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The effect of ovariectomy on the γ -aminobutyric acid content in the cerebral hemispheres of young rats

The removal of some endocrine glands affects the γ -aminobutyric acid (GABA) content in the rat brain. The GABA content is decreased after castration, but returns to normal after injection of testosterone propionate (Tzu-Yu Li & Chang-Hua Wu, 1964). Adrenalectomy results in decreased GABA content of rat brain (Rindi & Ventura, 1961), cerebral cortex (Vernadakis & Woodbury, 1959; Pandolfo & Macaione, 1964), and subcortex (Sutherland & Rikimaru, 1964). Thyroidectomy also decreased the brain GABA content (Nishioka, 1960). I have examined the effect of ovariectomy on the GABA content in the cerebral hemispheres of young rats.

Young female rats, 40–48 g, were divided into 4 equal groups of 9 each. One group received no treatment. The other animals were ovariectomized and left for 2, 15 and 30 days before being killed by guillotine for the estimation of GABA content in their cerebral hemispheres. At that time the rats weighed 60–70 g. For the analysis the cerebral hemispheres of three animals were pooled.

GABA was quantitatively determined using a chromatographic and colorimetric method. Within one min of death the brain was isolated and the cerebral hemispheres were separated and frozen to -4 to -6° . The frozen pooled sample was quickly weighed, triturated to a homogeneous mixture, and 60 ml of ethanol 75% was used as a solvent. The mixture was centrifuged and the supernatant fluid was evaporated to dryness. The residue was cooled and dissolved in distilled water. This extract was centrifuged and an amount of cerebral hemisphere extract equivalent to 60 mg of the original wet tissue was applied by an Agla micrometer syringe to a 20×46 cm band of chromatographic paper.* The chromatogram was developed with the descending technique by the one dimensional method using the solvent phenol-water (4:1 v/v) for 18 to 20 h, after which the solvent was removed. The paper was sprayed on both sides with 0.1% ninhydrin in butanol and left suspended for 30 min at 93° for maximal colour development (Roberts & Frankel, 1950). The GABA spots were eluted by distilled water, and the extinction read in a Unicam SP1300 colorimeter using filter No. 4. The relation between the extinction and the concentration of pure GABA† was determined under the experimental conditions. The formula of the curve was obtained by the least square method (Waugh, 1952) and used to convert the colorimetric readings into the equivalent concentrations of GABA.

The relation between different amounts of an authentic sample of GABA when used in between 5 and 35 μ g, with the extinction of the ninhydrin-stained chromatographic spots had the general formula $Y = 0.011 + 0.0051 X$.

Table 1. *GABA content in the cerebral hemispheres of 2, 15 and 30 days ovariectomized young female rats compared with the normal content*

Sample number and statistical data	GABA content (mg/100 g wet tissue) of cerebral hemispheres			
	Control	Ovariectomy		
		2 days	15 days	30 days
1	16.5	16.5	18.0	18.4
2	17.6	18.4	18.4	19.5
3	18.0	18.5	18.5	19.7
\bar{x} *	17.4	17.8	18.3	19.2 ($P < 0.025 > 0.001$)†
\pm s.e.	0.5	0.7	0.2	0.4

* Each mean is the average GABA content obtained from 3 pooled samples each of 3 rats.

† By *t*-test.

The results obtained for GABA contents in the cerebral hemispheres of young female rats and in similar animals 2, 15 and 30 days after ovariectomy are in Table 1.

The GABA content did not change significantly in the cerebral hemispheres of young female rats after their ovariectomy by 2 and 15 days but there was a significant increase in GABA content 30 days after ovariectomy amounting to 10.6% from the control value. This appears to be reverse of the effect produced after castration (Tzu-Yu Li & Chang-Hua Wu, 1964).

The induced change in GABA content in the cerebral hemispheres is of importance since GABA appears to be the main inhibitory transmitter in cerebral cortex (Iwama & Jasper, 1957; Krnjević, Randić & Straughan, 1966; Krnjević & Schwartz, 1967).

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* Selecta-Filter Paper No. 2316, Carl Schleicher and Schull Dassel K. R. Einbeck, Pachtbetrieb der Büttenpapierfabrik Habnemühle GMBH, West Germany.

† γ -Aminobutyric acid, m.p. 202–204°, L. Light and Co. Ltd., Colnbrook, Bucks., England.

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