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Concentration of catecholamines in human cardiac muscle[1]

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SUMMARY A trihydroxyindole fluorescent technique was used to measure the concentration of catecholamines in heart muscle taken from 42 patients undergoing open heart surgery. Noradrenaline was the dominant catecholamine and its depletion in the failing heart was confirmed. The distribution of catecholamines within the heart was such that the highest value, 2·64 ± 0·68 μg/g, was found in the non-failing right atrium and the lowest, 0·53 ± 0·40 μg/g, in the failing left ventricle.

The effects of sympathetic stimulation on the heart are mediated by the neurotransmitter, noradrenaline. This catecholamine is present in relatively large quantities in heart muscle but its concentration is reduced in patients with heart failure (Chidsey *et al.*, 1963; Gertler *et al.*, 1970; DeQuattro *et al.*, 1973). Little is known about the concentration and distribution of catecholamines in normal heart muscle. The data presented in this paper suggest that the myocardial catecholamine concentration may be higher than had been supposed in the past and that the distribution is similar to that found in the hearts of experimental animals.

Patients and methods

Samples of heart muscle (atrial appendage or ventricular papillary muscle unless stated) were obtained from 42 patients undergoing open heart surgery at the National Heart Hospital. There were 25 men and 17 women; their ages ranged from 3 to 66 years (mean 43·5).

The patients were grouped either according to their surgical diagnosis or into those with, and those without, cardiac failure. The diagnosis of failure was established by the referring physician and all patients with this diagnosis had received treatment with digoxin and diuretics; patients with coronary artery disease had all received treatment with beta-adrenergic antagonists, usually propranolol. Other drugs which were given routinely included the premedicating agents papaveretum and scopolamine, the anaesthetics methohexitone and nitrous oxide, and the antibiotics gentamycin and flucloxacillin.

Catecholamines were assayed by a trihydroxyindole fluorescent technique similar to that described by Anton and Sayre (1962). The only important difference was that sodium carbonate was used to adjust the *p*H when the catecholamines were being adsorbed onto the alumina. The differential assay for adrenaline and noradrenaline was performed by developing the fluorescence at *p*H 6·5 and 3·5. No attempt was made to differentiate dopamine from noradrenaline. Some drugs cause fluorescence *in vitro* and thus interfere with this technique. Carruthers *et al.* (1970) showed that this did not apply to such drugs as propranolol, digoxin, and papaveretum, but this point was examined using a variety of blank preparations and the absence of extraneous fluorescence was confirmed (Petch, 1977).

The assays were performed in duplicate using healthy fat-free myocardium; the absence of fat and fibrous tissue was confirmed by histological examination in a random sample of specimens. The results are expressed as the mean ± standard deviation per gram myocardium (wet weight). Student's t test was used to determine the significance of the results except in Table 3 where the difference between the groups was examined using a one-way analysis of variance.

Results

In all samples the dominant catecholamine was noradrenaline; only small amounts of adrenaline were found usually amounting to about 10 per cent of the total. The depletion of total catecholamines

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Table 1 *Concentration of catecholamines in human right atrium μg/g (mean ± SD)*

	Number	*Total*	*Noradrenaline*	*Adrenaline*
No cardiac failure	11	2·38 ± 0·65	2·16 ± 0·63	0·22 ± 0·12 (n = 11)
Cardiac failure	19	1·17 ± 0·45	1·06 ± 0·51	0·11 ± 0·08 (n = 19)
Probability		< 0·001	< 0·001	NS

Table 2 *Right atrial catecholamine concentration—effect of atrial fibrillation μg/g (mean ± SD)*

	Number	*Total*	*Noradrenaline*	*Adrenaline*
Sinus rhythm	7	1·39 ± 0·34	1·34 ± 0·53	0·19 ± 0·09
Atrial fibrillation	12	1·08 ± 0·44	0·90 ± 0·42	0·14 ± 0·09
Probability		< 0·01	< 0·01	NS

Table 3 *Right atrial catecholamine concentration μg/g (mean ± SD)*

Diagnosis	*Number*	*Total*	*Noradrenaline*	*Adrenaline*
Coronary artery disease	7	2·64 ± 0·68	2·38 ± 0·67	0·25 ± 0·12
Open mitral valvotomy	2	2·06	1·81	0·25
Aortic valve replacement	7	1·85 ± 0·70	1·64 ± 0·52	0·21 ± 0·09
Mitral valve replacement	5	1·11 ± 0·49	1·01 ± 0·49	0·09 ± 0·04
Multiple valve surgery	5	1·04 ± 0·28	0·83 ± 0·37	0·21 ± 0·10
Atrial septal defect	3	0·97 ± 0·29	0·88 ± 0·27	0·09 ± 0·01
Probability		< 0·001	< 0·005	< 0·05

Table 4 *Right atrial catecholamine concentration—effect of surgery μg/g (mean ± SD)*

	Number	*Total*	*Noradrenaline*	*Adrenaline*
Before	4	2·29 ± 1·06	2·01 ± 0·99	0·27 ± 0·09
After	4	2·25 ± 0·81	1·92 ± 0·82	0·33 ± 0·09
Probability		NS	NS	NS

Mean bypass time = 63 minutes.

Table 5 *Left ventricular catecholamine concentration μg/g (mean ± SD)*

Diagnosis	*Number*	*Total*	*Noradrenaline*	*Adrenaline*
Mitral valve replacement	11	0·53 ± 0·40	0·46 ± 0·37	0·07 ± 0·04
Aortic valve replacement	1	1·89	1·57	0·32
Coronary artery disease	1	1·11	1·07	0·04
Hypertrophic cardiomyopathy	1	1·06	0·98	0·08
Hypertrophic cardiomyopathy + failure	1	0·27	0·24	0·03

Table 6 *Right ventricular catecholamine concentration μg/g*

Diagnosis	*Number*	*Total*	*Noradrenaline*	*Adrenaline*
Fallot's tetralogy	1	1·06	0·91	0·15
Ventricular septal defect	1	2·19	2·06	0·13
Pulmonary valve replacement	1	1·61	1·57	0·04
Hypertrophic cardiomyopathy	1	0·81	0·71	0·10
Trucuspid valve replacement + mitral valvotomy (+ failure)	1	0·65	0·62	0·03

and of noradrenaline in the right atrial samples taken from patients with cardiac failure was confirmed (Table 1). In the cardiac failure group the presence of atrial fibrillation was associated with a lower catecholamine concentration (Table 2). If the patients are grouped according to their surgical diagnosis (Table 3) then it can be seen that those patients with coronary artery disease, who were deliberately selected because they had normal cardiac function, had the highest values, whereas those undergoing surgery for haemodynamic reasons had lower values. In addition to those results given in Table 3, the right atrial catecholamine concentration was examined in patients with two other conditions. A patient with hypertrophic cardiomyopathy who did not have cardiac failure had values for the total noradrenaline and adrenaline concentrations of 2·29, 2·20, and 0·09 μg/g, respectively. In 2 patients with an atrial myxoma and cardiac failure, the corresponding values were 0·87, 0·75, and 0·12 μg/g, respectively.

The absence of any significant change in the right atrial catecholamine concentration during cardiopulmonary bypass (Table 4) allowed a direct comparison of the right atrial and left ventricular catecholamine concentration to be made in 9 patients undergoing mitral valve replacement. In these 9 patients the total right atrial and left ventricular catecholamine concentrations were 1·22 $\pm$ 0·59 and 0·66 $\pm$ 0·50 μg/g, respectively ($P < 0·01$); the corresponding figures for the noradrenaline concentrations were 1·11 $\pm$ 0·59 and 0·55 $\pm$ 0·43 μg/g, respectively ($P < 0·01$). A comparison of right and left atria in 3 patients showed the mean total catecholamine concentration in both chambers to be similar at 1·03 and 1·40 μg/g, respectively. In 2 patients samples were obtained from the right atrium and right ventricle and in both the catecholamine concentration was higher in the right atrium.

In the left ventricle the lowest catecholamine concentrations were found in those patients undergoing mitral valve replacement (Table 5). A higher value was found in one patient with coronary artery disease. Here the sample was taken from a rim of healthy myocardium surrounding an area of fibrous tissue that was being resected during a coronary bypass operation. Higher values were also found in 3 other patients whose samples were taken from the hypertrophied outflow tract of the left ventricle though in these 3 patients the lowest value was again found in the patient with cardiac failure. The same pattern was observed in the right ventricle (Table 6). The highest value was found in a child undergoing an elective closure of a ventricular septal defect in whom an anomalous parietal muscle band was removed; the lowest value was found in a patient with chronic rheumatic heart disease undergoing mitral and tricuspid valve surgery.

Discussion

The sympathetic neurotransmitter, noradrenaline, is the dominant catecholamine in all peripheral mammalian tissues (Holzbauer and Sharman, 1972). Though there are important species differences the catecholamine concentration in experimental animals is higher in the right heart as compared with the left, and higher in the atria relative to the ventricles (Shore *et al.*, 1958; Muscholl, 1959; Angelakos *et al.*, 1963). The distribution of catecholamines within the walls of the cardiac chambers is uniform, and there is good evidence to show that the noradrenaline is stored in the adrenergic neurones so that the noradrenaline concentration reflects the density of sympathetic innervation (Angelakos, 1965; Iversen, 1967).

Any sustained increase in cardiac work leading to hypertrophy or failure is associated with a depletion of the myocardial catecholamine stores. This was first recognised by Meerson in rabbits subjected to aortic constriction and has been amply confirmed for a variety of experimental animal models (Meerson *et al.*, 1963; Fischer *et al.*, 1965; Spann *et al.*, 1965; Vogel *et al.*, 1969). The catecholamine depletion affects all cardiac chambers and is related to the severity and duration of the haemodynamic load; it appears to be peculiar to the adrenergic neurones of the myocardium since other tissues are not affected. In the experiments of Vogel *et al.* (1969) the noradrenaline depletion was shown to be reversible.

In patients with cardiac failure the myocardial catecholamine concentration is reduced. Chidsey *et al.* (1963) found a mean value of 0·53 μg/g in right atrial specimens taken from patients with chronic congestive failure, compared with a mean value of 1·82 μg/g in a 'control' group of patients undergoing surgery for such lesions as atrial and ventricular septal defects. Gertler *et al.* (1970) and DeQuattro *et al.* (1973) reported that the noradrenaline depletion was more pronounced in those patients with more severe symptoms. However, all these patients from previous studies had some haemodynamic lesion and only those patients with coronary artery disease in the present study had normal cardiac function. If their right atrial catecholamine concentration of 2·64 μg/g approaches the normal value for human right atrium, it implies a rich sympathetic innervation. The administration of beta-adrenergic antagonists is unlikely to have affected this value as the evidence

from experimental animals suggests that propranolol has little effect on the myocardial noradrenaline concentration (Westfall, 1967; Lemner and Saller, 1974).

The variation in the right atrial catecholamine concentration found in the present study is best explained by postulating that in man, as in experimental animals, a sustained increase in cardiac work is associated with progressive noradrenaline depletion. Thus, those patients undergoing aortic valve replacement had higher values than those undergoing mitral valve replacement because they were operated upon earlier in the natural history of their disease. Similarly patients with an atrial septal defect, though not in 'cardiac failure', had had at least a twofold increase in blood flow through the right heart from birth and consequently low right atrial catecholamine concentrations. This hypothesis may also explain the variation in catecholamine concentration found in ventricular muscle. Nevertheless, the left ventricular levels were much lower than the right atrial. Since this distribution is similar to that found in experimental animals, it seems likely that the right atrium is more richly supplied with adrenergic neurones than the left ventricle.

The cause of the noradrenaline depletion in the failing heart is not clear but a number of factors may be involved including depressed synthesis, as shown by reduced tyrosine hydroxylase activity (Pool *et al.*, 1967; DeQuattro *et al.*, 1973), impaired neuronal uptake of noradrenaline (Spann *et al.*, 1965; Petch and Nayler, 1979), and excessive noradrenaline release after intense sympathetic stimulation. Furthermore, the administration of digoxin may exacerbate the noradrenaline loss since it is a known inhibitor of the neuronal uptake process (Iversen, 1973) though its effect on the myocardial catecholamine concentration in experimental animals with normal hearts is slight (Forster and Rosler, 1967; Harvey, 1975). These abnormalities may be regarded as part of a general depression of sympathetic nerve function which occurs in the failing heart. This depression is also reflected in the physiological behaviour of the sympathetic neurone since the failing heart is unable to respond normally to sympathetic stimulation despite its ability to respond to exogenous catecholamines (Covell *et al.*, 1966; Goldstein *et al.*, 1975).

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