Examination of these cysticerci proved that they were fully mature. Experimental human infection did not seem desirable or necessary, but several of the larva were fed to each of 2 laboratory monkeys. More than 3 months later the monkeys were still negative. There is therefore no evidence that any primate other than man can serve as definite host of this tapeworm.

The experimental proof of the life cycle of *Taenia confusa* directs attention to the undoubted existence of this species of tapeworm in the Southern and Southwestern United States, both in the adult state as a human parasite and in the larval stage in cattle.

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Effects of Bacteriophage Upon Cultures of Entamoeba Histolytica.*

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The cultivation of *Entamoeba histolytica* has been made of practical and almost routine laboratory usefulness through the Boeck and Drbohlav[1] method of procuring cultures and the modification of Dobell and Laidlaw[2] has enhanced its success. However, the maintenance of continuous generations is often impaired and the strains frequently lost through the concurrent bacterial growth. As Dobell and Laidlaw have pointed out, the conjoint protozoal and schizomycetic microorganisms may attain an adaptation or adjustment that makes the amoebic growth luxuriant and the propagation of the culture simple. However, in continuing strains through many generations, the contamination or bacterial overgrowth feature is a most annoying factor and often threatens to destroy or actually causes the loss of the Entamoeba culture. As far as known the cultures of amoeba obtained by this method are always accompanied by bacterial growth and these accompanying microorganisms are regarded by many as obligatory commensals or symbionts to the protozoal culture. Many efforts have been made, however, to reduce the bacterial contamination both as regards numbers and species. Since nearly

* I wish to extend my thanks to Dr. W. H. Harris for his assistance and the suggestion of the problem.

[1] Boeck, W. C., and Drbohlav, J., *Am. J. Hyg.*, 1925, v, 371.

[2] Dobell, C., and Laidlaw, P. P., *Parasitol.*, 1926, xviii, 283.

all cultures have been obtained from the intestinal lesions, the bacterial accompaniment is naturally extensive.

Dobell and Laidlaw's modification of cultivation includes a weak solution of Acriflavine for the purpose of inhibiting overgrowth of bacteria especially of the starch splitting variety. This chemical must, however, be used in very weak strengths as otherwise it will destroy the amoeba themselves. In our hands the bacterial growth was only slightly influenced by these weak solutions, although the rice starch was better preserved.

In the present experiments the employment of bacteriophage in conjunction with the cultivation of *Entamoeba histolytica* was carried out with the view to inhibiting the bacterial growth occurring in the supernatant fluid medium. The culture of amoeba was obtained by Harris and Friedrichs[3] from an amoebic abscess of the liver procured at autopsy. It was thought that this culture would contain bacteria consisting only of the colon group; it was, however, found that many Gram positive microorganisms were also present.

In the first experiments, a race of phage was used, obtained several years previously from Yale. As this material was of rather low titer and could not be exalted by repeated passage, it was later abandoned. A sample of sewage was then collected from the main sewer of the Veteran's Hospital and passed through a Seitz-Wertz filter. From this was procured a bacteriophage with a final potency of one to ten billion for strains of both *B. coli communior* and *communis*. While this phage was highly potent for the laboratory stock cultures employed, it acted only feebly upon the colon strains grown from the culture of Entamoeba.

Numerous experiments were carried out employing this colon phage in conjunction with the entamoeba cultures. The phage was introduced in varying amounts at the time of transplanting the amoebic cultures. As a whole the results indicated that definite inhibition of bacterial growth was often demonstrated for a short period of time, but this was only transitory. The recurrent bacterial growth showed masses of gram positive microorganisms as well as some gram negative bacilli. It was, therefore, found that no clear cut or consistent phagic action could be obtained with this particular bacterial flora.

An attempt was next made to supplant the microorganisms present in the supernatant fluid of the medium with the strains of *B. coli*

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

upon which the phage employed had proven highly effective. Heavy suspensions of these colon strains were inoculated into the supernatant fluid at the time of transplanting the amoeba, hoping through this method to crowd out those present in the primary culture and thereby procure a flora vulnerable to the phage. Very little, if any, better results were obtained and apparently the original microorganisms could not be displaced.

The employment of bacteriophage appears desirable in reducing the bacterial growth present in amoebic cultures. While certain bacteria, especially *B. coli,* may be essential in the *in vitro* growth of *Entamoeba histolytica* it is likely that the phaged microorganisms may likewise supply the necessary food factor. Again if whole bodies of microorganisms are needed, killed cultures could be added. The only mitigating factor that is therefore to be considered, is whether or not the activities of the bacteria themselves are obligatory.

While it is true that for practical laboratory purposes the presence of a heavy bacterial growth in amoebic cultures is of no especial importance, from the experimental standpoint a pure culture of amoeba or one containing a minimum bacterial content is greatly to be desired. It is likely that such cultures will prove more easy to propagate.

Though varying amounts of a bacteriophage of considerable potency were introduced into the amoebic cultures, in no instance was evidence of interference with this protozoal growth manifested.