made to use it to oxidize dextrose under the same conditions. The attempts were without success even when air or oxygen was passed through the solution. It was observed, however, that the pH of the solution was lowered, which indicated that some oxidation had taken place, presumably in the phospholipid. The amount of oxidation was not great. Oxidation of phospholipid was then attempted with hydrogen peroxide at 37° C. and pH 7.4. Purified brain lecithin and cephalin were suspended in neutralized 30 per cent. hydrogen peroxide and kept at 37° C. for 24 to 48 hours. The solutions were then extracted with ether and the extracts analyzed. It was found that under these conditions about 75 per cent. of the lecithin had disappeared but only about 25 per cent. of the cephalin. The residue from the cephalin consisted largely of insoluble material, while that from the lecithin resembled the original lecithin, although the iodine numbers of the fatty acids had been greatly reduced. The products of the oxidation consisted largely of carbon dioxide and volatile acids the nature of which has not yet been determined.

*A chemical study of the balance sheet of fat absorption in the rat:* MATHIAS F. F. KOHL (introduced by D. D. Van Slyke). Balance sheet studies on the absorption of elaidic acid, the stereo-isomer of oleic acid, have been carried out on male rats weighing approximately 100 grams. The studies were made over hourly and daily absorption periods using two dietary régimes, the feeding of elaidin as the sole food source and as 40 per cent. of the caloric intake. The rates of absorption and disappearance of the absorbed elaidic acid are different under the two régimes but are found to be relatively continuous and constant. When elaidin alone is fed, most of the absorbed fat is apparently consumed in the vital processes of the organism with the deposition of only a small part of the absorbed fat. When ample protein and carbohydrate are supplied along with the elaidin, the amount of the absorbed fat disappearing is markedly reduced with a consequent greater storage of ingested fat. Elaidin when once deposited in the adipose tissue disappears at a slow rate; over thirty days are required to clear the depots of the elaidic acid that is deposited when elaidin is fed over a three-day period as 40 per cent. of the caloric intake. The conclusion is drawn that the elaidin that is continually being absorbed from the intestine is used to supply the energy requirements of the tissues for fat. This demand is reduced by an ample supply of protein and sugar which the cells catabolize preferentially to fat. The absorbed fat in excess of this cellular demand for fat is removed from the circulation into the fat depots, where it remains as a relatively inert deposit until periods of under-nutrition, when it is called forth again to supply the metabolic needs. Definite regulatory mechanisms of fat metabolism are in evidence, but nothing is known of their nature.

*Experimental diabetes insipidus:* ROWLAND T. BELLOWS and WILLIAM P. VAN WAGENEN (introduced by Harvey Cushing). Diabetes insipidus is generally considered to be an abnormally increased secretion of urine. There are other phenomena observed in the disease, however, which have been largely ignored. These suggest that it is a fundamental disorder in the water metabolism, in which the abnormally increased thirst, or polydipsia, is of prime consideration. In the experimentally produced disease in dogs the water intake appears to govern the amount of urinary output, and when the water intake is restricted the animals appear to remain in water balance. Dogs with diabetes insipidus gain weight and become obese when their thirst is satisfied, but fail to do so when the water intake is restricted to the normal amount. When diabetes insipidus is produced in dogs with fistulae of the esophagus, it is found that the animals continue to be thirsty, even though they are maintained in water balance by the administration of water per esophagus. These considerations suggest that the excessive thirst plays the leading rôle in diabetes insipidus rather than the polyuria and that the satisfaction of the polydipsia leads to the chronic retention of water. Two forms of diabetes insipidus are generally recognized—temporary and permanent. Our studies reveal a peculiar relationship between them. They appear to possess different characteristics than mere duration and to depend upon a different type of injury in the pituitary region for their production.

*Biographical Memoir of Edward Salisbury Dana:* ADOLPH KNOPF. (Read by title.)

*Biographical Memoir of Edwin Brant Frost:* OTTO STRUVE. (Read by title.)

*Biographical Memoir of Robert Simpson Woodward:* F. E. WRIGHT. (Read by title.)

# SPECIAL ARTICLES

## ANTIRABIC IMMUNIZATION WITH CULTURE VIRUS RENDERED AVIRULENT BY ULTRA-VIOLET LIGHT

PREVIOUS investigators have shown that rabies and poliomyelitis viruses, among others, may be rendered avirulent by ultra-violet light.[1] But reports in the literature indicate that a virus thus made avirulent is unable to immunize animals against a test inoculation; according to these experiments, immunizing potency is retained only when virulence has not been completely destroyed.

Evidence is presented in this communication showing that rabies virus may be exposed to ultra-violet

[1] G. Sankaran and W. A. Beer, *Indian Jour. Med. Res.*, 22: 581, 1935; John E. Gordon and Thomas P. Hughes, *Jour. Immunol.*, 30: 221, 1936; J. A. Toomey, *Amer. Jour. Dis. Child.*, 53: 1490, 1937.

light in such a way that its virulence may be destroyed without complete loss of its immunizing power.

Thirty cc of clear rabies culture virus,[2] containing 33,000 mouse intracerebral lethal doses per cc, were placed in a quartz flask 12.5 cm from a quartz mercury vapor lamp. The intensity of the ultra-violet light was measured by means of a photronic cell and filter. During the period of irradiation the temperature of the virus was kept at 23° C.

The virus was irradiated 45 to 60 minutes and then tested for virulence by injecting 0.03 cc intracerebrally into each of eight or ten mice. The immunizing potency of the irradiated virus was determined[3, 4] by vaccinating mice each with 0.25 cc intraperitoneally. In all, six vaccinating doses were given, one every other day. About 3 weeks later the immunity of the vaccinated mice was tested by injecting each of them intracerebrally with 0.03 cc of mouse-brain rabies fatal dose of test virus, as contrasted with only one of 19 vaccinated mice; 12 of 12 unvaccinated mice died after inoculation with 10 fatal doses, as contrasted with 3 of 11 vaccinated mice; and 8 of 8 unvaccinated mice succumbed to 100 fatal doses, as contrasted with 2 of 8 vaccinated animals. These differences are significant according to the $\chi^2$ test, $P = < .01$.

Further experiments showed that culture virus irradiated only 30 minutes immunized well but remained virulent to a slight degree, while virus irradiated 2 hours became inert both as to immunizing potency and virulence. Finally, culture virus in a glass flask, wrapped with tinfoil, which is impervious to ultra-violet light, and exposed to the mercury vapor lamp for 2 hours, showed no loss of virulence.

It is concluded, therefore, that rabies culture virus, exposed to a proper dose of ultra-violet light, becomes avirulent and yet retains enough of its immunizing power to protect mice against 10 intracerebral lethal doses of test virus.

TABLE I
IMMUNIZING POTENCY OF IRRADIATED, NON-VIRULENT RABIES CULTURE VIRUS

| Treatment of culture virus | Virulence following irradiation | Mice | Fate of mice receiving test virus: Dilutions of virus $10^{-4}$ | $10^{-5}$ | $10^{-6}$ | $10^{-7}$ | Protection afforded by vaccination in minimal lethal doses |
|---|---|---|---|---|---|---|---|
| Test 3 | | | | | | | |
| Irradiated | | Controls | ... | 4/4† | 2/4 | 1/4 | |
| 60 minutes | None 0/8* | Vaccinated | ... | 0/3 | 0/3 | ... | 10 |
| Test 4 | | | | | | | |
| Irradiated | | Controls | 4/4 | 7/13 | 1/4 | ... | |
| 45 minutes | None 0/8 | Vaccinated | 0/4 | 1/8 | ... | ... | 10 |
| Test 5 | | | | | | | |
| Irradiated | | Controls | 4/4 | 4/4 | 3/4 | 1/4 | |
| 45 minutes | None 0/10 | Vaccinated | 1/4 | 2/4 | 0/4 | 0/2 | 10 |
| 50 minutes | None 0/10 | Vaccinated | 1/4 | 1/4 | 0/4 | 0/4 | 100 |

* 0 of 8 injected mice succumbed.
† 4 of 4 injected mice succumbed.

virus. Some received 1,000 lethal doses, some 100, some 10 and some one lethal dose of the test virus. Unvaccinated mice of the same age received like amounts of the virus, respectively.

These experiments (Table I) show that the irradiated, avirulent culture virus immunizes against 10 or more intracerebral lethal doses of test virus. In each experiment, taking one fatal dose as the greatest dilution killing 50 per cent. or more of the unvaccinated mice, it appears that 10 to 100 times this dose was withstood by 50 per cent. or more of the vaccinated mice. Combining results of the three tests, it is noted that 12 of 21 unvaccinated mice succumbed to one

[2] L. T. Webster and A. D. Clow, *Jour. Exp. Med.*, 66: 125, 1937.

[3] L. T. Webster, *Amer. Jour. Pub. Health*, 26: 1207, 1936.

[4] L. T. Webster, *Amer. Jour. Pub. Health*, 1937 (in press).

The technique described above is being further developed and applied.

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## CHANGES IN HUMAN BRAIN POTENTIALS DURING THE ONSET OF SLEEP

WHEN a person goes to sleep, the pattern of his brain potentials alters systematically. Five clearly defined stages have already been described[1] as follows:

A—*alpha:* the normal waking 10-per-second rhythm
B—*low voltage:* the alpha rhythm is lost
C—*spindles:* short groups of 14-per-second waves ap-

[1] A. L. Loomis, E. N. Harvey and G. Hobart, *Jour. Exp. Psychol.*, 21: 127, 1937.