Effects of feeding level on nutrient digestibility and enteric methane production in growing goats (Capra hircus hircus) and Sika deer (Cervus nippon hortulorum)

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Objective: Two experiments were conducted to determine the effects of feeding level on nutrient digestibility and enteric methane (CH4) emissions in growing goats and Sika deer.

Methods: Three growing male goats (initial body weight [BW] of 22.4±0.9 kg) and three growing male deer (initial BW of 20.2±4.8 kg) were each allotted to a respiration-metabolism chamber for an adaptation period of 7 d and a data collection period of 3 d. An experimental diet was offered to each animal at one of three feeding levels (1.5%, 2.0%, and 2.5% of BW) in a 3×3 Latin square design. The chambers were used for measuring enteric CH4 emission.

Results: Nutrient digestibility decreased linearly in goats as feeding level increased, whereas Sika deer digestibility was not affected by feeding level. The enteric production of CH4 expressed as g/kg dry matter intake (DMI), g/kg organic matter intake, and % of gross energy intake decreased linearly with increased feeding level in goats; however, that of Sika deer was not affected by feeding level. Six equations were estimated for predicting the enteric CH4 emission from goats and Sika deer. For goat, equation 1 was found to be of the highest accuracy: CH4 (g/d) = 6.2 (±14.1)+10.2 (±7.01)×DMI (kg/d)+0.0048 (±0.0275)×dry matter digestibility (DMD, g/kg)–0.0070 (±0.0187)×neutral detergent fiber digestibility (NDFD; g/kg). For Sika deer, equation 4 was found to be of the highest accuracy: CH4 (g/d) = –13.0 (±30.8)+29.4 (±3.93)×DMI (kg/d)+0.046 (0.094)×DMD (g/kg)–0.0363 (±0.0636)×NDFD (g/kg).

Conclusion: Increasing the feeding level increased CH4 production in both goats and Sika deer, and predictive models of enteric CH4 production by goats and Sika deer were estimated.

Keywords: Capra hircus hircus; Cervus nippon hortulorum; Methane; Greenhouse Gas; Feeding Level

INTRODUCTION

Enteric methane (CH4) production in ruminants is recognized as one of the major sources of greenhouse gas emissions associated with agriculture [1]. Although a large number of goats and Sika deer inhabit East Asia [2], few studies have estimated enteric CH4 production of goats and Sika deer. Enteric CH4 production by ruminants is generally influenced by various dietary factors such as the forage-to-concentrate ratio [3], carbohydrate type [2,4], forage processing [5], fat addition [6], and ionophore addition [7]. Fundamentally, because enteric methane production is a part of the energy utilization process of ruminants, the amount of feed intake has been suggested as the major factor driving enteric CH4 production. Changes of feed intake level affect passage rate and rumen fermentation characteristics and, thereby, can alter nutrient digestibility and CH4 production of the animal [8]. Therefore, the aims of the present study were i) to determine the effects of feeding level on nutrient digestibility and enteric CH4 production and ii) to estimate the for the respective enteric CH4 produc-
tion prediction equations for growing goats and Sika deer.

MATERIALS AND METHODS

Experiments were conducted to determine the nutrient digestibility and enteric CH$_4$ and CO$_2$ emissions in goats (*Capra hircus hircus*) and Sika deer (*Cervus nippon hortulorum*) in accordance with the Institutional Animal Care and Use Committee of Konkuk University.

Animals, diets, and experimental design

Three growing male goats (initial body weight [BW] of 22.4±0.9 kg) and three growing male deer (initial BW of 20.2±4.8 kg) were used in this study. Each animal was housed individually in a respiration-metabolism chamber as described by Li et al [9] for a diet adaptation period of 7 d and a data collection period of 3 days [10,11]. The same experimental diet was prepared and used for both goat and deer experiments (Table 1), and animals were randomly assigned to one of three feeding levels (1.5%, 2.0%, or 2.5% of BW), which were applied in a 3×3 Latin square design. Experimental diets were fed daily at 1100 h, and water and mineral block (containing 3,250 K IU/kg vitamin A, 500 K IU/kg vitamin D, 2,500 IU/kg vitamin K, 200 mg/kg vitamin B$_6$, 200 mg/kg vitamin B$_2$, 250 μg/kg vitamin B$_12$, 3,500 mg/kg niacin, 9,000 mg/kg choline chloride, 48,000 mg/kg methionine, 10,200 mg/kg glycine, 100 mg/kg β-carotene, 254 g/kg calcium, 38 g/kg phosphorus, 4,000 mg/kg manganese, 9,050 mg/kg zinc, 4,500 mg/kg iron, 200 mg/kg iodine, 45 mg/kg cobalt, 11,000 mg/kg magnesium, 2,300 mg/kg copper, 92 mg/kg selenium) were freely accessible. Sampling unit was feces which collected daily (n = 3 d/animal). Total fecal samples were collected at 1000 h and immediately dried for subsequent chemical analysis. Digestibility coefficients were calculated by the following formula: nutrient digestibility (%) = [nutrient intake (g) – nutrient in the fecal (g)]/nutrient intake (g)×100. Experimental unit was animal (n = 3 animals/treatment).

Chemical analysis

Feed ingredients and fecal samples were analyzed in duplicate for dry matter (DM), organic matter (OM), crude protein (CP), and ether extract as described by AOAC [12]. Neutral detergent fiber (NDF) was analyzed using heat stable α-amylase (Sigma A3306; Sigma Chemical Co., St. Louis, MO, USA) according to the method described by Van Soest et al [13]. Gross energy (GE) was analyzed using a bomb calorimeter (C5000; IKA, Staufen, Germany).

Gas production measurement

A respiration-metabolism chamber system was used to measure enteric CH$_4$ and CO$_2$ production [9], and a gas flow meter (GF557, Aalborg Instruments & Controls Inc., Orangeburg, NY, USA) and a sample pump (Columbus Instruments, Columbus, OH, USA) were used to collect inlet and outlet gas samples. Collected gas samples were passed through a desiccant (CaSO$_4$) before the samples flew into the gas analyzer. A non-dispersive infrared gas analyzer (VA-3000; Horiba Stec Co., Kyoto, Japan) was used to analyze the concentrations of CH$_4$ and CO$_2$.

Statistical analysis

All variables were analyzed using SAS PROC MIXED (Version 9.2; SAS Institute Inc., Cary, NC, USA). The statistical model included treatment as a fixed effect and animal and period as random effects. The orthogonal polynomial contrast was tested using the CONTRAST statement to examine the linear and quadratic effects of feeding level on the response variables. Significant differences were declared at p<0.05, and trends were defined at 0.05≤p<0.10. SAS PROC REG (Version 9.2; SAS Institute Inc., USA) was used to estimate simple and multiple linear equations. Equations were evaluated based on the root mean square error (RMSE), adjusted-R$^2$, and the p-value.

RESULTS

Dry matter intake (DMI) of both goats and Sika deer increased linearly (p<0.05) as feeding level increased (Table 2). The DM, OM, CP, and neutral detergent fiber digestibility (NDFD) of goats decreased linearly (p<0.05) as the feeding level increased. However, the DM, OM, CP, and NDFD of Sika deer did not differ significantly among feeding level treatment groups.

In goats, enteric emission of CH$_4$ expressed as g/d and g/kg BW$^{0.75}$ increased linearly (p<0.05) with increasing feeding level (Table 3); however, when expressed as g/kg DMI, g/kg organic matter intake (OMI), and % of gross energy intake (GEI), it decreased linearly (p<0.05) with increasing feeding level. No difference in enteric CH$_4$ production when expressed as g/kg digested dry matter intake (DDMI) and g/kg digested organic matter intake (DOMI) was observed among feeding

Table 1. Ingredients and nutrient composition of experiment diets

| Items                     | Experimental diet |
|---------------------------|-------------------|
| Ingredients (%; DM basis) |                   |
| Ground corn               | 25.5              |
| Soybean meal              | 24.5              |
| Tall fescue, hay          | 50.5              |
| Nutrient composition      |                   |
| DM (%)                    | 89.3              |
| OM (%) DM                 | 92.1              |
| CP (%) DM                 | 16.0              |
| NDF (%) DM                | 44.0              |
| GE (MJ/kg DM)             | 15.9              |

DM, dry matter; OM, organic matter; CP, crude protein; NDF, neutral detergent fiber; GE, gross energy.
levels. In Sika deer, enteric emission of CH$_4$ expressed as g/d and g/kg BW$^{0.75}$ increased linearly (p<0.05) with increasing feeding level, while it was not affected by feeding level when expressed as g/kg DMI, g/kg OMI, or % of GEI. In goats, CO$_2$ production expressed as g/d and g/kg BW$^{0.75}$ increased linearly (p<0.05) with increasing feeding level, and in Sika deer, enteric CO$_2$ production expressed as g/kg was not affected by feeding level. However, CO$_2$ production expressed as g/kg BW$^{0.75}$ increased linearly (p<0.01) with increasing feeding level.

For goats, the model that used the DMI, DM digestibility (DMD), and NDFD as independent variables (equation 1), was most accurate (Table 4; R$^2$ = 0.69, RMSE = 1.05, and p = 0.096), whereas for Sika deer, the model that used the DMI, DMD, and NDFD as independent variables, was most accurate (R$^2$ = 0.95, RMSE = 0.98, and p = 0.001).

DISCUSSION

The amount of feed intake is fundamentally important for animals because it directly affects animal productivity. In general, feeding level and nutrient digestibility in ruminants are negatively correlated [14,15] because a change in feeding level can alter the passage rate and fermentation pattern of the reticulo-rumen [16]. In the current experiment, the nutrient

| Items       | Feeding level (% of BW) | p-value |
|-------------|-------------------------|---------|
|             | 1.5  | 2.0  | 2.5  | SEM  | Linear | Quadratic |
| Dry matter intake (g) |       |       |       |       |        |           |
| Goats       |      |       |       |       |        |           |
| Sika deer   |      |       |       |       |        |           |
| Digestibility (%) |       |       |       |       |        |           |
| Goats       |       |       |       |       |        |           |
| Sika deer   |       |       |       |       |        |           |

BW, body weight; SEM, standard error of the mean; DM, dry matter; OM, organic matter; CP, crude protein; NDF, neutral detergent fiber.

| Items       | Feeding level (% of BW) | SEM  | p-value |
|-------------|-------------------------|------|---------|
|             | 1.5  | 2.0  | 2.5  | Linear | Quadratic |
| Goats       |       |       |       |        |           |
| Sika deer   |       |       |       |        |           |
| CH$_4$ production |       |       |       |        |           |
| CH$_4$ (g/d) |      |       |       |       |        |           |
| CH$_4$ (g/kg BW$^{0.75}$) |   |       |       |       |        |           |
| CH$_4$ (% of GEI) |   |       |       |       |        |           |
| CH$_4$ (g/kg DMI) |   |       |       |       |        |           |
| CH$_4$ (g/kg OMI) |   |       |       |       |        |           |
| CH$_4$ (g/kg DOMI) |   |       |       |       |        |           |
| CO$_2$ production |       |       |       |        |           |
| CO$_2$ (g/d) |      |       |       |       |        |           |
| CO$_2$ (g/kg BW$^{0.75}$) |   |       |       |       |        |           |

BW, body weight; SEM, standard error of the mean; CH$_4$, methane; GEI, gross energy intake; DMI, dry matter intake; DDMI, digested dry matter intake; OMI, organic matter intake; DOMI, digested organic matter intake; CO$_2$, carbon dioxide.
digested by intermediate-type and grass/roughage eaters, it seems that the overall effect of feeding level on enteric CH₄ production can be diminished in Sika deer. In both goats and Sika deer, enteric CH₄ production expressed as g/kg DDMI and g/kg DOMI did not differ statistically. Although digestibility is difficult to measure and requires complex equipment, Na et al [3] reported that digested nutrient intake is a critical factor that explains CH₄ production in goats and Sika deer. Therefore, digested nutrient intake can be used as a CH₄ prediction factor when the experimental condition allows. Mineral salt supplementation can influence the enteric methane emission and methanogenic archaea population in rumen [30]. In the current study, mineral block intake did not measure, however, it appears that the effect of mineral block was slight because the animals can freely access to the mineral block.

The models that used nutrient digestibility as a factor exhibited the greatest accuracy in this as well as other experiments [3,31]. However, equations 3 and 4, which used the DMI as the only factor, can also be used to predict enteric CH₄ because the model without digestibility is more practical. To our knowledge, only one other study has predicted CH₄ production in goats and Sika deer. Therefore, the models evaluated in the current study contribute to our understanding of enteric CH₄ emissions in Sika deer.

**CONCLUSION**

Nutrient digestibility of goats decreased linearly as the feeding level increased, whereas the digestibility of Sika deer was not affected by feeding level. The enteric production of CH₄ expressed as g/kg DDMI, g/kg OMI, and % of GEI decreased linearly with increasing feeding levels in goats. However, enteric production of CH₄ expressed as g/kg DDMI, g/kg OMI, and % of GEI in Sika deer were not affected by feeding level.

**CONFLICT OF INTEREST**

We certify that there is no conflict of interest with any financial
organization regarding the material discussed in the manuscript.

ACKNOWLEDGMENTS

This work was supported by Korea Institute of Planning and Evaluation for Technology in Food, Agriculture, Forestry and Fisheries (IPET) through Agri-Bio Industry Technology Development Program, funded by Ministry of Agriculture, Food and Rural Affairs (MAFRA) (314010-4).

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