Short communications

The effect of drugs on stress-induced changes of myocardial glycogen and blood glucose concentration in rats

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Rats that were subjected to restraint stress for 18 h were found to have reduced myocardial glycogen and blood sugar levels and showed histological changes in heart and adrenals. The effects of α-methyl-*p*-tyrosine, α-methyl-dopa, disulfiram and actinomycin D on these stress-induced changes were examined. Of these drugs, only α-methyl-*p*-tyrosine was able to prevent the fall in myocardial glycogen and blood sugar and none of the drugs prevented the histological changes induced by stress.

Introduction.—The catecholamines released in stressful situations presumably fulfil a physiological function. However, excessive amounts in the circulation might be responsible for some of the pathological changes seen in various tissues after prolonged stress (Raab, Stark, MacMillan & Gigee, 1961). This was indicated by the observation that chlorpromazine, guanethedine or mecamylamine prevented changes in myocardial glycogen contents that are normally induced by cold stress (Sharma & Parmar, 1967). The present paper describes experiments designed to investigate whether drugs that block catecholamine synthesis are able to prevent the fall in cardiac glycogen and blood glucose that occurs after prolonged immobilization.

Methods.—Eighty albino rats of either sex (90–120 g) were divided into 10 groups of 8 rats each and treated as described in Table 1. To stress the rats, they were tied to a flat board in a prone position for 18 h (Renaud, 1959). α-Methyl-*p*-tyrosine (α-MPT) was used as an inhibitor of tyrosine hydroxylase, α-methyl-dopa as a dopa-decarboxylase inhibitor and disulfiram as an inhibitor of dopamine β-hydroxylase. Actinomycin D was used to inhibit (in addition to protein synthesis) the enzyme phenylethanolamine-N-methyl transferase. The drugs were given either 18 h before death or immediately before the start of the stress period. The electrocardiograms of the rats were recorded during the first hour of stress and just before terminating the experiment, when the rats were anaesthetized with ether and the hearts dissected out. Myocardial glycogen was estimated by the method of Kemp & Kits van Heijningen (1954). Blood glucose was determined by the method of Somogyi (1952). For histological studies sections of the myocardium and the adrenals were stained with haemotoxylin eosin.

Results.—After 18 h of immobilization the heart rate of the rats was decreased by 14% ($P<0.001$) and the QRS and QT intervals in the ECG increased. α-MPT itself decreased the heart rate by 8% ($P<0.01$) and α-methyl-dopa by 6% ($P<0.05$). In none of the rats treated with these drugs, whether stressed or unstressed, were there any changes in the ECG. Disulfiram alone caused a fall in the heart rate of 8% ($P<0.05$) but in the rats which were stressed in addition there was also an increase in the PR and QT intervals of the ECG. In three out of eight animals the T wave was flat and the heart rate decreased by 34% ($P<0.001$). No ECG changes were seen in the stressed or unstressed rats treated with actinomycin D.

The observations on myocardial glyogen and blood glucose concentrations are shown in Table 1. A significant decrease in the myocardial glycogen content (−42%) was observed in rats subjected to 18 h restraint stress. α-MPT alone caused a 20% increase in the glycogen content of the heart. In rats treated with α-MPT immobilization did not result in a fall in the concentration of myocardial glycogen, in contrast there was a further rise of 85%. α-Methyl-dopa alone caused a 50% increase in the myocardial glycogen content. In the rats which were treated with α-methyl-dopa and stressed the heart glycogen fell by 37%. However, the cardiac glycogen content in the stressed rats was equal to that in unstressed, untreated rats. Disulfiram alone did not produce any significant change in the cardiac glycogen content, but additional stress lowered it as in the controls. Actinomycin D itself nearly doubled the glycogen content but it did not completely

TABLE 1. *Heart glycogen and blood glucose contents in stressed and unstressed rats treated with inhibitors of enzymes involved in catecholamine synthesis*

Group No.	Drug treatment†	Immobilization stress*	Cardiac glycogen					Blood glucose				
			Cardiac glycogen concentration (mg/100 g±S.E.)	Difference between unstressed and stressed rats %	*P*	Difference from unstressed controls %	*P*	Blood glucose concentration (mg/100 g±S.E.)	Difference between unstressed and stressed rats %	*P*	Difference from unstressed controls %	*P*
I	Control	0	313± 9·0	−42	<0·001	—	—	143·5±9·28	−49	<0·001	—	—
II	Control	+	182± 7·3			−42	<0·001	72·6±10·12			−49	<0·001
III	α-methyl-*p*-tyrosine 200 mg/kg, orally	0	372± 7·0	+85	<0·001	+16	<0·001	163·1±6·95	+10	0·2–0·1	+14	0·2–0·1
IV		+	690±10·1			+115	<0·001	180·0±7·98			+25	0·05–0·02
V	α-methyl-dopa 120 mg/kg, i.p.	0	477± 8·3	−37	<0·001	+52	<0·001	200·4±7·80	−31	<0·001	+40	<0·001
VI		+	300± 7·0			−4	0·2–0·3	138·0±6·52			−4	0·7–0·6
VII	Disulfiram 400 mg/kg, orally	0	341± 5·9	−58	<0·001	+8	<0·02	114·0±9·21	−40	0·1–0·01	−21	0·05–0·02
VIII		+	144± 8·7			−54	<0·001	68·0±8·26			−53	<0·001
IX	Actinomycin D 500 mg/kg, i.p.	0	591±10·1	−30	<0·001	+88	<0·001	201·0±8·78	−44	<0·001	+40	<0·001
X		+	143± 9·3			+32	<0·001	112·0±6·92			−22	0·02–0·01

* 8 rats in each group.
† Drugs given either 18 h before death (0) or immediately before the start of the 18 h immobilization period (+).

prevent the fall after stress (−30%). The cardiac glycogen in the stressed rats was, however, still 120% higher than in the untreated, stressed controls (Group II).

Like cardiac glycogen, the blood glucose concentration fell by about 50% during prolonged immobilization. The drugs used affected the blood glucose in the same way as the cardiac glycogen, but to a lesser extent.

The histological investigation of the hearts of some animals subjected to 18 h restraint stress showed congestion of blood vessels, focal areas of fragmentation of muscle fibres and haemorrhage. The adrenal glands were enlarged, appeared to be depleted of lipids and showed focal degenerative changes. In α-MPT pretreated rats (Group II) the myocardium and the adrenal glands appeared normal. α-MPT did not prevent the stress-induced myocardial changes, however, the adrenals appeared normal except for some lipid depleted areas in four glands. The hearts of two rats treated with α-methyl-dopa (Group V) alone showed congestion of blood vessels and focal areas of fibre fragmentation and haemorrhage; the adrenal glands of one rat showed lipid depletion and in three rats haemorrhaging infarctions. Similar changes were observed in the myocardium and adrenal glands of the rats which were treated with α-methyldopa and stressed. After disulfiram alone two rats showed thickened vessel walls and another two showed vascular prominences in the heart. The adrenal glands appeared normal. Disulfiram did not prevent the tissue damage caused by stress. Actinomycin D (Group IX) produced fragmentation of the fibres and vascular prominences in the myocardium. The adrenal glands of three rats showed medullary hyperplasia. Similar changes were seen in the heart and adrenals when the animals were stressed in addition (Group X).

Discussion.—In the present experiments four drugs which are known to cause inhibition of enzymes involved in catecholamine synthesis were used in order to try to antagonize biochemical and histological changes caused by the stress of prolonged immobilization. All drugs used are reported to have long acting (up to 24 h) effects as seen by the duration of tissue noradrenaline depletion (Hess, Connamacher, Ozakap & Udenfriend, 1961; Maftre, 1965; Musacchio, Goldstein, Anagnoste, Poch & Kopin, 1966).

The decrease in myocardial glycogen seen after immobilization was not only prevented by α-MPT but the hearts of rats treated with this drug while stressed contained 85% more glycogen than those of unstressed rats treated with this drug. The fall in blood glucose seen after stress was also absent in the α-MPT treated rats. The protective effect of α-MPT on heart function might be due to its depressant action on behaviour. However, the drug did not prevent the occurrence of signs of tissue damage seen on histological examination of heart slices of stressed rats. None of the other drugs used were able to inhibit biochemical changes seen after stress.

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(*Received August* 24, 1971)