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Received November 19, 1956. P.S.E.B.M., 1957, v94.

Differential Concentration of Porphyrin in Various Divisions of the Central Nervous System.* (22943)

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The observation that white matter of the central nervous system exhibited a band fluorescence spectrum centered at 625 m led to the conclusion that the nervous system contained coproporphyrin(1). From further evidence based on solubility characteristics, Klüver(2) identified protoporphyrin and coproporphyrin type III isomer. Chu and Watson(3), using the fluorescence quenching technic, also identified the coproporphyrin isomer as type III and reported the average amount of this porphyrin in 10 determinations of normal rabbit brain to be 3.56 γ /100 g brain. The isolation of crystalline coproporphyrin III from beef brain was carried out by Blanshard(4). His method of extraction yielded 2 γ /100 g brain.

Earlier observation,[‡] on the fluorescence of various parts of the central nervous system of mice and rats, gave the impression that red fluorescent material was more abundant in the medulla oblongata and brainstem than in other areas. The quantitative distribution of coproporphyrin III in gross anatomical subdivisions of the central nervous system was

studied to ascertain whether porphyrins exhibit a gradient distribution in the nervous system and to obtain a clue as to the possible functional significance of the nervous system porphyrins.

Methods. Pig brains were collected at the abattoir, stripped of membranes, and washed to remove blood. Equal amounts (250 g) of cerebrum, midgrain, medulla oblongata, cerebellum, and spinal cord were homogenized with 4:1 ethyle acetate-glacial acetic acid, (4) and cooled to -15°C . The supernate was decanted and filtered under suction. Extractions were repeated until no color was observed in the supernate. The material remaining was mixed with Solka-Floc BW 40, a purified wood cellulose filter aid, and placed in a large Büchner funnel, and covered with 200 ml 3N HCl which filtered off slowly. This solution was neutralized with sodium acetate and extracted with ethyl acetate-glacial acetic acid(4:1). The extract was added to the filtered supernate decanted previously. Extractions were carried out 5 times on subdivisions of the central nervous system with exception of the medulla oblongata which was repeated 6 times. The extracted porphyrins were measured fluorimetrically.

Results. Highest concentration of copro-

* Aided by grants from Anna Fuller Fund, Am. Cancer Soc., Md. Div. and Grant of Div. of Allergy and Infectious Diseases, P.H.S.

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[‡] Figge, unpublished.

TABLE I. Coproporphyrin III Extracted from Subdivisions of Pig Central Nervous System.

γ of coproporphyrin III/100 g of tissues*					
Cerebrum	Midbrain	Medulla oblongata	Cerebellum	Spinal cord	Total CNS avg
3.05 ± 0.1	$4.23 \pm .27$	$4.85 \pm .31$	3.10 ± 0	$3.16 \pm .22$	3.68

* Mean $\pm$ S.E.

porphyrin occurs in the medulla oblongata (Table I). This is only slightly higher than the amount found in the midbrain. Values for midbrain and medulla oblongata are highest, while values for the cerebrum, cerebellum and spinal cord are slightly lower than the average for the total central nervous system. The average for all 5 subdivisions of the nervous system of the pig is in close agreement (3) with the quantitative determination of coproporphyrin in total rabbit brain (3.56 γ /100 g).

Discussion. Klüver(1) observed the 625 $m\mu$ fluorescence band in funiculi of the spinal cord, and other fiber tracts of the central nervous system. Examination revealed that the 625 $m\mu$ band was absent in peripheral parts of cranial nerves.

Subsequent investigations have been limited to porphyrin content of total brain. Our results indicate that coproporphyrin III concentration varies at different levels of the central nervous system. This observed variation or gradient in coproporphyrin III concentration with a maximum in the medulla oblongata, lends support to the hypothesis that

the central nervous system porphyrins have some functional significance. It should be emphasized that our values represent only the coproporphyrin III content of parts of the central nervous system.

Summary. The central nervous systems of pigs were collected and divided into the various gross anatomical subdivisions. Equal weights of cerebrum, midbrain, medulla oblongata, cerebellum, and spinal cord were extracted for coproporphyrin. A higher concentration of coproporphyrin III was found in the medulla oblongata and brain stem than in cerebrum, cerebellum, or spinal cord. The observed differential coproporphyrin III concentration in various subdivisions of the central nervous system supports the hypothesis that porphyrins in the nervous system may have functional significance.

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Received October 18, 1956. P.S.E.B.M., 1957, v94.

Effects of Varying Concentrations of Platelets and Their Lyophilized Derivatives on Generation of Thromboplastin.*† (22944)

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It has previously been reported that high concentrations of platelet material, fresh,

stored at -15°C , or lyophilized, inhibit the coagulation of recalcified, platelet-depleted plasma. Adjustment of the concentration of platelet material to approximate the equivalents of physiological platelet levels, brought the recalcification time into the normal range (1). The nature of these phenomena was

* Presented in part at Second Conference on Platelets, N.R.C., Nov. 18, 1955, Washington, D.C.

† This investigation was supported by Atomic Energy, by Children's Cancer Research Foundation, and Natl. Cancer Inst.