Catecholamines in the Adrenals of August and Wistar Rats with Acute Emotional Stress

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Acute emotional stress caused by immobilization and cutaneous electrical stimulation increases the relative weight of adrenals in Wistar rats and decreases it in August rats. The epinephrine and norepinephrine contents of the adrenals in control and stressed August rats are higher than in Wistar rats. Acute stress lowers the levels of these biogenic amines in the adrenals of both strains, particularly in Wistar rats. The left adrenal gland of control and stressed August rats, but not of Wistar rats, has a higher content of biogenic amines than the right, and both adrenals of stressed August rats contained higher dopamine concentrations than those of stressed Wistar rats. Presumably, epinephrine and norepinephrine are resynthesized in the adrenals of stressed August rats at higher rates than they are released from these glands, while the adrenals of Wistar rats respond to stress by rapidly releasing these catecholamines and resynthesizing them at a slow rate.

Key Words: *emotional stress; August rats; Wistar rats; adrenals; epinephrine; norepinephrine; dopamine*

Humoral and hormonal mechanisms of emotional stress (ES) involve activation of adrenocorticotropic hormone synthesis [11] followed by elevation of catecholamines (epinephrine and norepinephrine) in the blood as a result of their release by the adrenals.

Different rat strains differ in the susceptibility to ES [5]. For example, Wistar rats are less susceptible than August rats. The epinephrine content is higher in the hypothalamus of Wistar rats compared with than of August rats, and conflict immobilization cause smaller changes in this parameters in Wistar rats [6].

Susceptibility to ES may also vary widely among different animals of the same species [5]. Genetically determined activity of enzymes catalyzing catecholamine synthesis, primarily dopamine β-hydroxylase, is an important determinant of individual resistance to ES [3].

The aim of the present study was to evaluate and compare the effects of acute ES on catecholamine, (epinephrine, norepinephrine, and dopamine) levels in the adrenals of August and Wistar rats.

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MATERIALS AND METHODS

Male August (222.6±5.0 g; n=19) and Wistar (260.4±5.9 g; n=14) rats were used. They were kept at 20-22°C, 4 animals in cage, with natural light and free access to water and food.

Ten August and seven Wistar rats were immobilized for 1 h with subsequent cutaneous electrical stimulation. For this purpose a rat was placed in a plexiglass tube (16.5 cm long and 5.5 cm internal diameter) where the base of its tail was stimulated with alternating current for 30 or 60 sec using a stochastic scheme (4-6 V, 50 Hz, pulse duration 1 msec). Control group consisted of intact August (n=9) and Wistar (n=7) rats.

After a 1-h immobilization, the rats of both groups were decapitated, and the adrenals were dissected, weighed, homogenized in 1 ml of 0.4 M $HClO_4$ (without removing the connective tissue capsule), and stored at -20°C.

Epinephrine, norepinephrine, and dopamine were measured by high-performance liquid chromatography (125 mm×3 mm I.D. column; Nucleosil 100C18; 3

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μM). The mobile phase consisted of 0.1 M phosphate, 5% methanol, 240 mg/liter octyl sulfate, and 100 mg/liter EDTA, pH 2.5; flow rate, 0.7 ml/min; pressure, 150 bars.

The results were evaluated by multivariational analysis of variance (stress/control×left adrenal/right adrenal×rat strain), followed by a multiple comparison of the groups. Numerical values in the text and tables are expressed as the mean±standard error of the mean.

RESULTS

Body weight of Wistar rats was significantly ($p<0.0001$) higher than that of August rats, both in the control (263±7 and 223±8 g, respectively) and ES-exposed (258±10 and 222±7 g, respectively) groups. Emotional stress led to a decrease in body weight.

The relative weight of adrenals (Table 1) in control August rats was higher than in Wistar controls (the left and right adrenals weighed 1.28 and 1.49 times more, respectively). The relative weight of the left adrenal was 1.33 times greater in Wistar rats and 1.14 times greater in August rats than that of the right adrenal.

In Wistar rats, ES increased the relative weight of right and left adrenals by 1.45 and 1.18 times, respectively, compared with the controls. In the stressed August rats, the relative weight of the right adrenal was 1.06 times lower and that of the left 1.07 times lower than in the controls (statistically insignificant differences). The epinephrine level in the left adrenal was 1.1 times higher than in the right adrenal in August rats and 1.06 times higher in Wistar rats (Table 2).

In stressed August rats, the epinephrine level was 1.15 times lower in the right adrenal and 1.1 times lower in the left than in the controls, and the content of epinephrine in the left adrenal was 1.15 times higher than in the right.

In stressed Wistar rats, epinephrine levels were also lower than in the controls: by 1.29 times in the right adrenal and 1.33 times in the left; there were virtually no differences in epinephrine content between the left and right adrenals.

After ES, the epinephrine content of left and right adrenals of August was 1.5 and 1.33 times higher, respectively, than in Wistar rats.

Norepinephrine levels in the adrenals of August rats were also higher. In August controls, the level was 1.54 times higher in the right adrenal and 1.27 times higher in the left than in Wistar controls. In the latter, the left adrenal contained 1.29 times more norepinephrine than the right, whereas the left and right adrenals of August controls contained this corticosteroid at almost equal levels (Table 2).

In stressed August rats, the norepinephrine content in the right adrenal was 1.13 times and in the left adrenal 1.04 times lower than in the controls (Table 2), and their left adrenal contained 1.16 times more of this biogenic amine than did the right.

In the stressed Wistar rats, the left adrenal contained 1.25 time less norepinephrine and the right 1.1 times less as compared to the controls, and the difference between its levels in the left and right adrenals was insignificant.

After ES, the norepinephrine content in left and right adrenals of August rats was 1.52 and 1.5 times was higher, respectively, than in Wistar rats.

Dopamine levels in the adrenals of control August rats were insignificantly higher than in Wistar controls, and no significant differences were found in dopamine content between the left and right adrenals in the rats of these two strains (Table 2).

After ES, dopamine levels in both adrenals of August, but not of Wistar rats, were increased significantly by 1.3 times in the left adrenal and 1.35 times in the right *vs.* 1.19 and 1.15 times, respectively, in Wistar rats (Table 2). In stressed August rats, the left adrenal contained 1.31 times more dopamine and the right 1.29 times more than the respective adrenals of stressed Wistar rats. No significant differences in dopamine content between the left and right adrenals of stressed August or Wistar rats were found.

Thus, acute ES increased the relative adrenal weight in Wistar rats and decreased it in August rats. We also observed reductions in the adrenal weight in rats stressed by water immersion [4]. This finding does not agree with classical Selye's triad, which may be due to the polyphasic nature of hypothalamo-

TABLE 1. Relative Adrenal Weights (mg/100 g Body Weight) in August and Wistar Rats (*M*±*m*)

Rats	Left adrenal		Right adrenal	
	control rats	stressed rats	control rats	stressed rats
August	8.73±0.63^{+}	8.09±0.71	7.58±0.42^{++}	7.18±0.53
Wistar	6.78±0.41	8.01±0.30	5.13±0.38**	7.40±0.33$^{\circ\circ}$

Note. Here and in Table 2: $^{+}p<0.05$, $^{++}p<0.01$, $^{+++}p<0.001$ in comparison with Wistar rats; $^{*}p<0.05$, $^{**}p<0.01$, $^{***}p<0.001$ in comparison with the left adrenal; $^{\circ}p<0.05$, $^{\circ\circ}p<0.01$, $^{\circ\circ\circ}p<0.001$ in comparison with the control group.

TABLE 2. Epinephrine, Norepinephrine, and Dopamine Levels (nmol) in the Left and Right Adrenals of August and Wistar Rats (*M±m*)

Rats	Epinephrine		Norepinephrine		Dopamine	
	left	right	left	right	left	right
Control						
August	154.82±6.63**	140.45±5.99*****	46.25±1.57***	43.50±1.51***	1.34±0.13	1.25±0.09
Wistar	124.94±11.91	118.09±12.86	36.61±3.23	28.34±2.99***	1.12±0.16	1.14±0.13
Stressed						
August	140.76±7.20***°	122.45±8.05******°°	44.46±1.86***	38.50±3.22*******°	1.74±0.17**°°	1.69±0.21**°°
Wistar	93.63±9.49°°°	91.71±8.81°°°	29.16±4.25°°°	25.70±4.23	1.33±0.12	1.31±0.14

adrenocortical system [7] or inadequate stimulation of adrenocortical activity during ES.

Epinephrine and norepinephrine levels in the adrenals of control August rats were higher than in Wistar controls. Acute ES decreased adrenal levels of these biogenic amines in both stains, particularly in Wistar rats, the decrease being greater in the right adrenal of August rats and in the left adrenal of Wistar rats. As in the control animals, the levels of both catecholamines in the adrenals of stressed August rats were higher than in stressed Wistar rats.

Stress elevates peripheral blood acetylcholine [2]. It has been shown that acetylcholine and its agonists stimulate in a dose-dependent manner the secretion of catecholamines [13] and their release [12] by the adrenals. Lowered epinephrine and norepinephrine levels in the adrenals of stressed rats in our study were due presumably to elevation of blood concentration of acetylcholine and subsequent enhancement of catecholamine release by the adrenals.

Emotional stress rises blood concentration of interleukin-6, tumor necrosis factor, and interleukin-1 [9] which activate the hypothalamus-pituitary-adrenal axis [10]. Interleukin-1β stimulates the release of glucocorticoids [8] and catecholamines from the adrenals [14]. This is probably another mechanism by which catecholamine levels are lowered in the adrenals during ES.

Asymmetry in the immune and endocrine systems has been demonstrated [1]. In our study, the levels of biogenic amines in the left adrenal of August rats, which are less resistant to ES than Wistar rats, were higher than in the right adrenal, both in the control and stressed groups. In both groups of Wistar rats, which are resistant to stress, the catecholamine levels in left and right adrenals were practically the same. Presumably, the differences in epinephrine and norepinephrine levels between the adrenals in rats with high resistance to ES are less pronounced than in rats with low resistance to ES.

Although ES lowered epinephrine and norepinephrine levels in Wistar rats to a greater extent than in August rats, the increase in the dopamine concentration (an indicator of the catecholamine synthesis rate) in the adrenals was greater in August rats, and dopamine levels in both adrenals of these rats were higher than in Wistar rats. During ES, the rate of catecholamine resynthesis in the adrenals of August rats is probably higher than the rate of their release from these glands. In contrast, Wistar rats may respond to ES by fast release of epinephrine and norepinephrine from the adrenals, where these catecholamines are resynthesized at a slower rate.

The mechanism(s) by which acute ES influences the adrenal glands will be investigated in further studies.

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