subjected in nature. In order to study this question, oil was prepared from the livers of puffer fish which had been maintained in an aquarium in a tank for a period of 3 months. The fish were of the same batch which had been previously tested. During this period of captivity no active ultra-violet radiations had access to the fish, as the windows of the building were closed at all times. In spite of the fact that the fish were deprived for 3 months of activating rays, quantitative tests showed that their livers contained oil equal in potency to that which had been prepared shortly after they had been removed from the sea. The diet of these fish consisted largely of herring, which is very rich in antirachitic substance.

That the oil in the livers had been elaborated during the period of observation and was not merely residual oil which had been in the liver previous to captivity, is demonstrated by the fact that the livers became exceptionally rich in oil during the time the fish were deprived of ultra-violet rays. The average amount of oil which could be extracted initially was less than 5 per cent, whereas three months later 55 per cent of oil was extracted.

From this experiment we infer that the antirachitic factor can be elaborated in undiminished potency by fish deprived of sunlight for long periods, and that this characteristic quality of fish liver oil is dependent on the diet of the fish, rather than on the action of ultra-violet radiations.

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Action of ultra violet light upon bacteriophage and filterable viruses.

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The theories concerning the nature of bacteriophage are well known. D'Herelle[1] has maintained from the very beginning that

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[1] d'Herelle, F., The Bacteriophage, Fr. Edition, Oct., 1921; Eng. Edition, March, 1922.

the bacteriophage is a living ultramicroscopic virus. In this contention he has been opposed chiefly by the Bordet school, who consider the bacteriophage an inanimate agent which is developed by the microbe as a result of a reaction between the invading organism and the tissue cells of the host; that by the hereditary vitiation of the organism a lytic principle is developed.

Recently one of us has brought forward further evidence[2] in support of the Bordet conception and the lytic principle we have employed in these experiments is fully described in these publications.

This principle is markedly lytic for a strain of *B. coli* which we have designated *B. coli* "D" because of its diffuse growth in broth as contrasted with another strain which agglutinates spontaneously. Various dilutions of this bacteriophage were made in measured quantities of beef extract broth of pH 7.6. The materials to be exposed to the ultra violet light were placed in quartz test tubes which were obtained from the General Electric Co. To some of the tubes three to ten drops of a twenty-four hour broth culture of *B. coli* "D" were added. Other tubes contained both culture and lytic principle in amounts varying from three to ten drops. All of the tubes contained exactly 5 cc. of broth. The experiments were further controlled by exposing the medium alone to the action of ultra violet light before organisms or lytic principle were added. (Table 1.)

TABLE 1.

Tube containing 5 cc. broth	24-hour culture *B. coli* "D"	Lytic Principle	Exposure to ultra violet light at 1 ft. distance	Test for Lytic Principle.		
				Exp. 1.	Exp. 2.	Exp. 3.
I	3 drops	0 drops	40 min	—	—	—
II	6 "	0 "	40 "	—	—	—
III	10 "	0 "	40 "	—	—	—
IV	3 "	3 "	40 "	—	—	—
V	6 "	6 "	40 "	—	—	—
VI	10 "	10 "	40 "	—	+	—
VII	*3 "	10 "	40 "	—		
VII	†3 "	10 "	40 "		+	+
VIII	3 "	0 "	40 "	—	—	—
IX	3 "	3 "	0 "	+	+	+
X	†3 "	†3 "	40 "		+	+

[2] McKinley, E. B., *C. R. de la Soc. de Biol.*, 1925, xciii, 1050-2.

*Introduced 18 hours after irradiation.

†Introduced immediately after irradiation.

All tubes, except the lytic principle control tube, were exposed to ultra violet light radiation as projected from the Alpine Sun Lamp,† of 4.5 amperes at a distance of one foot for forty minutes. The apparatus was allowed to heat for five minutes before the exposures were begun. The quartz tubes were laid in open Petri dishes at an angle of about 5 degrees and their contents agitated every eight minutes during the exposure. During exposure the temperature did not exceed 30 degrees C. After the exposure to ultra-violet light the tubes were incubated at 37.5 degrees C. for twenty-four hours. The cultures were then tested for the presence or absence of lytic principle by a simple method which will be described in another publication.

It will be noted in Table 1 that in three experiments the three and six drop quantities of lytic principle were effectively destroyed; that in two experiments under the conditions of the experiment, ten drop quantities of the bacteriophage were destroyed. Exposure of *B. coli* "D" to the ultra violet light under conditions of this experiment, results in no apparent effect upon this organism. These conditions are quite the reverse with other physical agents, for example heating to 56.5 degrees C. At this temperature *B. coli* "D" is killed but there is no action upon the lytic principle.

Experiments were also made with regard to the action of ultra violet light, under precisely the same conditions we employed with bacteriophage upon herpes virus and Levatidi's so-called encephalitis virus. In former experiments, by repeated passage from one animal to another, we had obtained a herpes virus (originally isolated from a case of *Herpes genitalis* by LeFèvre de Arric) which, when injected into rabbits intracerebrally or into the spinal canal in .2 cc. amounts of a 1:100,000 dilution would produce Herpetic encephalitis and death of the animal always within five days and usually within three days. The Levatidi so-called encephalitis virus, after repeated passage through many animals, obtained a fixed virulence, when .2 cc. of a 1:10,000 dilution of brain emulsion was employed, of five or six days. In both instances the original brain emulsion employed consisted approximately of one gram of brain emulsified in 2 cc. of physiological salt solution.

In these experiments the original brain emulsions were not

† We are indebted to Dr. A. F. Hess for the use of his Alpine Sun Lamp.

diluted. The concentrated emulsions were centrifuged at first to throw down any tissue particles which were present and the supernatants withdrawn for our experiments. Emulsions were made in both normal rabbit serum and in physiological salt solution. Two cc. of the supernatant containing each virus in serum and salt solution were placed in quartz tubes. These tubes were exposed to ultra violet light radiation in the same manner as were the tubes containing bacteriophage, for forty minutes at a distance of one foot, the tubes being agitated every eight minutes. After exposure of the viruses to ultra violet light, rabbits were injected subdurally with 0.2 cc. of the material from each tube. Control animals were injected with the untreated supernatants. (Table 2.)

TABLE 2.

Date	Rabbit No.	Brain emulsion tested	Amt. injected	Time exposed to ultra violet light	Date of Death	No. of days	Date Living	No. of days	Remarks
				minutes					
Experiment 1.									
1/27/26	2549	H2548 Serum	0.2cc.	40	2/2/26	6	2/26/26	30	
"	2550	H2548 Saline	"	40					
"	2551	E2547 Serum	"	40	2/1/26	5			
"	2552	E2547 Saline	"	40	2/18/26	22			
"	2553	H2548 Saline	"	0	1/31/26	3			Control
"	2554	H2548 Serum	"	0	1/31/26	3			"
"	2555	E2547 Saline	"	0	2/1/26	5			"
"	2556	E2547 Serum	"	0	2/2/26	6			"
Experiment 2.									
2/6/26	2565	H2554 Saline	"	0	2/11/26	5	2/26/26	20	Control
"	2566	"	"	20			"	20	
"	2567	"	"	20			"	20	
"	2568	"	"	40			"	20	
"	2569	"	"	40			"	20	

H = Herpes virus.
E = Levaditi encephalitis virus

It will be noted that the control animals died within the specified time in experiments one and two; that the animals which received herpes and so-called encephalitis virus emulsified in serum and exposed to ultra violet light died in six and five days respectively; that the animal which received herpes virus emulsified in saline and exposed to ultra violet light was living and apparently normal thirty days after injection, while the rabbit which received the Levaditi virus which had been emulsified in salt solution and exposed to the ultra violet light, died twenty-two days after injection, as contrasted with five days for the control.

In Experiment 2 only the herpes virus was exposed to the action of ultra violet light and the emulsions were in saline. The control animal in this case died on the fifth day, while the animals receiving herpes virus which had been exposed to ultra violet light for twenty and forty minutes (the other conditions remaining the same as in the previous experiment) were living and apparently normal on the 20th day following injection.

While the series of animals in these two experiments is small, it is suggested that ultra violet light under the conditions we have worked is capable at least of attenuating both herpes virus and the Levatidi virus, and possibly destroys them completely under certain conditions. Further experiments are now in progress bearing on this problem. Since both herpes and Levaditi virus in serum brain emulsions are apparently protected from the ultra violet light, we thought it of interest to test bacteriophage under the same conditions. Experiments show that bacteriophage is also protected when serum is substituted for broth. The three, six and ten drop dilutions all remain active after exposure to the ultra violet light. Increase in density of the substrate may explain this.

Conclusions. One may say that under the conditions of these experiments, a lytic principle active for *B. coli* "D" is acted upon by ultra violet light in much the same way as are two strains of known filterable viruses, *i. e.,* herpes and Levaditi's so-called encephalitis virus. Exposure to ultra violet light at a distance of one foot for forty minutes is sufficient to attenuate or destroy both the bacteriophage and the two filterable viruses employed in these experiments. In a substrate of normal rabbit serum all three are protected from the action of ultra violet light.