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## Evaluating the effects of pelleting Deoxynivalenol-contaminated dried distillers grains with solubles in the presence of sodium metabisulfite on analyzed DON levels

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# Evaluating the Effects of Pelleting Deoxynivalenol-Contaminated Dried Distillers Grains with Solubles in the Presence of Sodium Metabisulfite on Analyzed DON Levels<sup>1</sup>

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## Summary

Deoxynivalenol (DON), also known as vomitoxin, was prevalent in the 2009 U.S. corn crop and subsequently present in dried distillers grains with solubles (DDGS), in which DON levels are about 3 times higher than the original corn source. One method shown to reduce DON levels was by increasing moisture and temperature when sodium bisulfite was added to DON-contaminated corn (Young et al., 1987<sup>4</sup>). Therefore, a pilot study aimed first to replicate these results by placing DON-contaminated DDGS in an autoclave (60 min at 250°F) in the presence of sodium metabisulfite (SMB). The study used 6 treatments: (1) control, (2) 0.5% SMB, (3) 1.0% SMB, (4) 2.5% SMB, (5) 5.0% SMB, and (6) 5.0% SMB with 100 mL/kg water added to evaluate the role of water. After drying, samples were analyzed at North Dakota State University Veterinary Diagnostic Laboratory (NDSU; Fargo, ND). Autoclaving reduced DON levels ( $R^2 = 0.99$ ) with increasing SMB, justifying a follow-up study that aimed to assess whether SMB has the same detoxifying effects on corn DDGS in a commercial pellet mill.

For this study, batches of 450 lb DDGS were prepared from DDGS with a known DON concentration (23.4 ppm). The pellet mill was set to a production rate of 1,000 lb/h so retention rate and conditioning temperature could be altered within each batch. Within each batch, 4 samples were collected at conditioning temperatures of 150 and 180°F and retention times of 30 and 60 sec within each temperature. Samples were sent to NDSU for full mycotoxin analysis. No differences ( $P > 0.15$ ) were found in conditioning temperature or retention time on total DON, DON, or acetyl-DON; however, pelleting DDGS reduced (quadratic;  $P < 0.01$ ) DON and total DON as SMB increased. Based on these results, the reduction in DON and total DON levels appear to plateau somewhere between SMB levels of 2.5 and 5.0%. These results imply that pelleting in combination with SMB may allow pork producers to utilize DON-contaminated DDGS more effectively, but additional research is required to determine the effect of pelleting SMB in DON-contaminated diets on growth performance of pigs.

Key words: deoxynivalenol, pelleting, sodium metabisulfite, vomitoxin, nursery pig

<sup>1</sup> Appreciation is expressed to Hubbard Feeds (Mankato, MN) for supplying the DDGS used in this study.

<sup>2</sup> Hubbard Feeds, Mankato, MN.

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<sup>4</sup> Young, J. C., H. L. Trenholm, D. W. Friend, and D. B. Prelusky. 1987. Detoxification of deoxynivalenol with sodium bisulfite and evaluation of the effects when pure mycotoxin or contaminated corn was treated and given to pigs. *J. Agric. Food Chem.* 35:259-261.

## Introduction

The 2009 corn crop presented challenges for livestock producers due to high concentrations of mycotoxins, particularly DON. Deoxynivalenol, also known as vomitoxin, is one of the most abundant members of the group of mycotoxins known as trichothecenes, which are produced by fungi of the *Fusarium* genus. Toxin production is strongly dependent on environmental conditions, especially temperature and humidity, so contamination cannot be avoided completely. In growing pigs, concentrations of DON above 1 ppm are associated with reductions in voluntary feed intake, abnormal digestive morphology, and subclinical immune suppression. Nevertheless, swine producers are interested in finding ways to incorporate DON-contaminated feedstuffs into swine diets. When DDGS is produced from DON-contaminated corn, swine producers encounter substantial problems because the DON level in DDGS is 2 to 3 times more concentrated than the original corn source. Due to the particularly high levels and prevalence of DON found in DDGS and other feed grains during so-called *Fusarium* years such as 2009, the traditional strategy of diluting concentrations during ration formulation may not be a viable option. Furthermore, using adsorbent materials to bind mycotoxins in the digestive tract have proven largely ineffective against mycotoxins in the trichothecene family.

Studies by Young et al. (1987<sup>5</sup>) and Danicke et al. (2004<sup>6</sup>) have both shown significant reductions in DON concentrations when sodium metabisulfite ( $\text{Na}_2\text{S}_2\text{O}_5$ ) is added prior to hydrothermal treatment of the feedstuff, such as in an autoclave or laboratory conditioner environment. Young et al. (1987) found that pure DON or DON in naturally contaminated feedstuffs reacts readily with sodium bisulfite, the aqueous form of SMB, to form a 10-sulfonate adduct that showed no acute toxic effects when fed to pigs at levels that caused emesis with DON. The initial goal of this study was to attempt to mimic results seen by Young et al. using an autoclave environment and DDGS as the contaminated feedstuff. Following the proof of concept at the autoclave level, we hypothesized that a commercial pellet mill could provide similar hydrothermal conditions through manipulation of retention time and temperature at the conditioner stage of the process. Ultimately, pelleting the original feedstuff or final diet with SMB could be a viable way to decontaminate DON levels and thereby increase the inclusion rate of the DON-contaminated feedstuffs in livestock diets. If SMB is able to detoxify DON effectively, titrating varying concentrations of SMB may reveal the optimal dose for use in livestock diets.

## Procedures

**Autoclave study.** All samples used in this pilot study were prepared at the Kansas State University Swine Nutrition Laboratory, with the samples autoclaved at the K-State Food Science Laboratory.

<sup>5</sup> Young, J. C., H. L. Trenholm, D. W. Friend, and D. B. Prelusky. 1987. Detoxification of deoxynivalenol with sodium bisulfite and evaluation of the effects when pure mycotoxin or contaminated corn was treated and given to pigs. *J. Agric. Food Chem.* 35:259-261.

<sup>6</sup> Danicke, S., H. Valenta, M. Gareis, H. W. Lucht, and H. von Reichenbach. 2005. On the effects of a hydrothermal treatment of deoxynivalenol (DON)-contaminated wheat in the presence of sodium metabisulfite ( $\text{Na}_2\text{S}_2\text{O}_5$ ) on DON reduction and on piglet performance. *Anim. Feed Sci. Tech.* 118:93-108.

The experiment used 6 DDGS treatments: (1) control, (2) 0.5% SMB, (3) 1.0% SMB, (4) 2.5% SMB, (5) 5.0% SMB, and (6) 5.0% SMB with 100 mL/kg distilled water added to evaluate the role of water in the reaction. Each treatment had a final weight of 500 g per sample, except treatment 6 (550g with water). Samples were split into two replicates and placed in aluminum trays with foil covers, but were not sealed airtight to allow steam interaction and gas release. Samples were autoclaved at 250°F for 60 min. After autoclaving, samples were dried in a 131°F drying oven to convert remaining sample to a DM basis before being sent for mycotoxin analysis.

All samples were sent for full mycotoxin analysis at NDSU and were analyzed using a combination of mass spectrometry, enzyme-linked immunosorbent assay (ELISA), and high-pressure liquid chromatography (HPLC). Samples were prepared from a previously identified, uniform source of DDGS with a known total DON (DON and 15-Acetyl DON) concentration of 23.9 ppm. The DDGS were homogenized thoroughly prior to sample preparation to eliminate variation in mycotoxin content due to “hot spots” that could cause a discrepancy in initial DON levels. Total DON reflects a combination of DON and 15-Acetyl DON, because both mycotoxins elicit similar effects and are typically combined to form an overall value. Total DON values were adjusted by the proportion of DDGS in the sample to show the actual magnitude of reduction from the original sample.

***Pelleting study.*** This study was conducted at the K-State Grain Sciences and Industry Feed Mill. Initially, a 450-lb batch of clean DDGS was pelleted to verify retention times and practice procedures. All personnel involved were required to wear respirators during the pelleting process because sodium metabisulfite gives off toxic sulfur dioxide gas in the presence of heat and moisture.

Treatments comprised 450-lb batches of DDGS after the addition of SMB. DDGS were sourced from 3 tons of bagged contaminated DDGS (23.9 ppm DM), which was provided by Hubbard Feeds (Mankato, MN). The experiment used 4 DDGS treatments: (1) control, (2) 1.0% SMB, (3) 2.5% SMB, and (4) 5.0% SMB. Prior to the addition of SMB, each batch was mixed for 4 min in a paddle mixer (Forberg 500 L double-shaft) to homogenize the DDGS and eliminate any variation in initial DON concentration. After adding SMB, each batch was mixed for an additional 3 min before pelleting. The pellet mill (CPM Master Model 1000HD, Crawfordsville, IN) was set to a production rate of 1,000 lb/h so conditioning temperature and retention time could be manipulated within each batch of DDGS. Within each treatment, the pellet conditioner was adjusted so 5-lb samples could be collected at temperatures of 150°F and 180°F and condition times of 30 and 60 sec within each temperature. Pellets were cooled prior to sampling, and the 4 corresponding samples from each batch were ground, homogenized, and sent for mycotoxin analysis at NDSU.

Data were analyzed for linear and quadratic effects of SMB and interactions with temperature and retention time using GenStat Release 11.1 (VBN International, 2009). For all statistical tests, significance and tendencies were set at  $P < 0.05$  and  $P < 0.10$ , respectively.

## Results and Discussion

**Autoclave study.** The results of the DON analysis are shown in Table 1. Mycotoxin analysis verified the expected reduction in DON with increasing SMB ( $R^2 = 0.99$ ; Figure 1). Deoxynivalenol concentration was reduced by 13.9% by autoclaving the DDGS (control treatment), and the addition of SMB elicited further detoxifying effects, with 5.0% SMB reducing DON by 76.7%. Adding 10% water to the 5.0% level of SMB also aided in detoxifying DON, reducing it by 88.1%, an 11.4% increase from the 5.0% level alone. Overall, the results of the autoclave experiment confirm the results shown by Young et al. The addition of SMB to DON-contaminated DDGS in an autoclave environment can effectively reduce analyzed DON levels, and the volume of water appears to affect the extent of the detoxification. Although autoclaving DDGS with SMB can substantially decrease DON levels, whether similar results will be seen in commercially viable conditions, such as by adding SMB to DDGS prior to pelleting, remains uncertain. Additionally, whether reductions in analyzed DON levels translate to improvements in animal performance remains unclear.

**Pelleting study.** Results from DON analysis are shown in Table 2. During the pelleting process, the addition of SMB caused a considerable amount of gas to be produced from the pellet mill, and the gas generated a very strong odor and irritation to both the eyes and respiratory tract. Whenever utilizing SMB at levels used in this study in combination with hydrothermal treatment, wearing a full respirator within direct vicinity of the pellet mill is critical.

Conditioning temperature had no effect on total DON, DON, or acetyl-DON levels. Additionally, total DON, DON, and acetyl-DON levels were similar across both 30- and 60-sec retention times in the pellet conditioner, so these data are not presented.

Pelleting DDGS reduced (quadratic,  $P < 0.001$ ) DON and total DON levels as SMB inclusion rate increased. According to these results, the reduction in DON and total DON levels appears to plateau somewhere between 2.5% and 5.0% SMB. These results imply that pelleting in combination with SMB may allow swine producers to utilize DON-contaminated DDGS more effectively. Additionally, DON concentrations appear to be reduced in the presence of SMB without requiring adjustment of retention time or conditioning temperature at the pellet mill, simplifying procedures for operators; however, additional research is required to determine if pelleting with SMB can reduce vomitoxin levels in final diets rather than only in individual ingredients such as DDGS. Also, further investigation needs to be conducted into the effects of pelleting with SMB in DON-contaminated diets on the growth performance of pigs.

**Table 1. Effects of sodium metabisulfite (SMB) and autoclave on deoxynivalenol (DON) levels in dried distillers grains with solubles (DDGS; DM basis)<sup>1</sup>**

Sample	SMB <sup>2</sup> added, %	Water added,		DON, ppm	15-acetyl DON, ppm	Total adjusted DON <sup>3</sup> , ppm	DON remaining, %
		mL/kg of DDGS					
1	0	---		18.2	2.4	20.6	86.1
2	0.5	---		16.5	2.3	18.9	79.0
3	1.0	---		14.5	2.0	16.7	69.7
4	2.5	---		7.5	2.0	9.7	40.7
5	5.0	---		3.5	1.8	5.6	23.3
6	5.0	100		1.2	1.5	2.8	11.9
DDGS <sup>4</sup>	0	---		20.6	3.3	23.9	100.0

<sup>1</sup> DDGS samples were autoclaved for 60 min at 250°F. After autoclaving, samples were dried in a 131°F drying oven. Mycotoxin analysis took place at the North Dakota State University Veterinary Diagnostic Laboratory (Fargo, ND) and included mass spectrometry, ELISA (enzyme-linked immunosorbent assay), and HPLC (high-pressure liquid chromatography) methods.

<sup>2</sup> Sodium metabisulfite (Samirian Chemicals, Campbell, CA); 100% by weight.

<sup>3</sup> Total adjusted DON = (DON + 15-acetyl DON)/% DDGS in sample (needed to correct for the dilution effect of the addition of SMB). Both DON compounds have similar toxicity, and are typically combined to form an overall DON value.

<sup>4</sup> Original DDGS sample (90.1% DM). DON levels are converted to a DM basis.

**Table 2. Effect of pelleting temperature (Temp) and dose of sodium metabisulfite (SMB) on deoxynivalenol (DON) and acetyl-DON in diets containing corn dried distillers grains with solubles (DDGS) naturally contaminated with DON<sup>1</sup>**

	Temp, °F	Sodium metabisulfite, %				SED <sup>3</sup>	Probability, <i>P</i> < <sup>2</sup>		
		0	1.0	2.5	5.0		Temp	Linear <sup>4</sup>	Quad <sup>4</sup>
Total DON, ppm <sup>5</sup>	150	23.2	12.8	8.2	6.0	1.52	0.25	0.001	0.001
	180	21.5	11.5	7.0	6.5				
DON, ppm <sup>5</sup>	150	20.5	10.2	5.6	3.3	1.29	0.15	0.001	0.001
	180	18.7	9.0	4.2	3.6				
Acetyl DON, ppm <sup>5</sup>	150	2.7	2.6	2.7	2.8	0.42	0.74	0.45	0.64
	180	2.8	2.5	2.8	3.0				

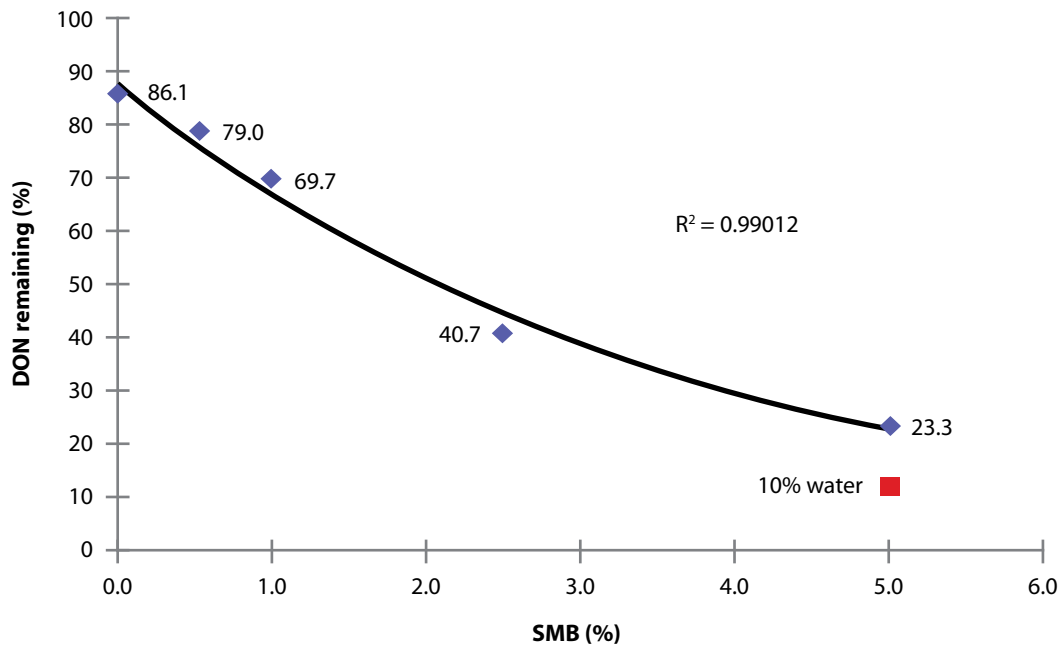
<sup>1</sup> No significant effect (*P* > 0.40) for retention time in pellet conditioner, thus data are not shown.

<sup>2</sup> No significant temp × SMB interactions (*P* > 0.69).

<sup>3</sup> Standard error of the difference for the temp × SMB interaction. For SED for effect of Temp and SMB, multiply by 0.50 and 0.71, respectively.

<sup>4</sup> Linear and quadratic effects of SMB.

<sup>5</sup> Samples analyzed at North Dakota State University Veterinary Diagnostic Laboratory (Fargo, ND) using a variety of mass spectrometry, ELISA (enzyme-linked immunosorbent assay), and HPLC (high-pressure liquid chromatography).



**Figure 1. Deoxynivalenol (DON) content of contaminated dried distillers grains with solubles following treatment with sodium metabisulfite (SMB) in an autoclave environment.**