RESEARCH PAPERS

PHARMACOLOGICAL AND BIOCHEMICAL EFFECTS OF 5-HYDROXYTRYPTAMINE IN ADRENALECTOMISED RATS

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Received March 3, 1961

In adrenalectomised rats 5-hydroxytryptamine induces a fall of temperature and blood pressure which is more marked than in intact animals. This hypersensitivity is not related to changes in tissue monoamine oxidase or serum ceruloplasmin. Administration of 5-HT raises the levels of tissue 5-HT in adrenalectomised rats to the same extent as in control rats.

IN adrenalectomised rats and mice 5-hydroxytryptamine (5-HT) is several times more toxic than in intact animals (Canal and Maffei-Faccioli, 1958; Garattini, Gaiardoni, Mortari and Palma, 1961; Munoz, Schuchardt and Ferwey, 1958). This enhanced toxicity is not shared by partially hepatectomised animals (Garattini and others, 1961). Furthermore adrenalectomy does not induce unspecific sensitivity to chemical treatment since tryptamine (Palma, unpublished results), methacholine and adrenaline (Loew and Woodman, 1956) are similarly toxic in intact or adrenalectomised animals.

This increased toxicity of 5-HT may be a direct effect, that is increased sensitivity to the pharmacological activities of the amine, or the result of an impaired metabolism of 5-HT.

To test these hypotheses several experiments have been carried out. The results show that the decrease of body temperature and blood pressure induced by 5-HT in adrenalectomised rats is not related to major changes in the metabolism of 5-HT.

MATERIALS AND METHODS

Female Sprague-Dawley rats of the weight of 150–180 g. fed with a balanced diet were used. Adrenalectomy was performed under ether anaesthesia (Bomskov and Bahnsen, 1935). Animals received saline for drinking; treatment was begun 72 hr. after the operation. Sham-operated rats were also employed; the effect of 5-HT in these animals was not significantly different from that observed in intact rats.

5-HT was given subcutaneously or intravenously at the doses indicated in the Tables. Body temperature was measured with a thermocouple inserted in the rectal cavity. Blood pressure was determined, after anaesthesia with ethylurethane 1 g./kg. subcutaneously, by cannulation of the carotid artery and using a Sanborn 150 electromanometer.

Brain, spleen, lung and kidney 5-HT were extracted by the method of Shore (1959) and measured spectrofluorimetrically by the method of Bogdanski, Pletscher, Brodie and Udenfriend (1956). Monoamine oxidase activity in brain, liver, lung and kidney was measured by the method recently described by Weissbach, Smith, Daly, Witkop and Udenfriend (1960) using as substrate kynureninamine (obtained from Regis Chemical Co., Chicago, Ill.).

Serum ceruloplasmin activity was determined by the method of Ravin (1956) with minor modifications using *p*-phenylendiamine as substrate (Garattini, Giachetti and Pieri, 1960).

TABLE I

EFFECT OF 5-HT ON BODY TEMPERATURE IN INTACT AND IN ADRENALECTOMISED RATS (TEMPERATURE OF UNTREATED CONTROLS, 37·1 ± 0·3°C)

No. of rats	Experimental condition	5-HT dose (mg./kg.)	5-HT route	Changes in body temperature (° C. ± S.E.) 1 hr.	3 hr.	6 hr.	24 hr.
10	intact	5	s.c.	−1·9 ± 0·2	−1·2 ± 0·2	−0·6 ± 0·1	−0·8 ± 0·2
17	adrenalectomised	5	s.c.	−3·7 ± 0·3	−1·8 ± 0·3	−2·0 ± 0·5	−0·8 ± 0·4
10	intact	10	s.c.	−2·8 ± 0·3	−1·0 ± 0·2	+0·3 ± 0·1	−0·9 ± 0·3
18	adrenalectomised	10	s.c.	−3·8 ± 0·3	−4·0 ± 0·3	−3·6 ± 0·4	−2·3 ± 0·3
10	intact	25	s.c.	−5·2 ± 0·3	−3·8 ± 0·5	−0·3 ± 0·3	−0·8 ± 0·1
34	adrenalectomised	25	s.c.	−6·0 ± 0·3	−9·7 ± 0·5	−10·1 ± 0·7*	−12·5 ± 1·0†
10	intact	3·75	i.v.	−0·5 ± 0·2	+0·4 ± 0·1	−0·3 ± 0·1	−0·9 ± 0·2
8	adrenalectomised	3·75	i.v.	−3·5 ± 0·4	−3·2 ± 0·6	−3·5 ± 0·7	−2·0 ± 0·2
9	intact	75	i.v.	−5·0 ± 0·5	−2·0 ± 0·6	−0·6 ± 0·4	−0·5 ± 0·2

* 11 rats died.
† 26 rats died.

5-Hydroxytryptamine creatinine sulphate was obtained through the generosity of Vister Co., Casatenovo (Como).

RESULTS

5-HT *on body temperature of intact and of adrenalectomised rats.* The effect of 5-HT on body temperature of intact and of adrenalectomised rats is summarised in Table I. The amine is more active in adrenalectomised than in intact animals in decreasing body temperature. This effect

TABLE II

EFFECT OF 5-HT ON BLOOD PRESSURE IN INTACT AND IN ADRENALECTOMISED RATS. READINGS TAKEN 1 HR. AFTER ADMINISTRATION

Group	No. of rats	Experimental condition	Subcutaneous dose of 5-HT (mg./kg.)	Blood pressure (mm. Hg ± S.E.) Max.	Min.	Difference Max.	Min.
1	12	intact	—	143 ± 4	101 ± 6	—	—
2	13	intact	10	104 ± 3	62 ± 4	39	39
3	7	sham-operated	—	144 ± 4	104 ± 2	—	—
4	7	sham-operated	10	112 ± 5	54 ± 5	32	50
5	6	adrenalectomised	—	130 ± 7	103 ± 6	—	—
6	11	adrenalectomised	10	66 ± 6	45 ± 5	64	58
Significance for groups 1–2				$P<0.01$	$P<0.01$		
2–6				$P<0.01$	$P<0.02$		
1–5				$P>0.05$	$P>0.1$		
5–6				$P<0.01$	$P<0.01$		

is not only more marked immediately after administration, but it is also longer lasting. When it is injected intravenously, the effect of 75 mg./kg. in intact animals corresponds to that of 3·75 mg./kg. in adrenalectomised rats.

5-HT *on blood pressure of intact and of adrenalectomised rats.* Table II shows the effect of 5-HT on the blood pressure of intact and adrenalectomised rats weighing about 300 g.

5-HT decreases the blood pressure of intact, sham-operated and adrenalectomised rats. However the effect is more marked in the latter group than in the first two experimental groups.

5-HT *on tissue* 5-HT *of intact and of adrenalectomised rats.* The content of 5-HT in brain, lung, spleen and kidney of intact and of adrenalectomised rats before and after 5-HT administration is reported in Table III.

The results show that the 5-HT content of tissues of adrenalectomised rats does not differ markedly from that of tissues of control animals. After 5-HT administration, the increases in the content of lung, kidney and

TABLE III

TISSUE 5-HT OF INTACT AND ADRENALECTOMISED RATS BEFORE AND 1 HR. AFTER ADMINISTRATION OF 5-HT

No. of rats	Experimental condition	Treatment mg./kg., s.c.	5-HT base (μg./g. ± S.E.)			
			Brain	Lung	Kidney	Spleen
8	intact	—	0·38 ± 0·01	0·56 ± 0·02	0·18 ± 0·01	3·03 ± 0·15
8	intact	5-HT 10	0·39 ± 0·01	1·61 ± 0·17	1·42 ± 0·11	5·27 ± 0·17
10	sham-operated	—	0·37 ± 0·03	1·26 ± 0·04	0·33 ± 0·02	2·48 ± 0·09
10	adrenalectomised	—	0·28 ± 0·01	0·85 ± 0·04	0·26 ± 0·01	2·40 ± 0·07
8	adrenalectomised	5-HT 10	0·28 ± 0·02	1·97 ± 0·19	1·09 ± 0·08	3·32 ± 0·03

TABLE IV

MONOAMINE OXIDASE (MAO) ACTIVITY IN TISSUES OF INTACT, SHAM-OPERATED, AND ADRENALECTOMISED RATS, MEASURED AS THE DIFFERENCE IN OPTICAL DENSITY AT 360 mμ AFTER INCUBATION AT 37° FOR 20 MIN.

No. of determinations	Experimental condition	MAO activity (difference ± S.E.)			
		Liver	Brain	Kidney	Lung
7	intact	152 ± 17	123 ± 10	34 ± 8	57 ± 9
7	sham-operated	154 ± 12	120 ± 12	49 ± 12	52 ± 8
7	adrenalectomised	132 ± 32	116 ± 6	48 ± 16	57 ± 4

spleen of the two groups are also similar. That the metabolism is not changed by adrenalectomy is shown by the results obtained for the activity of monoamine oxidase (see Table IV) and by the fact that ceruloplasmin activity in serum is also unchanged.

DISCUSSION

5-HT decreases the body temperature of rats and mice (Hoffman, 1958; Lessin and Parker 1957) but its effect is more marked after adrenalectomy. The data obtained in the present investigation largely agrees with that recently reported by Hoffman (1959). Further, the fall in blood pressure exerted in rats by 5-HT (Erspamer, 1954) is enhanced by adrenalectomy. Both effects may be due to alterations in the mechanisms regulating body temperature and blood pressure. It is not likely that 5-HT is more active in adrenalectomised rats because of an impaired degradation. In the present experiments, the brain, kidney, spleen and lung 5-HT levels were not changed after adrenalectomy. Other authors found a raised level in the tissues of adrenalectomised rats but only when these were maintained on drinking water (Hicks and West, 1958) instead of receiving saline as in our experiments. As far as the brain 5-HT is concerned these results are in agreement with data recently reported (Towne and Sherman, 1960), but at variance with those reported by De Maio (1959).

The administration of 5-HT induces an increase of lung, spleen and kidney 5-HT, but no changes in the brain concentration (Costa, Rinaldi and Himwich, 1957; Sjoerdsma, Weissbach and Udenfriend, 1956; Woolley and Shaw, 1957). In adrenalectomised rats this increase is less than in normal rats and this may be due to a replacement of the stores of blood 5-HT which have been lowered by adrenalectomy (Medaković and Spužić, 1959).

The measurement of monoamine oxidase activity, the major enzyme responsible for 5-HT degradation (Brodie, Spector and Shore, 1959; Zeller, Blanksma, Burkard, Pacha and Lazanas, 1959), confirms that liver, brain, lung and kidney of adrenalectomised animals should metabolise the amine at rates comparable with those of intact or sham-operated rats. Ceruloplasmin oxidase activity, recently suggested as important for the metabolism of 5-HT in blood (Blaschko, 1960; Martin, Eriksen and Benditt, 1958; Porter, Titus, Sanders and Smith, 1957), is also unchanged by adrenalectomy. The significance of these results in explaining the enhanced toxicity of 5-HT in adrenalectomised rats is consistent with the hypothesis that an impairment of 5-HT metabolism is not the major cause. It may be that there is an increased sensitivity to the pharmacological effects of 5-HT, probably because there is a lack of the compensatory mechanisms. The fall in blood pressure and particularly in body temperature may be sufficient to account for the increased toxicity of 5-HT.

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