ened, but in very young animals delayed gasps of reduced amplitude and increased rate occasionally occur.

Morphine tends to obliterate the interval between the first and second series and to reduce slightly the frequency; however, the total duration of gasping, as well as the total number of gasps, is usually greatly increased by this drug.

In very young animals (10 days or less) having a long second series, the frequency of gasping is increased by warming the isolated head and decreased by cooling; the duration of gasping is reduced by warming, increased by cooling. The total number of gasps however, appears to be little altered by temperature unless extreme ranges are employed.

If the area over the medulla and cerebellum of an isolated head of a young animal is quickly cooled by ethyl chloride or by immersion in cold Ringer's, gasping ceases. Upon warming, gasping is resumed and a more or less normal series ensues. Heads of 1 to 4-day-old rats have been revived after more than 30 minutes of cooling.

TABLE I.
Mandibular Movements of the Isolated Heads of Rats of Various Ages.*

	Age in days							
	1	2	7	14	21	28	35	90
Gasps in Series I	12.7	13.2	12.3	8.1	7.2	6.9	6.0	5.3
Series II	40.3	30.5	24.0	19.5	9.2	4.9	2.3	0.0
Total gasps	53.0	43.7	36.3	36.3	16.4	11.8	8.3	5.3
Total duration, sec	1630	1450	745	278	117	75	56.3	14.5

*Values represent averages for 10 animals.

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Immunizing Capacity of Virus of Eastern Equine Encephalomyelitis Inactivated by Ultraviolet Light.

ISABEL M. MORGAN AND GEORGE I. LAVIN.
(Introduced by Peter K. Olitsky.)

From the Laboratories of the Rockefeller Institute for Medical Research, New York City.

The inactivating effect of ultraviolet light on viruses as well as bacteria has been observed repeatedly. That viruses so inactivated may be effective as immunizing antigens has already been shown for at least 2 viruses capable of causing disease in man. With a

Cerebral Resistance to E.E.E. Virus in Mice after Vaccination with Virus Inactivated by Means of Ultraviolet Light (U.V.) or Formalin (F.V.)

Virus dilution	Mice: Vaccinated U.V.	F.V.	Virus dilution	Non-vaccinated
10^{-2}	■■■	■■□	10^{-7}	■■■■
10^{-3}	□□□□	■□□□	10^{-8}	■■■□
10^{-4}	■■□□	■□□□	10^{-9}	■□□□

■ = 1 mouse died □ = 1 mouse survived

FIG. 1.

vaccine prepared by irradiating mouse-brain infected with rabic virus, Webster and associates[1] successfully immunized mice and dogs against a subsequent injection of active virus. Salk, Lavin, and Francis[2] compared the antigenic potency of epidemic-influenza virus following irradiation with that of active virus. In high concentrations, irradiated virus was nearly as effective an immunizing antigen as active virus; when lower concentrations were tested, a hundredfold loss in immunizing capacity was found to have occurred during irradiation. Ultraviolet light has been applied to the virus of equine encephalomyelitis, Eastern strain (E.E.E.), by Sharp and associates;[3] they studied the molecular stability of ultraviolet-treated virus. The preparation of an immunizing antigen produced by irradiation of E.E.E. virus with ultraviolet light is reported here.

Chick embryos 7-days-old were inoculated with 0.1 cc 10^{-3} suspension of E.E.E. virus-infected embryo in 0.85% saline solution. Embryos removed from the egg 18-20 hours after inoculation were rinsed in saline solution, ground in a mortar and made to a 10% suspension in saline or Tyrode's solution. This suspension was centrifuged at 2000 rpm for 10 minutes; the centrifugation was repeated with the supernate; the supernate then obtained was spun in a Swedish angle-centrifuge at 4000 rpm for 45 minutes. About

[1] Hodes, H. L., Lavin, G. I., and Webster, L. T., *Science*, 1937, **86**, 447; Webster, L. T., and Casals, J., *J. Exp. Med.*, 1941, **73**, 601.

[2] Salk, J. E., Lavin, G. I., and Francis, T., *J. Exp. Med.*, 1940, **72**, 729.

[3] Sharp, D. G., Taylor, A. R., Beard, D., Finkelstein, H., and Beard, J. W., *Science*, 1940, **92**, 359.

30 cc of the final supernate were transferred to a quartz test-tube, with an internal diameter of 2.1 cm, which was placed in the center of a quartz-mercury resonance lamp in the form of a spiral* with an internal diameter of 9 cm. The spiral, 15 cm in height, consists of 7 coils. The tube containing viral suspension was so placed that there was at least one complete coil of the lamp above and below the suspension. About 85% of the energy emitted by this type of lamp is in the line of 2537 Ångström units. The lamp operates at 30 milliamperes and 15,000 volts transformed from 110 volts A.C. The titer of virus before irradiation was regularly 10^8 when injected intracerebrally in albino mice, Rockefeller Institute strain. After beginning of irradiation, samples of virus were removed at 5-minute intervals; the suspension was stirred at each sampling; 0.03 cc of each sample was tested for infectivity by intracerebral injection of each of 4 mice. Inactivation was complete after 15-25 minutes of irradiation.

The immunizing capacity of E.E.E. virus inactivated by ultraviolet light was compared with that of formalin-inactivated virus prepared from the same viral suspension, since formalin-inactivated virus has come to be accepted as a standard immunizing antigen. In Fig. 1 the result of one experiment is charted. A group of 11 mice was vaccinated by means of 3 intraäbdominal injections on alternate days of 0.25 cc 10% suspension inactivated by 20 minutes' irradiation. Another group was vaccinated with similar doses of virus inactivated with 0.5% formalin. Non-vaccinated mice served as controls. Two weeks after beginning of vaccination, all mice were injected intracerebrally with a broth-suspension of mouse-brain infected with E.E.E. virus in dilutions indicated in Fig. 1. At least half of each group of vaccinated mice survived 10,000 and 100,000 cerebral units. There was no difference between the two vaccinated groups in the degree of immunity induced. It should be added that the antigenicity of such irradiated E.E.E. virus was found to diminish rapidly with further irradiation, an effect already noted for rabic virus.[1]

In summary, a non-infective immunizing antigen has been produced by ultraviolet irradiation of the virus of Eastern equine encephalomyelitis.[4]

* Manufactured by the Hanovia Chemical Company.

[4] Drs. Casals and Palacios find that E.E.E. virus inactivated by ultraviolet light serves as a highly specific complement-fixing antigen against anti-E.E.E. serum. Their results will be published.