능공으로 근무하고 1981년 9월 귀국하여 약 8개월 후인 1982년 5월 경부터 심한 피로감, 전신쇠약, 복부의 腫物등을 느끼기 시작하였다. 환자가 국립서울병원을 거쳐 서울대학교병원에 1982년 9월 17일 입원할 당시에는 간과 비장이 모두 14cm 정도 만지질 만큼 부어있었고, 심한 체중 감소(4개월간 14kg의 감소), 적혈구, 백혈구, 혈소판등 모든 혈구의 감소등이 관찰되었다. 肝生檢을 통해 amastigote가 관찰되고 이는 투사전자현미경을 통해서 典型的인 amastigote型으로 확인되었다. 환자는 국외에서 구한 약제인 sodium antimony gluconate(Pentostam®)와 각종 보조요법에 의해 치료를 받았다. 퇴원 직전에 시행한 肝生檢에서 괴사된 간 조직이 섬유화로 대치된 것이 확인되었고 蟲體는 관찰되지 않았다.

이 Kala-azar 例는 1952년 만주에서 流入되어 보고된 3例 이후로는 처음 관찰된 것으로 中東에서 流入된 것으로는 첫번째 報告例이다. 최근 급격히 늘어난 해외 인력진출을 감안하면 해당 지역의 열대 풍토병에 대한 다각적인 대책이 절실히 요청된다.

LEGENDS FOR FIGURES

Fig. 1. Photograph of the patient's abdomen to show hepatosplenomegaly. The lower margins of the liver and the spleen are outlined with black ink.

Fig. 2. Low power view of the first liver biopsy before treatment. Note patchy area of necrosis and disorganization of lobular architecture in the right half. H&E ×100.

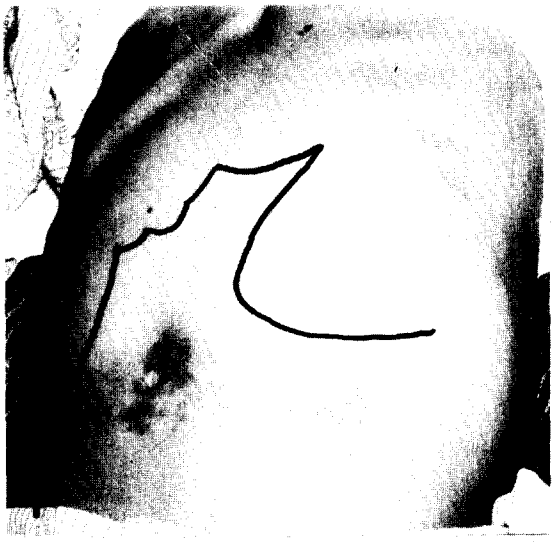
Fig. 3. High power view of amastigote in Kupffer cells of the junction of necrotic zone and relatively preserved area. H&E ×1,000.

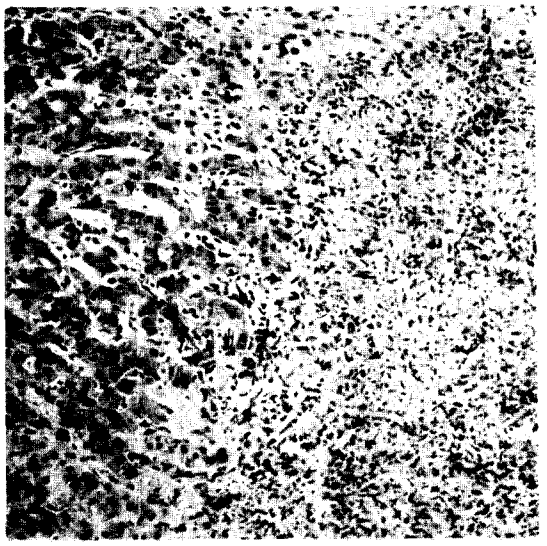
Fig. 4. Liver biopsy picture after treatment showing foci of dense fibrosis and otherwise well preserved liver tissue. No organism is demonstrated. H&E ×100.

Fig. 5. TEM picture of typical leishmania (amastigote) ×47,600, abbreviations as follows; N: nucleus with a central karyosome and peripheral chromatin, SM: subpellicular microtubule, M: mitochondria, F: flagellum, KP: kinetoplast, B: basal body, R: ribosomes.

Fig. 6. Cross sectioned flagellum showing 9+2 pattern of microtubules, ×42,000.

Fig. 7. An amastigote with a protruded flagellum, ×32,000.





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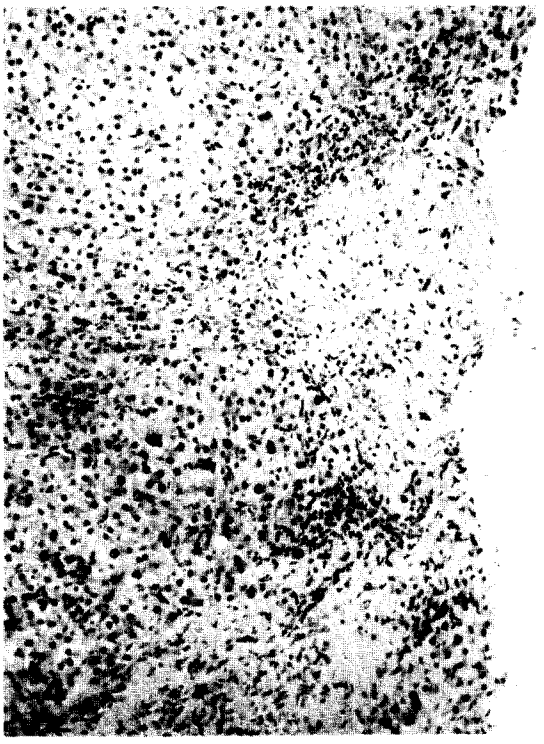

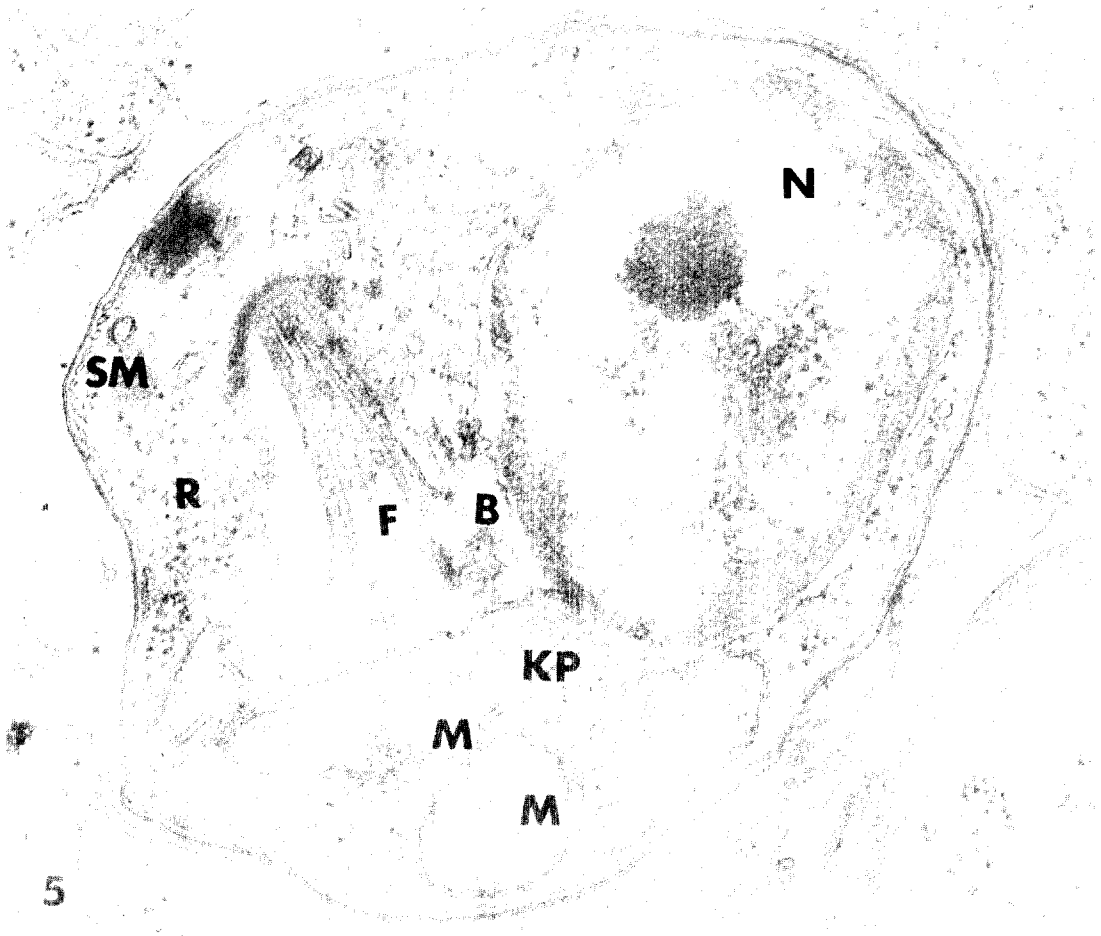

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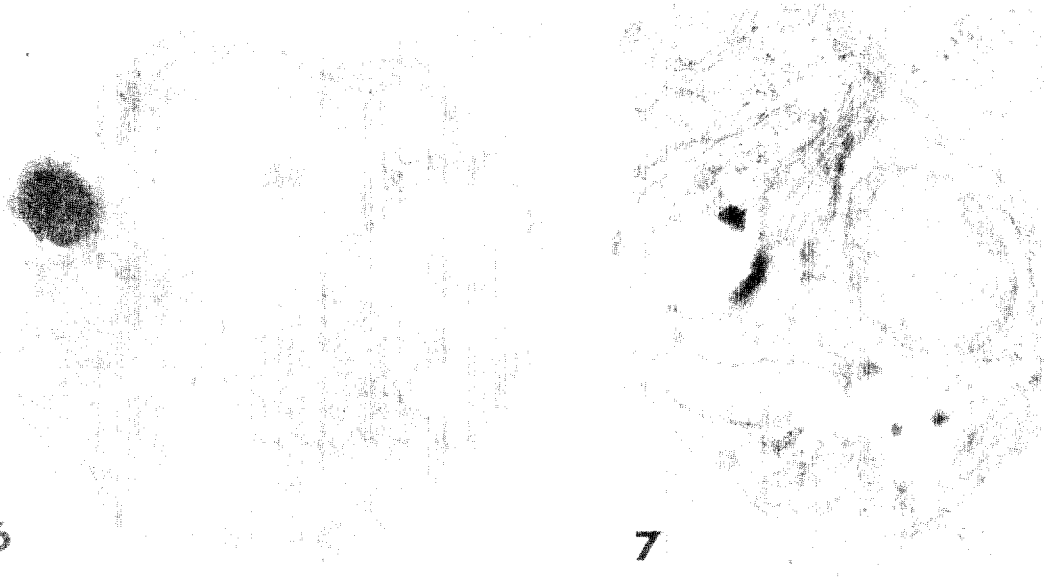

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