Influence of folic acid on noradrenaline stimulation of rat brain synaptosomes

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Many antiepileptic drugs may produce a megaloblastic anaemia in man. These drugs impair the absorption of folic acid, but administration of folic acid, which corrects the anaemia, may aggravate the epilepsy. It is thus possible that folic acid increases neuronal excitability and that the antiepileptic drugs may antagonize this.

In these experiments rat brain synaptosomes were exposed to a range of concentrations of formyl tetrahydrofolic acid (f-THF) (0·0016–0·635 mM) and of noradrenaline (NA) (0·015–0·15 mM). The rate of oxygen consumption in a glucose medium was measured in an oxygen electrode. Neither f-THF nor NA affected respiration rate, but when both substances were present the rate of oxygen consumption was increased. Adrenaline with f-THF also stimulated respiration, but isoprenaline, pteroyl glutamic acid, acetylcholine, histamine and dopamine did not affect oxygen uptake in this system.

The NA plus f-THF stimulation was blocked by concentrations of phenobarbitone which did not affect the control respiration rate. The effect was also inhibited by administration of 5-fluorouracil (100 mg/kg) *in vivo* for 3 days before the *in vitro* experiment.

This experiment suggests that, in the presence of extraneuronal NA, f-THF but not pteroyl glutamic acid can stimulate brain function. This provides a possible explanation of the antiepileptic properties of drugs which block folate reductase.

Distribution of octopamine in nervous tissues of *Octopus vulgaris*

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Octopamine (*p*-hydroxyphenylethanolamine) was first discovered in the posterior salivary glands of *Octopus vulgaris* (Erspamer & Boretti, 1951). A sensitive enzymatic assay (Molinoff, Landsberg & Axelrod, 1969) for octopamine has been developed and used to show that octopamine occurs normally in the sympathetic nerves of rat organs (Molinoff & Axelrod, 1969). The high concentration of octopamine in *Octopus* salivary glands suggested the possibility that it might also be present in neural tissues of octopods.

Tissues, dissected out as previously described (Juorio, 1971), were weighed and then quickly frozen on dry ice. After heat denaturation of the proteins the concentration of octopamine was estimated. Significant amounts of octopamine were found throughout the *Octopus* nervous system (Table 1).

The concentration of octopamine in most neural tissues is similar to the concentrations of catecholamines and of 5-hydroxytryptamine (Juorio, 1971). The concentration of octopamine in the posterior salivary gland was more than 1 mg/g. The administration of reserpine (4 mg/kg, i.m.) led to a fall in the octopamine concentration in most neural tissues but did not affect the concentration in the posterior salivary gland.

TABLE 1. *Distribution of octopamine in* Octopus *neural tissues*

	Weight of tissue (mg)	Octopamine (μg/g)
	Supraoesophageal ganglia	
Vertical lobes	8·8±1·6 (5)	0·72±0·19 (6)
Superior frontal lobe	4·9±0·5 (6)	0·55±0·20 (6)
Inferior frontal lobe	2·0±0·3 (5)	1·30±0·08 (5)
Superior buccal lobe	3·7±1·1 (6)	4·57±1·07 (6)
Posterior buccal lobe	2·6±0·7 (6)	2·05±0·76 (5)
Basal lobes system	31±6 (6)	1·23±0·41 (5)
Optic lobes	152±22 (6)	0·74±0·15 (7)
	Suboesophageal ganglia	
Anterior region	31±5 (3)	1·00±0·15 (3)
Median region	23±10 (3)	0·70±0·09 (3)
Posterior region	36±8 (3)	0·57±0·07 (3)
	Other tissues	
Stellate ganglia	15±2 (9)	0·24±0·07 (10)
Posterior salivary gland	932±139 (10)	1390±220 (10)
Anterior salivary gland	290 (1)	0·04, 0·02 (2)
Systemic heart	261±72 (5)	0·03±0·02 (5)

Values are means±S.E.M. in μg/g of fresh tissue. Number of experiments in parentheses.

When homogenates of the optic lobes were subjected to centrifugation on density gradients (15% glucose to 45% sucrose, wt/wt) some of the octopamine was found to be bound to particles. The octopamine-containing particles were found in the same regions of the gradient (21 and 28% sugar) as were noradrenaline and dopamine.

The fact that the concentration of octopamine varies over a 20-fold range in *Octopus* neural tissues, the decrease after treatment with reserpine, and the similarity in the amount and subcellular distribution of octopamine, noradrenaline and dopamine all suggest that octopamine is contained in neurone where it may function as a neurotransmitter.

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Hypnogenic properties of succinic semialdehyde and its fatty acid derivatives

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Several short chain fatty acid derivatives, including 4-hydroxybutyric acid (GHB) and 1,4-butanediol (1,4BDL), have hypnogenic properties in various species (Gessa, Spano, Vargiu, Crabai, Tagliamonte & Mameli, 1968). The experiments described