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学位論文内容の要旨

博士の専攻分野名称：博士（農学）　　氏　名：Kodithuwakku Arachchilage Heshan Taraka Kodithuwakku

学位論文題名

Study on the effects of early fibrous diet feeding via oral administration on gastrointestinal environment in pre-weaned calves

（繊維飼料の早期経口投与が哺乳子牛の胃腸内環境に及ぼす影響）

Calf health in the preweaning period is an important aspect for the future production of cattle. To ensure the healthy growth of neonatal animals, anatomical and functional development of the gastrointestinal tract is key not only for nutrient acquisition but also for preventing infectious diseases. Establishment of the gastrointestinal microbiota is vital for the development of gastrointestinal functions, such as feed digestion, provisioning of nutrients to the host, and developing the immune system. The supplementation of roughage to calves during the pre-weaning period shows beneficial effects, such as improvement in feed intake, growth performance, rumination, and rumen environment. In my master's study, it was confirmed that oral administration of a fibrous diet from 3 days of age improves the body weight and fecal microbiota (i.e., higher abundance of *Lactobacillus* spp. and *Prevotella* spp.; lower abundance of *Clostridium perfringens*) of calves within the first 3 weeks of life. This study aims to further assess the effects of oral fiber administration on hindgut microbiota and investigate the impacts on rumen microbiota.

1. Investigation of the beneficial effects of fibrous diet on hindgut microbiota.

Timothy hay and psyllium, which were the ingredients of fibrous diet for oral administration to calves, were separately evaluated by in vitro batch culture experiment using the calf feces as inocula. *Lactobacillus* spp. were enhanced solely by timothy hay, whereas *Prevotella* spp. were enhanced solely by psyllium. Furthermore, these effects

were more obvious by adding the water-soluble fraction compared to the insoluble fraction. The species composition of the *Lactobacillus* spp. enhanced by fibrous diets was characterized by the near full-length sequencing of the 16S rRNA gene. A higher proportion (> 60%) of the detected *Lactobacillus* species were represented by Uncultured (g) *Lactobacillus*, and they were phylogenetically closely related to *L. johnsonii*, *L. gasseri*, and *L. taiwanensis*.

2. Alterations in rumen microbiota via oral fiber administration during early life in dairy cows

This study evaluated the effects of oral fiber administration during the pre-weaning period on the development of rumen microbiota from pre-weaning to the first lactation period. Twenty female calves were assigned to control and treatment groups (n = 10 each). Animals in both groups were reared using a standard feeding program throughout the experiment, except for oral fiber administration (50–100 g/day/animal) from 3 days of age until weaning for the treatment group. Rumen content was collected during the pre-weaning period, growing period, and after parturition. Amplicon sequencing of the 16S rRNA gene revealed that oral fiber administration facilitated the early establishment of mature rumen microbiota, including a relatively higher abundance of *Prevotella*, *Shuttleworthia*, *Mitsuokella*, and *Selenomonas*. The difference in the rumen microbial composition between the dietary groups was observed even 21 days after parturition, with a significantly higher average milk yield in the first 30 days of lactation. Therefore, oral fiber administration to calves during the pre-weaning period altered rumen microbiota, and its effect might be long-lasting until the first parturition.

In conclusion, this study demonstrated that the fibrous diet enhances beneficial gut bacteria and the establishment of rumen microbiota in pre-weaned calves. These modulations of gastrointestinal microbiota could lead to improved host productivity, such as growth and milk yield.