THE SELECTIVE ACTION OF NICOTINE ON THE CENTRAL NERVOUS SYSTEM OF THE SQUID, LOLIGO PEALII.

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(Received for publication, February 19, 1919.)

Juvenile squid, *Loligo pealii*, are excellent indicators of the action of neurophil alkaloids. Excitation is shown by spasms of the musculature, in tentacles and mantle, and expansion of the chromatophores. This latter effect is caused by contraction of the radial muscle fibers attached to the sac-like chromatophores. The central nervous system of the squid is formed by the large cerebral ganglion to which are connected by nerve strands the two mantle or stellar ganglia. The musculature of the mantle may be excited either by the activity of the stellar ganglia or by that of the cerebrum. By means of observations on decapitated specimens it is thus possible to determine the seat of the nervous excitation. Strychnine in concentrations as low as 1: 500,000 causes the entire musculature to show spasms, with accompanying extension of the chromatophores in tentacles, head, and mantle. Since this occurs in decapitated as well as in entire specimens, it must be concluded that strychnine acts upon both groups of ganglia.[1] Camphor acts in a similar way, causing mantle spasms in decapitated animals, the excitatory action of camphor on the stellar ganglia being thereby proven.[2]

Nicotine is said to have an action in vertebrates similar to that of other excitants of the central nervous system.[3] With respect to its central action, this alkaloid falls in Maxwell's second group of brain stimulants since it acts upon ganglia alone.[4] In view of this selective

[1] Baglioni, S., *Z. allg. Physiol.*, 1905, v, 43.

[2] Moore, A. R., *Proc. Nat. Acad. Sc.*, 1917, iii, 598.

[3] Meyer, H., and Gottlieb, R., Pharmacology, Philadelphia, 1914, 373.

[4] Maxwell, S. S., *Am. J. Physiol.*, 1918, xlvii, 283; *J. Biol. Chem.*, 1907, iii, 21.

action, it seemed of interest to determine further whether nicotine is equally active on the stellar and cerebral ganglia of squid. For this purpose the freshly hatched animals were immersed in sea water solutions of Kahlbaum's alkaloidal nicotine. Typical effects were observed with dilutions as great as 1: 5,000,000, but stronger concentrations were found to be more satisfactory since they gave prompter results. It was noted also that ordinary commercial preparations of nicotine gave results identical with those of the purified alkaloid.

Upon being put into a solution of nicotine in sea water, the squid, in 3 or 4 seconds, falls to the bottom of the dish, with mantle shortened and broadened, quiescent, chromatophores relaxed, but with head drawn in and chromatophores of head and tentacles extended. After several minutes a secondary effect appears. If the dish is tapped, head and mantle are extended, accompanied by muscular spasms and maximum play of chromatophores over the entire body, while ink is often shot.

In order to determine whether the action of nicotine in producing such mantle spasms is due to its action on the head or mantle ganglia or both, experiments were carried out with decapitated mantles. These showed neither spasms nor play of the chromatophores, whether the decapitation took place before or after placing them in the nicotine solution. In order to remove any doubt in the matter, animals were kept in the solution of the alkaloid until the mantle spasms had developed, and were then decapitated, with the result that the mantles immediately relaxed and the chromatophores closed. Since in control experiments, in which decapitated mantles were immersed in sea water containing strychnine or camphor, spasms and chromatophore extension occurred, it is necessary to conclude that nicotine produces mantle spasms by acting upon the cerebral ganglion alone.

In a sea water solution of nicotine 1: 500,000 the mantle spasms develop after 6 to 7 minutes at 24°C. It made no difference in the time required for the onset of this effect whether the animals were removed from the solution and put into the sea water at the end of 1, 2, 3, 4, or 5 minutes or simply remained in the nicotine solution; the spasms in all cases appeared promptly 6 or 7 minutes after immersion in the poison. This somewhat astonishing result may be

accounted for if we assume it to be due to the formation of a compound between the nicotine and some substance of the cerebrum in critical concentration, and that sufficient nicotine is taken into the squid's body during the first minute to accomplish this result, and further that the uncombined nicotine is retained during the 6 or 7 minutes necessary for the reaction to reach the desired point. The assumption that this reaction or series of reactions is essentially chemical in nature is supported by the effect of the temperature on the length of the latent period.

Temperature. C.	Time. min.	Temperature coefficient.
24° / 14°	7 / 20	2.86
24° / 14°	6 / 17	2.83
23° / 13°	8 / 22	2.75

The rate of the reaction is increased approximately 2.8 times for an increase in temperature of 10°C. This value falls in well with the temperature coefficients of other life phenomena.

The effect of the concentration of the nicotine on the latent period of the mantle spasm is shown below. $T° = 24°$. t = time in minutes. v = the velocity of the process.

Concentration of nicotine.	Concentration ratio.	t	$v = 16.2 \times \frac{1}{t}$	$\sqrt[3]{\text{concentration}}$
1: 50,000	100	3	5.4	4.75
1: 500,000	10	7	2.3	2.2
1: 1,000,000	5	11	1.5	1.7
1: 5,000,000	1	14	1.1	1.0

From these results it appears that the velocity of the process varies approximately as the cube root of the concentration of the nicotine.

SUMMARY.

1. In specimens of freshly hatched squid, *Loligo pealii*, nicotine acts upon the cerebral ganglia alone.

2. After 1 minute in the nicotine solution 1: 500,000, the latent period for the mantle spasm is independent of the time spent in the solution.

3. The mantle spasm is conditioned by a chemical reaction, since the temperature coefficient of the process has a magnitude of about 2.8.

4. The velocity of the process which brings about the mantle spasm varies as the cube root of the concentration of the nicotine.