LOW-PHYTATE CORN: A GENETIC APPROACH TO MANURE-P MANAGEMENT¹

T.A. Doerge²

ABSTRACT

Managing nitrogen (N) and phosphorus (P) contained in manure produced by monogastric animals (primarily poultry and swine) is complicated because the N:P in corn grain is 6:1 but only 3:1 or less, in the manure. The P content in manure from monogastric animals becomes elevated because corn grain and many other feed sources contain most of their P as phytic acid (phytate) which is unavailable to these species because they lack the intestinal enzyme, phytase, needed to metabolize phytic acid-P. Repeated manure applications can lead to accumulation of soil P if the loading rates are based on N needs, or to greatly increased costs for spreading manure on larger areas of land if loading rates are based on P removal in the harvested crop. The typical practice of adding supplemental inorganic P to the feed ration exacerbates these problems. Strategies to reduce the P content in manure include the use of low-phytate, highphosphate feed grains, the use of added phytase enzyme with normal and low-phytate feed, or the genetic engineering of the grain seed or animal to produce their own phytase. Pioneer Hi-Bred International has obtained access to a corn mutant that exhibits a 54% decrease in phytate-P in the seed, and a corresponding increase in phosphate-P. Animal feeding studies with poultry and swine have documented a 10 - 40% increase in the bio-availability of P in low-phytate corn plus the expected 25 - 30% decrease in excreted P. Similar results have been reported for the supplementation of normal feed with microbial phytase. Field studies suggest that low-phytate hybrids have slightly reduced grain yield, but utilize soil P and partition plant-P similarly to their normal hybrid counterparts. Greenhouse studies suggest that the plant availability of P from lowphytate manure is the same as that of P derived from regular manure and inorganic P fertilizers. Pioneer is committed to bringing technologies to the market that enhance environmental quality although the availability of low-phytate products is probably about two to three years away.

INTRODUCTION

The potential pollutants that are derived directly from applied plant nutrients are nitrogen and phosphorus (Figure 1). Consequently, most of the research funding in soil fertility is directed toward characterizing the fate and transport of N and P, with much less support for the other essential, but non-polluting plant nutrients. Much of the agronomic work on N and P being done today is being supported by "environmental" research dollars. Innovative plant breeding work now underway at Pioneer Hi-Bred International, Inc. may provide a new genetic tool for more effective manure P management. Managing N and P is challenging enough when the two nutrients are applied as separate commercial fertilizers. It is even more difficult when managing manure-derived N and P since they are obviously contained in the same material, but in

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²Precision Farming Agronomist, Agronomy Sciences Dept., Pioneer Hi-Bred International. Inc., 7100 N.W. 62nd Ave., Johnston, IA 50131-1150

proportions that aren't necessarily optimum. The N:P in corn grain is about 6:1, while that in the manure from monogastric animals, primarily poultry and swine, has a N:P of about 3:1, or even less in some cases. With repeated manure applications, this can lead to some serious environmental and/or economic consequences. First, if manure loading rates are based on supplying an agronomically optimum amount of N, then in some cases, P levels in these fields may become elevated over time. Conversely, basing loading rates on P content will usually result in having to spread the manure on a much larger acreage, which is an obvious economic liability (Bosch et al., 1998). But why are the N:P ratios out of balance in the manure produced by monogastric animals ?

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Figure 1. The periodic table of the elements showing the essential plant nutrients and the two potential pollutant elements that can be derived from fertilizer materials.

For most feed grains, and for corn in particular, most of the P in the seed is stored as calcium or magnesium phytate, or phytic acid (Figure 2). This molecule is insoluble and unavailable to both plants and non-ruminant animals (Ravindran et al., 1994). Thus, swine and poultry feeders have traditionally added supplemental inorganic P, usually di- or mono-calcium phosphate, to the feed ration. The unutilized phytate-P was then excreted by the animals, resulting in manure that was enriched in P content relative to N. In nature, the partitioning of seed P to phytate is an ingenious way to protect the vital supply of slow-release P fertilizer, that is used by the germinating embryo and later by the young seedling. Phytate also chelates divalent cations such as Ca. Mg, and Zn and preserves these nutrients within the germinating seed. Phytate is simply myo-inositol phosphate with a phosphate group attached to each of the six carbon atoms in the cyclohexane ring. As previously mentioned, phosphorus bound as phytate is unavailable to monogastric animals unless the phosphate ester bond is cleaved, releasing free inorganic phosphate.

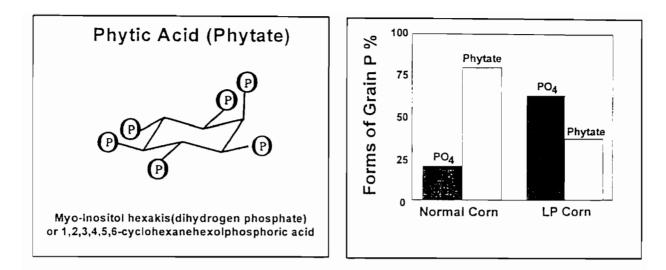


Figure 2. Structural formula of phytic acid and distribution of forms of grain-P in normal and low-phytate corn.

The group of enzymes that catalyzes the cleavage of the phosphate moiety from phytate is referred to collectively as <u>phytase</u> (Raboy and Gerbasi, 1996). Phytase is really a group of phytic-acid-specific phosphohydrolases. Thus, under natural conditions, not all of the phosphate ester bonds in phytate are normally cleaved due to the absence of all of the needed specific phytase enzymes. Phytase enzymes occur throughout nature in plants, animals and microorganisms. Phytase is produced in germinating seeds, in the intestines of ruminant animals, and in microbes such as *Aspergillus niger* (Harper et al., 1997). Unfortunately, humans and other monogastric animals lack phytase, and therefore can only poorly exploit the total phosphorus reserves in many grains. Particularly important with respect to the interests of environmental soil scientists is the lack of phytase enzyme in swine and poultry.

In 1992, Dr. Victor Raboy of the USDA-ARS National Small Grains Germplasm Research Facility in Aberdeen, Idaho developed a non-lethal corn mutant that stored most of its seed P as phosphate rather than as phytate (Raboy et al., 1994). Thus was born the concept of low-phytate corn. Now the challenge to plant breeders is to incorporate this desirable grain quality trait into potentially commercial hybrids with an acceptable suite of agronomic traits. The total concentration of P in low-phytate corn was the same as found in normal corn, but there was an equi-molar substitution of inorganic-P for phytate-P within the seed (Figure 2). Fortunately, some of the seed P in the mutant corn remained bound as phytate. This is because phytate is required for many other nutritional and developmental functions within the germinating seed and young seedling, including maturation, initiation of dormancy. and as a source of P and cations for use during germination (Raboy, 1990; Ravindran et al., 1995).

Several strategies now exist for reducing the phosphorus content in the manure from monogastric animals, and all involve improving the efficiency with which the animal extracts P from its feed ration. Reducing the phytate level in the feed grain(s) in the ration is one obvious strategy (Ertl et al., 1998). Likewise, commercially produced phytase enzyme could be added as

a supplement to either normal or low-phytate feed (Kornegay, 1996). This enzyme is commercially available under the name of Natuphos from BASF. A third strategy would be to genetically alter normal corn to contain phytase in the seed. Finally, the animal itself could be genetically engineered to produce the phytase enzyme in sufficient quantities to adequately increase the availability of the phytate-P contained in normal feed.

PRELIMINARY STUDIES WITH LOW-PHYTATE CORN

Pioneer was the first seed company to gain access to the *lpa1-1* gene from the USDA-ARS. In the process of evaluating this trait, several research questions had to be answered before the commercial value of this trait could be determined. First, what is the nutritional value of low-phytate corn, and is it at least equal to the nutritional value of normal corn plus phosphate additives ? Second, does the lower percentage of phytate-P in the seed actually translate into lower P levels in the swine and poultry manure that is produced ? Third, is the P remaining in manure derived from low-phytate fed animals available to plants once it has been applied to the soil ? Several agronomic questions also had to be addressed: Do low-phytate hybrids require any special management, and do they have a package of agronomic traits, particularly harvestable yield, that will be marketable (Ertl et al., 1998)?

Pioneer has conducted over 20 feeding trials with both swine and poultry in cooperation with several universities and the USDA. Overall, it was found that P availability to animals was indeed increased when low-phytate corn was consumed. There are several ways to estimate P bio-availability, but an average increase of ~35% appears typical (Ertl et al., 1998). Similar gains in P availability have been achieved with the addition of commercial phytase to normal feed (Kornegay, 1996). Likewise, P excretion was also reduced 25-30%, and up to a maximum of 60% in one study where both phytase and low-phytate corn were used (Baxter et al., 1998). In this study, the reduction in P excretion probably represents a "best case scenario" for P removal from typical feeds. The animals were young pigs that were on a high protein diet, which contained a relatively high percentage of (high-phytate) soybean meal in the ration. This may have accentuated the apparent effectiveness of the phytase treatment in boosting P availability. One interesting side note is the technique of in-vitro analyses used by animal nutritionists to estimate the P availability of different feed types. In concept, this is very similar to the in-vitro analyses used by soil scientists for many decades to assess the plant availability of soil P.

In one study, Joern and his co-workers at Purdue looked at the fractionation of P in manure derived from normal and low-phytate feed, both with and without added phytase (Baxter et al., 1998). The inclusion of low-phytate grain and phytase in the ration progressively increased the percent of manure-P in the phosphate form, and decreased the concentration of phytate-P (Figure 3). This finding is important for two reasons. First, the increased partitioning of P to phosphate should insure that P in the manure will be readily available to plants once applied to the soil. Second, decreased levels of phytate-P in the manure will favor more complete adsorption of inorganic P on soil colloids which may make the phosphate-P less susceptible to downward movement in soil drainage (Anderson et al., 1974).

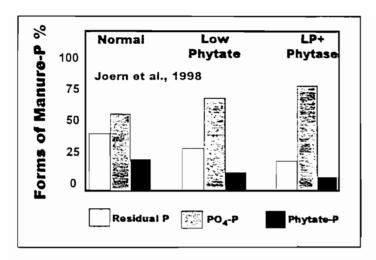


Figure 3. Distribution of the forms of P in manure obtained from young pigs fed normal feed, low-phytate feed, and low-phytate feed + phytase enzyme.

Some preliminary agronomic studies have also provided few surprises. Mikkelsen at NCSU reports that corn growth for the first six weeks was equivalent when inorganic-P, normal manure, and low-phytate manure were applied to a P deficient Norfolk s.l. soil at equivalent P rates (Spencer et al., 1998). Schmidt and Randall (1998) in Minnesota found that P uptake in grain and stover were the same for low-phytate hybrids and their normal counterparts. Grain yields for two converted low-phytate hybrids were 5-9% lower, although this was not unexpected for these experimental lines. They will still undergo several more backcrossing cycles to improve the genetic purity of the converted hybrids. At that time, further agronomic testing and characterization will be conducted.

ENVIRONMENTAL BENEFITS OF LOW-PHYTATE CORN

From the sample calculations shown in Table 1, the use of low-phytate manure has the potential for significant environmental benefits both when manure loading rates are based on N content or in the more restrictive case where they are based on the P content of the manure (Zublena et al., 1993). When manure loading rates are based on N requirement, the use of lowphytate manure will reduce applied P by 30%. When manure rates are restricted to just replace P removed in the harvested grain, the use of low-phytate manure allows for a 44% increase in loading rate compared to what would be allowed with regular manure. This directly affects the number of acres required to accommodate a given tonnage of manure, and hence, the economy of manure management. In this example, low-phytate manure could be spread on 30% fewer acres than the same number of tons of normal manure. The restriction of manure loading rates to crop removal of P is currently used in the Netherlands on soils with high soil test P levels (Parry, 1998). If low-phytate corn is commercialized, the areas of the U.S. where it will be needed will occur primarily in regions where soil test P values are high to very high and swine and broiler production is heavy. Animal producers should be able to source local low-phytate corn in all of the major production areas except perhaps in eastern North Carolina and the Delmarva Peninsula.

Manure	N-Based Rec	commendations ¹	P-Based Recommendations ¹					
Туре	Manure	$N P_2O_5$	Manure	$N P_2O_5$				
	t/a	lbs/a	t/a a/t	lbs/a				
Regular	21	175 187	7.3 0.14	61 66				
Low-Phytate	21	175 131	10.5 0.10	88 66				
% Advantage	-	- 30	44 30	- 44 -				
for LP manure								

¹Assumes: 175 bu/a yield goal, N requirement of 1 lb/bu, Fresh swine manure @ 0.60% N, 0.20% P or 0.14% P in LP manure, Soil incorporation (N Availability Coefficient = 0.7)

After: Zublena et al., 1993.

Table 1. Comparison of suggested swine manure loading rates using a nitrogen- or a phosphorusbased recommendation system for manure derived from normal corn feed and low-phytate feed.

THE FUTURE OF LOW-PHYTATE CORN

The commercialization of low-phytate corn depends on the value it commands in the market place. Contributing to the value of low-phytate corn are the replacement cost of feed supplements, primarily di-calcium phosphate, other nutritional benefits related to Ca, Mg. Zn. and N utilization, reduced transportation costs for hauling manure to fewer acres for application, and a largely undefined environmental benefit. The recent national policy on manure management announced in September, 1998 by the Clinton Administration may have an effect on the value of environmentally-friendly products like low-phytate corn, but that effect, again, is very difficult to define. Pioneer is very reluctant to push low-phytate corn adoption in such a way that they are viewed as "driving" regulation for the intent of increasing product sales. For now, it appears that the best fit for low-phytate corn will be in highly environmentally sensitive areas that are characterized by serious soil erosion, high soil test P levels, and/or high concentrations of swine or poultry feeding operations. The Delmarva poultry-producing area is one excellent example. Future industry recommendations may well include the use of low phytate corn, phytase enzymes, and possibly the use of low-phytate soybeans in the feed rations. Ultimately though, the commercialization of the LP trait will happen only if there is sufficient value available in the marketplace for this specific grain trait. Stacking of low-phytate with other value-added traits such as high-oil are also being considered. The bottom line is that Pioneer and it's partner, Optimum Quality Grains LLC, are committed to bringing technologies to the market that enhance environmental quality. Low-phytate is one of those products. The availability of low-phytate products is probably about 2-3 years away.

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