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Isolation of Virus of Lymphogranuloma Venereum from Blood and Spinal Fluid of a Human Being.*

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Although the virus of lymphogranuloma venereum has been isolated frequently from inguinal buboes, genital and rectal lesions, there have been few successful isolations of this agent from other parts of the body.[1] Sabin and Aring[2] unequivocally demonstrated the virus in the spinal fluid of a patient with meningoencephalitis. Von Haam and D'Aunoy[3] also reported its isolation from the spinal fluids of 2 patients suffering from lymphogranuloma venereum, but identification of their agents is open to some question, since it was based principally on positive skin reactions with infected mouse brain in patients with clinical lymphogranuloma.

The agent of lymphogranuloma venereum has been found in the blood of infected experimental animals,[1] but it has not previously been reported in the blood of a human being. The present communication reports the isolation of this virus from the blood and the spinal fluid of 1 of 8 proved cases of the disease.

Methods. The technic of isolation and identification of the virus of lymphogranuloma venereum used in this laboratory is given in detail elsewhere.[4] The salient features are as follows: Fresh material is inoculated intracerebrally in 0.03 cc amounts into each of 6 young adult Swiss mice, and in 1 cc amounts into the yolk sac of 6 6-day-old fertile eggs. Mice usually show signs of illness on the first passage, whereas blind passages frequently have to be made with yolk sac material before infection becomes evident. The virus is established in each host by a number of successive passages of infected material. Elementary bodies are readily found in spreads of infected mouse brains and yolk sacs stained by the Macchiavello method.

Further identification of these agents as the virus of lymphogranuloma venereum is based on tissue tropisms and sulfonamide inhibition. Of the various agents of the psittacosis-lymphogranuloma group, only lymphogranuloma venereum has the 2 characteristics of infectivity for mice by the intracerebral route of inoculation and of susceptibility to sulfonamide inhibition.[5]

Results. The 8 patients concerned in this report were young colored adults, 7 males and one female, and were all studied in the early stage of lymphogranuloma venereum infection. Four were tested within the first 4 days, one on the sixth day, one on the 14th day and 2 on about the 20th day of disease. Systemic manifestations in these patients were no more pronounced than in other patients with this disease. Furthermore, none of them showed evidence of meningeal involvement, and spinal fluid examinations in all 8 cases gave normal cell counts and protein levels.

The virus of lymphogranuloma venereum was isolated from bubo pus from every one of the 8 patients. Blood was examined for the presence of virus in 6 of them, and similar studies were carried out on the spinal fluid

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1 Literature summarized by: Stannus, H. S., *A Sixth Venereal Disease*, Baillière, Tindall, and Cox, London, 1933, Ch. 11; van Rooyan, C. E., and Rhodes, A. J., *Virus Diseases of Man*, Oxford University Press, London, 1940, Ch. 16; Koteen, H., *Medicine*, 1945, **24**, 1.

2 Sabin, A. B., and Aring, C. D., *J. A. M. A.*, 1942, **120**, 1376.

3 von Haam, E., and D'Aunoy, R., *J. A. M. A.*, 1936, **106**, 1642.

4 Wall, M. J., *J. Immunol.*, in press.

5 Hamre, D., and Rake, G., *J. Infect. Dis.*, 1944, **74**, 206.

of 5 patients, at the time when virus was obtained from the buboes. The virus of lymphogranuloma venereum was isolated from both blood and spinal fluid of one patient (No. 161), a 20-year-old woman, tested on the sixth day of the disease. The agents from her blood and spinal fluid produced signs of infection in chick embryos on the first passage, whereas material from her bubo required one blind passage before infection was evident. From 8 to 17 passages were made on all 3 agents. Elementary bodies were demonstrated in spreads of infected mouse brains and yolk sacs, and the tissue tropisms and susceptibility to sulfonamide therapy, characteristic of the virus of lymphogranuloma venereum, were found to be properties of these agents. In addition, serologic identification of the viruses was established by neutralization of their toxins by hyperimmune serum prepared in chickens against a known strain (J.H.) of the virus of lymphogranuloma.[4,6,7]

Summary. Eight patients with acute lymphogranuloma venereum, proved by isolation of the virus from buboes, were examined for the presence of virus in blood or spinal fluid, or both. The virus was found in the blood and also in the spinal fluid of one patient. The clinical features in this case were not significantly different from those of the other 7 cases. This is the first reported instance of isolation of the agent of lymphogranuloma venereum from the blood of a human being.

6 Rake, G., and Jones, H. P., PROC. SOC. EXP. BIOL. AND MED., 1943, **53**, 86; *J. Exp. Med.*, 1944, **79**, 463.

7 Hilleman, M. R., *J. Infect. Dis.*, 1945, **76**, 96.

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Effect of Diet on the Response of Chicks to Folic Acid.*

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Previous studies[1] in our laboratory indicated that chicks showed less growth, feather development and hemoglobin production when preparations of impure folic acid (obtained from Texas University) and the third *L. casei* factor (obtained from Lederle Laboratories, Inc.) were added to our basal ration containing sulfasuxidine than when added to the basal ration alone. Scott *et al.*[2] found the third *L. casei* factor to be somewhat active in the presence of sulfasuxidine, particularly in the presence of the pyridoxic acids. Subsequent work[3] showed that the activity of synthetic folic acid is apparently decreased when it is fed in diets containing sulfasuxidine. In an attempt to determine the function of folic acid by measuring its activity under different dietary conditions it was found that changes in diet may markedly alter the response observed when a given amount of folic acid is fed to chicks.

Experimental. Day-old White Leghorn chicks weighing between 40 and 50 g were divided uniformly into groups of 10, placed in electrically-heated cages with raised screen bottoms and fed the diets described in Table I. It is important to note that the level of vitamins was considerably higher than that normally used. The largest and smallest

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1 Briggs, G. M., Jr., Luckey, T. D., Elvehjem, C. A., and Hart, E. B., *J. Biol. Chem.*, 1945, **158**, 303.

2 Scott, M. L., Norris, L. C., Heuser, G. F., and Bruce, W. F., *J. Biol. Chem.*, 1945, **158**, 291.

3 Luckey, T. D., Moore, P. R., Elvehjem, C. A., and Hart, E. B., *Science*, 1946, **103**, 682.