HEPATIC AND CARDIAC WEIGHT ADJUSTMENTS OF YOUNG FEMALE CHICKS SUBJECT TO ESTRONE AND HYPERGRAVITY

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Abstract. Rhode Island Red female chicks, 2 weeks post hatching, were maintained for 2 weeks at either earth gravity or 2 g hypergravity. Control animals were injected with 0.2 ml saline and estrone treated groups with 0.2 mg or 0.4 mg of estrone daily. Animals were sacrificed following the last injection on day 15 and their livers and hearts were removed immediately, dissected free of connective tissue, weighed and frozen. Exposure to the effects of the 2 g hypergravity for 2 weeks resulted in decreased total body weight regardless of whether the animal was supplemented with or lacked hormonal treatment. Estrone at 0.2 or 0.4 mg had little effect on the non-centrifuged bird's weight. Liver weight was increased with estrone treatment in the normogravity animals but not in animals exposed to 2 g, except when treated with 0.4 mg estrone. Heart weight was unaffected by the experimental procedures used.

Clavert and Randavel (1947) have shown that estrogens increase both hyperplasia and hypertrophy of the pigeon liver. Yu and Marquardt (1973) observed that estrogen induced enlarged livers in pullets by stimulating hyperplasia, cellular hypertrophy and increasing the lipoprotein fraction of the organ, while Kudzman et al (1973) considered that the estrogen-induced hepatic lipogenesis in birds resulted from larger incorporation of acetate into triglycerides by the animal's liver. Few studies have attempted to reveal the interaction between steroid hormones and organs of animals exposed to acute and/or chronic hypergravity states. The present study was undertaken to determine the effects of the steroid estrone, administered at relatively high doses, and the 2 g hypergravity state on liver, heart and total body weight of young female birds at 2 weeks post hatching. Young chicks were selected for this study because they are virtually free of estrogen during the first few weeks post hatching, as indicated by Kudzman et al (1973).

MATERIALS AND METHODS

Ninety-six healthy Rhode Island Red female birds were exposed to the following treatments, for a 2 week period, at 2 weeks post hatching:

- Daily injection of 0.2 ml saline, controls (n=19)
- 0.2 mg estrone in saline daily (n=19)
- 0.4 mg estrone in saline daily (n=14)
- 2 g hypergravity with 0.2 saline daily (n=18)
- 2 g hypergravity with 0.2 mg estrone in saline daily (n=14)
- 2 g hypergravity with 0.4 mg estrone in saline daily (n=12)

Chicks were maintained in cages at 27°C with a 12 hr dark and 12 hr light cycle. Centrifuged animals were placed 3 to a cage under similar temperature and light conditions and mounted on an 11 ft (radius) centrifuge. A free-swinging yolk assembly held the cages so that the animals were subjected to a resultant centrifugal and gravitational force of 2 g when rotated at 22 rpm.

Animals were fed Purina Startina Mash and watered ad libitum. The centrifuge was stopped once daily for about 20 minutes to record body weights, and distribute food, water and estrone injections as required. The injection site for the hormone, the posterior thigh muscles of the left or right leg, was alternated daily for 14 days. The daily hormone treatment consisted of 0.2 mg or 0.4 mg of estrone per 100 gram body weight (Parke-Davis, Theelin) in 0.2 ml saline (Travenol, Bacteriostatic Sodium Chloride U.S.P.). Controls were injected with 0.2 ml of sterile isotonic sodium chloride only.

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FIGURE 1. Effects of estrogen treatment and hypergravity on daily body weight of chicks from 2 to 4 weeks post hatching.
Animals were sacrificed by decapitation (Harvard Apparatus, decapitator for small animals) after 2 weeks of experimentation. Livers and hearts were removed immediately after sacrifice, dissected free of connective tissue, weighed and frozen. The data were analyzed statistically by an F test and a Scheffe test (Glass and Stanley 1970).

RESULTS

Compared to controls, animal body weight was significantly decreased (p < 0.001) by the 2 g hypergravity state regardless of whether the animal was provided with or lacked a superimposed hormonal treatment. Hypergravity-exposed animals, with or without estrone treatment, showed a similar and significant weight loss (p < 0.001) compared to earth gravity animals treated with 0.2 mg or 0.4 mg estrone. Body weight differences between untreated or centrifuged control animals and their estrone treated normo- or hypergravity counterparts were not significant (figs. 1 and 2).

Liver weight of non-centrifuged birds was significantly increased by estrone at either 0.2 mg (p < 0.01) or 0.4 mg (p < 0.001) levels. The 0.4 mg estrone level was more effective in increasing the

Figure 2. Comparative effect of estrogen treatment and hypergravity on body weight, liver weight and heart weight at 4 weeks post hatching (after the 2 week experimental period) (••• = p<0.01, •••• = p<0.001).
weight of the liver than the lower hormonal level in both normogravity and hypergravity-exposed animals (p<0.01). Liver weights of hypergravity-exposed animals without hormonal treatment and hypergravity animals maintained at daily estrone levels of 0.2 mg were not different from those of control animals. Hypergravity significantly decreased liver weight of chicks lacking hormonal treatment compared to hepatic weight of earth gravity animals supplemented with either 0.2 mg (p<0.01) or 0.4 mg (p<0.001) of estrone. Differences in liver weights of estrone-treated earth gravity and hypergravity exposed animals subjected to estrone treatment were not significant. The combination of estrone treatment (0.4 mg) and hypergravity increased liver weight significantly (p<0.02) over that of the hypergravity state alone. Heart weight was unaltered by the animals subjected to the 2 hormonal levels were also exposed to the hypergravity state (p<0.05). The liver/heart weight ratio was decreased by the hypergravity state without hormonal treatment (p<0.05) as compared to that of earth gravity animals exposed to either 0.2 mg or 0.4 mg estrone treatment (table 1). High estrone levels (0.4 mg) increased liver weight of chicks maintained at earth gravity by 45% and for the animals subjected to a 2 g environment by 30%. The lower level of estrone (0.2 mg), although increasing the liver weight of earth gravity chicks by 30%, had little or no effect on the weight of the organ under centrifuged conditions. Livers of the estrogen treated animals were larger than those of the controls, and when palpated were relatively soft. Livers from non-estrone treated chicks appeared reddish-brown as compared to the yellowish color of the estrone treated group.

**DISCUSSION**

The ability of estrogenic compounds to increase total animal body weight by inducing sodium and water retention and increasing deposition of subcutaneous fat is known (Goodman 1974). The daily administration of either 0.2 mg or 0.4 mg estrone to non-centrifuged birds, for 2 weeks, did not result in a significantly altered total body weight, suggesting that estrone treatment, as given, was ineffective in changing total body weight of young chicks at 4 weeks post hatching. The significantly decreased total body weight found in hypergravity-exposed animals was probably due to the stressful experimental procedure involving either estrone or centrifugation alone or combined (fig. 2).

The ratio of body/liver weight was significantly decreased (p<0.001) for all experimental animals. The body/heart weight ratio was decreased by centrifugation alone (p<0.05) or combined with either 0.2 mg or 0.4 mg of estrone treatment (p<0.001). The ratios of these 3 experimental animal groups were significantly decreased (p<0.001) compared to earth gravity animals receiving 0.2 mg of estrone treatment. The liver/heart weight ratio was increased by 0.2 mg (p<0.05) and 0.4 mg estrone treatment (p<0.01) in the earth gravity group of animals. It was similarly increased when

| TABLE 1 |
|---|---|
| **Comparative effect of estrogen treatment and hypergravity on the ratio of body weight to liver and heart weight, and liver weight to heart weight at 4 weeks post hatching.†** |  |
| **Ratio of Weight** | **Earth Gravity** | **Hypergravity** |
|  | Saline controls | Estrone | Estrone | Estrone |
|  |  | 0.2 mg | 0.4 mg | Saline | 0.2 mg | 0.4 mg |
| Body/Liver | 36.2 | 28.7* | 26.1* | 29.4* | 23.3* | 21.8* |
| Body/Heart | 114.1 | 120.0 | 106.3 | 98.9** | 93.8* | 89.6* |
| Liver/Heart | 3.2 | 4.2** | 4.2** | 3.3 | 4.0** | 4.1** |

†After the 2 week experimental period.
*Significant at 0.001 to 0.005 level.
**Significant at 0.01 to 0.05 level.
factors induced by prolonged centrifugation (Negulesco 1976). This finding is in agreement with studies of Smith and Kelly (1963) who exposed birds to 1.5 to 3.0 g hypergravity states. The absence of an estrogenic effect upon total body weight of hypergravity animals was similar in nature to that suggested for earth gravity chicks by Negulesco and Clark (1976) and Negulesco (1976). It is interesting to note that all centrifuged animals, regardless of hormonal treatment, lost, on the average, 30 g of body weight during the initial 48 hours of centrifugation. Following this initial induced weight-loss period, centrifuged animals remained lighter and showed increased weight deviation from earth gravity animals during the 2 week experimental period (fig. 1). Weight differences were probably due to the increased demand of energy required by the hypergravity-exposed animals to maintain posture in the hyperkinetic state. Water and food consumption by centrifuged animals was equivalent to that of earth gravity animals with little or no waste found on the floor of the cage. It is worth mentioning that over the 2 week experimental period we had no animal mortality due to continuous centrifugation such as that reported by Smith et al (1959) for birds exposed to 1.5 to 3.0 or higher g forces.

Estrogenic side effects in non-centrifuged birds, as studied over the years, indicate hepatic hypertrophy and hyperplasia (Clavert and Randavel 1947), increased hepatic lipoprotein (Luskey et al 1974; Talwar et al 1973; Yu and Marquardt 1973), and hepatic lipogenesis (Balnave 1969; Kudzman et al 1973; Ranney and Chaikoff 1951). The increased hepatic weight of our earth gravity animals due to estrone treatment concurs with the hepatomegaly described by most of the investigators mentioned above. Hepatic weight of earth gravity animals was significantly elevated by the administration of 0.2 mg, and more so by 0.4 mg, estrone treatment, suggesting a dose-dependent response. Estrone administration not only changed liver weight but also affected gross morphology. High estrone levels (0.4 mg) resulted in hepatomegaly, yellow hepatic discoloration, loss of organ firmness and spongy stroma in hormone treated animals (except 2 g hypergravity chicks).

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LITERATURE CITED