The role of blood glucose in activity related self-starvation¹

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Rats in the experimental group lived in activity wheels on a 23-h deprivation schedule, while those in the control group lived in laboratory cages on the same feeding schedule. Blood-glucose determinations made on the 1st, 3rd, and 6th days of deprivation indicated that, while both groups were hypoglycemic on Days 1 and 2, by Day 6 the experimental Ss were still hypoglycemic, while control Ss' blood-glucose levels approached normal ad lib levels. The results are in opposition to the hypothesis of glucocorticoid involvement in activity-related self-starvation.

Routtenberg & Kuznesof (1967) reported that rats confined to activity wheels and placed on a 23-h food-deprivation schedule progressively lost weight yet dramatically increased their activity until starvation occurred. All control Ss, living on the deprivation schedule and in home cages, were able to adapt and maintain weight. In addition, the experimental Ss ate significantly less than did controls.

Routtenberg (1968) reported that adaptation to the deprivation schedule prior to introduction of the above conditions was successful in mitigating the self-starvation of experimental Ss. Leveille & O'Hea (1967) have shown that animals adapted to a 22-h food-deprivation cycle run significantly less than do animals on an ad lib schedule. Adaptation to the wheel, however, was found by Routtenberg (1968) to ameliorate the effect on initial food-intake suppression but not on self-starvation.

Routtenberg (1968) postulated that two factors are contributing to the self-starvation phenomenon: "deprivation stress" and "novelty stress." It was hypothesized that the endocrine system may play an important role and, more specifically, that "self-starvation is brought about by an adreno-cortical response driven by the posterior hypothalamus [p. 238]." The mechanism would presumably operate through the hypothalamic control over the adenohypophysis via the hypothalamic-hypophyseal portal system and the consequent secretion of ACTH followed by an increased output of glucocorticoids from the adrenal cortex.

Many kinds of stress have been shown to activate this system. The increased output of glucocorticoids under conditions of novelty and deprivation stress would increase the rate of gluconeogenesis, thus producing increased blood-glucose levels, which could signal satiety to the hypothalamus. The increased glucocorticoid output would make glucose available for the energy expenditure required in running, yet would serve to suppress food consumption (Mayer, 1953).

The purpose of the present experiment was to replicate the self-starvation phenomenon and to investigate the possibility that adrenocortically mediated gluconeogenesis might underlie the suppression of appetite through the elevation of blood-glucose levels.

METHOD

The Ss were 30 male Sprague-Dawley (Charles River) rats bred in the Franklin and Marshall laboratory. They were 67-70 days of age and weighed 246-333 g at the beginning of the experiment. Water was available ad lib to all Ss.

Fifteen standard Wahmann activity wheels were used. Adjoining the wheel was a 10 x 6 x 5 in. cage connected by a sliding door that could be opened to permit and closed to prevent movement between the cage and wheel. Water was available in this adjoining cage. The Ss were fed in standard laboratory cages located in a nearby room. The food consumed was Rockland Mouse/Rat Diet (complete), and the homeroom temperature was maintained at $74 \pm 2^\circ\text{F}$.

From weaning (age 21 days) until the beginning of the experiment, all Ss were housed in the same room with a 12-h light/12-h dark cycle. This light-dark cycle was maintained throughout the experiment. All Ss were weighed weekly during the time between weaning and the beginning of the experiment. The Ss were then divided into two equal groups, balanced according to body weight and litter affiliation. Fifteen experimental Ss lived in individual activity wheels, while the remaining Ss served as controls and lived in standard laboratory cages. The Ss were removed from wheels or home cages and fed in the feeding room for 1 h per day during the middle of the light cycle, i.e., 6 h after light onset. All Ss were weighed daily before their respective feeding times. Activity, food intake, and water intake, all in the homeroom and feeding room, were recorded daily.

On the 1st, 3rd, and 6th day of

deprivation, a tail-blood sample was taken from each S just prior to the feeding time. The blood-glucose determinations were made by the glucose oxidase method (Glucostat, Worthington Biochemical Corp.).

The criterion of weight stabilization was the same as described by Routtenberg & Kuznesof (1967), which required that the weight on Day 4 of any 4-day period had to be equal to or greater than the weight on Day 1 of that 4-day period. The experiment ended when either weight stabilization or death occurred for all Ss.

RESULTS

Fourteen of 15 experimental Ss starved in a mean of 10.1 days, while 1 experimental S achieved weight stabilization in 25 days. Ten of the 15 control Ss reached weight stabilization in a mean of 19.7 days, while 5 control Ss died in a mean of 20.2 days. Analysis by t test indicated that, among Ss who died, experimental Ss died significantly sooner than did control Ss ($t = 5.93$, $df = 17$, $p < .002$).

Figure 1 shows the per cent of activity increase above 1st-day running scores for the experimental group over the last 6 days of the study. The characteristic increase of activity is present, and there is a significant days effect ($F = 3.11$, $df = 5/83$, $p < .025$). This effect emerges at the same significance level when the raw activity scores are analyzed.

The body weight of experimental Ss falls dramatically over the last 6 days of the study, while control Ss show a gradual weight loss and then increase. Analysis done according to Myers (1966, p. 176) indicates that, over these days, control Ss weigh significantly less than experimental Ss ($F = 4.86$, $df = 1/28$, $p < .05$), and there is a significant days effect ($F = 135.25$, $df = 5/140$, $p < .01$) and a significant Treatment by Days interaction effect ($F = 49.06$, $df = 5/140$, $p < .01$).

The mean food intake over the last 6 days of the study is presented in Fig. 1. Control Ss consume significantly more food than do experimental Ss ($F = 26.10$, $df = 1/28$, $p < .01$). The food consumption of experimental Ss drops markedly over the last 2 days, and there exists a significant days effect ($F = 13.75$, $df = 5/140$, $p < .01$) and a significant Treatment by Days interaction effect ($F = 5.42$, $df = 5/140$, $p < .01$).

The total water intake of experimental Ss shows a gradual then sharp decline over the last 6 days of the study, while the water intake of control Ss rises gradually over this period. The water intake is nearly the same for the two groups overall, except for the last 2 days. A significant days effect ($F = 3.21$, $df = 5/140$, $p < .01$) as well as a

Fig. 1. Mean per cent activity increase and mean food intake in grams over the last 6 days of the study.

significant interaction effect ($F = 3.35$, $df = 5/140$, $p < .01$) is present.

The mean blood-glucose levels of experimental and control groups are presented in Fig. 2. The points have been connected by straight lines to form a continuous function over days. No significant between-group differences exist on Days 1 or 3, but on Day 6, the control Ss have increased blood-glucose levels that approach those found in ad lib fed animals. A significant between-group difference exists on Day 6 ($t = 3.22$, $df = 27$, $p < .01$). A one-way analysis of variance (subjects by treatments) indicates a significant change in control blood-glucose levels over days ($F = 6.80$, $df = 27$, $p < .01$), and post hoc analysis by the Scheffé test indicates that the increase between Days 1 and 3 and between Days 3 and 6 is not significant, but the overall increase in Days 1 to 6 is significant. A one-way analysis of variance (subjects by treatments) indicates that the decrease in experimental blood-glucose levels is not significant.

DISCUSSION

The results of the present study show clearly the existence of the "self-starvation" phenomenon, originally reported by Routtenberg & Kuznesof (1967). The experimental Ss show the

characteristic increase in activity as well as the suppression of food consumption. The experimental Ss self-starve, while most control Ss are able to adapt, over days, to the deprivation schedule.

The present data are in opposition to the hypothesis of Routtenberg (1968) that

"the hyperglycemia caused by corticoid mediation of gluconeogenesis [p. 238]" is the critical mechanism in activity-related self-starvation. There was no significant difference in blood-glucose levels between active and nonactive Ss on the 1st and 3rd days of exposure to food deprivation and

Fig. 2. Mean blood-glucose levels (mg/100 ml) on the 1st, 3rd, and 6th days of the study.

significantly lower blood-glucose values in the active Ss on Day 6.

Thus, it would appear that the suppression of food intake found in the active Ss was occurring in spite of lowered blood-glucose values rather than as a result of corticoid-mediated gluconeogenesis and "hyperglycemia." The tail-blood samples in the present study were taken immediately before feeding, when the active animal would have to be relatively hyperglycemic if this mechanism was operative in the suppression of eating. It is of interest to note that control Ss adapted to the deprivation schedule both behaviorally, by increasing their food intake, and physiologically, by showing increased blood-glucose levels.

The lowered blood-glucose levels in the experimental group may be attributed to the combined effect of increasing activity with consequent increase in energy expenditure and the reduced food intake. The mechanism underlying the reduced

food intake remains unidentified from this study. It should be noted, however, that the tail-blood sample consisted of venous blood, so that the present data do not refute the hypothesis (Mayer, 1953) that feeding behavior is regulated by the difference between arterial and venous blood and the rate of glucose utilization. It would be valuable if these ratios could be obtained from active self-starving rats, although arterio-venous differences are negligible (Oser, 1965, p. 1055) in the resting fasted state.

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