# RECENT ADVANCES IN THE USE OF PHYTASE ENZYME IN DIETS FOR GROWING PIGS

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# SUMMARY

The effect of phytase enzyme on the digestibility and availability of phosphorus (P) in soya-bean meal was investigated with grower pigs. In addition, the relative effectiveness of different parameters for assessing P availability in slope-ratio assays were compared. The addition of phytase increased growth rate (741 vs 835 g/d; P < 0.05), lowered the FCR (2.37 vs 2.16; P < 0.01), increased protein deposition (108 g vs 123 g/d; P < 0.05), and increased the apparent digestibility of P (0.45 vs 0.69; P < 0.01). There were considerable differences in the estimates of P availability depending on whether bone bending moment, total P retention or digestible P intake was the criterion of response. Phytase supplementation increased the availability of P in soya-bean meal from 0.11 to 0.69, when bone bending moment was the criterion of availability.

## **INTRODUCTION**

The availability of P in vegetable sources varies as the P is bound as phytate P, which renders it unavailable. The degree of availability depends on the level of phytase activity naturally found in the feed ingredients (Pointillart *et al.* 1984, 1987). The phytase releases the bound P, making it available to the pig. Recently, phytase has been developed commercially for use in diets to release the bound P (Anonymous 1989). If completely successful, their use would make estimates of availability of P in feed sources no longer necessary in dietary formulations. In addition, any increase in the availability of P would decrease the concentration of undigested P in the faeces, thereby reducing environmental pollution.

There are two general systems for assessing P availability. Values recommended by National Research Council (1988) were based on slope-ratio assays with growing pigs and using bone bending moment as the criterion of response. By contrast, many European countries use P digestibility as an estimate of P availability. Previous work at Wollongbar had indicated inconsistencies in the availability of P in soya-bean meal depending on whether bone bending moment or P retention in the empty body was the criterion of response.

This paper reviews two experiments that were conducted to (1) determine the effectiveness of supplementation of phytase on the digestibility and availability of P in soyabean meal, and (2) to compare the effectiveness of bone bending moment, total P retention and P digestibility as parameters of response for assessing P availability.

# MATERIALS AND METHODS

#### Experiment 1 Slope-ratio assav for P availability

The aims of this experiment were to (1) determine the effect of phytase supplementation on the availability of P in soya-bean meal and (2) to compare bone bending moment and total P retention in the empty body as criteria of response for assessing P availability.

With the slope-ratio assay, the slope of the response of growing pigs to graded levels of the test P (soya-bean meal) is compared to the slope of the response to standard P (monosodium phosphate [MSP]). In this work, the basal - sugar - soya-bean meal diet

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contained 2.5 g P/kg and two levels of P (3.25 and 4 g/kg) were contributed from either the soya-bean meal or MSI?

One of the principles of the slope-ratio assay is that the addition of the test meal does not interact with the basal diet. This principle was not possible with the use of phytase as supplementation with the enzyme would affect the availability of P in both the test and the basal diet. Thus, two separate slope-ratio assay treatments were necessary to determine the effect of phytase on the availability of P in soya-bean meal. However, both assay treatments were run within the one experiment.

In the first slope-ratio assay, the availability of P was determined in soya-bean meal. In the second, all diets were supplemented with phytase (1,000 units/kg) and the availability of P in the soya-bean meal, with phytase supplementation, determined. Thus, the change in the availability of P in soya-bean meal would be a measure of the effect of the enzyme on P availability.

Thus there were 10 diets for this assay (five without phytase and five with phytase) and the diets were offered *ad lib* to growing pigs for 35 days.

The pigs were then slaughtered, a number of bones were removed for bone bending moment determination from the right hand side of the carcass, and the left hand side, together with the blood and washed viscera, were frozen and ground for subsequent analyses.

The effects of treatments on growth, bone and empty body parameters were determined by a linear regression analysis with parameters being regressed against P concentration in the diets. The estimate of P availability was expressed as a ratio of the slope of the response to the soya-bean meal diet relative to the slope of the response to the MSP diets. In many cases, particularly with soya-bean meal, the responses were non linear. Rather than delete these analyses, the estimates of P availability were still calculated for comparative purposes.

## Experiment 2 P digestibility

The aims of this experiment were to (1) determine the availability of P in soya-bean meal using P digestibility as the criterion of availability, and (2) to determine the effect of phytase supplementation on P digestibility.

For the slope-ratio assay, the five diets were as for the unsupplemented diets in Experiment 1. In addition, a sixth diet incorporated phytase supplementation (1,000 units/kg) of the diet containing 4.0 g P/kg from soya-bean meal only (diet 5).

The pigs were fed once daily at a feeding rate of three times maintenance and P digestibility was determined over a ten day collection period.

#### RESULTS

### Experiment 1

The addition of phytase increased gain/d (741 vs 835), decreased the FCR (2.37 vs 2.16) but had no effect on feed intake (Table 1). The addition of soya-bean meal depressed feed intake and gain/d and increased the FCR relative to the effects of the addition of MSP.

Phytase supplementation significantly increased protein deposition (108 g vs 123 g/d) (Table 1) whereas the addition of soya-bean meal depressed the rates of deposition of both protein and energy.

The availability of **P** in soya-bean meal, using the average of the bone bending moment responses, was 0.11 (Table 2). The addition of phytase increased the estimate of availability to 0.69.

When availability of **P** was based on total **P** retention in the empty body, the estimate was 0.28. Supplementing the soya-bean meal with phytase increased the estimate of availability to 0.41.

The mean retention of **P** from soya-bean meal was 0.28 (without phytase) and 0.46 (with phytase).

Diet	P level (g/kg)	Feed intake (g/d)	Live-wt gain (g/d)	FCR	Protein deposition (g/d)	Energy deposition (MJ/kg)
Without phytase						
Basal diet	2.50	1844	700	2.64	100	10.9
MSP	3.25	1912	833	2.32	122	11.1
	4.00	1692	794	2.14	121	9.9
Soya-bean meal	3.25	1659	691	2.43	97	9.2
,	4.00	1584	687	2.31	102	7.2
With phytase						
Basal diet	2.50	1913	841	2.27	121	11.4
MSP	3.25	1801	826	2.19	125	11.3
	4.00	1785	915	1.96	134	11.5
Soya-bean meal	3.25	1719	760	2.28	114	9.4
,	4.00	1722	833	2.08	120	9.2
Mean						
Without phytase		1738	741	2.37	108	9.7
With phytase		1788	835	2.16	123	10.6
Statistical significance						
Phytase						
(with vs without)		NS	*	**	*	NS
P source (MSP vs soya)		*	**	NS	**	**
P level (3.25 vs 4.00)		NS	NS	**	NS	NS

Table 1 The effect-of phytase on growth responses and protein and energy deposition in growing pigs in Experiment 1

Table 2 The effect of phytase on the availability of P in soya-bean meal using bone bending moment (kg-cm) or P retention in the empty body (g) as the criteria for P availability in Experiment 1

Basal Total P (g/kg) diet		Monosodium			Soya-bean		Statistics		Availability*	
	phosphate		m	meal		Linearity				
	2.5	3.25	4.00	3.25	4.00	MSP	Soya	-		
Bone bending										
moment										
Without phytase										
Metacarpal <sub>3</sub>	26	32	39	25	27	**	NS		(0.05)	
Metacarpal₄	27	35	44	26	27	**	NS		(-0.03)	
Metatarsal <sub>3</sub>	24	33	42	26	27	**	NS		(0.13)	
Metatarsal	24	34	45	26	30	**	NS		(0.27)	
Femur	211	294	377	234	226	**	NS		(0.09)	
								Mean	(0.11)	
With phytase										
Metacarpal <sub>3</sub>	36	42	42	36	39	NS	NS		(0.53)	
Metacarpal <sub>4</sub>	38	48	50	41	45	**	NS		(0.57)	
Metatarsal <sub>3</sub>	35	51	49	39	46	**	*		0.82	
Metatarsal₄	35	57	53	44	48	**	*		0.72	
Femur	326	401	457	371	433	**	**		0.82	
								Mean	(0.69)	
P retained										
Without phytase	50	78	93	45	62	**	NS		(0.28)	
With phytase	85	123	121	85	100	**	NS		(0.41)	

\* Values within brackets were based on non-linear responses

## Experiment 2

When digestible P was used as the criterion of availability, the estimate of availability of P in soya-bean meal was 0.66 (Table 3). The addition of phytase decreased P in the faeces, and increased P digestibility from 0.45 to 0.69 (Table 4). Phytase supplementation had no effect on the dry matter or crude protein digestibility but decreased energy digestibility slightly.

Table 3 The availability of P in soya-bean meal using digestible P intake (g/10 d) as the criterion for availability in Experiment 2

	Basal diet	М	SP		-bean eal	Stati	stics	Availability
Total P (g/kg)	2.5	3.25	4.00	3.25	4.00	Line	arity	
Diet no.	1	2	3	4	5	MSP	Sova	
Digestible P	9	17	26	14	20	**	**	0.66

Table 4 The effect of phytase on P content in the faeces and the digestibility of P, dry matter, crude protein and energy in the diet containing soya-bean meal in Experiment 2

	Soya-be	Significance		
Total P (g/kg)	4	4		
Diet no.	5 - phytase	6 + phytase		
P intake (g)	46	47	NS	
P in faeces (g)	25	14	**	
Digestible P intake (g)	20	33	**	
P retained (g)	20	33	**	
Digestibility (g/g)				
P	0.45	0.69	**	
Dry matter	0.92	0.91	NS	
Crude protein	0.92	0.91	NS	
Energy	0.92	0.91	*	

Table 5 The effect of phytase supplementation on the availability of P in soya-bean meal, as assessed by various criteria, together with the effect of phytase on apparent digestibility and mean retention of P by growing pigs

	Soya-bean	Soya-bean meal*			
	Without phytase	With phytase			
Estimate of P availability					
Based on digestible P	0.66	-			
Based on bone bending moment	(0.11)	(0.69)			
Based on P retained in the empty body	(0.28)	(0.41)			
Apparent digestibility of P	0.45	0.69			
Mean P retention in the empty body	0.28	0.46			

\* Values within brackets were based on non-linear responses

### DISCUSSION

The results indicate two major effects, (1) that there were considerable inconsistencies in the estimates of availability of P depending on the criteria used to estimate availability (results summarised in Table 5), and (2) that phytase supplementation had substantial effects, not only on bone development, but also on growth responses.

The availability of P in soya-bean meal was 0.11 using bone bending moment as the criterion of response. This was lower than estimates of 0.25 - 0.38 for soya-bean meal reported by the National Research Council (1988). It was also considerably lower than the estimate of 0.28 based on total I? retention in the empty body and 0.66, based on the digestibility of I? This latter estimate of 0.66 is actually higher than the apparent digestibility of P in soya-bean meal (0.45). This, in part, reflects the fact that the P in MSP that was used as the standard in the slope-ratio assay was not fully digested (estimated at 0.87).

These inconsistencies in estimates of availability also occured with phytase supplementation. Phytase supplementation increased the availability of P from 0.11 to 0.69, using bone bending moment as the criterion of response. This was substantially greater than the availability estimate based on total P retention (0.41) but similar to the apparent digestibility of P in the diet (0.69).

It is difficult to explain the reasons for the inconsistencies in the estimates of P availability. Part of the reason may be due to the fact that a number of the estimates were based on nonlinear responses. In order to improve the chances of a linear response, there is a need to increase the intake of available I? This, however, is difficult with protein concentrates such as soya-bean meal, which have a low available P content. It is necessary in the slope-ratio assay to ensure that the basal diet is adequate in protein but deficient in available P, to make the assay sensitive. The addition of substantial quantities of a protein concentrate to the basal diet is likely to depress pig performance due to excess protein in the diet (as happened in Experiment 1). It is assumed such effects do not affect P utilization. However, it would be difficult to increase the quantity of soya-bean meal substantially above that used in Experiment 1, thereby making it difficult to achieve a linear response.

On the other hand, the estimates of apparent digestibility of P were not effected by the differences in P concentration from the addition of soya-bean meal. It is possible that digestible P may be a more suitable estimate of P availability than slope-ratio assays. Values for apparent digestible P would seem suitable provided that dietary P concentration does not effect digestibility. The assumption also has to be made that all of the digestible P is absorbed in a form that is fully available to the pig. If these two conditions can be met, then the experimentation associated with digestibility estimates is less than that required for slope-ratio assays, based on growth responses.

The results indicate that phytase supplementation has substantial effects on the digestibility and availability of I? The effects on growth rate, feed efficiency and protein deposition are more difficult to explain. One explanation is that the phytase enzyme exerted a proteolytic effect, possibly by liberating amino acid - phytate bonds, thereby resulting in increased protein deposition. However, there was no evidence of phytase supplementation improving nitrogen digestibility in Experiment 2 to support this hypothesis. The alternative is that the diets were so P deficient that protein deposition and growth rate was limited. The release of available P would have stimulated skeletal development, which in turn would have stimulated protein deposition and improved growth and feed efficiency. It would be necessary to examine the effect of phytase supplementation of diets adequate in P to confirm this hypothesis. Phytase supplementation also had a small negative effect on energy digestibility, but this seems most likely due to chance.

Overall, the results indicate there is a need for additional research to clarify techniques for assessing P availability, over a wider range of feeds. However, provided phytase supplementation is cost effective, then its routine use would reduce the importance of accurate estimates of P availability in feeds. Further work is needed to verify this aspect over a wider range of dietary formulations.

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