

EFFECTS OF HEAT-STRESS IN RATS*

III. ON PREGNANT RATS, MATERNAL ORGANS AND FOETAL TISSUES

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Summary: Exposure of intact and ovariectomized rats to high ambient temperature (34.4°C, 60-65% relative humidity) resulted in reduction of their body weight, increased water consumption. The feed consumed which was lowered by 25% initially, did not alter later. Intact rats were also subjected to a still higher ambient temperature of 36.6°C and 70 to 75% RH and the effects were observed.

When compared with 'controls', no significant changes in the weights of ovaries or pituitary gland were seen due to heat stress in intact rats. But, the latter weighed heavier in ovariectomized rats. Adrenal glands were found enlarged and weighed more both in intact and ovariectomized rats. The average foetal weight due to high ambient temperature in intact rats was lower, but in ovariectomized rats it weighed more. Similarly, the average weight of the placenta was lower in intact rats but higher in ovariectomized rats due to thermal stress.

Key words: heat-stress ovariectomized rats pituitary weight
adrenal hyperplasia foetal weight placental weight

INTRODUCTION

The high ambient temperature having a critical period to cause deleterious effects on reproduction and basic causative factor(s) were reported earlier. In this study, the effects of heat stress on pregnant rats in terms of changes in body weight, feed and water consumption, weights of maternal pituitary gland, adrenal glands and ovaries (if *in situ*) are elucidated besides the effects of the thermal stress on the foetal tissues.

MATERIALS AND METHODS

A number of 166 intact (ovaries *in situ*) pregnant rats that were heat stressed under Trial 1 and Trial 2 in the first paper published by us (3) and 134 ovariectomized rats in whom gestation were maintained with various levels of gonadal hormones besides some of them being heat stressed in another work (Experiment 1) and reported by us later (4) formed the total number of animals used. The ambient temperature and relative humidity (RH) for both the control and experimental groups remained the same as in the previous studies (3, 4). Initial weights of the rats and their

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weights on the day of killing were made, besides they were weighed at random each day of the study period. The animals were euthanized on day-18 (day-0=day of mating) when the weights of the pituitary, adrenal glands and ovaries (if *in situ*) from these bred rats were taken. The weight of the foetus and placenta of each rat was also taken and recorded separately.

Results were analysed statistically using student 't' test for unequal number.

RESULTS AND DISCUSSION

Rats exposed to high ambient temperature revealed heat stress through an increase in their average rectal temperature to 40.5°C from 37.2°C which was observed in about two hours after exposure to thermal stress.

Heat stress was found to result in the reduction of the body weight of the rats. On an average, rats left at control temperature gained 75 g during the experiment, while the heat stressed animals lost 86 g in them. But it was noticed that animals exposed only for a short duration of 24 hours did not lose their body weight significantly and also they regained the slight loss very quickly. No differences were noticed between the intact bred rats and ovariectomized rats in this parameter.

The feed consumption in heat stressed rats was 25% less than the control for the first few days of the experiment, however there was no significant difference subsequently. This was probably due to the adaptation effort on the part of the animals to maintain normal feeding habits even under high ambient temperature. The water consumption was found to be increased by 80% in those rats exposed to high altered temperature during the study period.

Maternal organs : The average weight of maternal organs (ovaries, pituitary and adrenals) from heat stressed intact rats (ovaries *in situ*) are given and compared with 'control' in Table I and II. Similar data from heat stressed ovariectomized rats and the comparison with 'ovariectomized control' is shown in Table III.

Ovaries : Though the weights of the ovaries were different within the rats whether they were heat stressed or not, no significant difference was noticed between the control and thermally stressed rats. The variations in the weights of the ovaries is attributed due to differences in corpora lutea number.

Pituitary gland : Weight of the organ in the intact 'control' and heat stressed rats did not vary significantly. But, the weight of the gland in ovariectomized heat stressed rats were more than the ovariectomized control (1.21 and 1.03 mg respectively).

Adrenal gland : Thermal stress seemed to have resulted in heavier adrenals both in the intact bred and ovariectomized bred rats (Tables I, II and III), especially when the exposure to heat was continued for a longer duration, particularly up to day-18. But, a mere 24 hours

thermal stress either on day-0 or -1 was not sufficient to induce adrenal enlargement at day-18 (Table II). When the weights of the adrenal glands were compared between the intact (ovaries *in situ*) bred and ovariectomized bred rats, the following informations were elucidated: 1) Under 'control' ambient temperature adrenal weighed less in the intact rats than in ovariectomized rats (66.3 and 71.8 mg respectively). 2) Heat stress had same effect on the weight of adrenal both in intact and ovariectomized animals (79.4 and 76.5 mg respectively). The weights were altered to higher values, but the intact rats had heavier adrenals than ovariectomized animals (79.4 and 76.5 mg respectively).

TABLE I: The effect of heat stress (34.4°C, 60 to 65% RH) on the various maternal organs of intact rats, foetus and placenta^a.

Exposed to heat stress day (s)	No. of rats	Maternal organs				
		Av. wt. of ovaries ±SE (mg)	Av. wt. of pituitary ±SE (mg)	Av. wt. of adrenals ±SE (mg)	Av. wt. of foetus ±SE (g)	Av. wt. of placenta ±SE (g)
Control	51	98.60 ±3.8 ^b	10.32±0.26	66.35±1.6 ^b	1.79±0.03(462) ^c	0.95±0.01
8 to 18	35	100.20±4.7	10.07±0.29	77.45±1.6**	1.65±0.04(338)	0.80±0.02**
0 to 18	10	99.60 ±5.9	11.40±0.52	79.40±2.7*	1.57±0.04(32)**	0.72±0.04**
0 to 8	10	104.10±8.7	11.10±0.34	77.30±6.6	1.49±0.03(23)**	0.86±0.06**
1 to 18	10	89.80 ±2.8	10.60±0.48	80.40±3.4*	1.72±0.02(116)*	0.75±0.05**
0	13	97.30 ±6.7	10.90±0.35	66.20±3.6	1.60±0.04(93)**	0.84±0.02*
1	13	98.20 ±4.9	11.10±0.31	68.2 ±2.4	1.38±0.06(115)**	0.91±0.03

^aAll weights taken 18 days after mating

^bTotal of paired organs

^cFigures in parenthesis indicate the total number of foetuses

*Different than control (P<0.05)

**Different than control (P<0.01)

However, comparison of the adrenal gland weight between ovariectomized control and ovariectomized heats stressed, showed that the former had lighter weights compared to the latter (71.8 and 76.5 mg respectively). The adrenal enlargement observed in this study agreed with the established fact of 'Adaptation syndrome' (5), occurring through adrenal cortical hyperplasia at high body temperature (1).

The interesting observation made in this study was that, while ovariectomized and heat stressed rats had both heavier adrenals and pituitary glands, heat stress in intact rats resulted only in enlarged adrenals and the pituitary gland remained unaltered. Though, histochemical studies were not done by us, it is suggested that the presence of ovaries *in situ* in the latter group,

had in some way inhibited the hyperplasia of pituitary without affecting the activity of the pituitary cells resulting in the hyperplasia of the adrenals. In other words, it is possible to presume that ACTH producing basophils of the pituitary in this group were kept hyper-active during the thermal stress.

TABLE II: The effect of exposure of bred intact rats to 36.6°C and 70 to 75% RH on weight of the various maternal organs, foetus and placenta^a.

Day of exposure ^b	Number of rats	Maternal organs				
		Average weight of ovaries ±SE (mg)	Average weight of pituitary ±SE (mg)	Average weight of adrenals ±SE (mg)	Average weight of foetus ±SE (g)	Average weight of placenta ±SE (g)
Control	51	98.60±3.8	10.32±0.26	66.35±1.6	1.79±0.03(462) ^c	0.95±0.01
0	12	106.71±3.0	11.30±0.30	63.10±3.3	1.47±0.03(8)**	1.50±0.30
1	12	109.26±6.0	10.90±0.20	73.9 ±4.1	1.37±0.06(56)**	1.00±0.08

^aAll weights taken 18 days after mating.

^bRats were placed in a chamber at 36.6°C and 70 to 75% RH between 9.00 and 10.00 AM on the day shown and removed 24 hours later.

^cFigures in parentheses indicate the total number of foetuses. Fifty-one rats were pregnant in the control group, 4 in the day-0 group and 11 in the day-1 group.

**Different than control ($P < .01$).

Foetus and Placenta : The average weight of the foetus and placenta due to different stages of heat stress during pregnancy is shown in Table I, while Table II reveals the same at a still higher ambient temperature. Similar data for the ovariectomized rats is provided in Table III.

TABLE III: The effect of elevated ambient temperature on weight of the various maternal organs, foetus and placenta in ovariectomized rats^a.

Environmental temperature and relative humidity (RH)	No. of rats	Maternal organs			
		Average weight of pituitary ±SE (mg)	Average weight of adrenals ±SE (mg)	Average weight of foetus ±SE (g)	Average weight of placenta ±SE (g)
21.1°C and 60 to 65% RH	67	9.90±0.19	71.80±1.8	1.70±0.03(358) ^b	1.03±0.02
34.4°C and 60 to 65% RH	67	10.8±0.20*	76.5 ±1.3*	1.87±0.03(291)*	1.21±0.03*

^aAll rats were ovariectomized on day-4, rats were then injected twice daily with progesterone and estrone (Table I(4) until killed at day-18. All weights were taken at day-18 (day 0 = day of mating).

^bFigures in parentheses indicate the total number of foetuses.

*Different than rats at 21.4°C and 60 to 65% RH ($P < .05$).

Heat treatment significantly depressed foetal weights in all intact groups except those that were treated from day-1 to day-18. But, it appeared that there was no correlation between

foetal weights and duration of heat stress. As a matter of fact, rats exposed to the too high ambient temperature merely for 24 hours early in gestation (either on day-0 or day-1) had lighter foetus than rats stressed for longer periods or those in which heat exposure began on day-8 (Tables I and II). The heat stressed rats had on an average 1.65 g as the foetal weight, where as foetus of 'control' rat weighed 1.79 g at 34.4°C and 60 to 65% RH, while the same was 1.47 g at 36.6°C and 70 to 75% RH (Tables I and II). It was observed however, that heat stress significantly increased foetal weight in ovariectomized rats. The average foetal weight was 1.87 g compared to 1.70 g noticed in the corresponding control (Table III). Our results were different from the reports of (2), where the foetal weight was less in ovariectomized heat stressed rats.

Reduction of placental weights were seen due to thermal stress in intact rats at 34.4°C and 60 to 65% RH. While the 'control' had on an average 0.95 g, the heat stressed rats showed on an average 0.86 g as the placental weight. The weight of the placenta at a still higher ambient temperature (36.6°C and 70 to 75% RH) was 1.50 g, which though was higher than the 'control' was not statistically different (Table I and II). With regard to the placental weight of heat stressed ovariectomized rats, our results showed significant increase. While the thermally stressed rats had on an average 1.219 g as placental weight the corresponding 'control' showed on an average of 1.03 g as the placental weight. Even, (2) have reported that ovariectomized heat treated rats had heavier placental weight.

From these results, it is suggested that the ovariectomized rats on heat stress having a heavier foetus and placenta compared to intact rats may be due to the anabolic effects of the exogenous gonadal hormones administered. They have produced these effects, though they failed to reduce embryonic mortality rate (4).

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