

made post-mortem from the scrapings of the abscess wall showed definite contamination composed in greater part of members of the colon group. It is apparent that bacterial invasion had occurred post-mortem during the interval between death and the autopsy, which was approximately 12 hours.

The examination of the material from the scrapings of the liver wall showed many actively motile amoeba, the general features of which corresponded to those of the histolytica type. Smear preparations were made from this material and from the cultures and were stained by Heidenhain's iron haematoxylin method. A study of these stained specimens revealed the morphological features that have been regarded as indicative of the species histolytica.

The microscopic study of sections of the liver showed the characteristic autolysis of the hepatic structure as evidenced by massive necrosis with karyorhexis and karyolysis and few, if any, inflammatory cellular elements were present. Many amoeba were present in the tissue sections and were found especially at the margin of the necrotic and partially injured liver tissue.

While cultures from the intestinal tract may present a varied assortment of non-pathogenic amoeba, there seems but very remote likelihood of such condition occurring in the liver lesion wherein a definite clinical and pathological picture of amoebic infection exists, as was seen in the present instance.

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Injection into the Rabbit of Cultures of *Entamoeba Histolytica*.

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Cultures of *Entamoeba histolytica* obtained from the stools of human cases of amoebic infection have been introduced into the intestinal tract of kittens by Cutler¹ and Boeck and Drbohlav² and the disease apparently successfully reproduced in this type of animal.

A rather generalized systemic invasion by *Entamoeba histolytica* has also been described by other workers, but there are many who question the cellular structures considered as amoeba in such re-

¹ Cutler, D. W., *J. Path. and Bact.*, 1918, xxii, 22.

² Boeck, W. C., and Drbohlav, J., *Am. J. Hyg.*, 1925, v, 371.

ports. The invasion, however, of these protozoa into the liver with the resulting autolysis of hepatic tissue and the formation of the so-called amoebic "abscess" has been generally accepted as occurring through the portal system. The study of the histopathologic lesion in the liver often reveals numerous amoebas in the vascular pathways, suggesting this system as the route of invasion.

In the present experiments, having obtained at autopsy a satisfactory culture of *Entamoeba histolytica* from a hepatic "abscess," as herein reported³ we were desirous of ascertaining the effects that might be produced by hypodermic injections of such a culture into the lower animal through various routes of introduction.

The culture contained a varied bacterial flora in conjunction with the luxuriant growth of the amoeba. The presence of the bacterial microorganisms in the injection material naturally somewhat confuses the results.

Twenty-five rabbits were employed.

Series 1.—Eight rabbits were laparotomized and fresh cultures rich in amoeba were introduced with a hypodermic syringe into the branches of the mesenteric veins. Such suspensions were naturally carried through the portal system and into the liver. Five of these animals were killed at varying periods of from 2 to 5 weeks. At autopsy evidences of localized peritonitis were present about the site of inoculation but resolution and organization of exudate were shown in such areas. The remainder of the animals died of generalized peritonitis in from 24 to 72 hours. In these, some of the inoculation material had dropped into the cavity at the time of the injection. The livers of all 8 rabbits were sectioned serially and no evidences of gross changes were found. Smear preparations were made from the cut surfaces and while a few bodies suggestive of cysts were seen no definitely motile amoeba were present. It is remarkable that the presence of the inoculum in the peritoneal cavity provoked at all times a reaction and in some instances caused death, whereas the bulk of such material introduced intravenously produced little if any response when carried to the liver by the portal system. This serves as additional evidence of the defensive mechanism of the liver structure to invading microorganisms.

Series 2.—Seven rabbits were injected directly into the liver substances. In 2 of these, the needle was introduced from without the body into the liver and in the other 5, the animal was laparotomized and the needle introduced directly into the liver structure. One of

³ Friedrichs, A. V., and Harris, W. H., *PROC. SOC. EXP. BIOL. AND MED.*, in press.

the animals injected through the wall died after 3 days and a peritonitis was found in the upper portion of the peritoneal cavity. The remaining 6 were killed after 2 to 6 weeks and again evidences of local reaction due to the injected material were found over the surface of the liver about the point of inoculation. Sections of these livers revealed no localized abscess formations, and smears and cultures for amoeba were negative.

Series 3.—Four rabbits were inoculated into the marginal ear vein with a culture suspension containing numerous amoebae and the accompanying microorganisms. Two of these died in 3 to 6 hours and 2 survived and were eventually killed. Smear preparations were made from the lungs, heart, liver and spleen of all animals and in no instance could the amoeba be found.

Warthin⁴ states that spermatozoa were found within the amoeba in human testes. It was thought therefore that in this organ lesions might be formed in which the amoeba would flourish *in vivo*.

Series 4.—Six rabbits were inoculated into the testis with an amoebic culture suspension. A very marked acute inflammatory reaction followed, due, no doubt, to the bacteria present in the suspension. Considerable local tumefaction occurred for all animals and 2 died in 10 days and 2 weeks respectively from a septicemia. Three died within 2 months and one still survives 4 months after the inoculation and presents a marked tumefaction of the testis and scrotum. Specimens of fluid were aspirated from the inflammatory masses either with a hypodermic syringe or a glass capillary pipette with rubber teat attached. These examinations have been made frequently and at repeated intervals but in no instance were actively motile amoeba found. At certain times when encysted amoeba were found cultures were prepared but no growth of amoeba was obtained.

Although freshly isolated cultures rich in *Entamoeba histolytica* were introduced directly into the liver tissue and also into this organ through the portal system, in no instance did this parasite produce a lesion. It is possible that the bacteria present provoked a transitory response on the part of the tissues causing destruction of the amoeba. It might also be a fact that the animal chosen, namely the rabbit, is not susceptible to this infection.

⁴ Warthin, A. S., *J. Infect. Dis.*, 1922, xxx, 559.