# EVALUATION OF THE EFFICACY OF A COMMERCIAL PURIFIED PHYLOSILICATE TO REDUCE THE TOXICITY OF ZEARALENONE IN GILTS.

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#### **INTRODUCTION**

Zearalenone (ZEA) is a naturally occurring mycotoxin produced by *Fusarium* species, mainly *Fusarium graminearum*, on a variety of different grains, such as corn, wheat, rice, sorghum, barley and rye. (4) The *Fusarium graminearum* also survives in crop residues, which in turn provide the inoculum for the next year's crop. (3) The primary effect of ZEA is estrogenic, and prepubertal female pigs are clearly the most affected farm animals. The basis for the estrogenic effect is well established and is due to a close structural similarity between ZEA and estradiol. (6, 9) Clinical signs of intoxication in pigs can occur with levels above 0.1 mg of ZEA/kg of complete feed (100 ppb) (4)

Practical methods to detoxify mycotoxins contaminated grain or feed on a large scale and in a cost-effective manner are not currently available. At present, one of the most practical approaches to prevent mycotoxicosis consist of using adsorbent materials in the feed that reduce the absorption of mycotoxins from the gastrointestinal tract. Myco-Ad A-Z has been effective in preventing the toxic effect of 1.25 ppm of T-2 when added at 1 kg per metric ton of broiler feed. In addition, the product did not affect performance or bone mineralization of broilers, demonstrating its lack of nutrients absorption. (5)

Reduction in the bioavailability of ZEA by adsorbents has also been investigated but at present the results are unsatisfactory and none of them have proven to be effective in the treatment of hyperestrogenism in swine (1). Therefore, the objective of this research was to determine the efficacy of a low inclusion modified phylosilicate, Myco-Ad<sup>®</sup> A-Z, to ameliorate the estrogenic effect of zearalenone in young prepubertal gilts.

## MATERIALS AND METHODS

This experiment was conducted at the Experimental Farm of Trilogy Analytical Laboratory. Feed was experimentally contaminated with crystalline ZEA isolated from a *Fusarium* culture that was prepared by the University of Missouri Veterinary Diagnostic Laboratory. Spectrophotometric and chromatography method determined that the ZEA was over 99% pure. The product used was a commercial modified phylosilicate, Myco-Ad<sup>®</sup> A-Z, produced in Texas (Special Nutrients, Miami, FL, USA)

A total of eighteen 20-day-old recently weaned Yorkshire Cross gilts were used in this experiment, after a 4-day adaptation period. Pigs were individually housed and reared under uniform management conditions, with feed and water provided *ad libitum*. Two commercial base diets containing or exceeding NRC (5) recommended nutrient levels were fed for 30 days: ADM Alliance Nutrition Momentum 15-25 for the first 10 days and Momentum 25-50 for the remainder of the trial. The feeds were analyzed for mycotoxins, E. coli, salmonella and listeria prior to feeding.

Gilts were randomly distributed into three treatments with 6 replications each. Dietary treatments were as follows: 1) control diet; 2) control diet with the addition of 750  $\mu$ g of ZEA/kg of feed; and 3) control diet with 750  $\mu$ g of ZEA/kg of feed plus 0.1% Myco-Ad A-Z. At the end of the experiment pigs were weighed individually, total feed consumption recorded, and vulva area observations recorded. All gilts were sacrificed to carry out an exhaustive necropsy and weighing the internal reproductive organs.

Data were evaluated with ANOVA for a complete randomized design, using the general linear models procedure of SAS software; SAS Institute (13). When the ANOVA showed significance, Duncan's significant-difference test was applied. Statistical significance was accepted at  $P \le 0.05$ .

# **RESULTS AND DISCUSSION**

The commercial base diets testing indicated no mycotoxins above the detection limits and they were negative for E. coli, salmonella and listeria.

The effects of dietary treatments on pig performance from 24 to 54 days of age are presented in Table 1. Results indicated no significant differences in body weight gain, feed intake and feed conversion ratio among treatments. These results are in agreement with the literature on ZEA (3, 4, 8, and 9) that reports no influence of this mycotoxin on performance. In field conditions, it is possible to observe feed rejection or decreased feed intake due to the off smell and taste produced by the fungal contamination. (4) As demonstrated before, (2) no adverse effects of the addition of Myco-Ad A-Z on ZEA diets were observed.

After two weeks of consuming 750 ppb ZEA contaminated diet with or without adsorbent all gilts started to present some degree of vulvovaginitis; but after day 23 swelling of the vulva was greater in all gilts receiving ZEA contaminated diet without adsorbent; suggesting that the addition of Myco-Ad A-Z had some benefits in reducing vulvovaginitis. None of the gilts fed the control diet presented any external sign of hyperestrogenism.

Results of necropsy showed normal liver and kidneys in all the experimental animals. Pigs fed the control diet had a normal general appearance; however all gilts receiving ZEA contaminated diet with or without adsorbent presented an enlargement of the reproductive organs. In addition, the 6 pigs fed 750 ppb ZEA had enlarged internal inguinal lymph nodes and 2 of them enlarged lymph nodes in the neck. Only 4 of 6 pigs showed a mild enlargement of the internal inguinal lymph nodes when Myco-Ad A-Z was added to the ZEA contaminated diet.

Table 2 and Figure 1 show the results of internal reproductive organs' weight. Gilts fed 750 ppb zearalenone contaminated diet had a significant heavier ovary + bursa weight (40%), uterus weight (93%), cervix weight (260%) and total reproductive organs weight (98%) than gilts fed the control diet. The addition of Myco-Ad A-Z to the contaminated diet resulted in gilts with a statistically significant reduction in ovary + bursa weight (12%), uterus weight (25%), cervix weight (32%) and total reproductive organs' weight (24%) than those fed 750 ppb zearalenone.

In spite of the effectiveness of Myco-Ad A-Z in reducing the abnormal growth of the internal reproductive organs of gilts fed diets contaminated with 750 ppb of ZEA, they were still heavier than those from gilts fed the control diet. It should be noted that feeding 750 ppb of ZEA for 30 days in succession does not illustrate what would occur in feed operations and represents a level much higher than those required to generate clinical signs of hyperestrogenism. (4)

#### CONCLUSIONS

The addition of 1.0 kg of Myco-Ad A-Z per metric ton of feed was effective in reducing the estrogenic effects of a high level of ZEA in young prepubertal gilts.

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### ABSTRACT

An experiment was conducted to study the efficacy of a very low inclusion commercial purified phylosilicate (Myco-Ad A-Z) in preventing the deleterious effects of Zearalenone in prepubertal gilts. Eighteen 20-day old recently weaned Yorkshire Cross gilts individually housed were randomly distributed into three dietary treatments with 6 replications each. After a 4-day pretrial period, pigs were fed a commercial basal diet containing or exceeding NRC recommended nutrients levels for 30 days. The feed was experimentally contaminated with crystalline Zearalenone, determined to be over 99% pure. Treatments were: (1) control basal diet; (2) control + 750 ppb zearalenone; and (3) control + 750 ppb zearalenone + 1.0 kg/mt Myco-Ad A-Z. At the end of the experiment all pigs were sacrificed and the internal reproductive organs weighed. Results indicated no significant differences in body weight gain, feed intake and feed conversion ratio among treatments. Gilts fed 750 ppb zearalenone contaminated diet showed significant heavier ovary + bursa weight (40%), uterus weight (93%), cervix weight (260%) and total reproductive organs weight (98%) than gilts fed the control diet. The addition of Myco-Ad A-Z to the contaminated diet resulted in gilts with a statistically significant reduction in ovary + bursa weight (12%), uterus weight (25%), cervix weight (32%) and total reproductive organs weight (24%) than those fed 750 ppb zearalenone. Even though the addition of 1.0 kg/mt of Myco-Ad A-Z to gilts diets contaminated with 2 to 3 times the level of zearalenone producing problems in the field did reduce the abnormal growth of the internal reproductive organs, they were still heavier than those from gilts fed the control diet. These results indicated that Myco-Ad A-Z at 1.0 kg/mt was effective in reducing the estrogenic effects of zearalenone in prepubertal gilts.

Key Words: Myco-Ad A-Z, zearalenone, gilts

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TREATMENT	BODY WEIGHT GAIN kg	TOTAL FEED INTAKE kg	FEED CONVERSION RATIO
Control	13.8 a	31.8 a	2.30 a
750 ppb Zearalenone	13.7 a	32.0 a	2.33 a
750 ppb Zearalenone + 1 kg/mt MYCO-AD AZ	14.4 a	33.2 a	2.30 a

 Table 1. Effects of Myco-Ad A-Z on body weight gain, total feed intake and feed conversion ratio of 54 day-old gilts exposed to test diets for 30 days.

**a** Means within columns with no common superscripts differ significantly ( $\not \le 0.05$ )

Table 2.	Effects of Myco-Ad A-Z on internal reproductive organs weight
	of 54 day-old gilts exposed to experimental diets for 30 days.

TREATMENT	OVARY + BURSA g/100 g PV x 1000	UTERUS g/100 g PV x 1000	CERVIX g/100 g PV x 1000	Total Reproductive System g/100 g PV x 1000
Control	<b>3.0</b> a	28.9 a	<b>3.0</b> a	37.1 a
750 ppb Zearalenone	4.2 b	55.9 b	10.8 b	73.5 b
750 ppb Zearalenone + 1 kg/mt MYCO-AD A-Z	3.7 ab	41.9 c	7.3 c	55.8 c

**a**, **b**, **c**. Means within columns with no common superscripts differ significantly ( $\cancel{P}0.05$ )

Figure 1. Relative internal reproductive organs weight of 54 day-old gilts exposed to experimental diets for 30 days.



# REPRESENTATIVE INTERNAL REPRODUCTIVE ORGANS FROM GILTS IN EACH TREATMENT



CONTROL

750 ppb ZEA

750 ppb ZEA + MYCO-AD A-Z