lowing experiments dealing with the estimation of the pH of the Rous chicken sarcoma No. 1 are given.

pH OF ROUS CHICKEN SARCOMA

Subcutaneous tumor No. 1		Subcutaneous tumor No. 2		Large intramuscular tumor No. 1	
Minutes	pH	Minutes	pH	Minutes	pH
1	6.76	3	6.89	2	6.42
3	6.76	5	6.87	4	6.49
5	6.73	7	6.85	5	6.47
8	6.76	10	6.84	9	6.42
11	6.76	16	6.82	13	6.32
13	6.78	30	6.79	16	6.32
		61	6.79	18	6.32
		95	6.80		
		125	6.82		

These results indicate that the Rous chicken sarcoma in the living animal is characterized by an extremely low pH, as compared with that of blood. Work with the Jensen rat sarcoma and the Walker rat carcinoma 256 has also revealed pH values considerably on the acid side of 7. These very significant observations have led us to a systematic investigation of the pH of a great variety of normal and malignant tissues in the living animal.

Since October, 1931, Dr. R. H. Fitch has joined us in this work and we are indebted to him for assistance in the final development of this new method.

CARL VOEGTLIN
H. KAHLER

NATIONAL INSTITUTE OF HEALTH,
U. S. PUBLIC HEALTH SERVICE

A CULTURE MEDIUM FOR PARAMECIUM

THE medium set forth in this article has proven itself to be very satisfactory in the culturing of various species of Paramecium in pure line cultures. The main result of the use of the medium is that the organisms do not exhibit a lowering of their normal metabolism after continuous culturing.

The basic part of the medium is the usual hay infusion of ten grams of chopped timothy hay boiled for fifteen minutes in a liter of well water. This infusion is filtered and sterilized in the Arnold sterilizer at 100° C. one hour a day for three days. It is diluted with nine volumes of sterile well water just before using. Two portions of this infusion are placed in sterile liter flasks with sterile cotton stoppers. One liter is inoculated with *Bacillus subtilis* and the second with *B. coli communis*. A third portion of the medium is made up as follows: Approximately thirty grains of wheat are boiled in a small amount of water for ten minutes. The wheat grains only are then placed in a third liter flask of sterile well water. The three portions are incubated at 37° C. for twenty-four hours, and then combined in one large sterile flask. The medium is now ready for use. The cultures may be used in almost any size of container, but that used has been the three hundred cc Erlenmeyer flask. These flasks are fitted with cotton stoppers and sterilized. Each flask is filled about two thirds full of the medium, and different species of Paramecium are transferred to the cultures with sterile pipettes.

The original basic infusion may be made up, sterilized and stored in a refrigerator until ready for use. Likewise, the medium, made of the three portions, may be stored in a refrigerator for later use.

Sterile precautions are maintained throughout the procedure, but after Paramecium has been transplanted, such strict precautions are no longer necessary. The essential part of the process is to provide a medium rich with a suitable food in which Paramecium will continue to grow normally. With these sterile precautions, other ciliates and flagellates are eliminated. A single organism placed in such a medium will produce a flourishing culture in seven to ten days. One should transplant every two to three weeks. *Paramecium multimicronucleatum, P. bursaria* and *P. aurelia* have been thriving for eight months in this medium.

LAUREN E. ROSENBERG

ZOOLOGY LABORATORIES
UNIVERSITY OF CALIFORNIA
BERKELEY

SPECIAL ARTICLES

SIZE OF INFECTION AS AN INFLUENCE ON THE PERSISTENCE OF ADULT TRICHINAE IN RATS[1]

THE statements that can be found in text-books and published work on trichiniasis regarding the length of life of adult *Trichinella spiralis* are usually quite vague and indicate that wide variations may occur. The usual statement is that the adult worms live for several weeks or longer. Ransom[2] says that adult trichinae of both sexes have been found in the intestine as late as 12 weeks after infection, and that they may commonly be found in large numbers for as long

[1] From the laboratory of parasitology, department of bacteriology, University of Rochester, School of Medicine and Dentistry, Rochester, N. Y. This investigation was aided by a grant from the Rockefeller Foundation.

[2] B. H. Ransom, "Trichinosis," Rep. U. S. Live Stock San. Assoc., pp. 147–165, 1915.

as 7 weeks. On the other hand, Christenson[3] in a series of experimentally infected rats did not find any adults after the sixteenth day and concluded that 16 days is about the normal duration of the worms in the intestine.

In the study of a large number of rats experimentally infected with trichiniasis the writer has found that the size of infection has a great influence on the length of time that the adult worms persist in the intestine. All rats were infected by stomach tube with known numbers of larvae obtained free from infected muscle by artificial digestion. The doses were administered according to the body weight of the animals.

In light infections of 20 larvae per gram, or less, the adult worms were never found in appreciable numbers after the fifteenth day after infection. When these rats were killed after 4 weeks only full-grown larvae could be found in the muscles. On the other hand following a much larger dose (40 or 50 larvae per gram) larvae in all stages of development could nearly always be found in the muscles for as long as 4 or 5 weeks after infection. Some of them were of a size which indicated that they had but recently migrated. Upon examination of the intestines of these rats considerable numbers of adult trichinae were usually found to be still present. These observations were consistently made in a large number of rats that died from trichiniasis 4 to 6 weeks after infection.

The following table gives the results of an experiment in which rats receiving doses of 20 and 50 larvae per gram were killed at intervals up to 3 weeks after infection. Very few adult worms were found in two rats given 20 larvae per gram and killed on the fourteenth day after infection. None at all were found in two more of this group killed on the twenty-first day. On the other hand, 25 to 50 per cent. of the original number of larvae fed were still present as adult worms in the intestines of the rats given 50 larvae per gram and killed after 14 and 21 days.

The most plausible explanation of these observations seems to be that a heavy infection breaks down a resistance in the rats which ordinarily limits the duration of the adult trichinae in the intestine. The size of infection which best illustrates this effect is one which does not kill the rats until after the fourth week. If the infection is too large, the animals die within two weeks in the intestinal stage of the disease. If it is not large enough, the resistance is not broken sufficiently and the animals recover.

No doubt there are other factors, such as diet, health of the animal, etc., which might influence the resistance of the rats and the length of life of the adult trichinae. Other experiments, also, indicate that the infection behaves differently in different hosts. Apparently guinea-pigs have very little resistance against trichiniasis, for the lethal dose of larvae is much smaller than for rats and the adult worms live for as long as 5 weeks following doses as small as 5 larvae per gram. The infection in rabbits, on the other hand, apparently behaves much the same as in rats. It will be interesting to compare the parasitological course of trichiniasis in a larger series of hosts, including man.

These observations that the rat possesses a "normal resistance" against trichina infection fall in line with other experiments[4] which show that rats can develop an immunity to reinfection with *Trichinella spiralis*. They also add to the evidence that has been piling up of late that host factors can play an important rôle in determining the course of helminth infections. In any event, they offer a probable explanation of the variations in the reported length of life of adult trichinae.

O. R. McCOY

TABLE I

SURVIVAL OF ADULT TRICHINAE IN RATS GIVEN DIFFERENT SIZED DOSES OF LARVAE

20 larvae per gram	No. of larvae fed	Killed—days after infection	No. of adults in intestine	Per cent. of original dose
Rat no. 1	4,200	5 days	2,960	71
" " 2	4,140	5 "	2,920	71
" " 3	3,540	5 "	2,660	75
" " 4	3,600	14 "	30	1
" " 5	3,900	14 "	570	15
" " 6	4,920	21 "	0	0
" " 7	5,800	21 "	0	0
50 larvae per gram				
Rat no. 8	10,800	5 days	5,220	48
" " 9	9,100	5 "	4,640	51
" " 10	11,500	5 "	6,420	56
" " 11	10,400	14 "	5,700	55
" " 12	12,400	14 "	5,850	47
" " 13	9,800	21 "	4,800	49
" " 14	9,700	21 "	2,400	25

[3] R. O. Christenson, "The Sex Ratio of Adult Trichinae," SCIENCE, 66: 259, 1927.

[4] O. R. McCoy, "Immunity of Rats to Reinfection with *Trichinella spiralis*," *Am. Jour. of Hyg.*, 14: 484–494, 1931.