## 論文の内容の要旨

獣医学専攻 平成30年度博士課程入学

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論文題目: Study on the usefulness of tannin-related substances as feed additives in pig farming

(養豚におけるタンニン関連物質の飼料添加物としての有用性に関する研究)

There are many vulnerable stages of pig growth that put pig farming at great risk, especially the weaning stage of piglets. Stress from weaning can lead to growth check, diarrhea or even death, causing serious economic losses. Adding antibiotics to the diet can protect them from intestinal pathogens. But with the increasing importance of antibiotics in the farming industry, drug-resistant strains have been triggered. Since 1986, the use of antibiotics in feed has been gradually banned in some countries, and the pork production efficiency has been influenced. Other feed additives or management programs such as probiotics, organic acids, mineral salts, and vaccines have emerged with more or fewer deficiencies in terms of efficacy, stability, and economic applicability. Therefore, a sustainable and effective antibiotics alternative is urgently needed to prevent diarrhea in early-weaned piglets and to promote growth performance.

Tannins have great potential for development as natural antimicrobial agents, due to their structural complexity, and the ability to bind indiscriminately to proteins which makes it more difficult for microorganisms to develop resistance. In addition, it has antioxidant, anti-inflammatory, and antiparasitic activities, and so on. Notably, its ability to bind to proteins allows them to also bind to feed proteins or digestive enzymes,

interfering with the digestion and absorption of nutrients, thus creating anti-nutritional properties. Therefore, how to strike a delicate balance between anti-nutritional properties and beneficial biological activity is our research focus.

Due to the instability of hydrolyzed tannins, and considering the effect of tannins in complex environments, such as feed and intestine, we finally chose more stable condensed tannin (quebracho tannin) for the study. Past experience shows that the antinutritional properties of tannins are usually associated with high dosage. In view of this, we started with a small amount to determine the feasibility of quebracho tannin as a piglet feed additive, and tried various addition levels to determine the appropriate dosage and the possibility of replacing antibiotics. In experiment 1 (Chapter 2), the feasibility of using low-levels MGM-P (quebracho tannin product) addition as feed additives for weaned piglets was assessed; it investigated MGM-P effects on growth performance, diarrhea, and overall health in early-weaned piglets. In total, 24 piglets (Duroc × Landrace × Yorkshire, 6.51±0.17 kg) were allocated to three treatment groups fed basal diets supplemented with 0, 0.2%, or 0.3% MGM-P for 20 days. The addition of 0.3% MGM-P to the diet of early-weaned piglets improved diarrhea incidence, hematological parameters, and intestinal mucosa structure. Furthermore, the addition of 0.2% or 0.3% MGM-P to the diet of early-weaned piglets did not affect their growth performance. In experiment 2 (Chapter 3), higher additive amounts were used, and an antibiotic control group was set up. 36 early-weaned piglets (Duroc × Landrace × Yorkshire, 5.84±0.21 kg) were allotted to one of four treatments and given either a control diet free of MGM-P (NC, negative control) or diets containing 0.5% (LT, lowlevel treatment), 1.0% (HT, high-level treatment) MGM-P, or antibiotics (PC, positive control). Growth performance and blood parameters were monitored during the feeding period. The results showed that in the first 14 d after weaning, the 0.5% MGM-P group had the highest feed conversion ratio (FCR) and higher (p < 0.05) than that in the 1.0%

MGM-P group. Although there was no diarrhea, the average fecal scores in the whole experimental stage were HT > NC > PC > LT. Therefore, in combination with experiment 1, it can be concluded that around 0.5% is probably the appropriate additive amount of the MGM-P. To further validate the effect of 0.5% MGM-P supplementation in promoting growth in pigs, we conducted a trial for grow-finish pigs. In experiment 3 (Chapter 4), a total of 24 pigs (Duroc × Landrace × Yorkshire,  $37.60 \pm 0.86$  kg) were divided into two groups (3 replicates of 4 pigs per treatment), and they were fed a control diet or 0.5% MGM-P supplementary diet for 16 weeks. Our results indicated that the average daily gain (ADG) of pigs in the MGM-P supplementary group was significantly higher (p < 0.01) compared to the control group at 8-12 w. Pigs received MGM-P had lower (p < 0.05) glutamic oxaloacetic transaminase (GOT) levels and higher kidney weight (p < 0.05) at the end of the experiment compared with the control group. All these reasons could potentially promote growth performance in pigs.

The use of tannins in pig feed dates back centuries, Iberian pigs in the Mediterranean region, known for their high-quality meat, were often foraged for tannin-rich acorns or chestnuts. This may be due to the antioxidant activity of tannins, which helped improve the antioxidant capacity and unsaturated fatty acid ratio of the pork. And chestnut shells, which are discarded in chestnut food production, are very rich in tannins. Therefore, we tried to reuse these shells to explore the possibility of using the waste as feed to ensure the quality of meat and improve economic profitability.

Experiment 4 (**Chapter 5**) investigated the availability of chestnut by-products in finishing pig production. In the 110-day experiment, 6 Duroc finishing pigs (103.37±2.03 kg, males castrated) were selected as experimental animals and evenly divided into control and treatment groups (three pigs each). The control group was fed a basal diet (C) and the treatment group received a basal diet containing 60% chestnut shell meal (T). The assessment of pork quality revealed that the addition of chestnut

shells effectively improved the sensory evaluation and color stability of the meat and increased the content of free amino acids. In order to maximize the use of chestnut byproducts, the development of reasonable storage means is necessary to overcome the limitations imposed by the extremely seasonal nature of chestnut harvesting, so the feasibility of silage chestnut shells, and its effects on growth, health, and meat quality of finishing pigs was studied in experiment 5 (Chapter 6). Two 110-day feeding trial was conducted with total 12 Duroc finishing pigs (105.62±2.85 kg). Each trial used 6 pigs from the same sow and was assigned into two diet dietary treatments (3 pigs each). The pigs in trial 1 were fed with basic diet (S3C) or basic diet plus 60% chestnut shells silaged for 3 months (S3T), and in trial 2 were fed with basic diet (S12C) or basic diet plus 60% chestnut shells silaged for 12 months (S12T). The results showed that silage chestnut shells had no effect on the growth performance of pigs. However, the tannin content in chestnut shells decreased with increasing silage time, and its effect on pork changed. The addition of short-term (3 months) silage chestnut shells to feed significantly improved pork tenderness, while the effect on free amino acids is not obvious. In contrast, the feeding of chestnut shells with long-term (12 months) silage had a positive effect on the free amino acid content of pork, but resulted in poor tenderness.

Collectively, quebracho tannin has shown effectiveness as a swine feed additive. 0.5% MGM-P supplementation can provide pigs with the right amount of quebracho tannin to improve health and growth performance, and has the potential to contribute to the mitigation of drug resistance by replacing antibiotic additives in today's pig industry. As for the substitution of some feeds by chestnut shell by-products, this waste food reuse method can improve the quality of pork on the one hand and reduce the cost of breeding on the other. Continuing to promote this kind of waste food reuse research in pig farming in the future will not only help to promote the sustainable development of pig farming

to increase economic benefits but also help to establish an environmentally friendly image for pig farming. For example, making the pig industry less involved in competition for natural resources compared to other livestock industries; enabling the pig industry to help reduce food waste, etc., ultimately increasing the competitiveness and market share of the pig industry in the livestock sector.