Tranquilizing Effect of "Substance P". (24917)

P. Stern and R. Milin* (Introduced by F. M. Berger)
Pharmacological Labs. of Sarjevo University, Yougoslavia

Because of synergism between "Substance P" (SP) on the one hand and mephenesin(1) (MF) and meprobamate(2) (MB) on the other hand, we have postulated that SP may be a physiological tranquilizer(3). According to their mechanism of action, tranquilizers have been classified by Berger into 2 groups (4): 1) autonomic suppressants which comprise derivatives of phenothiazine, reserpine and diphenylmethane and, 2) central relaxants derived from propanediols such as MF and MB. Our studies indicate that there is a similarity between SP and the central relaxant group of tranquilizers. SP and central relaxants do not possess anticholinergic or antihistaminic action. They counteract strychnine convulsions and prolong hexobarbital sleep. They do not influence conditioned reflexes but relax spastic muscles. The autonomic suppressants on the other hand often produce an opposite effect. Previously we showed that tranquilizing drugs such as chlorpromazine(5), reserpine(6), as well as mephenesin and meprobamate (unpublished). have a marked sedative effect on wild hares kept in a state of fear. Our experiments were designed to find out whether SP was also capable of producing a tranquilizing effect in frightened hares. Wild hares are preeminently suited as experimental animal for evaluation of tranquilizing drugs. When kept in a cage with a dog nearby, the hares develop within one or 2 weeks distinct hyperthyrosis with marked exophthalmus(5,7). The animals make frantic efforts to escape, do not permit being touched or held and move around constantly. They become quiet when no visible observer is present. When exposed to strong light or in presence of an observer their restlessness returns. It has previously been shown that SP had a sedative effect on normal animals(8).

* We thank the Scientific Research Fund of Rectorate of Sarajevo Univ. for providing resources for these studies.

Methods. The hares were divided into 3 groups. Group 1 consisting of 6 control animals were hares hunted down in their natural habitat and sacrificed immediately after capture. Group 2 of 11 hares that were kept in a state of fear for 2 days by the presence of a dog. Group 3 of 7 hares kept in a state of fear for 2 weeks. Group 1 had 20 to 23 units of SP/g of brain. A similar amount was found in brains of hares belonging to Group 3. Hares of Group 2 were given doses of SP, prepared according to Pernow(9) and purified according to Euler's procedure(10). Our preparations of SP contained approximately 3 to 3.5 units/mg. The assay was carried out using a reference standard of SP kindly given to us by Dr. Pernow. SP was given in doses of 100 mg and 300 mg/3 kilos body weight, intravenously.

Results. After receiving SP, all animals became subdued within 5 to 10 minutes. They stopped jumping about and did not react to presence of man or barking of dog. Half an hour later the animals tolerated handling and permitted being held in hand like tame rabbits. Hares given the smaller dose of SP remained tranquilized for 7 hours while those receiving the larger dose remained tranquilized for about 24 hours. Some hares given 100 mg of SP were sacrificed 4 hours later and amount of SP in their brains determined. SP had increased to 30-31 units/g of brain. Similar tests were carried out with another group of hares given meprobamate. The tranquilizing effect of the drug was rapid and intense.

Discussion. It appears that SP has a definite tranquilizing effect. It is important to ascertain the mode of action of SP on the hypothalmus and hypophysis of experimental animals. We hold that SP exerts a depressive action on stress-produced stimulation of the hypothalmus. It is of particular interest that SP in the brain of rats tends to decrease under the influence of both MF and MB. Res-

erpine, on the other hand, a tranquilizer belonging to the group of autonomic suppressants, has no effect on SP of the brain of dogs (11). Our results showed that the amount of SP remains unchanged in hares subjected to fear for 14 days. No attempt was made to investigate the amount of SP in brains after shorter periods of time. That the amount of SP in brains of hares given SP intravenously is increased, indicates that this substance is able to pass the blood brain barrier. Presence of SP in the reticular formation(8,12,13) and especially the high amounts present in substantia nigra (which represents the condensation area of the reticular substance(14)), indicates that SP may represent an active principle closely connected with the function of nerve cells.

The marked tranquilizing effect of SP in hares lends support to the view that this substance belongs to the class of biological regulators of the neuro-endocrine system or that it performs the function of a physiological tranquilizer.

Summary. Wild hares have 20 to 23 units of Substance P/g of brain. This amount does not change when animals are kept 14 days in cages in a state of fear, although hyperthyrosis develops during this time. When Substance P is given intravenously its amount in the brain increases. Wild hares given Substance P intravenously behave like tame rabbits.

1. Stern, P., Dobric, V., *Arch. internat. Pharmacodyn.*, 1957, v112, 102.

2. Stern, P., *Radovi, Izdanje Naučnog drustva NRBIH,* 1958, v9, 23.

3. ———, in Garratini, S., Ghetti, V. (eds.) *Psychotropic Drugs,* Elsevier. Amsterdam, 1957, pp448.

4. Berger, F. M., *Ann. New York Acad. Sci.,* 1957, v67, 685.

5. Milin, R., Stern, P., Dzinic, B., *Compt. rend. assn. anat.,* 1955, v42, 1041.

6. Stern, P., Milin, R., Scepovic, M., *Schweiz. med. wchnschr.,* 1956, v86, 415.

7. Kracht, J., Kracht, U., *Virchows. Arch.,* 1952, v321, 238.

8. Zettler, G., *Arch. exp. Pathol. u. Pharmakol.,* 1956, v228, 513.

9. Pernow, B., *Acta physiol. scand.,* 1953, v29, suppl. 105.

10. Euler, V. U., *ibid.,* 1942, v4, 372.

11. Paasonen, N., Vogt, N., *J. Physiol.,* 1956, v131, 617.

12. Kopera, H., Lazzarini, V., *Arch. exp. Pathol. u. Pharmakol.,* 1953, v219, 214.

13. Zettler, G., Schlosser, L., *ibid.,* 1955, v224, 159.

14. Roussy, G., Mosinger, M., *Traité de Neuro-endocrinologie, Masson,* 1946.

Received February 25, 1959. P.S.E.B.M., 1959, v101.

Quantitative Study of Estrogen-Induced Atherosclerosis in Cockerels.* (24918)

CLYDE T. CALDWELL AND DELBERT E. SUYDAM (Introduced by M. H. Kuizenga)
Dept. of Nutrition and Metabolic Diseases, The Upjohn Co., Kalamazoo, Mich.

Hyperlipemic atheromatosis has been produced in chickens by subcutaneous implantation of diethylstilbestrol(1). Starting with 3 mos old cockerels, aortic lesions developed in 6 to 7 mos which resembled more closely those occurring spontaneously in birds than the cholesterol-induced type. The 3 types of lesions were fundamentally similar and appeared to differ mainly in concentration of various lipid components(2). Use of 6 wk old birds decreased the possibility that the observed lesions were of spontaneous origin(3). These observations suggested the possibility of developing a quantitative method for study of atherosclerosis. This has been accomplished through time and dose response studies of the atherogenic effect of estrogenic material on young cockerels. Study of the sequence of artery and blood serum changes was made concomitantly.

Material and methods. Approximately 4

* The authors wish to thank Harris D. Webster, Dept. of Pathology, for assistance in evaluating the arteries.