Effects of Short Term Heat Exposure on Physiological Response and Plasma Substrate Concentration in Laying Hens

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Abstract Feed intake levels, physiological response and plasma substrate concentration were measured to assess the effect of heat on substrate metabolism during physiological acclimation for heat balance over a three-day period of heat exposure and/or feed reduction in laying hens. 1) Decreased daily feed intake on the 1st day of heat exposure assumed a definite level on the 3rd day of heat exposure. 2) Body temperature and respiration rate attained maximum levels on the 1st day of heat exposure, and then steadily declined during heat exposure. Heart rate was slightly low during heat exposure. 3) Plasma concentrations of uric acid, cholesterol ester, phospholipid and β-lipoprotein showed low levels during heat exposure and/or even following heat exposure. The decrease in these concentrations appeared higher during heat exposure than feed reduction. Thus possibly, changes in plasma lipid and/or protein metabolism due to heat, regardless of reduction in feed intake, may accompany physiological acclimation for heat balance during a period of heat exposure.

Key words: laying hen, heat exposure, physiological response, plasma substrate concentration

General features of physiological response and productive performance in laying hens subjected to high environmental temperature were first studied by HUTCHINSON and SYKES7). In this study, the scope of attention has been extended to thermal environment2) and diet component control14) as well as thermoregulation and heat production18). The major purpose sought here is technological improvement in productivity on a mass scale by increasing energy intake. This study was prompted by lack of information on metabolic acclimation to heat which may depend on substrate metabolism, i.e. the origin of energy metabolism.

Observation of decreased heat production during heat acclimatization18) and a definite decreased feed intake within 2 or 3 days following heat exposure9) indicated a short period of heat exposure to affect energy metabolism. The decrease in energy metabolism may possibly lead to reduced egg production11) and lesser fat deposition in carcass and adipose tissue of ducklings exposed to heat3).

Differences in yolk lipid composition of laying hens subjected to one week of heat exposure during the fourth and fifth weeks posthatching and laying hens whose feed was reduced during the same period22) indicate the metabolism of lipids to be possibly altered by the experience of heat exposure.

General features of plasma concentrations of glucose, protein and lipid were examined in this
study to determine if such change in substrate metabolism accompanies physiological acclimation for heat balance during a short period of heat exposure.

Materials and Methods

Three experiments were conducted in this study. In Experiment 1 (Exp. 1), hens were exposed to heat, as was also done in Experiment 2 (Exp. 2). In Experiment 3, feed quantity was reduced. Each of 4 commercial laying hens (Shaver, Starcross 288), 9 months of age, weighing 1.7 ~ 1.8 kg was used in all three experiments. Each hen was housed separately in a cage placed in a temperature controlled chamber and provided commercial mash (S-SEBUN, NIHON-NOSANKOGYO, ME; 2820 kcal/kg, CP; 17%) and water at 9:00. Feed and water were available ad libitum in Exps. 1 and 2. Feed was reduced to the level observed during heat exposure (Exps. 1 and 2) in Exp. 3. The light was on from 6:00 to 20:00. Carbon-powder pick-up was used to measure respiration rate (RR), 3 electrodes were used for heart rate (HR) measurement by ECG induction and a vinyl tube placed in the intra-abdominal position was used for body temperature (BT) measurement in all three experiments. A brachial vein cannula for blood sampling was attached to each hen in all experiments according to the method of JOHNSON8. These preparations were carried out at least 1 week before the measurements and following which, the hens were not handled throughout the experimental period, except bleeding and cannula maintenance. Each cannula was washed with 1% of EDTA solution once in a day at the sampling time throughout the study. Hens were made accustomed to 20°C constant environmental temperature at least for 2 weeks before starting the experiments. In Exps. 1 and 2, after a period of 3 days maintained at 20°C, the temperature was raised at 9:00 and kept at 35°C for the following 3 days, and lowered again to 20°C. In Exp. 3, the temperature was kept at 20°C during 9 days. Thus, the total period for all 3 experiments was 9 days. RR, HR and BT were measured every 4 hr from 13:00 to 9:00 during this period. Feed intake (FI) was measured every 1 hr in Exps. 1 and 2 and once a day at 9:00 in Exp. 3 by manually weighing each feed trough. Water intake was measured once a day at 9:00 in each experiment. Two ml of blood sample was taken from each hen at 14:00 each day of the experiment. Plasma was obtained immediately after bleeding, thereafter stored at −20°C untill determination of glucose (GL), total protein (PR), uric acid (UA), free fatty acid (FFA), cholesterol ester (CE), phospholipids (PL) and β-lipoprotein (βLP) concentration. This parameter for GL, PR, UA, FFA, CE, PL and BLP was measured using commercial kits (Glucose C-Test Wako, A/G B-Test Wako, Uric Acid C-Test Wako, NEFA C-Test Wako, Cholesterol CII-Test Wako, Phospholipids B-Test Wako, β-Lipoprotein C-Test Wako, WAKO PURE CHEMICAL INDUSTRIES, Tokyo). The paired Student t-test was conducted for statistical analysis of difference.

Results

The mean egg production rate of the hens used in this study was more than 90%, and was not affected by heat exposure or feed reduction. Changes in daily FI in Exps. 1 and 2 were essentially the same. As were also physiological response and plasma substrate concentrations. Thus, all the data for Exps. 1 and 2 were included in one group.

Mean daily FI decreased to 70.7±2.2% that of the control period during 3 days of heat exposure. Feed quantity dropped to 68.5±4.4 % that of the control period during 3 days of feed reduction. Hourly FI of the control period during heat exposure experiment was usually less than 9 g/hr., and was 58.1% of the total number of observation while the level 10-19 g/ hr. was 37.5% and more than 20 g/hr., 4.0%.
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Table 1 Changes of environmental temperature, body weight, feed intake and water intake during 9 days of heat exposure and feed reduction experiments

<table>
<thead>
<tr>
<th>Days</th>
<th>20°C</th>
<th>20 or 35°C</th>
<th>20°C</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Environmental temperature (°C)</td>
<td>Heat exposure</td>
<td>±1.6</td>
<td>±1.3</td>
</tr>
<tr>
<td></td>
<td>Feed</td>
<td>22.5</td>
<td>22.3</td>
</tr>
<tr>
<td></td>
<td>reduction</td>
<td>±0.3</td>
<td>±0.3</td>
</tr>
<tr>
<td>Body weight (kg)</td>
<td>Heat</td>
<td>1.74</td>
<td>1.74</td>
</tr>
<tr>
<td></td>
<td>exposure</td>
<td>±0.06</td>
<td>±0.06</td>
</tr>
<tr>
<td></td>
<td>Feed</td>
<td>1.80</td>
<td>1.78</td>
</tr>
<tr>
<td></td>
<td>reduction</td>
<td>±0.10</td>
<td>±0.11</td>
</tr>
<tr>
<td>Feed intake (g/hen · day)</td>
<td>Heat exposure</td>
<td>±5.6</td>
<td>±14.0</td>
</tr>
<tr>
<td></td>
<td>Feed</td>
<td>138.8</td>
<td>116.5</td>
</tr>
<tr>
<td></td>
<td>reduction</td>
<td>±5.7</td>
<td>±7.4</td>
</tr>
<tr>
<td>Water intake (g/hen · day)</td>
<td>Heat exposure</td>
<td>±118.1</td>
<td>±124.3</td>
</tr>
<tr>
<td></td>
<td>Feed</td>
<td>284.5</td>
<td>270.8</td>
</tr>
<tr>
<td></td>
<td>reduction</td>
<td>±123.9</td>
<td>±111.1</td>
</tr>
</tbody>
</table>

Environmental temperature was the mean of every 1 hour measurement. Each value indicates the mean ± S.D. of 8 hens in heat exposure experiment and 4 hens in feed reduction experiment. **, *: significant difference between the means of 3 days of the control period and each day following heat exposure or feed reduction (P < .01, P < .05).

On the 2nd day of heat exposure, less than 9 g/hr. was 81.8% and 10-19 g/hr., 18.2% of the total while more than 20 g/hr. was not observed. The same was noted on the 3rd day of heat exposure.

Mean daily BT and RR were maximum on the 1st day of heat exposure and decreased steadily thereafter. This decrease in mean daily BT was significant (P < .05) as was also that in the mean daily RR (P < .01) on the 2nd day of heat exposure. Decrease in the mean daily HR on the 1st day (P < .05) of heat exposure was much significant on the 2nd (P < .025) and 3rd days (P < .01) though the difference between these days was not significant. BT, RR and HR measured every 4 hours were high late in the day and low at night during the control period of heat exposure. BT and RR during feed reduction were essentially the same though the mean daily HR was significantly low (P < .01) on the 1st and 2nd days each relative to the mean of control period of feed reduction.

In some cases, substrate concentration showed considerable variation according to the day and hen. During the feed reduction experiment, this parameter was slightly higher than during the heat exposure experiment.

UA concentration was lower during heat exposure, being significant on the 1st day (P < .01) and 3rd days (P < .05) of heat exposure. Its decrease during heat exposure was higher than during the feed reduction period. The concentrations of CE, PL and \( \beta \)LP were less during and following heat exposure. Significant decrease was noted in PL (P < .05) on the 1st day of heat exposure and also in CE (P < .05), PL (P < .01) and \( \beta \)LP (P < .05) on the 2nd day of heat exposure. Concentrations of CE and PL were observed to decrease only slightly during feed reduction and not as much during heat exposure.

**Discussion**

The general features of physiological response and feeding behaviour were observed
during 3 days of heat exposure. Initially, there was an increase in BT, and then in RR, as has already been reported\(^{17}\). Increase in water intake\(^{26}\) and decrease in feed intake\(^{9}\) were also noted. Heat production\(^{18}\) may have decreased as a result of that in HR. Egg shell quality\(^{13}\) subsequently lessened as did also egg weight\(^{11}\) (data not shown).

RR, water intake, feed intake and HR observed in the present study on heat exposure indicated improvement in heat balance as a result of heat loss and decreasing HP as indicated by change in BT. The values determined for these should provide indication of acclimation to high temperature, even before this has been fully achieved. That is, the steady decline in BT to a constant level by the 3rd day of heat exposure was definite indication that heat balance had improved during the 3 days of exposure. That, RR as active heat loss steadily declined during this period is also evidence for the improvement of heat balance (Fig. 1).

Decrease in FI (Table 1) with hourly lowering FI and slight decrease in mean daily HR were observed during the exposure. Egg layers feed continually during the day\(^{6}\) and this produces heat\(^{10}\). HP was suppressed by the FI level during the 3 days of exposure.

There is presently no information on substrate metabolism to explain why lowering energy metabolism leads to improved heat balance and lesser productivity at high temperature. In this study, change in substrate concentration was observed during heat exposure, and also during feed reduction. This parameter varied considerably according to the day and hen, due possibly to the time of oviposition and random feeding during the day time, and thus difficult to avoid in the present study. However, the variation was less than that indicated in other papers\(^{4,5}\) since a cannula was used in this work\(^{8}\).

The concentrations of UA, CE, PL and \(\beta\)LP were low, and significantly so on some days during the 3 days of heat exposure and the period following (Fig. 2). So long as the plasma substrate concentration is maintained by the entry rate and the clearance rate for catabolic utilization and anabolic tissue deposition is controlled, the low concentration may
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Fig. 2. Changes of plasma substrate concentrations during 9 days of heat exposure and feed reduction experiments. Each point and vertical line indicate the mean±S.D. of 8 hens in heat exposure experiment (—■—) and 4 hens in feed reduction experiment (—○—). **, *: significant difference between the means of 3 days of the control period and each day following heat exposure or feed reduction (p < .01, p < .05).

have provided decrease in these two parameters of clearance. The slightly low HR in the present study would surely be indication of decrease in catabolism. Decrease in anabolic tissue deposition has also been reported. These findings appear to provide a basis for
concluding blood lipid fraction concentration and egg production are closely related\(^ {12} \).

Slightly low levels of UA, CE and PL concentration were observed during feed reduction (Fig. 2) but not following this and the decrease was less than that during heat exposure. Thus, plasma substrate concentration is much more affected by high temperature lessening the amount of feed and consequently there is the possibility that metabolic flux of anabolic tissue deposition may be further influenced by high temperature. Studies on fatty liver syndrome under high temperature\(^ {1, 16, 27} \) support this possibility. AKIBA et al.\(^ {1} \) examined the effect of feed fat level on egg production, liver lipid content and plasma lipid content, and found that liver lipid content was increased with lowered feed intake and lowered plasma lipid level at high temperature. There should thus be an active mechanism for clearing plasma lipid leading to lipid accumulation in liver tissue to suppress catabolic metabolism at high temperature. There seems to be no reason to indicate otherwise that the considerable decrease in the concentrations of CE, PL and \( \beta \)LP on heat exposure result from the same mechanism as that for fatty liver syndrome. The urgent need to clear plasma lipid may thus induce metabolic competition, resulting in further decrease in the flux of anabolic metabolism, as reported when exercising\(^ {24} \). Though the plasma PR concentration did not change, lesser catabolism of protein is also suggested by the decrease in UA concentration on heat exposure.

Studies on thermal environment\(^ {2} \), feed quality\(^ {14} \), training\(^ {20} \) and breeding\(^ {25} \) for heat tolerance at high temperature to improve productivity are based on data for heat balance and energy supply. Increase in CP content\(^ {16, 21} \) and ME content\(^ {19} \) may effectively lead to greater productivity by increasing the flux for anabolic metabolism. However, this would not prove successful in all cases since substrate metabolism is still not adequately understood.

Posthatching heat exposure affected on yolk lipid composition\(^ {22} \). The difference in plasma FFA profile for rest and exercise\(^ {23} \) indicated specific utilization of FFA fraction. AKIBA\(^ {13} \) found the inducement of lipoprotein lipase to depend on the diets. These data indicate specific metabolism for any nutritional situation and provide a basis for studies on dynamics of substrate metabolism at high environmental temperature. This in turn may lead to improved productivity at high environmental temperature through control of energy intake and the process of metabolism.

References

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産卵鶏の体温、呼吸数、心拍数ならびに血漿中の脂質、蛋白質およびグルコース濃度に及ぼす短期暑熱感作の影響

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暑熱順化過程における物質代謝を検索するために、20℃恒温環境下で飼育した約270日齢の産卵中の市販鶏を35℃恒温に3日間暴露し、採食量、体温、呼吸数、心拍数ならびに血漿中脂質、蛋白質およびグルコース濃度を、経時的に測定した。また、35℃暴露の間に低下した採食量の影響を検討するために、20℃恒温環境下で飼料摂取量を制限した実験区において、同様の測定を行なった。1) 暑熱暴露1日に低下した日採食量、1時間毎の採食量の低下を伴ないながら暑熱3日には安定した。2) 体温は暴露4時間までに、呼吸数は暴露8時間までに頂上値に達した後、低下を続けて暴露3日には安定した。心拍数は暴露3日間とも低水準を維持した。3) 血漿中尿酸、コレステロールエステル、リン脂質およびβ-リポ蛋白濃度は暴露3日間および暴露後2日間においてわずかに低下した。これらの血漿中成分の変化は、20℃環境下で飼料摂取量を制限した実験区での結果と異なる。以上の結果、暑熱への順化過程では、体温調節性の生理、行動反応だけではなく、血漿中の脂質や尿酸濃度に変化が認められ、暑熱の影響を解析するためには、物質の代謝過程に関する情報も必要であると考えられた。

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