Feed intake and metabolic use of nutrients are important factors to maximize milk production of lactating dairy cows, and they are partly regulated by metabolic hormones. Growth hormone (GH) is well documented for the increase in milk production through some metabolic regulation to promote lipolysis in the adipose tissue and to enhance gluconeogenesis and biosynthesize of insulin-like growth factor-1 (IGF-1) in the liver. IGF-1 has a galactopoietic effect with increasing blood flow into the mammary gland and with causing insulin resistance. Thus, GH plays a central role in regulating milk production in lactating cows, and hence, the approach to enhance the endogenous GH secretion potentially leads to the increase in milk production. However, such approach has been not established because GH secretion is strongly regulated by central nervous systems. Ghrelin, secreted from the gastrointestinal tract, was identified as a potent GH secretagogue in recent years. In addition, ghrelin is related to appetite regulation; ghrelin injection increases the food intake in laboratory animals. Because circulating ghrelin is sensitive to ingested nutrients, hence the nutritional management or feeding strategy to control ghrelin secretion may be able to adjust the GH secretion. The previous study demonstrated that plasma ghrelin concentration in ruminants was decreased by volatile fatty acids, and was not affected by glucose as a secretory inhibitor in non-ruminants. Another previous study revealed that plasma ghrelin concentration was increased by amino acids in sheep. However, there has been still little information of which nutrients affect plasma ghrelin concentration in ruminants. The objectives of this study were to determine the effects of some nutrients on plasma ghrelin concentration and ghrelin-induced metabolic changes, and to consider ghrelin’s role in controlling milk production of lactating cows.

In the 1st trial, the effects of amino acids on plasma ghrelin concentration and ghrelin action were investigated in lactating cows. A mixture solution of amino acids (AMI) or saline (CON) were intravenously infused to determine plasma ghrelin concentration, and after then, during the infusion of AMI or CON, ghrelin was intravenously injected to determine the ghrelin action. In this experiment, six lactating Holstein cows were randomly assigned to two infusion treatments (AMI or CON) in a cross-over design. There was no difference in
plasma ghrelin concentration between AMI and CON before the ghrelin injection. After the ghrelin injection, plasma GH, glucose, and non-esterified fatty acids (NEFA) concentrations in comparison with before injection immediately increased ($P < 0.05$) with no difference between two infusion treatments. Plasma insulin and glucagon concentrations were also increased by ghrelin injection in two infusion treatments ($P < 0.05$). The peak value of plasma insulin concentration was greater in AMI compared with CON ($P < 0.05$). Plasma glucagon concentration showed no difference at the peak value reached at 5 min in both treatments, and after then, showed sustained higher values in AMI compared with CON ($P < 0.05$). After plasma glucose concentration reached the peak, the degree of decline was greater in AMI compared with CON ($P < 0.05$). These results indicated that the plasma amino acids directly do not affect plasma ghrelin concentration, but may modify the ghrelin actions; amino acids which stimulate glucagon, insulin, and glucose release into the blood circulation.

In the 2nd trial, the effects of medium-chain fatty acids (MCFA) on plasma ghrelin, GH, other metabolic hormone, metabolite concentrations, and milk production were investigated. Five early-lactating Holstein cows were randomly assigned to two dietary treatments in a cross-over design for 2-week periods. The diets were with a supplement of MCFA (MCFA-Ca, 1.5% added on dry matter basis) and without MCFA-Ca (CON). Plasma concentrations of hormones and metabolites in jugular vein were measured around the morning feeding on day 14 of each period. Dry matter intake (DMI) and metabolizable energy intake (MEI) were decreased, and milk yield tended to be decreased in the MCFA-Ca compared with CON diet ($P < 0.05$). Milk protein and lactose contents were decreased in the MCFA-Ca diet ($P < 0.05$). The MCFA-Ca diet increased plasma ghrelin concentration ($P < 0.05$), but did not affect plasma GH concentration, and decreased plasma IGF-1 concentration ($P < 0.05$). The MCFA-Ca diet did not affect plasma glucagon-like peptide-1 (7-36) amide (GLP-1) concentration. Plasma insulin concentration was decreased ($P < 0.05$), and plasma glucagon concentration was not changed with the MCFA-Ca diet. Plasma NEFA, total cholesterol (T-CHO) and beta-hydroxy butyrate (BHBA) concentrations were increased in the MCFA-Ca diet ($P < 0.05$). In conclusion, although plasma GH concentration did not link plasma ghrelin concentration, the MCFA diet increased plasma ghrelin concentration in lactating cows.

In the 3rd trial, the effects of long-chain fatty acids (LCFA) and methionine (Met) on plasma ghrelin, GH, other metabolic hormone, metabolite concentrations, and milk production were investigated. Four lactation Holstein cows were used in a 4 x 4 Latin square experiment for each 2-week period. Cows were fed diets with supplements of calcium salts of LCFA (LCFA-Ca, 1.5% added on dry matter basis), rumen-protected Met (RPM, 20 g/d), LCFA-Ca plus RPM and without supplements (CON). Jugular blood samples were taken from 1 h before to 2 h after morning feeding at 10 min intervals on day 12 of each period. The LCFA-Ca decreased DMI ($P < 0.05$), but RPM did not affect DMI. Both supplements of LCFA-Ca and RPM did not affect MEI and milk yield and composition. Plasma concentrations of NEFA, triglyceride (TG), and T-CHO were increased with LCFA-Ca alone
(\(P < 0.05\)), but the degrees of increases in plasma TG and T-CHO concentrations were moderated by LCFA-Ca plus RPM (\(P < 0.05\)). The LCFA-Ca increased plasma ghrelin concentration (\(P < 0.05\)) and the ghrelin concentration with LCFA-Ca plus RPM was the highest among the treatments. Similarly, plasma GH concentration tended to be increased by LCFA-Ca (\(P = 0.056\)) and showed the highest level in LCFA-Ca plus RPM. On the other hand, plasma IGF-1 concentration was decreased by LCFA-Ca and RPM (\(P < 0.05\)). Plasma GLP-1, glucagon, and insulin concentrations were decreased with LCFA-Ca (\(P < 0.05\)), whereas there was the interaction between LCFA-Ca and RPM in plasma glucagon concentration (\(P < 0.05\)); the LCFA-Ca plus RPM mitigated the decrease of plasma glucagon concentration by LCFA-Ca. These results showed that the supplementation of RPM together with LCFA-Ca increases plasma ghrelin and glucagon concentrations. Although the GH concentration was linked with plasma ghrelin concentration, plasma IGF-1 concentration was not associated with plasma GH and ghrelin concentrations.

In lactating cows, the exogenous ghrelin injection shows certain catabolic effects such as stimulating GH and glucagon secretions, associated with NEFA and glucose releases into the circulation in lactating cows. This study showed that amino acids did not affect plasma ghrelin concentration, but modified these ghrelin actions; ghrelin-induced glucagon and insulin secretions were augmented, and simultaneous glucose removal from the circulation was enhanced by amino acids. This suggests amino acids may counteract the catabolic action of ghrelin. The present study demonstrated that plasma ghrelin concentration varies according to the kinds of available nutrient. MCFA increased plasma ghrelin concentration because they are directly used for acylation of ghrelin. LCFA also increased plasma ghrelin concentration. When Met was fed with LCFA, plasma ghrelin concentration was further increased. This might be caused not by a direct action of Met, but by the indirect action of Met because Met modulates lipid metabolism. The incremental degree of plasma ghrelin concentration was greater in cows fed MCFA (43%) compared with cows fed LCFA plus Met (19%). Nonetheless, the endogenous increase in plasma ghrelin concentration did not increase plasma GH concentration in cows fed MCFA, but increased in cows fed LCFA plus Met. The present study showed that continuous hyper plasma ghrelin concentration did not increase GH secretion always, suggesting that keeping of plasma ghrelin concentration above threshold level is not effective for the stimulation of GH secretion. The increased plasma ghrelin concentration by LCFA plus Met might be also related to some metabolic changes such as the higher plasma glucagon and glucose concentrations.

In conclusion, the kind of available nutrients affect plasma concentration and action of ghrelin in lactating cows. Milk production is not enhanced by the feed supplements as MCFA and LCFA because of their adverse effect on feed intake. However, the changes in plasma ghrelin concentration induced by these feed supplements could mediate major metabolic hormones to control nutrients use in the whole body of lactating cows.