Summary of Doctoral Thesis

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Title	Effect of feeding whole crop corn silage on digestive functions, protein and energy metabolism in sheep exposed to heat

The rise in the environmental temperature due to climate change alters the basic physiology of the ruminants; hence negatively affect ruminant production. Thus exposure to the hot environment is considered as an expensive issue for the global livestock industries. Recently, in connection with the concern of global warming, research on feeding management of ruminant livestock during summer heat exposure is drawing more attention. In terms of nutrient intake and digestibility, dietary roughage is more vulnerable to thermal stress than concentrate. However, studies related to dietary roughage source on physiological response, and digestive and metabolic functions under heat exposure are scarce. Whereas, whole crop corn silage (WCS) is widely used as a feed for ruminant livestock due to its high starch content, excellent palatability, and better digestibility. Therefore, feeding WCS might improve the physiological response, and digestive and metabolic functions of sheep compared to feeding other roughages under heat exposure. Accordingly, a series of experiments were carried-out to clarify the feeding effect of WCS on physiological response, nutrient digestibilities, rumen fermentation characteristics, blood metabolites, protein and energy metabolism in heat exposed sheep compared to other roughages.

The first experiment was conducted with the aim to investigate the potential of feeding WCS diet compared to mixed hay of orchardgrass and reed canarygrass (MH diet) or orchardgrass silage (GS diet) based on their physiological response and digestive functions in heat exposed sheep. Six sheep were fed *ad libitum* with any of the three diets in a replicated 3×3 Latin-square design for 24 days. After 14 days adaptation, they were exposed to thermoneutral environment (20°C; TN) and then heat exposure (30°C; HE) with 70% relative humidity (RH) for 5 days each. Rumen

fermentation characteristics were performed on day 4, while physiological response and nitrogen (N) balance were carried-out for three successive days in each exposure period. Respiration rate (RR) and rectal temperature (RT) of WCS fed sheep were lower ($P<0.05$) than of MH and GS fed sheep and both were higher ($P<0.01$) during heat exposure. The intakes of DM and N and the digestibilities of N and neutral detergent fiber (NDF) were lower ($P<0.05$) for WCS diet than for MH and GS diets and they did not differ between two environmental exposures. However, WCS fed sheep had higher ($P<0.05$) digestibilities of DM and organic matter (OM), as well as metabolizable energy intake than the sheep fed MH or GS diet, and there was no environmental effect on them. As DM and N intakes were lower for WCS diet, the variables of rumen fermentation were also lower ($P<0.05$) than the other two diets except propionate concentration, and they did not differ between environmental exposures except NH_3 concentration. In this experiment, WCS fed sheep were physiologically more tolerant to heat exposure compared to other diets. However, as results were inconsistent in terms of nutrient intake and digestibility and which might be attributed to the lower N content of WCS diet. Therefore, it was concluded for further investigation of feeding WCS with supplemented N.

In connection with the first experiment, second experiment was carried out to explore the protein metabolism potential of WCS diet compared to MH diet in heat exposed sheep under iso-nitrogenous condition. Six shorn sheep were fed either of the diets at maintenance energy level in a crossover design for 24 days, where soybean meal was added to WCS-diet to make it iso-nitrogenous. The adaptation with diets, method and duration of environmental exposures and sample collections were as same as for first experiment. An isotope dilution method using $[^2H_5]$Phe and $[^2H_2]$Tyr was performed on day 5 of each exposure to determine whole body protein synthesis (WBPS). The microbial nitrogen supply (MNS) was determined using urinary purine derivatives (PD) method. The RR and RT were higher ($P<0.01$) at heat exposure, while only RR was affected by diet ($P<0.01$). The RR was lower ($P<0.01$) for WCS diet and it was also lower ($P<0.05$) for WCS diet than the MH diet during heat exposure. The digestibility of DM and N were higher ($P<0.01$) for WCS diet than the

MH diet and both were also higher (P<0.05) for WCS diet at heat exposure compared to thermoneutral. Whereas, N balance was higher (P<0.01) in WCS fed sheep and did not differ between the exposures. Rumen pH was lower (P<0.01) for WCS diet than the MH diet and it was lower (P<0.05) during heat exposure. Rumen NH_3 concentration was higher (P<0.01) for WCS diet than the MH diet and did not differ between the exposures, while only for MH diet it was lower (P<0.05) during heat exposure. The total volatile fatty acid (VFA) and acetate concentrations were lower (P<0.05 and P<0.01; respectively) for WCS diet than the MH diet. The total VFA and propionate concentrations were lower (P<0.05 and P<0.01; respectively) during heat exposure compared to thermoneutral. Plasma glucose concentration was higher (P<0.01) for WCS diet and was lower (P<0.01) during heat exposure. Among plasma free amino acids (AAs), the major glucogenic AAs were higher (P<0.05) for WCS diet than the MH diet and some of these AAs were lower (P<0.05) at heat exposure. The total PD and total MNS did not differ between diets and both were lower (P<0.05) at heat exposure. The turnover rate of plasma Phe (PheTR) was higher (P<0.01) and plasma Tyr (TyrTR) tended to be higher (P=0.06) for WCS diet than MH diet. While PheTR tended to be lower (P=0.09) and TyrTR was lower (P<0.05) at heat exposure. However, the WBPS was higher (P<0.01) for WCS diet than MH diet and did not differ between the exposures. The results of this study indicate that WCS fed sheep was more heat tolerant than those fed MH diet. Although the digestibilties of DM and N were increased in WCS diet during heat exposure, some conflicting results such as reduced MNS were observed. Therefore, because protein metabolism is influenced by the energy supply in ruminants, it was decided to clarify the effect of feeding WCS on energy metabolism in heat exposed sheep based on the rumen fermentation characteristics, blood VFA concentration and plasma glucose metabolism.

The third experiment was carried out in iso-energetic and iso-nitrogenous condition to explore the potential of WCS as feed for heat exposed ruminants based on energy metabolism. Therefore, the effect of feeding WCS compared to timothy hay (Hay diet) on physiological responses, blood metabolites, rumen fermentation

characteristics, blood VFA concentrations and plasma glucose metabolism were measured under heat exposure. Six shorn sheep were fed either of the diets in a crossover design for 24 days, where soybean meal was added to both diets to make them iso-nitrogenous at maintenance energy level. The dietary adaptation, method and duration of environmental exposures and sample collections were as same as for first experiment. Blood VFA concentrations were measured on day 3, while plasma glucose turnover rate (GluTR) was determined by an isotope dilution method using [U-^{13}C]Glu on day 5 of each exposure period. Similar to the second experiment, RR was lower (P<0.01) for WCS diet and was higher at heat exposure (P<0.01); it was also lower (P<0.05) for WCS diet than the Hay diet during heat exposure. Serum thyroxine concentration tended to be higher (P=0.06) for WCS diet than the Hay diet and was lower (P<0.01) during heat exposure. The serum non-esterified fatty acid (NEFA) concentration did not differ between the diets and was lower (P<0.05) during heat exposure. However, the β-hydroxy-butyric acid (BHBA) and plasma lactic acid concentrations were higher (P<0.01 and P<0.05; respectively) for WCS diet than the Hay diet and did not differ between the exposures. The DM digestibility was higher (P<0.01) for WCS diet than the Hay diet and tended to be higher (P=0.06) during heat exposure; it was higher (P<0.05) for WCS diet at heat exposure compared to thermoneutral. Rumen pH tended to be lower (P=0.07) for WCS diet and was lower (P<0.05) during heat exposure. Rumen NH_3 concentration was higher (P<0.01) for WCS diet than the Hay diet and was also higher (P<0.01) during heat exposure. Rumen acetate concentration was lower (P<0.05) for WCS diet, while the total VFA and propionate concentrations did not differ between the diets. Total VFA concentration tended to be lower (P=0.08) during heat exposure; it was lower (P<0.05) for WCS diet during heat exposure compared to thermoneutral. Propionate was lower (P<0.05) during heat exposure, while acetate concentration did not differ between the exposures. Blood total VFA, acetate and propionate concentrations were lower (P<0.01) for WCS diet than the Hay diet. Though blood total VFA and acetate concentrations did not differ between the exposures, blood propionate was lower (P<0.01) at heat exposure; it was lower (P<0.05) in Hay diet during heat exposure.

The plasma glucose concentration and GluTR were higher ($P<0.01$) for WCS diet than the Hay diet and did not differ between the exposures. The results of this study suggest that physiological responses of heat exposed sheep were influenced by feeding WCS diet compared to Hay diet. The energy metabolism in terms of GluTR was enhanced by feeding WCS diet in comparison with Hay diet without effect of environmental exposures. So it could be suggested that feeding WCS diet might have positive influence on energy metabolism but its effect under heat exposure is not clear.

In this research, it is proved that the N supplemented WCS diet has better digestibility as indicated by DM and N digestibilties compared to other tested diets. It is also proved that the sheep fed WCS diet is more heat tolerant and the digestibility of WCS diet is enhanced by heat exposure. Moreover, WCS diet has the positive influences on both protein and energy metabolism. Therefore, WCS is excellent as livestock feed, and due to its resistance to heat exposure effects, it is concluded that it can be used as a feed for ruminant livestock under heat exposure.