# Relationships among Behavior, Physiological States and Body Weight Gain in Grazing Holstein Heifers

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**ABSTRACT :** This study examined the behavior of dairy heifers and the factors affecting the performance of them on pasture. Behavior of 10 Holstein heifers in a herd of 25 animals that rotationally grazed five 8-ha pastures was observed and recorded every 5 minutes during 24 hours; body weights were measured once a month from June to October. Blood and rumen fluid samples were collected from 5 of them bimonthly. Chemical composition was analyzed for the forage samples collected each month. CP content (DM basis) of herbage ranged from 12.2 (June) to 17.2% (October) and ADF from 31.1 (October) to 39.1% (July). Standing (posture) time was different significantly among months ($p<0.001$) ranging from 48.3 to 61.3% of 24 hours and was longer in July and August (61.3% and 58.3%, respectively) when ADF content of herbage was higher than in the other months. Grazing time which significantly differed among months ($p<0.001$) ranged from 29.1 to 41.6% of 24 hours and was shorter in June and September (29.1% and 33.0%, respectively) when ADF content was lower than in the other months. Average DG through the experiment period was 0.74 kg/day. August was the lowest in DG (0.41 kg/day) and the longest in rumination time and standing-rumination time among months. Animals of higher DG had a shorter standing time ($r=-0.36$, $p<0.01$) and a longer lying-rumination time ($r=0.55$, $p<0.001$) throughout the experiment. Total protein concentration in blood ranged from 9.04 to 9.64 g/dl and was negatively correlated with DG ($r=-0.65$, $p<0.05$). Phospholipid concentration of blood ranged from 119.66 to 156.40 mg/dl and was negatively correlated with DG ($r=-0.57$, $p<0.05$). VFA in rumen fluid, acetic acid proportion (ranging from 69.35 to 74.76%) and butyric acid proportion (ranging from 7.18 to 12.05%) showed significant differences among months ($p<0.05$, $p<0.001$, respectively). Butyric acid proportion was significantly related with DG ($r=0.60$, $p<0.05$). *(Asian-Aust. J. Anim. Sci. 2001. Vol. 14, No. 6 : 803-810)*

**Key Words :** Behavior, Heifer, Body Weight Gain, Physiological States, Pasture, Grazing

## INTRODUCTION

Raising dairy and beef cattle by grazing on pasture is a traditional practice in many parts of the world. Heifers grown on pasture have been found to have comparable growth rates to those grown in dry lots (Ando et al, 1986 and Iketaki et al., 1987). In Japan, because grassland areas possessed by dairy and beef farmers are small, many heifers are kept in confinement. There are many cooperative and public pastures being in use for raising dairy and beef cattle to reduce the labor and cost of personal farming. However, these are not fully utilized because satisfactory performance of heifers is not always obtained when they are raised there.

There are many factors affecting the performance of animals on pasture (eg., Caton and Dhuyvetter, 1997; Krysl and Hess, 1992; Poppi and McLennan, 1995; Seman et al., 1991). The present study aimed to investigate some behavioral characteristics, physiological states and seasonal herbage conditions in relation to body weight gain of heifers on a cooperative pasture.

## MATERIALS AND METHODS

This experiment was carried out at Kushinai Cooperative Pasture 800 meters above sea level in central Hokkaido, northern island in Japan. A herd of twenty-five Holstein heifers was rotationally grazed across 5 pastures each of which was approximately 8 ha. Rotation length was 3 to 5 days per pasture. For behavioral observation, 10 animals ($308 \pm 33$ kg BW) were chosen from the herd and were numbered individually using fluorescent paint. Their behavior was observed and recorded every 5 minutes during 24 hours once a month for 5 months from June to October. Behavior observed was categorized into grazing (G), rumination (Rum) and resting (R) with standing (S) and lying (L) as posture. Body weights of those 10 animals were measured two days before each behavioral observation. Samples of blood and rumen fluids were collected from 5 of them bimonthly. Bloods were analyzed for hematocrit, total protein, glucose, insulin, free fatty acid, total cholesterol, triglyceride and phospholipid and rumen liquid for volatile fatty acids (VFA) and ammonium

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Received April 28, 2000; Accepted February 6, 2001

nitrogen ($NH_3$-N). Chemical composition (CP and ADF) and mineral contents (Ca, P, Mg and K) of herbage samples taken one or two days before each 24-h behavioral observation were determined using AOAC methods. Data were analyzed by analysis of variance (ANOVA), using the general linear models procedure and student's t-test for comparison of the means when main effects were significant (SAS, 1985).

## RESULTS

The chemical composition of herbage is given in table 1. Among five months, legume percentage was greatest in October (23.3%) and dry matter (DM) content was smallest in June (21.2%). Crude protein (CP) content was greatest in September and October (16.7, 17.2%, respectively), acid detergent fiber (ADF) content was highest in July and August (39.1, 37.5%, respectively) and sward height was lowest in September and October (41, <15 cm, respectively). These data indicate that in June, pasture had a high nutritive value and large mass, while in July and August nutritive value was poor but mass was large. In September and October, nutritive value was high but mass was small.

Change in average body weight of heifers is shown in figure 1. At the beginning of the experiment on July, it was 318±33 kg, and at the end of experiment on October, 421±27 kg. Average body weight gain during 139 days was 103 kg.

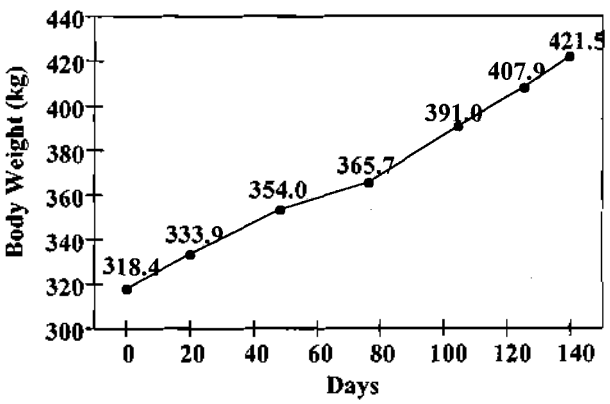


**Figure 1.** Body weight change of heifers

Seasonal changes in heifer behavior on pasture, average daily gain (ADG) and the results of analysis of variance are given in table 2. Overall, ADG of experimental animals through the period was 0.74± 0.12 kg. The greatest daily gain (DG) recorded was 0.91 kg in September and the smallest was 0.41 kg in August. Total time spent S was longer and total time spent L was shorter in July and August (S;61.3, 58.3%, L;38.7, 41.7%, respectively). Total time spent G was shorter in June and September (29.1, 33.0%, respectively) and longer in July and October. In August, total times spent Rum and Standing-Rum were longest (33.0, 10.5%, respectively) and total time spent R was shortest (26.3%) among five months.

On analysis of variance for behavior, values of DG in each month were sorted into two classes, higher and lower than the means. Data of behavior were analyzed with models containing Month, Class of DG and Heifer. Month had highly significant or significant effects on all behavioral categories (S, L, G, Standing Rum, R, Standing-R, Standing-R, Lying-R, $p<0.001$; and Rum, DG, $p<0.01$) except Lying Rum. Class of DG had no significant effects on any behavioral categories. Heifer had significant effects on S, L and Standing R ($p<0.01$) but not on DG.

Correlation coefficients between DG and behavior are given in table 3. Figure 2 shows relationships of DG of heifers to rumination behavior. In August and October, animals with higher DG had longer total time spent Rum ($r=0.69$, $r=0.73$, respectively, $p<0.05$) and Lying-Rum time ($r=0.85$, $p<0.01$, $r=0.65$, $p<0.05$, respectively), and in October had trends of shorter total time spent R and Lying-R ($r=-0.59$, $r=-0.56$, respectively, $p<0.1$). Through June to October, higher DG accompanied shorter total S ($r=-0.36$, $p<0.01$), longer total L ($r=0.36$, $p<0.01$), shorter Standing-Rum ($r=-0.45$, $p<0.001$), and longer Lying-Rum time ($r=0.55$, $p<0.001$).

Table 4 gives seasonal changes of frequency of behavior and the statistical results by ANOVA. Categories of behavior for frequency were Standing and lying (SL), G, Rum, Rum in bouts shorter than

**Table 1.** Changes in chemical composition, legume percent in fresh matter, and sward height of herbage

| Month | DM | CP | ADF | Ca | P | Mg | K | Legume | Sward height |
|---|---|---|---|---|---|---|---|---|---|
| | % | 10% of DM | | | | | | % of FM | cm |
| June | 21.2 | 12.2 | 34.5 | 0.40 | 0.18 | 0.12 | 1.37 | 18.1 | 71 |
| July | 24.8 | 13.4 | 39.1 | 0.37 | 0.20 | 0.15 | 2.27 | 16.8 | 67 |
| August | 25.6 | 13.9 | 37.5 | 0.34 | 0.19 | 0.12 | 2.29 | 17.6 | 73 |
| September | 27.2 | 16.7 | 33.9 | 0.33 | 0.23 | 0.15 | 2.24 | 13.8 | 41 |
| October | 25.7 | 17.2 | 31.1 | 0.44 | 0.21 | 0.15 | 2.62 | 23.2 | <15 |

**Table 2.** Seasonal changes in heifer behavior on pasture, average daily gain and the results of ANOVA

| | S[2] | L | G | Rum | (S | L) | R | (S | L) | ADG |
|---|---|---|---|---|---|---|---|---|---|---|
| Month[1] | % of 24 hour | | | | | | | | | kg/day |
| June | 53.0$^{b}$ | 47.0$^{b}$ | 29.1$^{c}$ | 28.8$^{b}$ | ( 4.1$^{b}$ | 24.7) | 37.3$^{a}$ | (15.2$^{a}$ | 22.1$^{a}$) | 0.78$^{ab}$ |
| July | 61.3$^{a}$ | 38.7$^{c}$ | 41.5$^{a}$ | 27.5$^{b}$ | ( 4.2$^{b}$ | 23.3) | 28.3$^{b}$ | (13.0$^{ab}$ | 15.3$^{c}$) | 0.72$^{ab}$ |
| August | 58.3$^{a}$ | 41.7$^{c}$ | 39.7$^{a}$ | 33.0$^{a}$ | (10.5$^{a}$ | 22.5) | 26.3$^{b}$ | ( 7.1$^{d}$ | 19.2$^{b}$) | 0.41$^{b}$ |
| September | 48.3$^{c}$ | 51.7$^{a}$ | 33.0$^{b}$ | 26.8$^{b}$ | ( 0.9$^{c}$ | 25.9) | 37.0$^{a}$ | ( 11.3$^{bc}$ | 25.8$^{a}$) | 0.91$^{a}$ |
| October | 51.7$^{bc}$ | 48.3$^{bc}$ | 41.6$^{a}$ | 27.5$^{b}$ | ( 1.8$^{c}$ | 25.7) | 30.2$^{b}$ | ( 7.8$^{d}$ | 22.4$^{ab}$) | 0.81$^{a}$ |
| Overall | 54.4 | 45.5 | 34.0 | 28.7 | ( 4.3 | 24.4) | 31.8 | (10.9 | 20.9) | 0.74 |
| Source of variance | | | | | | | | | | |
| MONTH | ***[4] | *** | *** | ** | *** | NS | *** | *** | *** | * |
| CLASS OF DG[3] | NS | NS | NS | NS | NS | † | NS | † | NS | *** |
| HEIFER | * | * | NS | NS | NS | NS | NS | * | NS | NS |

[1] Values in each month are expressed as the means of 10 observations. [2] S, standing; L, lying; G, grazing; Rum, rumination (with the % of time spent ruminating while S and L); R, resting (with the % of the time spent resting while S and L). [3] Values of daily gain (DG) in each month were sorted into two classes, higher and lower than the means. [4] NS: non significant, †: p<0.1, *: p<0.05, **: p<0.01, ***: p<0.001.

$^{abc}$ Means in a column without a common superscript letter differ significantly by student's t-test (p<0.05).

**Table 3.** Correlation coefficients between daily gain and behavior

| Month | S[1] | L | G | Rum | (S | L) | R | (S | L) |
|---|---|---|---|---|---|---|---|---|---|
| June | 0.02 | -0.02 | 0.49 | 0.13 | 0.11 | 0.00 | - 0.46 | -0.36 | -0.04 |
| July | -0.49 | 0.49 | -0.37 | 0.24 | -0.28 | 0.48 | 0.09 | -0.06 | 0.14 |
| August | -0.52 | 0.52 | -0.12 | 0.69** | -0.51 | 0.85** | -0.36 | -0.42 | -0.03 |
| September | 0.07 | -0.07 | 0.15 | 0.15 | -0.06 | 0.17 | -0.13 | 0.19 | -0.22 |
| October | 0.07 | -0.07 | 0.31 | 0.73** | 0.22 | 0.65* | -0.59† | -0.44 | -0.56† |
| Overall | -0.36** | 0.36** | -0.11 | 0.10 | -0.45*** | 0.55*** | 0.02 | -0.02 | 0.04 |

[1] S, standing; L, lying; G, grazing; Rum, rumination (with the % of time spent ruminating while S and L); R, resting (with the % of the time spent resting while S and L).

† p<0.1, * p<0.05, ** p<0.01, *** p<0.001.

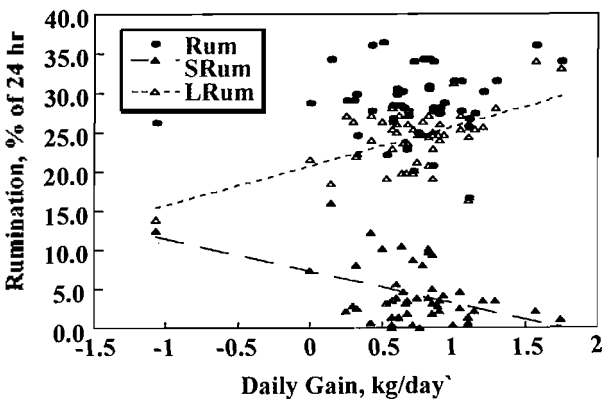


**Figure 2.** Relationships of daily gain (DG) of heifers to rumination behavior (Rum: total time of rumination, SRum: standing - rumination, LRum: lying - rumination, % of 24 hr). Correlation coefficients are r=0.10 (p>0.1) between DG and Rum; r=-0.45 (p<0.001) between DG and SRum; and r=0.55 (p<0.001) between DG and LRum.

10 min. (Rum1), Rum in bouts longer than 10 min. (Rum2), Standing - Rum (SRum), Lying - Rum (LRum), Standing - R (SR) and Lying - R (LR).

On analysis of variance for frequency of behavior, Month had highly significant effects on all frequencies of behavior except LR (p<0.001), and a significant effect on LR (p<0.05). CLASS of DG tended to affect the frequencies of SRum and LRum (p<0.1). Heifer had a significant effect on LRum (p<0.05) and tended to affect SR (p<0.1).

Table 5 gives correlation coefficients between DG and frequency of behavior. Higher DG had a lower frequency of SR (r=-0.78, p<0.01) in June; in August had a lower frequency of G (r=-0.82, p<0.01) and tended to have a lower frequency of SR (r=-0.60, p<0.1); in September had a higher frequency of Rum1 (r=-0.81, p<0.01); in October lower frequencies of SL and G (r=-0.61, r=-0.69, respectively, p<0.05). Through June to October, frequency of SRum had a significantly negative correlation with DG (r=-0.31, p<0.05).

Figure 3 shows seasonal change in standing

**Table 4.** Seasonal change of frequency of behavior and the statistical results by ANOVA

| | Frequency of behavior | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | SL[2] | G | Rum | Rum1 | Rum2 | SRum | LRum | SR | LR |
| Month[1] | Number of bouts in 24 hours | | | | | | | | |
| June | 23.9[b] | 18.1[a] | 19.4[a] | 4.4[a] | 15.0[a] | 7.3[a] | 16.6[a] | 18.2[a] | 15.8[a] |
| July | 31.3[a] | 19.2[a] | 20.1[a] | 5.1[a] | 15.0[a] | 7.5[a] | 16.9[a] | 18.8[a] | 15.1[a] |
| August | 14.3[c] | 13.0[b] | 16.3[b] | 2.4[b] | 14.2[a] | 6.6[a] | 12.8[b] | 10.3[c] | 11.9[b] |
| September | 16.0[c] | 11.9[b] | 12.9[c] | 1.4[b] | 11.8[b] | 1.9[b] | 12.4[b] | 13.8[b] | 14.8[a] |
| October | 12.5[c] | 11.2[b] | 15.3[b] | 3.0[b] | 12.3[b] | 2.4[b] | 13.3[b] | 9.4[c] | 14.7[a] |
| Overall | 19.6 | 14.7 | 16.8 | 3.3 | 13.7 | 5.1 | 14.4 | 14.1 | 14.5 |
| Source of variance | | | | | | | | | |
| MONTH | ***[4] | *** | *** | *** | *** | *** | *** | *** | * |
| CLASS OF DG[3] | NS | NS | NS | NS | NS | † | † | NS | NS |
| HEIFER | NS | NS | NS | NS | NS | NS | * | † | NS |

[1] Values in each month are expressed as the means of 10 observations.
[2] SL, standing and lying; G, grazing; Rum, rumination; Rum1, rumination in bouts shorter than 10 min.; Rum2, rumination in bouts longer than 10 min.; SRum, standing - rumination; LRum, lying-rumination; SR, standing-resting; LR, lying-resting.
[3] Values of daily gain (DG) in each month were sorted into two classes, higher and lower than the means.
[4] NS: non significant, †: p<0.1, *: p<0.05, **: p<0.01, and ***: p<0.001.
[abc] Means in a column without a common superscript letter differ significantly by student's t-test (p<0.05).

**Table 5.** Correlation coeffecients between daily gain and frequency of behavior

| Month | SL[1] | G | Rum | Rum1 | Rum2 | SRum | LRum | SR | LR |
|---|---|---|---|---|---|---|---|---|---|
| June | -0.31 | -0.33 | -0.15 | -0.42 | 0.29 | -0.52 | 0.41 | -0.78** | -0.01 |
| July | -0.01 | -0.08 | -0.28 | -0.19 | -0.19 | -0.17 | 0.25 | 0.35 | 0.02 |
| August | 0.41 | -0.82* | -0.09 | -0.54 | 0.48 | -0.42 | 0.27 | -0.60† | -0.04 |
| September | 0.37 | 0.10 | 0.14 | 0.81** | -0.38 | 0.26 | 0.14 | 0.15 | 0.35 |
| October | -0.64* | -0.69* | -0.38 | -0.39 | -0.06 | -0.29 | -0.23 | -0.18 | -0.29 |
| Overall | 0.07 | -0.18 | -0.16 | -0.18 | -0.05 | -0.31* | 0.14 | -0.01 | 0.17 |

[1] SL, standing and lying; G, grazing; Rum, rumination ; Rum1, rumination in bouts shorter than 10 min.; Rum2, rumination in bouts longer than 10 min.; SRum, standing-rumination ; LRum, lying-rumination; SR, standing-resting; LR, lying-resting.
† p<0.1, * p<0.05, ** p<0.01, *** p<0.001.

behavior. Triangles indicate sunset and sunrise time. time spent for S was longest in July and August. The weather in July was hot. In June and August it rained around noon and then was cloudy in the afternoon. Figure 4 shows seasonal change in grazing behavior. Grazing around midnight was longer in September and October when sward heights were shorter and grazing at morning in each month began around sunrise. Figure 5 shows seasonal change in rumination behavior. Time spent for Rum was longest in August, and frequency of Rum was greatest in June and July.

Table 6 gives blood constituents, the correlation coefficients of those with DG, and the statistical results by ANOVA. Figure 6 shows relationships of DG to blood concentrations of total protein and phospholipid. Total protein was lowest and glucose was highest in June (9.04 g/dl, 91.1 mg/dl, respectively) among the three months. Total cholesterol was highest in August (117.86 mg/dl) and phospholipid was lowest in June (119.66 mg/dl). DG was correlated negatively with total protein (r=-0.65, p<0.01) and phospholipid (r=-0.57, p<0.05), and also tended to correlate negatively with total cholesterol (r=-0.49, p<0.1). On analysis of variance for constituents of blood, Month had significant effects on total protein, glucose, total cholesterol and phospholipid (p<0.05). Heifer had significant effects on total protein (p<0.01), total cholesterol (p<0.01) and phospholipid (p<0.05).

Table 7 gives constituents of rumen fluid, correlation coefficients between DG and items of rumen fluid and the results of ANOVA. Figure 7 shows relationships of DG to total volatile fatty acids (VFA) concentration (nmol/dl) and to butyric acid (% of VFA). The molar proportion of acetic acid was highest and that of butyric acid lowest in August (74.76%, 7.18%, respectively). Acetic acid / propionic acid ratio was lowest and $NH_3$-N was highest in October (3.90, 19.69 mg/dl, respectively). Butyric acid correlated significantly with DG (r=0.60, p<0.05) but

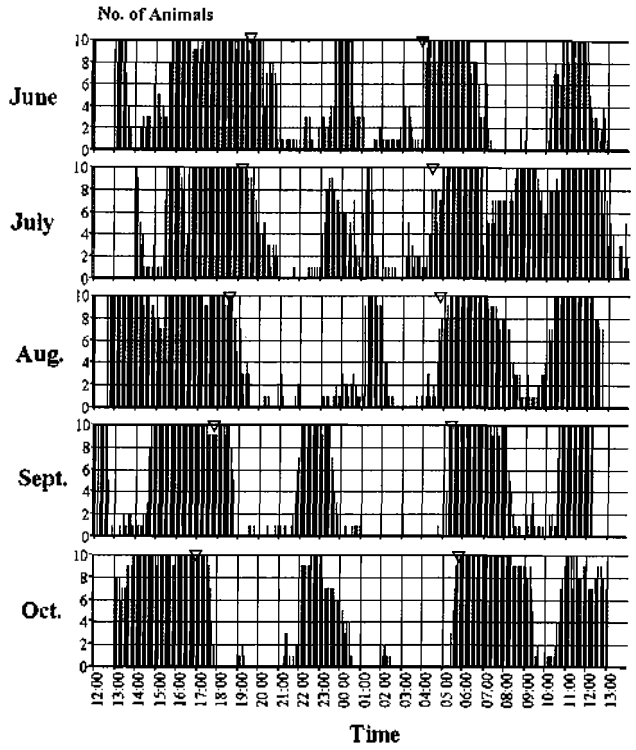


**Figure 3.** Seasonal change in standing (posture) behavior. Triangles indicate sunset and sunrise time.

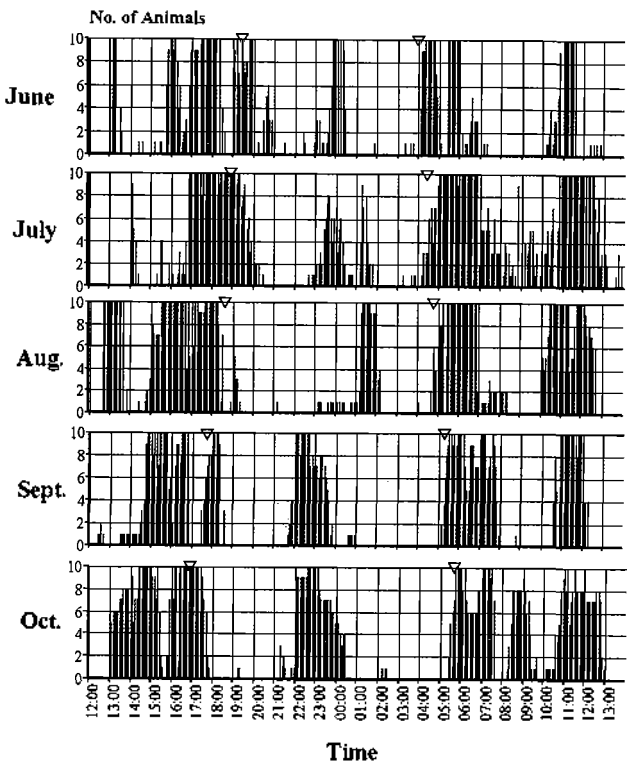


**Figure 4.** Seasonal change in grazing behavior. Triangles indicate sunset and sunrise time

VFA tended to have a negative correlation with DG ($r=-0.50$, $p<0.1$). On analysis of variance for constituents of rumen fluid, Month had significant effects on acetic acid ($p<0.05$), butyric acid ($p<0.001$) and $NH_3$-N ($p<0.05$) and tended to affect VFA and acetic acid/propionic acid ratio ($p<0.1$). Heifer had no effects on components of rumen fluid.

## DISCUSSION

The quality and quantity of pasture herbage seasonally changed. From spring to autumn, CP content and sward height of herbage increased and ADF content was higher in summer than in the other seasons. Pasture had high nutritive value and large mass in June, while in July and August the nutritive value was poor but the mass was large. In September and October, the nutritive value was high but the mass decreased drastically.

The behavior of heifers showed seasonal change and it is considered to be affected by such conditions of pasture. Low quality or small quantity of herbage prolonged G time. It was longer in July, August and October than in the other two months. It is considered that August was the poorest for nutritive value of herbage among months because of the higher ADF content and the smallest DG in spite of the longest Rum. The October result was due to the scarcity of herbage as shown by the shortest sward height. It is presumed that the energy requirement of animals increased in October since it was the coldest month. It

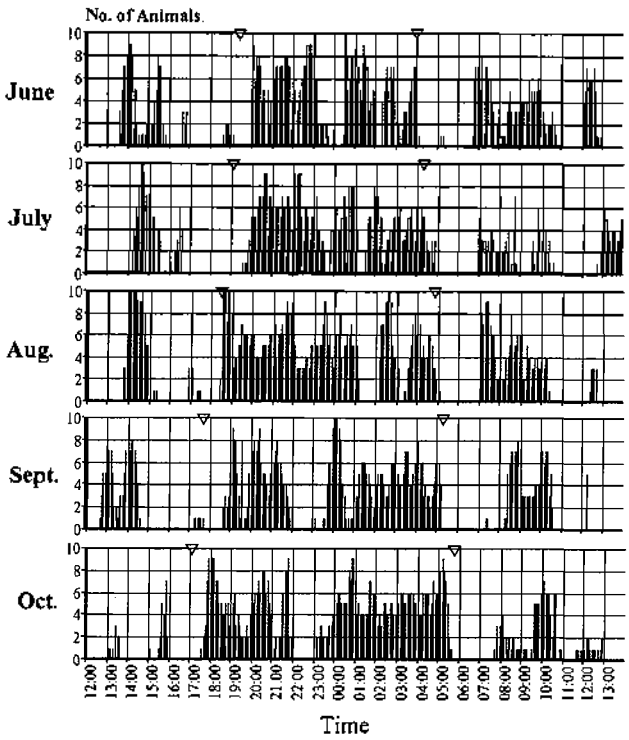


**Figure 5.** Seasonal change in rumination behavior. Triangles indicate sunset and sunrise time

**Table 6.** Blood constituents, correlation coefficients between daily gain and blood constituents and the statistical results by ANOVA

| Item | Month[1] | | | Correlation coefficient | Source of variance by ANOVA[2] | |
|---|---|---|---|---|---|---|
| | June | August | October | | Month | Heifer |
| ADG, kg/day | 0.83 | 0.50 | 0.61 | | NS | NS |
| Hematocrit, % | 32.1 | 32.1 | 32.1 | -0.02 | NS | NS |
| Total protein, g/dl | 9.04[b] | 9.64[a] | 9.38[ab] | -0.64** | * | ** |
| Glucose, mg/dl | 91.10[a] | 85.06[ab] | 82.40[b] | 0.09 | * | NS |
| Insulin, $\mu$U/ml | 2.74 | 6.34 | 6.14 | -0.36 | NS | NS |
| Free fatty acid, mEq/l | 0.403 | 0.372 | 0.323 | 0.28 | NS | NS |
| Total cholesterol, mg/dl | 105.31[b] | 117.86[a] | 107.76[a] | -0.49[†] | * | ** |
| Triglyceride, mg/dl | 36.90 | 21.08 | 17.28 | -0.36 | NS | NS |
| Phospholipid, mg/dl | 119.66[b] | 156.40[a] | 145.26[a] | -0.57* | * | * |

[1] Values in each month are expressed as the means of 5 determinations.

[2] NS: non significant, *: $p<0.05$ and **: $p<0.01$.

[ab] Means in a row without a common superscript letter differ significantly by student's t-test ($p<0.05$).

[†] $p<0.1$ and * $p<0.05$.

is considered that heifers compensated for reductions in bite size because of the shorter herbage by increasing grazing time, as reviewed by Forbes (1988).

S and Standing Rum were negatively and L and Lying Rum was positively correlated with DG. Suzuki et al. (1972) reported similar results in every 10 minute recording trials during daytime. They found that heifer growth was positively correlated with time spent Rum and negatively correlated with time spent G. They concluded that either greater herbage mass or lower nutritive value brought longer rumination time and that smaller herbage mass brought longer grazing time.

The frequencies of all behavioral categories in June and July were greater than in the other three months These results are thought to be related to the higher quality and the greater quantity of herbage in those months than in the other month. However, it might be related to group formation of heifers. In July, all animals moved together as one group, but September and October, they divided into small groups which moved separately. July was the hottest among the five months. It is considered that such multiple conditions brought the result of greatest frequencies of SL, G and Rum1.

DG had negative correlation with frequency of G in August and October. It is considered that under conditions of insufficient available herbage mass, heifers of higher DG had longer continuous grazing periods.

Among heifers, S time, Standing-R time and

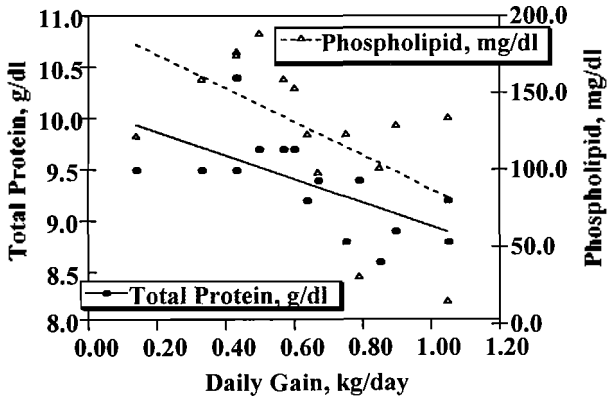


**Figure 6.** Relationships of daily gain (DG) of heifers to blood concentrations of total protein and phospholipid. Correlation coefficients are r=-0.64 (p=0.01) between DG and total protein; and r=-0.57 (p=0.03) between DG and phospholipid.

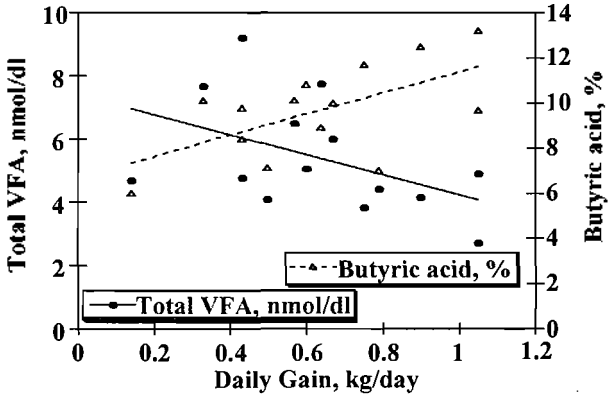


**Figure 7.** Relationships of daily gain (DG) of heifers to total volatile fatty acid (VFA) concentration (nmol/dl) and the rate of butyric acid (% of VFA) in rumen fluid. Correlation coefficients are r=-0.46 (p=0.096) between DG and total VFA; r=0.60 (p=0.023) between DG and butyric acid.

**Table 7.** Rumen fluid constituents, correlation coefficients between daily gain and rumen fluid constituents and the statistical results by ANOVA

| | Month[1)] | | | Correlation coefficient | Source of variance by ANOVA[2)] | |
|---|---|---|---|---|---|---|
| | June | August | October | | Month | Heifer |
| Total VFA, nmol/dl | 4.50 | 4.55 | 6.42 | -0.46[†] | † | NS |
| VFA molar proportions, % | | | | | | |
| Acetic acid | 69.35[b] | 74.76[a] | 69.52[b] | -0.32 | * | NS |
| Propionic acid | 16.23 | 15.74 | 17.80 | -0.14 | NS | NS |
| Butyric acid | 12.05[a] | 7.18[c] | 9.94[b] | 0.60* | *** | NS |
| Acetic acid/propionic acid | 4.29[ab] | 4.85[a] | 3.92[b] | -0.01 | † | NS |
| $NH_3$-N, mg/dl | 9.28[b] | 7.96[b] | 19.69[a] | -0.14 | * | NS |

[1)] Values in each month are expressed as the means of 5 determinations.

[2)] NS: non significant, †: p<0.1, *: p<0.05, **: p<0.01, and ***: p<0.001.

[abc] Means in a row without a common superscript letter differ significantly by students t-test (p<0.05).

† p<0.1 and * p<0.05.

frequency of LRum were significantly different, and frequency of SR tended to be different. Frequency of LRum was positively and that of SRum was negatively correlated with DG. Arave et al. (1974) observed that cows lower in dominance rank tended to have more activity in order to avoid dominant cows. Beilharz and Mylrea (1963) also reported that heifers with low dominance values tried to get out of range of superior heifers. Hasegawa et al. (1997) observed that ingestive time and frequency of lying in bouts shorter than 15 min. was greatest in subordinate among dominance rank. In this experiment, social hierarchy was not investigated, however, it is supposed that those results might show the effects of dominance order in the herd on behavior of heifers and that behavior of subordinates was often interfered and interruped by dominants.

Constituents of blood and rumen fluid changed among months. Greater DG was accompanied with lower blood total protein and phospholipid concentrations, and greater butyric acid proportion in rumen fluid. Other researchers observed higher (Holden et al.) or lower (Berzaghi et al.) acetic acid proportion and $NH_3$-N concentration in rumen fluid than those of this experiment. It is considered that our results were affected by seasonal changes of chemical composition of herbage and DM intake.

As a summary of this study, heifers of lower daily gain on pasture were characterized by having a longer standing time, a shorter total rumination and a shorter lying-rumination time, and the greater frequencies of standing-rumination. They also had a higher protein concentration in blood and a lower proportion of butyric acid in rumen fluid when the nutritive value of pasture was poor or the amount of pasture was short.

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