

Using the method employed by Kögl and Erxleben, we have isolated glutamic acid from two adenocarcinomas of the large intestine, and from a sample of normal intestinal tissue adjacent to one of the carcinomas. Our results indicate that approximately 41 per cent. of the glutamic acid in the first carcinoma, and approximately 26 per cent. of the glutamic acid in the second, were present as d(-) glutamic acid. These findings confirm the report of Kögl and Erxleben. Our experimental data are summarized below:

(1) *Adenocarcinoma of the sigmoid colon.* Initial weight, 97.5 g. Weight of dried combined protein, 14.0 g. Glutamic acid hydrochloride isolated, 97 mg. M. P., 205° C. $[\alpha]_D^{20} + 5.5^\circ$.

(2) *Adenocarcinoma of the cecum.* Initial weight, 108.5 g. Weight of dried combined protein, 15 g. Glutamic acid hydrochloride isolated, 140 mg. M. P., 201° C. $[\alpha]_D^{20} + 15.0^\circ$.

(3) *Normal tissue adjacent to adenocarcinoma of the cecum.* Initial weight, 150 g. Weight of dried combined protein, 19.1 g. Glutamic acid hydrochloride isolated, 19 mg. M. P., 205° C. $[\alpha]_D^{20} + 31.0^\circ$.

It will be noticed that the yield of glutamic acid obtained from the adenocarcinoma of the cecum was considerably greater than that obtained from adjacent normal tissue. No conclusion can be drawn from this, however, since the insolation of the glutamic acid was not quantitative.

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POLIOMYELITIC VIRUS IN SEWAGE

WITHIN the past two years it has become increasingly apparent that poliomyelitic virus may be readily isolated from the stools of human patients during acute and convalescent stages of this disease. Many reports now testify to the ease with which this can be accomplished.¹ Consequently, whenever poliomyelitis occurs within a city, there is at least an opportunity for this

virus to enter the local sewage system. But actually poliomyelitic virus has never been demonstrated in sewage, and there is no information as to the possible amounts which may be there, nor the length of time which this virus might survive in this medium.

An opportunity to test this situation has recently occurred in the City of Charleston, South Carolina, where poliomyelitis assumed epidemic proportions during the months of May, June and July of this year. From this same South Carolina epidemic a strain of virus was isolated from a specimen of feces received in our laboratory on July first. The stool was from a child (B.Cr.) three years old, who had contracted paralytic poliomyelitis in a small up-state town, but who had been hospitalized in Charleston. Part of the stool suspension was inoculated intraperitoneally into one rhesus monkey (No. 1213) on July fourth, and typical experimental poliomyelitis resulted after an incubation period of five days. This strain has been successfully carried to its third passage (monkeys No. 1230 and 1242) and is characteristic of poliomyelitic virus.

Subsequently, through the kindness of Dr. L. Banov, health officer of the City of Charleston, samples of sewage were collected by two of us (J. R. P. and J. D. T.) from a few sites throughout the city during the period of July 7 to 14, 1939. Particular attention was paid to one pumping station in which the sewage came not only from the hospital where poliomyelitis patients were isolated, but also from that part of the city where most of the cases had arisen. One sample (C) of this sewage, amounting to about 8 liters in volume, was collected on July 11 in a tall glass bottle and allowed to stand for a period of twenty-four hours. Ice was packed about the base of the bottle for part of this time. After the first two hours, a sample of sediment (C-1) amounting to 200 cc was removed and to it 24 cc of ether were added for bactericidal purposes. This sample was then taken to New Haven, where, on July 12 and 13, 75 and 45 cc, respectively, were inoculated intraperitoneally into one monkey (No. 1227). Another sample (C-2) amounting to 700 cc of the sediment, was removed from the original 8-liter specimen on July 12; 70 cc of ether were added to it, and the specimen was sent to New Haven, where one monkey (No. 1232) was inoculated intraperitoneally with 125 cc on July 14. Both of these animals developed experimental poliomyelitis. The incubation period in monkey 1227 was eight days from the last inoculation, and in monkey 1232 it was seven days. After a brief febrile period both animals developed quadriplegia, and both were prostrate on the third day of their disease. Monkey 1232 died on this day; no. 1227 was sacrificed. Spinal cord lesions, demonstrable histologically, were found to be very extensive in both animals. It is unusual in our experience to see rhesus

¹ (a) P. H. Harmon, *Jour. Am. Med. Assn.*, 109: 1061, 1937; (b) J. D. Trask, A. J. Vignee and J. R. Paul, *Jour. Am. Med. Assn.*, 111: 6-11, 1938; (c) P. Lépine and P. Sédallian, *Compt. rend. Acad. d. sci.*, 208: 129-130, 1939; (d) S. D. Kramer, B. Hoskwith and L. H. Grossman, *Jour. Exp. Med.*, 69: 49-67, 1939; (e) C. Kling, G. Olin, J. H. Magnusson and S. Gard, *Bull. Acad. méd.*, 121: 826-831, 1939; (f) J. R. Paul, A. J. Vignee and J. D. Trask, *Trans. Assn. Am. Phys.* (to be published). Also personal communications from: G. Y. McClure, Division of Laboratories and Research, N. Y. State Department of Health; and from C. Armstrong, National Institute of Health, Washington, D. C.

TABLE 1
TESTS FOR POLIOMYELITIC VIRUS IN SEWAGE SPECIMENS

Sewage specimen	Original amount	Interim between collection and inoculation	Amount of inoculum		Monkey No.	Result
			Intra-peritoneal	Subcutaneous		
(A) Pump	3.5 liters	6 and 7 days	36 cc	22 cc	1229	Abscess of abdominal wall; acute peritonitis. Killed on 4th day.
(B) Har.	0.5 liter	7 days	24 "		1231	Chronic and acute peritonitis. Died on 18th day.
(C) Pump (1)	8 liters	1 and 2 days	120 "		1227	Poliomyelitis. Killed on 10th day.
(2)		3 days	125 "		1232	Passage to monkey 1248 successful. Poliomyelitis. Died on 8th day.
(D) Hosp. (1)	7 "	5 "	75 "	125 "	1239	Passage not done. Infected abdominal wall. Died on 2nd day.
(2)		6 "	25 "	46 "	1240	Abscess of abdominal wall. Killed on 4th day.

monkeys develop such a severe form of the experimental disease when inoculated with material from a human source, and so we have suspected that the amounts of virus in the inocula here were not small.

The results of our experiments to date on this type of material appear in Table I. Here it will also be seen that our other attempts to isolate virus from Charleston sewage were unsuccessful as a result of the premature death of the monkey from bacterial infection. Obviously, methods have not yet been perfected for handling or testing this type of material.

Our criteria for the diagnosis of poliomyelitis in these experiments have been practically the same as those we have used in previous work² on the isolation of poliomyelitic virus from extra-neural human sources, namely, that the inoculated animal developed signs and symptoms compatible with those of the experimental disease; that typical lesions were found in the spinal

cord, and that the strain was successfully passed to another monkey. All these criteria were met in the case of monkey 1227; one of them is omitted in monkey 1232, for a passage of this strain was not attempted.

It is not evident from this work whether the presence of poliomyelitic virus in sewage is a direct or even an indirect link in the chain which leads this infectious agent from one patient to another in this disease. Our report merely calls attention to the fact that poliomyelitic virus may not only be present in urban sewage but also that it may possibly be present in appreciable quantities.³

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

DEMONSTRATION OF THE SHAPE OF CILIA IN NORMAL MOTION

It is often desirable to demonstrate the shape and motion of cilia without the use of methods which may cause distortion or prevent an estimation of the rate of beating. An easily made stroboscope meets this need.

To make a periodically moving object like a cilium appear stationary, it must be made visible repeatedly at only one point in its path. This can be achieved by a light flashing on the object at exactly the same frequency as its motion. If the synchronization is not quite perfect, the cilium will appear to be moving very slowly because each flash of light will reveal it at a slightly different position. Should the synchronization be much poorer, the entire effect will be lost. These principles constitute the basis of the stroboscope.

An obvious means of producing a flashing light is to have a disk with a number of regularly spaced radial

slits rotating in front of a fairly strong light source. Four slits have been found most suitable. In projection, it makes little difference where the disk is located; for convenience it may be placed between the ocular and the screen.

A satisfactory method for the adjustment of the disk's speed is an important element in the success of the stroboscope. A rheostat in series with the motor attached to the disk is adequate; however, finer adjustment is obtainable with two rheostats in series if the resistance per unit length of one is considerably larger than the other's. In the way of a motor, even those obtainable in metal construction kits are usable. A possible arrangement of the apparatus is shown in the diagram.

The rate of ciliary motion can easily be determined from the disk's speed when synchronization, as judged from the cessation of movement, is attained. In general, it will be found a simple matter to count the

² Criteria reviewed by A. J. Vignec, J. R. Paul and J. D. Trask, *Yale Jour. Biol. and Med.*, 11: 15-31, 1938.

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