(100-150% augmentation of ovarian weight with 1/3 of a 24-hour excretion). Histologic examination of the ovaries of our test animals revealed a heavy luteinization.

The presence of appreciable amounts of gonadotropic substance in urine of prepubertal children has been demonstrated. This hormone resembles "prolan" in its action and not the follicle-stimulating material found in castrate or menopausal urine. That it differs from the latter is also shown by the fact that, although Evans[4] has recently reported that the follicle-stimulating hormone of castration and menopause is synergistic with pituitary extracts, we were unable to obtain such effects with the pituitary extracts we prepared. Nevertheless this same pituitary preparation gave marked synergistic effects with pregnancy urine, as well as with the urine extracts of children as detailed above.

The presence of a prolan-like substance in the urine of normal children suggests the pituitary gland as its source.

I wish to thank Dr. Samuel Soskin for aid and direction.

8240 C

Some Effects of Ether on Bioluminescence in the Lampyrid, Photuris pennsylvanica.

GEORGE A. EMERSON. (Introduced by Paul D. Lamson.)

From the Department of Pharmacology, Vanderbilt University School of Medicine, Nashville.

In normal anesthetic concentrations ether has no direct chemical action on the dehydrogenase system involved in carbohydrate metabolism in mammalian brain, but through humoral effects ether anesthesia in the intact animal has a marked action indirectly on the rate of autoxidation of subsequently excised surviving brain, due presumably to the limiting of available carbohydrate.[1] The present paper demonstrates a striking corollary in the *Lampyridae,* for while ether has little action directly on the dehydrogenase system having to do with bioluminescence, it does exert various indirect effects in the intact insect which yield again quite unexpected results.

The light-organ of *Photuris pennsylvanica* generally flashes spon-

[4] Evans, H. M., and Simpson, M. E., PROC. SOC. EXP. BIOL. AND MED., 1935, **32**, 1047.

[1] Emerson, G. A., *J. Tenn. Acad. Sci.* In press.

taneously at irregular intermittent intervals. The characteristics of the flash have been studied photoelectrically by Snell[2] and the mechanism of control by Gerretsen, Snell and others,[2] it appears agreed that the flash is under nervous control. Access of adequate oxygen to the light-producing areas, which is a requisite for luminescence, is regulated by supposedly innervated[3] muscular tissue which normally between flashes almost completely occludes the entrance to tracheoles leading to the light-organ. Flashes may be artificially elicited through single shocks or short tetanic electrical stimulation, and the so-called pseudo-flash by rapidly raising the oxygen tension in an environment in which the oxygen concentration is low. Both artificial methods were found to be lacking in uniformity and to have potentialities of irreversibly injuring the flies, in confirmation of Snell's[2] work. Since no regular frequency of flashing can be induced in captured flies, it was obvious that little significant data could be obtained using flashing as a criterion for comparison of treated flies with controls.

Creighton's[3] observation, that injection of minute amounts of epinephrine hydrochloride into the fly results in an action on the occluding musculature of the tracheoles to permit the constant access of oxygen and a resultant constant bright glow of the light-organ, afforded an opportunity of preparing specimens for observation of the direct effects of gaseous agents on the luciferin-luciferase system *in situ*. The alternate dimming and brightening of the glow as observed by Creighton when the fly is alternately placed in atmospheres of nitrogen and oxygen was confirmed, and may be taken as evidence of the lack of the normal occluding mechanism, although both this and the postmortem anatomical evidence of action of epinephrine upon the tracheoles together are insufficient to answer the possibility of a direct chemical action of epinephrine upon the bioluminescence reaction. In lieu of experiments on an isolated luciferin-luciferase mixture which would show if epinephrine had any significant direct chemical effect, demonstration of the antagonistic action of the choline esters to the action of epinephrine and other sympathomimetic agents supports the hypothesis of indirect physical action in controlling the oxygen tension within the light-organ. Taylor's demonstration[9] of the effect of epinephrine HCl on bio-

[2] Gerretsen, F. C., *Biol. Zentralblatt*, 1922, **42**, 1; Harvey, E. N., *Physiol. Rev.*, 1924, **4**, 639; Snell, P. S., *J. Cell. Comp. Physiol.*, 1932, **1**, 37; *Science*, 1931, **73**, 372.

[3] Creighton, W. A., *Science*, 1926, **63**, 600.

[9] Taylor, G. W., *J. Cell. Comp. Physiol.*, 1932, **1**, 297; 1934, **4**, 329.

luminescence in bacteria is suggestive but not absolutely conclusive of lack of action in an *in vitro* system of luciferin-luciferase. If a direct chemical effect exists, applications of Creighton's observations on the occluding musculature, some of the present results and those obtained on the California singing fish, *Porichthys notatus*,[4] would be modified or negated.

Flies prepared through intraabdominal injection of 0.01-0.02 ml. of 10^{-3} epinephrine hydrochloride were exposed to various concentrations of ether vapor in air, made up by Fühner's[5] method. No immediate dimming of the constant bright glow was apparent except at the higher concentrations, so time of persistence of glow after 10 minutes' treatment with different tensions of ether vapor was chosen as a more sensitive criterion of action. Ten insects were used at each dosage level and the dimming end point arbitrarily taken as the time at which the bright glow, originally uniformly distributed over the 2 segments composing the light-organ, fell off so much that either of the segments appeared mottled or dark in spots when viewed through a 7 power hand lens. Results are given in Table I. Mortality was

TABLE I.
Action of Ether on *Photuris* Treated with Epinephrine to Produce a Steady Glow.

	Epinephrine Treated			Untreated		
Conc. Ether mM/l	Av. Time of Persistence of Glow min.	Mortality Ratio*	Motility	Av. Time of Persistence of Glow, min.	Mortality Ratio*	Motility
0.0	>240	0/10	+	0	0/10	+
0.5	>240	0/10	+	0	0/10	+
1.0	>240	0/10	—	0	0/10	—
2.5	>240	0/10	—	w	0/10	—
4.0	>240	0/10	—	0.8	0/10	—
7.5	>240	0/10	—	5.2	0/10	—
10.0	178	2/10	—	6.1	0/10	—
12.5	152	3/10	—	8.1	1/10	—
15.0	144	8/10	—	11.8	4/10	—
20.0	0	10/10	—	†	10/10	—

*No. dead at the end of one hour after anesthesia/no. anesthetized.
†Glow extinguished during anesthesia; 2/10 glowed 14 min. after anesthesia.
w = weak glow present during anesthesia.

determined by examining the number of flies of 10 alive one hour after exposure for 10 minutes to ether, and motility of the insects by loss of posture on disturbing the anesthesia chamber by a sharp blow or shake, which normally induces vigorous running about. The mortality curve obtained by Knoefel and co-workers[6] on mice is

[4] Green, C. W., and Green, H. H., *Am. J. Physiol.*, 1924, **70**, 691.
[5] Fühner, H., *Biochem. Z.*, 1921, **115**, 235.
[6] Knoefel, P. K., *et al.*, *J. Pharmacol. Exp. Therap.* In press.

of interest for comparison, the corresponding MLD_{50} for 10 minutes' exposure being about 4.5 mM/1 of ether.

The data collected in Table I demonstrate that ether has little direct action in anesthetic and sublethal tensions on the dehydrogenase concerned with bioluminescence in the intact insect, even when no barrier to its accessibility exists in insects in which the light-organ tracheoles are opened through the action of epinephrine.

The indirect action of ether on bioluminescence in normal *Photuris* is also shown by Table I. Insects placed in ether vapor at concentrations from 1.0 to 4.0 mM/1 show a weak constant glow similar to that produced by intraabdominal injection of 10^{-7} epinephrine hydrochloride. At ether concentrations of from 7.5 to 15 mM/1, a constant bright glow involving the whole of both segments of the light-organ and of approximately the same intensity as the glow produced by higher concentrations of epinephrine results. Above 15 mM/1, the bright glow is not maintained through the period of anesthesia, but on removing the killed insects from the ether, a permanent soft glow slowly develops. In the light of Gerretsen's[2] findings on the action of chloroform on *Luciola vittata,* a Javanese lampyrid, and his explanation of the 3 stages of effect through simply the lethal action of the agent, it is of interest to note that motility is lost with ether concentrations below that effective on the light-organ tracheoles but that a maximal effect of ether is obtained at concentrations below lethal concentrations.

Creighton dismisses the theory that there may be humoral control of bioluminescence in the *Lampyridae,* although tissue histologically similar to that of the medulla of mammalian suprarenals has been demonstrated in invertebrates.[3] The effect of ether in producing a constant glow is a peculiar analogy to the probable mediation of many of the biochemical effects of ether by suprarenal activity in mammals,[1, 7] however, and strongly suggests that some such endocrine tissue may be active in this case also. At any rate, the actions of ether and epinephrine are in the same direction here as elsewhere. The results[8] obtained on lampyrids with other sympathomimetic compounds and with narcotics which may have an indirect sympathomimetic effect in mammals are also suggestive of true humoral control of the flash, as is the partial antagonism to such action by parasympathomimetic compounds injected previously. The pertinent pharmacology and its bearing on observations of Gerretsen,[2] Taylor,[9]

[7] Emerson, G. A., *J. Pharmacol. Exp. Therap.,* 1935, **54**, 90.

[8] Emerson, G. A., and Knoefel, P. K. Unpublished.

and Shoup[10] on the effects of chloroform, urethane, epinephrine and other agents on various types of bioluminescence are discussed elsewhere.[8]

Summary. In the firefly, ether is effective in producing a constant glow through dilation of light-organ tracheoles only in concentrations well above the minimal anesthetic tension, while a maximum effect is produced at concentrations below the lethal tension; the effect of ether on bioluminescence is thus independent of both anesthesia and death. Ether inactivates the bioluminescence reaction *in situ* only at or above lethal tensions.

8241 P

Variations in Plasma Magnesium and Potassium in Epilepsy.

ARTHUR D. HIRSCHFELDER AND VICTOR G. HAURY.*

From the Department of Pharmacology, University of Minnesota.

McCollum and his collaborators,[1, 2] and also Greenberg and Tufts[3] have produced hyperirritability and convulsions in rats fed on a diet deficient in magnesium. Since we have found neuromuscular twitchings and convulsions in cases of clinical hypomagnesaemia,[4] since McQuarrie[5] believes that in epilepsy there is a "leakage of potassium" through the cell membrane, and since Hirschfelder[6] has

10 Shoup, C. S., *J. Gen. Physiol.*, 1929, **13**, 27; *J. Cell. Comp. Physiol.*, 1934, **5**, 269.

*Supported by a research grant from the Eli Lilly Co.

This investigation was conducted at the Minnesota Colony for Epileptics, whose clinical material and laboratory facilities were placed at our disposal by the Minnesota State Board of Control. To them, to Dr. D. E. McBroom, Superintendent of the Minnesota Colony for Epileptics, and to Dr. R. W. Brown we desire to express our thanks for their valuable cooperation.

1 Kruse, H. D., Orent, E. R., and McCollum, E. V., *J. Biol. Chem.*, 1932, **96**, 519; *Am. J. Physiol.*, 1932, **101**, 454.

2 Kruse, H. D., Schmidt, M. M., and McCollum, E. V., *Am. J. Physiol.*, 1933, **105**, 635.

3 Greenberg, D. M., and Tufts, E. V., PROC. SOC. EXP. BIOL. AND MED., 1935, **32**, 674.

4 Hirschfelder, A. D. (with the technical assistance of Victor G. Haury), PROC. SOC. EXP. BIOL. AND MED., 1933, **30**, 996; *J. Am. Med. Assn.*, 1934, **102**, 1138.

5 McQuarrie, I., *Ann. Int. Med.*, 1932, **6**, 497.

6 Hirschfelder, A. D., *J. Pharmacol. and Exp. Therap.*, 1929, **37**, 399.