Effect of Three Different Species of Hay on Dry-matter Intake and Serum Cortisol of Ewes

J. Sekine*, Mootaz A. M. Abdel-Rahman[1], A. El-Moez A. Ismail[1], R. M. Dosoky[1], H. E. M. Kamel[2] and M. Hishinuma

Department of Veterinary Sciences, Faculty of Agriculture, Tottori University, Tottori 680-8553, Japan

ABSTRACT : To determine whether the feeding of different species of hay affects the dry-matter intake and the serum cortisol level of sheep, 6 non-pregnant, non-lactating ewes were offered alfalfa (*Medicago sativa*) hay (Al), oats (*Avena sativa*) hay (Ot) and perennial ryegrass (*Lolium perenne*) hay (Pr) under 6 kinds of treatment including 1) change from Al to Ot, 2) Al to Pr, 3) Ot to Al, 4) Ot to Pr, 5) Pr to Al, and 6) Pr to Ot. The experimental design was a 6×6 Latin square with a 14 day period of which 9 days were a preliminary period and 5 days as a digestion trial period. The change in hay feeding was done abruptly on the first day of each period without an adaptation period. The blood from the jugular vein was collected on the first, third and the last day of each period through a sterile catheter for the analysis of cortisol. The dry-matter intake was recorded daily throughout the period. The mean daily intake of dry matter (DM, g/kg live weight) was significantly different among the 3 species of hay (Al>Ot>Pr; p<0.05). The digestibility of DM for Al and Ot was the same, but that for Pr was significantly lower than Al and Ot (p<0.05). The mean serum cortisol levels were significantly different among the hays (p<0.05). The level for Pr was the highest and that for Al was the lowest. The abrupt change of hay feeding of 6 treatments produced a significant change in the serum cortisol levels. The DM intake was inversely related to the change of the cortisol level. It is suggested that the animal's intake response to different species of hay may be partly motivated by the psychological feelings toward the hay offered. ***(Asian-Aust. J. Anim. Sci. 2003. Vol 16, No. 9 : 1297-1302)***

Key Words : Forage, Dry-matter Intake, Forage, Serum Cortisol, Sheep

INTRODUCTION

The factors affecting feed intake have been intensively studied on ruminants for a long time. The diverse theories proposed by several workers have been intensely reviewed by Forbes (1995a). Among them, the various nutritional and physical factors influencing forage dry-matter intake have been well documented (Forbes, 1995a). It is generally accepted that the intake of a forage is influenced by the capacity of the digestive tract to handle bulk (Forbes, 1998). The bulkiness of a forage is negatively correlated with the concentration of slowly digested nutrients in a diet such as fiber influencing the rate of digestion (Sekine et al., 1995). Yet, the chemical and physical characteristics of forage may explain only two third of the variation in the forage intake by sheep (Tomari et al., 1995). Forbes (1994) has postulated that ruminants ingest a quantity of feed that gives them the most comfortable feelings. Furthermore, intake is primarily a behavior that is influenced by hunger, which is a painful feeling and by satiety, which is generally a pleasant one (Reed, 1992). Faverdin et al. (1995), therefore, have postulated a hedonic behavior for feed consumption, which relates to the senses that animals anticipate the post-ingestive effects of feed. The central nervous system responding to signals from various receptors generates a total signal of discomfort that influences forage intake, and feedback signals to the central nervous system are divided into two groups that is, pre-ingestive and post-ingestive signals (Dynes et al., 2003). The pre-ingestive feedback signals may reflect the post-ingestive effect of the feed through the previous experience of it. Pre-ingestive feedback may give an adverse effect on intake of some forages (Dynes et al., 2003). Thus, forage intake may be partially influenced by some psychological condition of the animals. Forbes (1995a) has suggested that the degree of stress may have influenced the results of the experiments on the control of the intake. There is, however, no study reported on the direct relationship between the degree of stress and the forage intake, nor on the trial finding whether forage itself is to be a storessor to the animals. The review paper by Friend (1991) has shown that a single determination of plasma concentrations of glucocorticoids may be limited in investigations of chronic stress, but in more acute situations in which the initial release of glucocorticoids is monitored, plasma concentrations have been very useful.

The present study was to determine whether the feeding of different kinds of hay affects the dry-matter intake and the serum cortisol level of sheep.

* Corresponding Author: Junjiro Sekine. Tel: +81-857-31-5439, Fax: +81-857-31-5347, E-mail: sekine@muses.tottori-u.ac.jp

[1] Department of Animal Hygiene, Faculty of Veterinary Medicine, Assiut University, Assiut, Egypt.

[2] Department of Animal Production, Faculty of Agriculture, Alexandria University, Alexandria, Egypt.

Received January 3, 2003; Accepted May 13, 2003

Table 1. Chemical composition of three species of hay offered to ewes

Hay	DM	C P	Cellulose	Hemicellulose	ADL
	g/kg as is	g/kg DM			
Alfalfa	889	198	339	82	56
Oats	888	60	295	245	41
Perennial ryegrass	899	56	358	271	58

MATERIALS AND METHODS

Animals used were 6 non-pregnant, non-lactating adult ewes (mean live weight 61.7±2.9 kg) of which 3 were Suffolk and 3 were Corriedale. They were kept individually in a metabolism crate throughout the experimental period. Three species of forage offered to ewes as a sole diet were alfalfa (*Medicago sativa*) hay (Al), oats (*Avena sativa*) hay (Ot) and perennial ryegrass (*Lolium perenne*) hay (Pr) chopped by a precision cutter at a 5 cm length. Ewes were offered hay *ad libitum* twice a day at 08:00 and 16:00 h. Refusals were weighed immediately before the next morning feeding to measure the dry-matter intake. Drinking water and mineralized salt licks were offered with free access. Samples of hays offered and feed refusals were collected and pooled to each animal for further chemical analyses. To study the effect of previous hay eaten on the intake of the hay offered during the next period, 6 kinds of the treatments were prepared as follows: 1) change from Al to Ot, 2) Al to Pr, 3) Ot to Al, 4) Ot to Pr, 5) Pr to Al, and 6) Pr to Ot. The experimental design was a 6×6 Latin square with a 14-day period of which 9 days were a preliminary period and 5 days as a digestion trial period. Before starting the experiment, ewes were offered one of 3 species of hay randomly for 14 days as an adaptation period, and to estimate the daily intake of each kind of hay. The change in hay feeding was done abruptly on the first day of each period without an adaptation period. The dry-matter intake was recorded daily for 14 days for each animal. Animals were fitted with a sterile catheter in the jugular vein for blood sampling. The catheter was covered with protective elastic bandages and was flushed daily with a heparinized sterile physiological saline. About 10 mL blood samples were collected at 11:00 h on the first day (Day 1), 3rd day (Day 3) and the last day (Day 14) of each period. The sera were separated and stored frozen at -83°C till the analysis of the cortisol. Digestion trials were carried out by the total collection method. The 5% of total weight of fresh feces were collected daily as a sample, and were bulked for each period. The procedure for animal care and management followed the Japanese Law of Animal Care and Management.

Dry matter and nitrogen contents of the hay offered, feed refusals and feces were analyzed by the methods of AOAC (1984), and fibrous fractions were analyzed using the method of Georing and Van Soest (1970). The serum samples were sent to private sectors (FML Service, Farco-Biosystems, Japan) to determine cortisol level by radioimmuno-assay. Statistical analyses were carried out by the GLM procedure of SAS (1995) for completely random design. The analyses of correlation and regression have been carried out by the use of Statistix7 (2000).

RESULTS

Table 1 shows the chemical composition of alfalfa, oats and perennial ryegrass hays. Alfalfa hay contained the highest level of crude protein (CP) among the 3 species of hay. The CP concentration of Ot and Pr was about 60 g/kg DM. The cellulose concentration was around 300 g/kg DM for the 3 hays, but hemicellulose was lower in Al than the other two hays. Acid detergent lignin (ADL) tended to be lower in Ot than Al or Pr. The cell wall constituents consisted of cellulose, hemicellulose and ADL, were 477, 581 and 687 g/kg for Al, Ot and Pr, respectively.

The mean daily intake of DM (g/kg live weight) was significantly different among the 3 hays as shown in Table 2 ($p<0.05$). The DM intake for Al was the highest and for Pr, the least. The digestibility of DM for Al and Ot was same, but that for Pr was significantly lower than Al and Ot ($p<0.05$). The digestibility of CP was significantly greater in Al than the other 2 hays, as expected. The fibrous fractions, however, showed a similar digestibility among the 3 hays except for hemicellulose in Al, which was significantly lower than the other 2 hays.

The daily DM intake (g/kg live wt.) for the 3 species of hay was shown in Table 3 for each day throughout the experimental period together with the mean values for each hay. To study the effect of the previous hay offered on the intake of the following hay, the DM intake of each hay was shown as each period offered after the different hay offered in the previous period. There was no significant effect of the

Table 2. Mean daily intake of dry-matter (g/kg live wt.) and mean digestibility with a standard deviation for nutrients of 3 species of hay offered to ewes

	Dry-matter intake, g/kg wt.	Digestibility			
		DM	CP	Cellulose	Hemicellulose
Alfalfa	$28.5^{a1)}\pm3.7$	$0.626^{a}\pm0.022$	$0.791^{a}\pm0.031$	$0.542^{a}\pm0.020$	
Oats	$21.4^{b}\pm3.7$	$0.626^{a}\pm0.021$	$0.491^{b}\pm0.013$	$0.558^{a}\pm0.021$	$0.596^{b}\pm0.026$
Perennial ryegrass	$14.5^{c}\pm3.6$	$0.488^{b}\pm0.021$	$0.324^{c}\pm0.011$	$0.511^{a}\pm0.020$	$0.524^{b}\pm0.023$

[1)] Figures in the same column with different superscripts differ significantly ($p<0.05$).

Table 3. Effect of previous hay on daily intake of dry matter (g/kg live weight) for 3 species of hay offered to ewes

Hay previously offered	Oats	Perennial ryegrass	Alfalfa	Perennial ryegrass	Alfalfa	Oats
Hay offered	Alfalfa	Alfalfa	Oats	Oats	perennial	Perennial
Day 1	27.9±2.7	$25.1^{A,C1)}$±2.9	25.5^{A}±2.2	24.5^{A}±1.3	13.7±1.9	15.2±3.8
2	25.6±5.0	$24.2^{A,C}$±5.3	$23.7^{A,C}$±1.7	$20.2^{B,C}$±4.3	13.6±2.1	16.7±3.5
3	27.8±5.1	$23.6^{A,C}$±3.7	$22.9^{A,C}$±2.7	$20.3^{B,C}$±3.1	13.0±3.9	15.1±4.7
4	28.6±4.0	$25.4^{A,C}$±4.2	$23.2^{A,C}$±2.9	$19.1^{B,C}$±3.0	13.2±2.9	14.9±5.5
5	29.7±5.7	$25.3^{A,C}$±4.9	$21.7^{A,C}$±3.1	$19.4^{B,C}$±3.4	13.9±3.3	16.6±5.7
6	30.6±3.9	$28.4^{A,D}$±2.4	$21.9^{A,C}$±2.9	$20.1^{A,C}$±4.8	12.6±3.4	15.3±4.4
7	30.6±1.9	$29.3^{A,D}$±3.5	$21.6^{A,C}$±4.2	$20.1^{A,C}$±4.8	13.9±2.9	15.7±6.2
8	30.5±4.1	$28.1^{A,C}$±4.4	$21.8^{A,C}$±3.7	$20.9^{A,C}$±4.7	14.2±2.8	15.9±4.5
9	30.7±3.5	$30.4^{B,D}$±2.5	$21.7^{A,C}$±3.3	$20.1^{A,C}$±6.0	14.0±2.6	16.2±3.3
10	30.7±2.1	$32.1^{B,D}$±3.3	$21.4^{A,C}$±4.6	$19.8^{A,C}$±5.5	12.7±3.6	16.7±3.9
11	30.7±2.3	$32.2^{B,D}$±3.8	$21.7^{A,C}$±3.4	$20.3^{A,C}$±4.3	13.5±1.9	16.1±2.7
12	29.5±2.6	$32.4^{B,D}$±4.7	$21.6^{A,C}$±3.2	$21.1^{A,C}$±4.3	14.3±2.8	16.8±2.9
13	27.7±5.0	$31.3^{B,D}$±3.9	$22.2^{A,C}$±4.1	$20.5^{A,C}$±3.6	12.2±3.6	15.8±3.7
14	27.8±1.8	$32.6^{B,D}$±5.1	$20.2^{B,C}$±3.5	$20.2^{B,C}$±3.8	12.7±1.7	14.6±4.9
Mean	$29.2^{a2)}$±3.6	26.5^{a}±3.9	22.2^{b}±3.2	22.2^{b}±3.2	13.4^{c}±2.8	15.8^{c}±4.0

1) The values in the same column with different superscripts differ significantaly (p<0.05).
2) The mean values in the same row with different superscripts differ significantly (p<0.05).

Table 4. Mean serum cortisol level (ng/ml) for ewes given 3 species of hay

Day	Alfalfa	Oats	Perennial ryegrass
1	$32^{A1)}$±2	39^{A}±5	81^{A}±5
3	30^{A}±3	31^{B}±2	97^{B}±7
14	25^{B}±2	38^{A}±2	77^{A}±7
Mean	$29^{a2)}$±2	36^{b}±3	85^{c}±6

1) The values in the same column with different superscripts differ significantly (p<0.05).
2) The mean values in the same row with different superscripts differ significantly (p<0.05).

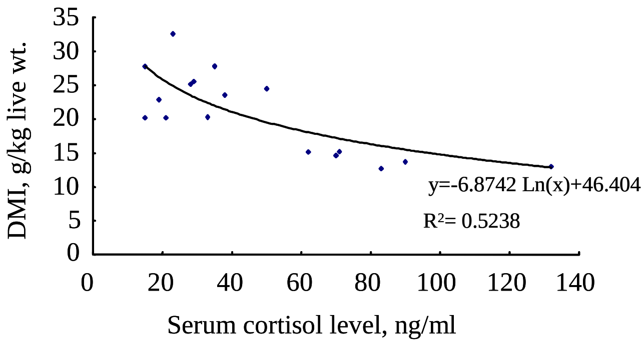

Figure 1. Relationship between DMI and serum cortisol level of ewes offered 3 species of hay.

previous hay on the mean DM intake of the following hay, irrespective of species of hay, although there was a significant difference among 3 hays. Ewes, however, tended to increase the intake of alfalfa offered after perennial ryegrass throughout 14-day period, while increasing tendency of intake ceased in 6 days and reduced to the beginning level in the treatment of alfalfa offered after oats. The level of DM intake for the treatment for Al to Ot tended to decrease in 5 days after oats hay offered, and then stayed at about the same level. For the treatment for Pr to Ot, the intake was the highest on Day 1, but decreased to the level for the rest of the period on Day 2. The intake of perennial ryegrass offered after alfalfa or oats was consistently low throughout the period, although perennial ryegrass offered after oats tended to be higher than that offered after alfalfa, although there was no statistical significance.

The mean serum cortisol levels of ewes were shown in Table 4 on the first, third and the last day of each period, together with the overall mean for each hay. The pattern of day-to-day changes of cortisol level differed among 3 hays. The level on Day 1 was significantly higher than that on Day 14 for alfalfa hay (p<0.05), but the difference was not significant between Day 1 and Day 14 for oats and perennial ryegrass hay (p>0.05). Ewes given Pr showed the highest level of serum cortisol among 3 hays (p<0.05). The Al showed the lowest cortisol level (p<0.05) and Ot was also lower than Pr (p<0.05).

To study the effect of the previous hay offered on the level of serum cortisol for the following hay, comparison was done between the serum cortisol level and DM intake on Day 1 and those on Day 14 of the previous period as shown in Table 5. For the treatments for Ot to Al and Pr to Al, the serum cortisol level significantly decreased on Day 1 immediately after Day 14 of the previous hay (p<0.05). For the treatments for Al to Ot, Al to Pr, and Ot to Pr, the level significantly increased on Day 1 immediately after Day 14 (p<0.05). The DM intake was inversely related to the change of the cortisol level, although significant differences were not found in the treatments for Ot to Al and Ot to Pr.

Table 5. Comparison between the serum cortisol level (ng/mL) and DMI (g/kg wt.) on the first day offered either one of 3 species of hay and on the last day of the previous period

Hay offered on day 14 of the previous period	Cortisol[1] ng/ml	DMI[2] g/kg wt.	Hay offered on day 1 of the previous period	Cortisol ng/ml	DMI g/kg wt.
Oats	42±2	20.2±3.8	Alfalfa	35±2	27.9±2.7
Perennial	78±6	14.6[a]±4.9	Alfalfa	28±2	25.1[b]±2.9
Alfalfa	26±2	32.6[a]±5.1	Oats	38±6	25.5[b]±2.2
Perennial	76±8	12.7[a]±1.7	Oats	40±4	24.5[b]±1.3
Alfalfa	24±2	27.8[a]±1.8	Perennial	80±4	13.7[b]±1.9
Oats	42±2	20.2±3.5	Perennial	81±6	15.2±3.8

[1] Cortisol levels in the same row differ significantly ($p<0.05$). [2] DMI in the same row with different superscripts differ significantly ($p<0.05$).

The relationship between DM intake and serum cortisol level was shown in Figure 1 by using pooled results. The DM intake and cortisol showed a significant and negative curvilinear relationship. The coefficient of determination, however, was as low as 0.5 as shown in the figure.

DISCUSSION

A lower concentration of CP in Ot and Pr was responsible for a lower digestibility of CP. The fibrous fractions for the 3 species of hay, however, were digested at about the same level. The results of *in vitro* experiment showed that the maximum level of rumen ammonia was 5 mg/mL for the maximum growth of the rumen bacteria (Satter and Slyter, 1974). This level has been confirmed *in vivo* (Satter and Roffler, 1981). Although we have not determined the level of ammonia in the rumen, it is speculated that the level of ammonia in the rumen would exceed the level described above. Thus, it is assumed that the nitrogen in the rumen may have been supplied to a level sufficient for microbial needs for the fermentation of fibrous fractions, even though CP content was low in Ot and Pr, since nitrogen in forage generally as a higher degradability in the rumen (ARC, 1980). Therefore, the digestibility of cellulose or hemicellulose was not significantly different among the 3 hays except for hemicellulose of Al, which was an extremely low concentration compared with the other 2 hays. Fibrous fractions are generally accepted as one of factors related to physical characteristics of forage such as bulkiness (Dynes et al., 2003). Forbes (1995a) has described that with ruminants, there is a positive relationship between the digestibility of forages and the level of voluntary intake, due to physical limitation. However, he pointed out that the capacity of the rumen is rarely the only factor controlling feed intake. The study on sheep has shown that the chemical and physical characteristics of forage may explain only two third of the variation in the forage intake (Tomari et al., 1995). In the present study, the DM intake was significantly greater in Al than Ot, nevertheless Al and Ot showed the same DM digestibility. Cattle consumed 22.6 g/kg wt. of Napier grass with similar CP but higher NDF contents to Ot, but ram ingested only 13.4 g/kg wt. (Gwayumba et al., 2002). Thus, DM intake of Ot may be satisfactory to ewes and that of Pr may be comparable to that reported by ram. The bulkiness of hay was negatively correlated with the hay intake by sheep (Tomari et al., 1995). The mean values of dry bulk density (DBD, g/mL) for Al, Ot and Pr reported by Tomari et al. (1995) were 0.189, 0.154 and 0.104 g/mL of each hay, respectively. The proportion of DBD for Al to Ot is calculated to be 1.23, which showed a fair agreement with that of DM intake for both hays, that is 1.33.

Similar agreement is found in the relation of Al to Pr or Ot to Pr. These facts show that the capacity of the rumen to accommodate the bulky food is one of limiting factors for DM intake of forage. Thus, the digestibility of the forage may be responsible for reducing the volume of the rumen occupied by forage. The mean intake of 3 species of hay was not affected by the species of hay offered immediately before, although the mean intake was significantly different among 3 hays (Table 3). When alfalfa was offered after oats hay or perennial ryegrass hay, DM intake was significantly higher than in the case that those two hays were offered after alfalfa (Table 3). This phenomenon was also found in the case of oats hay offered after perennial ryegrass hay (Table 3). However, DM intake was significantly lower in perennial ryegrass offered after alfalfa or oats hay than those of two hays. Therefore, DM digestibility may not be a main determinant for hay intake in the present study, but animals may have responded to factors other than DM digestibility.

Mean levels of serum cortisol for 3 species of hay were significantly different among hays used in the present study (Table 4), and were inversely related to mean intakes of 3 species of hay. These responses to different hays by animals may be caused by a psychological sensation to the different hays as suggested by Read (1992). Psychological factors are effective in increasing pituitary-adrenal activity determined by plasma cortisol concentrations as an index of sensitivity to environmental stimuli, and thus, the pituitary-adrenal response seems more related to emotional reaction (Dantzer and Mormede, 1983). Kronberg et al. (1993) also reported that the weed inducing aversion of ingestion increased blood cortisol levels of sheep, when they were dosed with leafy spurge at the level of 0.3% of body weight. Therefore, the difference in intake of 3 species of hay may have been caused by a difference in the comfortable feelings that have been produced in sheep by the hedonic effects of hays offered as postulated by Forbes (1995b). The difference in CP content of hays may have caused the hedonic sensation to animals accelerating ingestion of alfalfa because the serum cortisol showed the lowest level among the 3 species of hay. This is the case when hays were changed from alfalfa to the other two hays or vise versa. In each case, sheep increased the level of intake of alfalfa with a decreased level of serum cortisol, when they were offered alfalfa on the next day when they have been given either oats or perennial ryegrass hay for 14 days (Table 5). The situation was reversed when they were offered oats or perennial ryegrass hay after alfalfa. There was a significant negative correlation between the serum cortisol level and the hay intake level ($p<0.01$). Oats and perennial ryegrass hays contained CP at about 60 g/kg DM comparing about 200 g/kg DM for alfalfa. The change from oats to ryegrass caused a decrease in DM intake and a significant increase in cortisol level, while DM intake significantly increased and the cortisol level decreased significantly in the change from ryegrass to oats. Thus, the CP content may not play a sole determining role for hay intake. These results showed the same phenomenon described by Forbes and Kyriazakis (1995) as a paradox of two feeds with low protein contents. They speculated that animal will have consumed the less-limiting feed and have avoided the more-limiting low protein feed. They also described that in some instances, however, animals appeared to consume appreciable amounts of the more-limiting feed. Kyriazakis et al. (1990) postulated the expectation in the pig that an animal will have preferred the less-limiting feed, but it will also have consumed some from the more-limiting feed when it was given a choice between two limiting feeds. In the present study, the change from oats to perennial ryegrass hay or vise versa did not allow animals to make a choice of forage. But, they consumed oats hay or ryegrass hay at a fairly constant level for a 14-day period with a little day-to-day variation in intake (Table 3). Yet, the changes of intake occurred immediately after the change in species of hay offered. Therefore, it may not be responsible for an amplified sampling behavior of feed intake as described by Forbes and Kyriazakis (1995), nor differences in digestibility or bulkiness of the hay previously offered. The change in intake of a given hay may have been caused by some hedonic effects of its properties which may generate a sensation of preference like the effect of a starch flavor on rats (Ramirez, 1993). However, the serum cortisol level would not be a sole determinant for DM intake because the coefficient of determination for the regression analysis was fairly low as shown in Figure 1.

Pearson and Mellor (1976) demonstrated that untamed sheep have adjusted to a new environment within two weeks, when they were brought in directly from the field. The animal response observed in the present study may not be caused by environmental conditions other than species of hay offered, because the 14-day adaptation period was provided before starting the experiment.

It is suggested that the animal response to species of hay is partly motivated by factors other than the nutritional or physical characteristics of the hay. Therefore, the like or dislike feelings toward the hay being offered may have affected the DM intake of the different hays.

REFERENCES

Agricultural Research Council 1980. The Nutrient Requirements of Ruminant Livestock. Commonwealth Agricultural Bureaux, Slough Analytical Software Statistix7 2000. User's Manual. Analytical Software, Tallahassee, FL. USA.

Association of Official Analytical Chemists 1984. Official Methods of Analysis. 14th ed. AOAC. Washington DC.

Dantzer, R. and P. Mormede. 1983. Stress in farm animals: A need for reevaluation. J. Anim. Sci. 57:6-18.

Dynes, R. A., D. A. Henry and D. G. Masters. 2003. Characterising forages for ruminant feeding. Asian-Aust. J. Anim. Sci. 16:116-123.

Faverdin, P., B. Baumont and K. L. Ingvarsten. 1995. Control and prediction of feed intake in ruminants. (Ed. M. Journet, E. Grenet, M-H. Face, M. Theriez, C. Demarquilly) In: Recent Developments in the Nutrition of Herbivores. INRA, Paris. pp. 95-120.

Forbes, J. M. 1994. Physical control of feed intake in ruminants and its interactions with other controls. Proc. Soc. Nutr. Physiol. 3:119.

Forbes, J. M. 1995a. Voluntary Food Intake and Diet Selection in Farm Animals. CAB International, Wallingford, UK.

Forbes, J. M. 1995b. Physical limitation of feed intake in ruminants and its interactions with other factors affecting intake. (Ed. W. V. Engelhardt, S. Leonhard-Marek, G. Breves and D. Giesecke) In: Ruminant Physiology: Digestion, Metabolism, Growth and Reproduction. Ferdinand Enke Verlag, Stuttgart. pp. 217-232.

Forbes, J. M. 1998. Control of feed intake to enhance milk production in dairy cows. Proc. 8th WCAP, Seoul National University, Symposium Series 2:185-194.

Forbes, J. M. and I. Kyriazakis. 1995. Food preferences in farm animals: why don't they always choose wisely? Proc. Nutr. Soc. 54:429-440.

Friend, T. H. 1991. Behavioral aspects of stress. In Symposium: Response of Animals to Stress. J. Dairy Sci. 74:292-303.

Georing and P. J. Van Soest. 1970. Forage Fiber Analyses (Apparatus, Reagents, Procedures and Some Applications) Agriculture Handbook No. 379. USDA. Washington DC.

Gwayumba, W., D. A. Christensen, J. J. McKinnon and P. Yu. 2002. Dry matter intake, digestibility and milk yield by Friesian cows fed two Napier grass varieties. Asian-Aust. J.

Anim. Sci. 15:516-521.

Kronberg, S. L., J. W. Walker and J. A. Fitzgerald. 1993. Feeding behavior of grazing ruminants experiencing stress. Physiol. Behav. 54:1191-1194.

Kyriazakis, I., G. C. Emmans and C. T. Whittemore. 1990. Diet selection in pigs: choices made by growing pigs given foods of different protein concentrations. Anim. Prod. 51:189-199.

Pearson, R. A. and D. J. Mellor. 1976. Some behavioural and physiological changes in pregnant goats and sheep during adaptation to laboratory conditions. Res. Vet. Sci. 20:215-217.

Ramirez, I. 1993. Relative preference for starch and sugar in rats. Physiol. Behav. 54:1195-1200.

Reed, N. W. 1992. Role of gastrointestinal factors in hunger and satiety in man. Proc. Nutr. Soc. 51:7-11.

Satter, L. D. and R. E. Roffler. 1981. Influence of nitrogen and carbohydrate inputs on rumen fermentation. In: Recent Developments in Ruminant Nutrition. (Ed. W. Haresign and D. J. A. Cole). 115-139. Butterworth, London.

Satter, L. D. and L. L. Slyter. 1974. Effect of rumen ammonia concentration on rumen microbial production *in vitro*. Br. J. Nutr. 34:199-208.

Sekine, J., J. Ueda, Jin Hai, H. E. M. Kamel, R. Oura and Z. Morita. 1995. Effects of chemical and physical characteristics of forage on dry-matter intake of cattle. Ann. Zootech. 44(Suppl.):253. abstr.

Statistical Analysis System, 1995. User's Guide: Statistics. Ver. 6. 2nd ed. SAS Inst. Inc. Cary, USA.

Tomari, T., H. E. M. Kamel, Jin. Hai, R. Oura and J. Sekine. 1995. Effects of crude protein, neutral detergent fiber, dry bulk density, and brittleness of the hay on feed intake of sheep. Jpn. J. Sheep Sci. 32:7-13. (In: Japanese with English summary).