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EFFECTS OF γ-AMINO-β-HYDROXYBUTYRIC ACID ON CEREBRAL AMINES

BY

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γ-Amino-β-hydroxybutyric acid (GABOB), a hydroxy-derivative of γ-aminobutyric acid (GABA), is normally found in human and animal cerebral tissues. From that source, the substance was isolated by paper chromatography (Ohara, Sano, Koizumi & Nishinuma, 1959). GABOB has the property of blocking excitatory synaptic terminations, inhibiting experimentally induced convulsions in animals, and inverting the polarity of strychnine peaks (Savoldi, Maggi, Arrigo, Cosi & Tartara, 1962). These effects are elicited both by topical application and by systemic administration, indicating that the substance crosses the blood-brain barrier. Clinically, GABOB is definitely effective in preventing and alleviating epileptic seizures (Hayashi, 1959; De Maio, Madeddu & Faggioli, 1961; Buscaino & Ferrari, 1961; Floris, Morocutti, Gaggini & Napoleone-Capra, 1961), thus qualifying as a physiological antiepileptic agent.

Many substances endowed with anticonvulsant activity increase the concentration of 5-hydroxytryptamine in the brain (Bonnycastle, Giarman & Paasonen, 1957). More recently, Anderson, Markowitz & Bonnycastle (1962) tried to elucidate this problem and concluded that no rigorous relation exists between the anticonvulsant activity of a drug and its ability to increase cerebral 5-hydroxytryptamine; also, that substances undoubtedly capable of increasing cerebral 5-hydroxytryptamine are sometimes completely devoid of anticonvulsant activity.

In view of these circumstances we decided to investigate the effects of GABOB upon the concentration of cerebral amines, and more particularly, the effect of GABOB on the concentration of cerebral amines in normal animals; the effect of GABOB on the concentration of cerebral amines in animals affected by reserpine; and the effect of intracerebral administration of GABOB on the concentration of cerebral amines.

METHODS

Groups of albino rats of the Sprague-Dawley strain were used in these experiments. The average weight was 150 to 200 g, and the animals were maintained on a balanced diet. GABOB and reserpine were administered intraperitoneally, dissolved in distilled water. Intracerebral administration of GABOB was made with a blind technique through the petrosquamous foramen, the injected volume being 50 μl.

Rectal temperatures were measured with a thermocouple (Ellab). 5-Hydroxytryptamine and noradrenaline were assayed by spectrofluorimetry with the method of Bogdanski, Pletscher, Brodie & Udenfriend (1956), after extracting the cerebral matter as described by Shore (1959).

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RESULTS

Intraperitoneal administration of GABOB in normal animals

Doses of 500 to 600 mg/kg were administered. The substance did not produce ptosis, diarrhoea or sedation, and did not appear to modify the animals' behaviour. The results are shown in Table 1, and indicate that intraperitoneal administration of GABOB in rats does not produce significant changes in the concentrations of cerebral 5-hydroxytryptamine and noradrenaline or in body temperature.

TABLE 1

EFFECT OF INTRAPERITONEAL GABOB ON CEREBRAL CONTENT OF 5-HYDROXYTRYPTAMINE AND NORADRENALINE IN ALBINO RATS

Values are means with standard deviations

No. of rats	Treatment (mg/kg)	Time after treatment (hr)	Brain concentration of 5-Hydroxytryptamine (μg/g)	Brain concentration of Noradrenaline (μg/g)	Rectal temperature (° C)
6	Saline	1	0·45±0·03	0·43±0·02	36·8±0·5
6	GABOB (500)	0·5	0·50±0·06	0·49±0·03	37·1±0·8
		1			36·7±0·5
		2	0·45±0·05	0·42±0·06	36·8±0·3
		3			37·0±0·4
		4	0·45±0·03	0·36±0·02	37·1±0·5
		6			37·2±0·5
		8	0·47±0·05	0·49±0·03	37·1±0·5
		16	0·46±0·04	0·45±0·03	36·8±0·3
			$P>0·01$	$P>0·01$	$P>0·01$
6	GABOB (600)	0·5	0·55±0·05	0·37±0·03	37·1±0·4
		1	0·48±0·03	0·40±0·03	37·0±0·4
			$P>0·01$	$P>0·01$	$P>0·01$

Administration of GABOB and reserpine

This experiment was made in order to clarify whether GABOB, like monoamine oxidase inhibitors, could prevent or modify the depletion of cerebral amines produced by reserpine. GABOB was administered intraperitoneally at the dose of 500 mg/kg, and 2.5 mg/kg of reserpine was administered, again intraperitoneally, 30 min later. The cerebral amines were assayed 4 hr after the administration of reserpine when the rats showed sedation and ptosis. The results are shown in Table 2, and indicate that intraperitoneal administration of

TABLE 2

EFFECT OF INTRAPERITONEAL GABOB AND RESERPINE ON CEREBRAL CONTENT OF 5-HYDROXYTRYPTAMINE AND NORADRENALINE IN ALBINO RATS

Values are means and standard deviations

No. of rats	Treatment (mg/kg)	Brain concentration of 5-Hydroxytryptamine (μg/g)	Brain concentration of Noradrenaline (μg/g)	Rectal temperature (° C)
5	Saline	0·48±0·03	0·45±0·04	37·1±0·4
5	Reserpine (2·5)	0·19±0·03	0·08±0·03	34·8±0·4
		$P<0·01$	$P<0·01$	$P<0·01$
5	GABOB (500) +reserpine (2·5)	0·21±0·03	0·10±0·04	34·9±0·5
		$P>0·01$	$P>0·01$	$P>0·01$

GABOB in subsequently reserpinized rats does not significantly modify the depletion of cerebral 5-hydroxytryptamine and noradrenaline, or the hypothermia induced by reserpine.

Intracerebral administration of GABOB

In order to determine whether GABOB could influence the metabolism of cerebral amines when brought into direct contact with nervous tissues, we also administered the drug by intracerebral injection. The dose was 30 mg/kg. The animals showed no changes of behaviour with the exception of mild sedation and hypothermia, which disappeared

TABLE 3

EFFECT OF INTRACEREBRAL GABOB ON CEREBRAL CONTENT OF 5-HYDROXYTRYPTAMINE AND NORADRENALINE IN ALBINO RATS

Values are means and standard deviations

Treatment (mg/kg)	Time after treatment (min)	Brain concentration of 5-Hydroxytryptamine (μg/kg)	Brain concentration of Noradrenaline (μg/kg)	Rectal temperature (° C)
Saline	90	0·45±0·01	0·59±0·02	37·1
GABOB (30)	15			34·4±1·0
	30			35·2±1·4
	45			35·8±1·2
	60			35·8±0·9
	90	0·42±0·06	0·50±0·07	37·3±0·8
				$P>0·01$

within 1 hr. The results are reported in Table 3, and show that GABOB, at the dosage employed in these experiments, does not modify the concentrations of cerebral 5-hydroxytryptamine and noradrenaline, even upon direct intracerebral administration.

DISCUSSION

The results of this investigation indicate that GABOB is devoid of any appreciable effect on the concentrations of brain 5-hydroxytryptamine and noradrenaline. This finding agrees with the opinion of Anderson *et al.* (1962) that there is no direct relationship between the anticonvulsant activity of a drug and its ability to increase the concentration of 5-hydroxytryptamine in the brain. Indeed, in a recent paper, De Maio & Pasquariello (1963) stated that GABOB, administered intraperitoneally to rats at a dose of 500 mg/kg, caused a significant decrease of brain 5-hydroxytryptamine. Our results with the same doses, however, are not in agreement with the foregoing. The total lack of any action of GABOB on reserpine-induced depletion of cerebral amines, and the absence of significant changes in the concentration of cerebral amines following the direct intracerebral administration of relatively large doses of GABOB, confirm the findings obtained with GABOB alone, administered systemically.

It is therefore safe to assume that GABOB acts as an anticonvulsant drug not by way of modifying the concentration of cerebral amines, but rather by a direct action upon excitatory synaptic terminations.

SUMMARY

1. γ-Amino-β-hydroxybutyric acid (GABOB), an anticonvulsant substance normally present in brain tissues, had no definite effect either on brain 5-hydroxytryptamine or on noradrenaline content.

2. Animals treated with GABOB show no modifications of the effects subsequently induced by reserpine on brain amines.

3. The results confirm that there exists no relationship between the anticonvulsant activity of a drug and its ability to modify cerebral amines contents.

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