9492

## Effect of Ultraviolet Irradiation on Poliomyelitis Virus in Vitro.*

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The experimenter who is confronted with the problem of natural resistance to poliomyelitis finds two avenues open for methodical approach. He can either investigate the agencies which are liable to cause changes in virulence of the etiological agent or he can attempt to study and correlate the different factors which modify susceptibility of the host. While there are reasons to suspect that host factors are more important in determining the course of events, the environmental conditions which may affect the virulence of the virus obviously lend themselves more readily to experimental analysis. It is a simple matter, for instance, to expose the virus to various physical and chemical agencies and to test for its survival or destruction by intracerebral injection into monkeys. Such studies have already contributed valuable data that are helpful in explaining the nature of the infectious agent. Thus, it has been shown that poliomyelitis virus possesses a very low thermal deathpoint[1] and that it is extraordinarily susceptible to the oxidizing or reducing action of certain chemicals.[2] On the other hand, the virus has proved quite resistant to exposure, to freezing,[3] drying,[3] X-rays,[4] as well as sonic vibrations,[5] and remains relatively unharmed after contact with strong protoplasmic poisons.[6] Judging from these properties, the virus of poliomyelitis seems to resemble much more the toxins and ferments than the common animate agents of disease.

The literature offers very little information concerning the effect

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1 Shaughnessy, H. J., Harmon, P. H., and Gordon, F. B., *J. Prev. Med.*, 1930, **4**, 149.

2 (a) Jungeblut, C. W., *J. Exp. Med.*, 1935, **62**, 517; (b) *Poliomyelitis Monograph*, International Committee for the Study of Infantile Paralysis, Baltimore, Williams and Wilkins Co., 1932.

3 See 2b.

4 Lenz, M., and Jungeblut, C. W., PROC. SOC. EXP. BIOL. AND MED., 1932, **29**, 814.

5 Scherp, H. W., and Chambers, L. A., PROC. SOC. EXP. BIOL. AND MED., 1936, **35**, 495.

6 Flexner, S., Clark, P. F., and Amoss, H. L., *J. Exp. Med.*, 1914, **19**, 205. See also 2b.

of ultraviolet light on the virus of poliomyelitis *in vitro,* although the light sensitiveness of other filtrable viruses, toxins and ferments has been well established. According to Flexner,[7] poliomyelitis virus is rapidly killed by sunlight, and Amoss[8] states that light in the presence of eosin destroys the virus. Schultz[9] found that exposure to ultraviolet rays from a cold quartz lamp inactivates the virus in less than 3 minutes. It was, therefore, desirable to add to our knowledge on the properties of the virus by investigating in detailed experiments the influence of ultraviolet irradiation on poliomyelitis virus *in vitro.*

The experimental setup was as follows: Amounts of 2 cc. of a 1:20 dilution of a 10% centrifuged virus suspension (Aycock strain) were placed in rock crystal vessels which were covered with a lid made of the same material. This amount of virus was chosen as representing a dose which, while not excessively large, has proved to be sufficiently infective to bring down 95% of the controls with paralysis in routine intracerebral tests. These vessels were then exposed to ultraviolet irradiation for various lengths of time (5 seconds to 30 minutes), at a distance of about 25 cm. from the quartz burner. An ultraviolet lamp, energized by high frequency currents (Lepel) and emitting a spectrum in which the longer ultraviolet waves in the range from 2800 to 3100 Angstrom units predominate, was used for this purpose. The temperature was carefully checked

TABLE I.
Effect of Ultraviolet Irradiation on Poliomyelitis Virus *in vitro.*

| Exposure of virus | Test Monkey intracerebral injection (1 cc. 1:20 dil. 10% virus) Rayed | Result | Control Monkey intracerebral injection (1 cc. 1:20 dil. 10% virus) Unrayed | Paralysis days |
|---|---|---|---|---|
| 30 min. | W95 | No paralysis | W93 | 6 |
| " " | Y98 | " " | Y73 | 9 |
| 15 " | X27 | " " | X23 | 8 |
| " " | Y99 | " " | Y73 | 9 |
| 5 " | X61 | " " | X60 | 11 |
| " " | Y100 | " " | Y74 | 9 |
| 2 " | Y5 | " " | Y6 | 12 |
| " " | Z1 | " " | Y75 | 13 |
| 1 " | Z2 | " " | Y76 | 11 |
| " " | Z3 | " " | Y77 | 7 |
| 5 sec. | Z4 | Paresis, 15 days | Y76 | 11 |
| " " | Z5 | Paralysis, 14 days | Y77 | 7 |

[7] Flexner, S., Rockefeller Institute pamphlet, 1916.

[8] Rivers, T. M., *et al., Filtrable Viruses,* Baltimore, Williams and Wilkins Co., 1928.

[9] Schultz, E. W., *J. Ped.,* 1932, **1**, 358.

in the rayed fluid during the experiment and found not to exceed 38.5°C. after one hour exposure. It is important to mention this point since poliomyelitis virus is attenuated or killed by heating for 30 minutes between 42.5 and 50°C. After raying, one cc. of the virus suspension was injected intracerebrally into monkeys. Duplicate tests were done at all intervals and each experiment was accompanied by control animals injected with one cc. of the unrayed virus suspension. The results obtained are given in Table I.

It may be seen from Table I that raying for periods of time varying from 30 minutes to as little as one minute was evidently sufficient to destroy the virus completely. Even an exposure as short as 5 seconds had possibly caused a certain degree of attenuation of the virus as suggested by the slight paresis developing in one of the 2 monkeys which had received this material.

It follows from these experiments that poliomyelitis virus *in vitro* is incredibly sensitive to the effect of ultraviolet irradiation. The mechanism of this inactivation is unknown, but it is more than likely that oxidation plays an important part. In applying this observation to the epidemiology of the human disease caution is necessary since one is dealing with a complex problem involving operation of the same factor on the virus as well as the host—and the effects produced may not be the same. It is conceivable that exposure to ultraviolet light, under certain conditions, may tend to increase the susceptibility of the host while at the same time causing a diminution in the virulence of the virus.

After completion of this work, a paper by Toomey[10] appeared, who found in repeated tests that exposure for 75 minutes at 18 inches distance from a standard quartz lamp bulb inactivated a 1% suspension of poliomyelitis virus. No shorter periods of exposure were tested.

## 9493

## An Apparatus for Grinding Bacteria at Low Temperatures.

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The disrupting of bacteria by grinding in mills is a well established laboratory procedure, for which various forms of apparatus have been devised. The long grinding required to break most of the bac-

[10] Toomey, J. A., *Am. J. Dis. Child.*, 1937, **53**, 1490.