

学 位 論 文 の 要 旨

論文題目 **Studies on the Use of Persimmon Peel for Ruminant Feeds**
(反芻家畜に対する柿皮の飼料利用に関する研究)

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Fruit byproducts are abundantly available in Japan, and thus should be considered as alternative feed resources because of their economic and environmental impact through the food chain. One example, persimmon peel (PP), is a waste product from the producers of dried persimmon. The PP contains fiber, non-fiber carbohydrate (NFC), and protein as well as phenolic compounds, particularly tannins, which gives persimmons their astringent taste. This astringent taste would decrease palatability if used in animal feed. Fruit byproducts, especially PP, have a high moisture content, which leads to the problems of susceptibility to microbial spoilage when used and preserved. Mixing PP with other feeds when preparing total mixed ration (TMR) reduces these obstacles and may improve its utilization. Ensiling TMR is a practical method for preserving high moisture feeds over a long period. However, during the ensiling process, a part of nutritional compounds in PP can be lost and the tannin in PP would also be insolubilized. This would reduce the astringent taste because the ethanol and CO₂ that are used for the deastringency treatment of persimmon fruit are also produced during the ensiling process. Additionally, mixing PP in TMR silage may reduce the ruminal protein degradability because tannins bind to dietary protein and forms a less degradable complex in the rumen. Thus, the purpose of the present study was to evaluate the utilization of PP as an ingredient of TMR silage for ruminant feeds, with a particular focus on changes in the contents of tannins and nutrients during the ensiling process.

The first experiment (Experiment 1) was conducted using four sheep that each received four different diets in a 4×4 Latin square design with 15 days for each experimental period. The ingredients of the diets were as follows: 1) a basal non-ensiled diet containing oat hay, soybean meal, beet pulp, and barley (T1); 2) a basal non-ensiled diet with 1% Quebracho tannin on a dry matter (DM) basis (T2); 3) a control mixed silage diet with barley (T3); and 4) a PP mixed silage diet with barley (T4). The control and PP mixed silages were prepared with oat hay, soybean meal and beet pulp or PP and ensiled for about 6 months. The sheep were fed diets at 1.5 times their maintenance energy. There was no difference in the average daily gain and nitrogen balance between sheep fed the four different diet. The digestibility of DM, organic matter (OM) and neutral detergent fiber (NDF) in sheep fed the T4 diet was lower than those in sheep fed the other diets. The methane production in sheep fed the T3 diet was the highest among these diets. However, the total VFA concentration and its proportion in the rumen of sheep fed the T4 diet was similar to those in sheep fed the T3 diet. The ruminal ethanol concentration was higher for the T4 diet than that for the other diets at 2 hours after feeding. The ruminal ammonia-N ($\text{NH}_3\text{-N}$) and plasma urea-N concentrations were higher in sheep fed the T3 and T4 diets than those in sheep fed the T1 and T2 diets. The concentrations of essential amino acids in the plasma were not affected by the diets. For plasma metabolites, the T4 diet showed the highest concentrations of ketone bodies and triglycerides among the diets. In conclusion, the mixed silage containing PP showed lower digestibilities of DM and NDF but reduced methane emission from the sheep. Nitrogen utilization in the sheep was not affected by feeding the mixed silage although the ruminal protein degradation seemed to be enhanced by feeding the mixed silages.

To investigate the reasons for the lower digestibility of the PP mixed silages (T4 diet) observed in Experiment 1, the effect of the ensiling treatment on the nutrient contents and in vitro ruminal fermentation between PP and grape pomace were compared (Experiment 2.1). The PP, red grape pomace

(RGP) and white grape pomace (WGP) were ensiled for 0, 1, 2, 4 and 8 weeks. Freeze-dried samples of byproducts ensiled for 0 (fresh byproduct) and 4 weeks were used for the *in vitro* rumen fermentation with rumen fluid collected from sheep. For the *in vitro* study, polyethylene glycol (PEG) was added to determine the tannin activity because PEG can bind tannins and inhibit its activity. The NFC content in the byproducts decreased after ensiling, especially for the PP. The total tannin content was similar among the fresh byproducts. The proportion of soluble tannin in the PP decreased after ensiling but the grape pomaces were not affected by the ensiling treatment. The ruminal fermentation, indicated by the gas production, total VFA concentration and methane production parameters for all byproducts, was reduced by the ensiling treatment, but was increased by adding PEG, except for the PP silage. The gas production and VFA concentration for the PP silage was increased less by adding PEG compared with grape pomaces. In conclusion, reducing the NFC content by the ensiling treatment decreased the ruminal fermentability of the PP. The insolubilization of tannins that occurred in the PP during ensiling reduced its inhibitory effect on ruminal fermentability. In Experiment 2.2, the *in vitro* fermentability was compared between PP and beet pulp. Compared with beet pulp, PP silage showed lower gas and methane production and DM and NDF digestibility by the *in vitro* rumen incubation. The replacing lower digestible PP for beet pulp was the one of the reasons for the lower DM and NDF digestibility of the mixed silage diets containing PP.

In Experiment 3, the effect of including PP in TMR silages at different levels on their chemical composition, protein fraction, tannin fractions, and *in vitro* rumen fermentation were investigated. Four different types of TMR silage containing PP at 0, 50, 100, 150 g/kg DM were used in this experiment. The TMR silages were prepared in 500-mL plastic bottles then stored for 6 months. Freeze-dried samples of pre-ensiled and ensiled TMR were used for *in vitro* rumen fermentation. Increasing the PP level in the TMR silages decreased the soluble protein and increased the neutral detergent insoluble

protein. The total tannin content in the TMR silage was not affected by the PP level. However, increasing the PP level in the TMR silages decreased soluble tannin and increased the insoluble tannin contents. During the ruminal fermentation, the gas and methane production in the TMR silages was lower than that in the pre-ensiled TMR and decreased linearly with increasing PP level. The concentrations of NH₃-N and total VFA in the *in vitro* rumen were not affected by the ensiling treatment or the PP content. Including PP in the TMR silages affected protein breakdown during the ensiling process with a negative correlation being observed between the insoluble tannin and soluble protein contents. Although gas and methane production in TMR could be reduced by including PP, this decrease was more pronounced in the pre-ensiled TMR.

In conclusion, even though TMR silage containing PP can be preserved well for 6 months, the tannins of PP become inactivated and a part of the NFC of PP is lost during the ensiling treatment. TMR silage containing PP did not affect the nitrogen balance parameters in the sheep, even though the tannins in PP can reduce protein solubility in TMR silages. Furthermore, TMR silage containing PP reduced DM digestibility, which relates to the reduction of methane emission from animals, although this inhibitory effect on DM digestibility and methane emission seemed to have occurred through the loss of NFC during ensiling rather than the tannin activity in PP. The loss of NFC after ensiling in TMR containing PP means that additional energy sources might be required to cover this loss and to provide enough energy for the animals.