Mitigating Mycotoxin Contamination from Grains used in Swine Feed Through Nanotechnology

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INSIGHT FOR PRODUCERS

Mycotoxins can be detrimental to animal health, growth, and welfare. Using nanotechnology to mitigate mycotoxin contamination is a promising approach.

SUMMARY

Mycotoxin contamination can be a concern for producers resulting from adverse health issues in livestock and significant financial losses in the agriculture sector. Deoxynivalenol (DON) is a significant mycotoxin, contaminating common feed grains, such as wheat, barley and corn. Mitigation strategies for DON are limited in terms of binding efficiency, biosafety, and cost-effectiveness. This study investigated the potential application of nanotechnology to mitigate DON contamination in grains. Nanomaterials have been used to address contaminants in air, soil, and water due to their unique physical and chemical properties. Three nanomaterials were identified and selected for testing: chitosan polymeric nanoparticles (CS), montmorillonite nanocomposites (MN), and magnetic graphene oxide (MGO). It was determined that MGO had the greatest potential for mitigating DON contamination in grains (wheat).

In-barn feeding trials (nursery pigs) were conducted to determine the effectiveness of MGO nanoparticles in mitigating DON in swine diets and their effect on pig performance. Results showed that feeding pigs with MGO-treated diets had no adverse impact on their growth performance, nutrient digestibility and nitrogen retention. Economic analysis revealed using MGO to mitigate DON-contaminated feed is currently not viable due to the high cost of MGO. This could be significantly reduced by developing our own MGO materials or combining MGO nanotechnology with other environmental mitigation techniques such as photocatalytic degradation, which has been utilized to remove various environmental pollutants more effectively and at lower operational costs. Follow up research involving long-term feeding of MGO-treated diets to nursery and grower-finisher pigs with higher DON levels (more than 3 – 5 ppm) is recommended to fully assess the overall impact of this technology in mitigating mycotoxin contamination in diets as well as on the environment.

INTRODUCTION

Mycotoxins are secondary toxic chemicals produced by organisms of fungal origin found in contaminated grains. The occurrence of mycotoxin may start in the field, proliferating throughout subsequent handling, storage, and processing of the grains. The most prevalent mycotoxins identified in western Canada are deoxynivalenol (DON), HT-2 and T-2 toxins, and ergots. Use of mycotoxin-contaminated grains in livestock feed may result in feed refusal, and affect immune and health status.

Nanotechnology can be used to treat contaminants in air, soil and water. Findings from previous studies show that selected nanomaterials can effectively reduce the levels of hazardous gases (Alvarado and Predicala, 2017), growth of disease-causing microorganisms (Alvarado and Predicala, 2014), and Porcine Epidemic Diarrhea (PED) virus (Predicala et al., 2018) in swine barns. Nanomaterials are also known to have antifungal properties; several studies have shown the effective use of nanomaterials in mitigating mycotoxin contamination due to their high surface area and high reactivity, and the fact nanomaterials can be modified to enhance their physical and chemical properties (Tang and Kotov, 2005).

Based on assessment criteria including previous applications, cost effectiveness, safety, and availability, three nanomaterials were selected: chitosan (CS) polymeric nanoparticles, montmorillonite nanomaterials (MN), and magnetic graphene oxide (MGO) nanostructures. While the nano-adsorbent efficiency of CS, MN and MGO against the elimination of specific mycotoxins have been demonstrated, their effectiveness against the synergistic effects of multiple mycotoxins is unknown. *Fusarium graminearum* and *Fusarium culmorum* are known to produce several different fusariotoxins, including DON, which interact synergistically when fed to pigs. The best way to assess the effectiveness of nanomaterials against mycotoxins is under actual production conditions using the dose of mycotoxins found in feed.

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In general, research on the potential application of nanomaterials for mycotoxin decontamination in grains and in livestock feed is very limited, thus, its operational requirements such as effective dosage, binding capacity, optimum treatment application conditions, among others, are still unknown. Hence, this work has been conceptualized to fill these gaps by conducting a comprehensive evaluation of the application of nanotechnology for mitigating mycotoxin risk in grain and livestock industries.

EXPERIMENTAL PROCEDURES

Phase 1: Laboratory-scale testing

Phase 1 aimed to determine the efficacy of selected nanomaterials, particularly chitosan polymeric nanoparticles (CS), montmorillonite nanocomposites (MN), and magnetic graphene oxide (MGO). These three nanomaterials were identified based on a comprehensive literature review.

Of the three nanomaterials tested, MGO nanoparticles had the greatest potential to reduce DON contamination. MGO nanoparticles were investigated to establish the optimum application rate and operating conditions for mitigating the mycotoxin contaminated grain (i.e., wheat, barley and corn). Dry mixing of materials was employed to simulate the mixing process during feed processing. Hence, the sorption experiments were carried out in 50-mL flasks with each flask containing a mixture of 5 g of DON-contaminated ground grain and test nanomaterial. The operating conditions investigated included MGO nanoparticle application rate (or adsorbent dose), contact time and temperature.

Phase 2: In-barn testing

Phase 2 investigated the effects of using MGO nanoparticles as a DON binder in diets related to overall pig performance. The experiment involved feeding pigs with DON-contaminated diets treated with MGO nanoparticles at an appropriate inclusion rate, as determined in phase 1. In addition to evaluating the impact of MGO nanoparticles as a DON binder on nutrient digestibility and growth performance, as well as on DON content in the diet.

A total of 12 nursery pigs (initial BW of 22 ± 2 kg) were used in the 14-day experiment and fed their assigned diets for 10 days (days 1-10). The daily feed allocation for all pigs was at 2.8x maintenance metabolizable energy requirements (197 kcal/kg BW0.60/d; NRC, 2012) and fed in two equal meals. Water was freely available. Following the 10-day feeding period, feces (days 11-14) and urine (days 11-12) were collected.

The three dietary treatments (in mash form) were:

1. Negative Control (NC): Pigs were fed a basal wheat-barley-soybean diet that was not contaminated with DON.
2. Positive Control (PC): Pigs were fed a basal wheat-barley-soybean diet that was contaminated with DON.
3. Positive Control + 1% MGO (PC+MGO): Pigs were fed a DON-contaminated basal wheat-barley-soybean diet that was added with 1% MGO as a DON binder.

Upon completion of the experimental work, PC+MGO pigs were euthanized using captive bolt in compliance with CFIA guidelines, while the remaining NC and PC pigs were returned to the normal production herd at PSC, where they were fed conventional wheat-barley-soybean-based diets until attaining market weight and eventually sent to a commercial abattoir.

Digestibility and retention of nutrients

Digestibility and nutrient retention were measured from fecal and urine samples collected on days 11- 14 and days 11-12 of the trial, respectively, from individual pigs in each treatment group. Fecal samples were analyzed for energy, dry matter, and fiber content. Both feces and urine were analyzed for nitrogen content, allowing overall nitrogen balance to be determined to estimate protein accretion.

Growth performance

The growth performance of the nursery pigs was assessed using average daily gain (ADG), average daily feed intake (ADFI), and gain-to-feed ratio (G: F) throughout the testing period. ADG was determined by pig weight at days 0, 7, and 14.

RESULTS AND DISCUSSION

Phase 2

The growth performance of nursery pigs fed diets not contaminated with DON (NC), diets contaminated with DON (PC,) and DON-contaminated diets treated with MGO (PC+MGO) are presented in Table 1. Pigs fed PC and PC+MGO had slightly higher ADG compared to NC pigs. The difference, however, was not statistically significant ($p>0.05$). For average daily feed intake (ADFI), pigs fed PC+MGO had the highest ADFI among the three treatments, but the difference was not statistically significant ($p>0.05$). Similarly, there is no significant difference ($p>0.05$) in gain to feed ratio (G:F) between the dietary treatments. Overall, the growth performance results suggested that feeding pigs MGO had no significant impact on its ADG, ADFI and G:F.

Table 1. Growth performance of nursery pigs fed diets contaminated with DON and added with MGO as a DON binder.

Treatments	ADG1	ADFI	G:F
NC (n=4)	0.99 ± 0.2	1.37 ± 0.1	0.72 ± 0.1
PC (n=4)	1.04 ± 0.1	1.35 ± 0.1	0.77 ± 0.04
PC + MGO (n=4)	1.04 ± 0.03	1.40 ± 0.1	0.74 ± 0.04

There was no significant difference ($p>0.05$) in the levels of aflatoxin B1, fumonisins B1 and B2, HT-2, T2, nivalenol, and ochratoxin A among the three dietary treatments. However, the NC diet showed significantly lower ($p<0.05$) levels of DON, 3+15ADON and zearalenone compared to PC and PC+MGO diets. This was expected given that the NC diet is the control diet and should be free from any mycotoxin contamination. On the other hand, levels of DON, 3+15ADON and zearalenone in the PC diet were not significantly different from those in the PC+MGO diet ($p>0.05$). DON levels in the PC diet were very low (i.e., 647 ppb), making it a challenge to assess the impact of MGO on DON levels in the PC+MGO diet.

The effect of MGO inclusion on nutrient digestibility in nursery pigs is shown in Table 2. The digestibility of dry matter, gross energy, total digestible nutrients (TDN), fat and crude protein was significantly lower ($p<0.05$) in pigs fed DON-contaminated (PC) diet and PC+MGO diet than those fed the negative control (NC) diet. There was no significant difference ($p>0.05$) in dry matter, gross energy, TDN, fat and crude protein digestibility was found in piglets fed PC+MGO diet compared to those fed PC diet. The reduced digestibility of nutrients observed in pigs fed PC diet and pigs fed PC+MGO diet could be attributed to DON contamination in the diet as both diets had comparable levels of DON. The inclusion of MGO in the diet had no apparent effect on nutrient digestibility.

Table 2. Effect of MGO inclusion on nutrient digestibility in nursery pigs.

Treatments	Calculated nutrient digestibility (%)				
	GE	DM	TDN	Fat	CP
NC	86.75a	96.03a	92.76a	65.61a	86.93a
PC	83.25b	95.10b	91.11b	56.80b	82.25b
PC+MGO	82.96b	94.61b	90.44b	55.37b	82.23b

[a b]Means without a common superscript within a column are significantly different ($p<0.05$).

Phase 3: Economic analysis and recommendations

An economic analysis was conducted to determine the costs and requirements for the proper implementation of the nano-mitigation treatment in a typical pork production facility. The analysis included all costs associated with the nano-mitigation strategy, treatment cost, and labour and operating costs. The estimates were applied to a 100-head nursery room as nursery pigs are more susceptible to DON contamination compared to other stages of production. In addition, the DON-contaminated barley, which comprised 10% of the diet was treated with MGO to reduce its mycotoxin contamination.

At an application rate of 200 mg MGO per 5 g of barley, the total amount of MGO needed to treat about 4,000 kg of feed is 3.14 kg. At the current cost of the commercially-available MGO used in this experiment, this translates to a treatment cost of $62.8 per pig room turn. Furthermore, the duration of each treatment application would be about 1 hour per cycle, and the labour cost was assumed to be $15.0/hour. The operating cost to prepare the MGO-treated barley is $0.03 per pig per room turn. Summing up all these cost estimates, the total incremental cost of treating DON-contaminated feed using MGO nanoparticles is about $62.98/ pig, which is extremely high mainly due of the cost of the MGO nanoparticles.

MGO nanoparticles used in this project are research materials which are relatively costly at present because these are currently custom ordered from a commercial supplier which produced them for us in small quantities; however, similar to other nanomaterials we have used in our other previous studies, the cost for MGO nanoparticles is expected to decrease as demand for this material increases with new applications for the product. Alternatively, options for synthesizing custom batches of MGO nanoparticles that are tailor-made for this application can be explored in university laboratories as well as in small chemical companies, which can greatly reduce the cost of the nanomaterial compared to current commercial suppliers. Another potential alternative for lowering treatment cost is to integrate this nano-mitigation strategy with other environmental remediation techniques, such as photocatalytic degradation, which has been proven effective in the removal of various environmental pollutants due to its inherent properties and low cost (Wu et al., 2019).

IMPLICATIONS

This study demonstrated that nanotechnology has great potential for mitigating mycotoxin contamination in feed grains such as wheat, barley and corn, however, are not cost effective. Among the nanomaterials tested, the addition of magnetic graphene oxide (MGO) in wheat grains resulted in the highest reduction in DON levels.

In-barn tests revealed that feeding MGO-treated diet to nursery pigs had no adverse impact on their growth performance, nutrient digestibility and nitrogen retention. The average daily gain, feed intake and gain-to-feed ratio of pigs fed MGO-treated diet were not significantly different from the control pigs. Nutrient digestibility, nitrogen retention and protein deposition were not significantly affected by MGO inclusion in the diet.

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