Strategies to Reduce Environmental Pollution from Animal Manure: Nutritional Management Option* - Review -

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ABSTRACT : The first option in manure management is developing an environmentally sound nutritional management. This includes proper feeding programs and feeds which will result in less excreted nutrients that need to be managed. Critical components that should be controlled are N, P and minerals that are used at supranutritional levels. Amino acid supplementation and protein restriction reduce N excretion in the monogastric animals. Supplementation with enzymes, such as carbohydrases, phytase and proteases, can be used to reduce excretion of nutrients and feces by improving digestibility of specific nutrients. Growth promoting agents, such as antibiotics, beta-agonists and somatotropin, increase the ability of animals to utilize nutrients, especially dietary protein, which results in reduced excretion of N. Some microminerals, such as Cu and Zn, are supplemented at supranutritional level. Metal-amino acid chelates, metal-proteinates and metal-polysaccharide complexes can be used at a much lower level than inorganic forms of metals without compromising performance of animals. Deodorases can be used to avoid air pollution from animal manure. Nutritional management increases costs to implement. It is necessary to assess the economics in order to find an acceptable compromise between the increased costs and the benefits to the environment and production as well. *(Asian-Aus. J. Anim. Sci. 1999. Vol. 12, No. 4 : 657-666)*

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INTRODUCTION

The animal industry must be environmentally sound to ensure its long-term sustainable growth. Livestock wastes, mostly manure, can be a valuable resource as a fertilizer or soil conditioner, but can also be a potential hazard to the environment. Environmental concerns relate to water quality, soil degradation, air pollution and rural-urban interface issues. Land applications of manure are subject to surface run-off and leaching that may contaminate ground or surface waters. Nitrate leaching has been considered a major nitrogen (N) pollution concern with livestock farms. Ammonia toxicity to fish and altered effectiveness of chlorination are other concerns. Phosphorus (P) entering surface waters can stimulate growth of algae and water plants. Decomposition of these results in an increased oxygen demand, which may interfere with the well-being of fish and wildlife. Excessive contributions of some minerals from animal manure can create high salt concentration in the soil. High concentrations of copper in pig diets can cause accumulation of copper in the soil. Manure can be a major source of methane and nitrogen oxides which contribute to the accumulation of greenhouse gas. Volatilization of ammonia causes acid rain which results in forest dieback in western Europe (ApSimon et al., 1987). Emissions of nitrous oxide (N_2O) during nitrification and denitrification cause depletion of the stratospheric ozone layer (Christensen, 1983). Manure can be a source of odours which contribute to friction between urban and rural residents.

As described above, animal manure can be a valuable resource while it can be a major obstacle in the future development of animal industry if the impact on the environment is not properly controlled. Major efforts are required to adopt all best available technologies capable of reducing pollutants from the animal industry before further restrictive legislation is enacted to control the problem. There are a number of possible solutions to this problem.

The first option in manure management is developing an 'environmentally sound' nutritional management, that is, a feeding program and feeds to result in less excreted nutrients that need to be managed. Once the manure is produced it can be best utilized as a fertilizer or a soil conditioner. In many countries the amount of manure that can be spread on land depends on the nutrient requirements of the crop being grown. The laws specify maximum application rates and not animal stocking rates. Farmers who reduce the N and P component of manure can release pressure on the environment without having to reduce the number of animals. There are alternative systems for housing and manure treatment which generate manures that are easier to handle and have less pollutants or more economic value. Treated animal wastes may also be used as a feedstuff or fuel source.

The present paper reviews and reports the results of experiments on the nutritional management for dealing with animal manure to reduce environmental pollution.

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NUTRITIONAL MANAGEMENT TO REDUCE POLLUTION FROM ANIMAL MANURE

A feeding strategy, i.e. nutritional management, is the beginning point in reducing environmental pollution from animal manure. In the past, feeding programs and diet formulations were aimed at maximizing production performance without special concern for nutrient oversupply. Environmental constraints now necessitate a close adjustment of nutrient supply to requirements so as to obtain the lowest level of nutrient excretion. There are measures that can be taken to reduce the amount of pollutants excreted by farm animals, particularly N and P in poultry and swine. Some feed additives can be used to control odour from manure. Minerals in special forms can be used at lower levels than those in inorganic forms, resulting in reduced excretion of minerals. The potential reductions in N and P pollution that could be achieved by employing the various measures are shown in table 1.

Table 1. Potential effects of nutritional strategies to reduce the N and P content of manure

Factors	Estimated % reduction in manure	
	N	P
Supplements;		
Synthetic amino acids and reduced protein in feeds	20-25	
Enzymes - Cellulases	5	
Phytases		25-30
Growth promoting substances	5	5
Systems;		
Formulation closer to requirements	10-15	10-15
Phase feeding	10	10
Use of highly digestible feed ingredients	5	5

(Federation Europenne des Fabricants d'Adjuvants pour la Nutrition Animale, 1992).

Amino acids supplementation and protein restriction

1. Pigs

When trying to identify possible methods for reducing the level of N excretion by pigs it is important to understand where the excreted N comes from. Data reviewed by Jacob (1995) indicate that of the total N consumed by a growing-finishing pig, about 20% is excreted in the feces and 50% in the urine. Fecal N is of three main forms: undigested dietary protein, microbial protein and endogenous protein. Up to 80% of fecal N in the pig is in the form of bacterial protein.

On the basis of the information obtained to date, the following methods can be identified as having potential for reducing N excretion:

- Meeting the animal need for amino acids without providing excesses (i.e. providing ideal protein)
- Increasing protein digestibility
- Reducing endogenous N excretion

A one percentage unit reduction in total protein content of growing finishing diets is possible with the use of synthetic lysine and methionine. Such a reduction in dietary protein content can result in up to an 8.5% decrease in N excretion. With the use of synthetic threonine and tryptophan in addition to lysine and methionine, it is possible to lower the crude protein level by two percentage units, thereby reducing N excretion by approximately 20%. Koch (1990) demonstrated that in pigs, by using a diet low in protein (11 vs. 13.9%) but supplemented with the four most limiting amino acids (lysine, methionine, threonine and tryptophan), there was no reduction in live-weight gain or feed conversion efficiency but N excretion was reduced by nearly 30%.

The amino acid requirements of a pig will change as it grows. This has led to the introduction of phase feeding. For example, the use of a grower diet from 45-70 kg live wt and a finishing diet from 70-106 kg instead of one growing-fattening diet from 45-106 kg results in a reduction in excretion of N. Multiphase feeding with weekly mixing of two feeds (17 and 13% crude protein) reduces N output by 10-20% compared with a two-phase feeding. Table 2 shows an example of the reduction of N excretion through protein restriction, amino acids supplementation and change of feedstuffs.

Table 2. Options in feeding program and protein level (%) of pig diets and N excretion (kg/pig)

Feeds	Present situation, 1990	Protein restriction	Supplementation with 2-4 synthetic AAs[1]	Additional AAs plus selected feedstuffs
Starter feed	17.5	16.0	15.0	14.0
Grower feed	17.0	15.5	14.5	13.5
Finishing feed	16.0	14.5	13.5	12.5
Fattening feed	16.6	-	-	-
N excretion	4.5	3.9	3.5	3.1

[1] Amino acids. (Jongbloed and Lenis, 1992)

2. Poultry

Considerable work has been carried out to determine the levels to which dietary protein can be reduced without adverse effects on animal performance. The possibilities of reducing N excretion and environmental pollution from poultry by dietary supplementation with individual amino acids was reviewed by Leclercq and Tesseraud (1993). These authors calculated that poultry used feed protein with an overall efficiency approaching 45% but that the conversion by poultry of dietary protein to human consumable protein was in the order of 27%. They calculated that, in theory, by the use of pure amino acids and reducing the protein content of a grower diet from 22 to 16%, it was possible to achieve a 72.5% efficiency of utilization of protein with a concurrent reduction in N excretion by a factor of 1.7. However, results of experiments indicated that it is difficult to achieve this level of improvement in practice.

Table 3 shows the effect of dietary protein and amino acid levels on nitrogen excretion by broilers during the grower phase (Jacob et al., 1994). Broilers fed the reduced protein diets (20.5% vs. 18%) with increased amino acid levels performed as well as, if not better than, the broilers on the control diets and showed a significant reduction in total N output. The N of the excreta was composed of urea/ammonia N (9.67%), uric acid N (61.05%) and residual N (29.27%). On a daily basis, the reductions in N excretion by lowering protein content were 22.8% for total N, 25.4% for uric acid N, 17.2% for urea/ammonia N and 18.2% for other N components.

Table 3. The effect of dietary protein and amino acid levels on the growth and nitrogen excretion of broilers during the grower phase

Dietary protein, %	Amino acid supplement	Final body weight, g	Feed:Gain Ratio	Total N in feces, % DM	N excreted, g/bird/day
20.5	+ 10%	2,230	2.26	5.16	2.87
	Control	2,217	2.29	5.04	3.09
	- 10%	2,224	2.22	5.53	3.25
	Average	2,221	2.26	5.25	3.07
18.0	+ 10%	2,230	2.17	5.60	2.08
	Control	2,200	2.34	4.46	2.45
	- 10%	2,172	2.43	4.10	2.58
	Average	2,201	2.31	4.72	2.37

(Jacob et al., 1994)

A similar experiment with laying hens indicated that dietary protein can be reduced from 17 to 13.5% with no loss in egg production or egg quality, as long as essential amino acid levels are maintained (table 4). The reduction in dietary protein resulted in more than a 30% reduction in daily N output.

Table 4. The effect of dietary protein and amino acid levels on the egg production and nitrogen excretion of laying hens

Dietary protein, %	Amino acid supplement	Egg production, %	Total N output, g/bird/day
17.0	+ 10%	90.3	6.96
	Control	89.0	6.90
	- 10%	90.2	6.88
	Average	89.8	6.91
13.5	+ 10%	89.4	4.60
	Control	87.7	4.17
	- 10%	88.6	4.42
	Average	88.6	4.40

(Blair et al., 1995)

Enzymes

1. Carbohydrase

Non-starch polysaccharides (NSP) in some feed grains (e.g., pentosans or arabinoxylans in wheat and rye, and β-glucans in barley and oat) are soluble fibers. Their presence can either block digestion of other nutrients (e.g., protein and starch), or can seriously inhibit absorptive capacity. Therefore, the digestibility of NSP is low in monogastric animals (table 5).

Table 5. Content of non-starch polysaccharides (NSP) in feedstuffs and digestibility of NSP in young chickens

Feedstuff	NSP, % DM	Digestibility, %
Barley	15	14
Wheat	10	12
Soybean	20	0
Pea	22	18
Bean	23	19
Rapeseed	24	7
Wheat bran	34	9
Sunflower seed	28	17
Rice bran	25	3
Grass	28	5
Corn gluten feed (20%)	31	17

(Cited by Charlton, 1996)

It was found that the results of using enzymes (xylanase or β-glucanase) did not stem from complete hydrolysis of the non-starch polysaccharides but that relatively minor hydrolysis altered the ability of the medium to form a viscous solution and act as a barrier to endogenous enzyme activity.

In the past few years a number of different feed enzymes have been developed. The use of multi-enzyme preparations in traditional wheat-based poultry diets was examined (Graham, 1992). The results demonstrated that with a diet based on 60% wheat, a mixed enzyme preparation was capable of increasing the rate of liveweight gain (+17%) and at the same time reducing feed conversion ratio (1.46 to 1.29). There was also an increase in the N utilization percentage (37.4 to 45.3%). Such improvements were attainable even after pelleting which in itself was capable of solubilizing starch (Pettersson et al., 1991). A commercial multi-enzyme preparation from *Trichoderma viride* contained 11,150 U/g cellulase, 27,600 U/g glucanase and 37,150 U/g xylanase. This multi-enzyme product was tested with layers fed a barley-based diet (Brufau et al., 1994) and a wheat-based diet (Um et al., 1998). The results showed that barley and wheat can replace corn as an energy source in layer diets if the enzyme is properly supplemented.

For the better utilization of enzymes in feed industry, commercial enzyme preparations should be customized depending on the animal species, age of animals and major feed ingredients. Enzyme products which contain β-glucanase and xylanase in different proportions were produced from *Trichoderma longibrachiatum* and *Bacillus subtilis*. They were used with different diets (wheat-based or barley-based) in different animal species of different ages (poultry, starting pigs or growing-finishing pigs). Enzyme products added to the respective diets reduced the viscosity caused by non-starch polysaccharides and increased amino acid availability as well as energy and P availability (Creswell, 1994). Low and Longland (1990) reported that N retention of pigs was slightly increased by enzyme supplementation.

Contents of moisture and N were lower in the litter of birds given diets supplemented with β-glucanase. Measurement of ammonia release from the litter indicated that when a second flock of birds was raised on the same litter, the presence of a glucanase in the diet reduced the level of ammonia release by 80% (Williams and Kelly, 1994).

An experiment was conducted to test the possible interaction of an enzyme complex and feed antibiotics on growth and metabolic parameters of broilers. The basal diet contained barley at a level of 40%. Both supplements, when added together in the diet, had almost an additive effect on growth parameters, and on energy, fat and N utilization (Vukic Vranjes and Wenk, 1993).

Overall nutritional management can result in the reduction of manure output. A proven and more direct method is enzyme supplementation. Reducing the DM content of the digesta in the intestinal tract with supplemental feed enzymes has a marked impact on excreta volume and composition. In a trial offering wheat or wheat/barley-based diets to broilers, excreta weight was reduced by 17-28% in fresh or 12-15% in DM by supplementation of a multi-carbohydrases enzyme product (table 6). The direct production benefits of lower excreta output and reduced fecal DM are seen in some broiler trials where observations on the frequency of hock lesions and breast blisters are recorded. Reductions in manure output and water content will improve litter quality, and possibly decrease carcass downgrade.

Table 6. Excreta output of broilers fed wheat or wheat/barley-based diets with and without multi-enzyme (carbohydrases) supplementation at 19 - 21 days of age

	Wheat Control	Wheat+ Enzyme	Wheat/ Barley Control	Wheat/ Barley +Enzyme
Fresh excreta, g	221	184	258	185
Excreta DM, %	42.4	45.2	39.8	47.3
Dry excreta, g	94.0	83.0	102	87.0

(Lakeside Research Center, Canada cited by Wyatt, 1995)

2. Phytase

Feedstuffs of plant origin contain adequate levels of P but about two thirds of it is in the form of phytate-P (myo-inositol hexakisphosphate) which has a low availability in non-ruminant animals. This low availability is due to the absence of phytase in the gastrointestinal tract of non-ruminant animals.

Phytase is an example of a specific enzyme which is used to target phytate to release free phosphorus with excellent results in pigs and broilers (Simons et al., 1990; Keteran et al., 1992). There are several options available for increasing the phytase activity of diets - either the diet can be supplemented with phytase-rich cereals, or phytase of microbial origin can be added.

Plant phytase activity, especially that of wheat, has been known for some considerable time. Seed phytase activity varies greatly from one species to another. In fact, few dormant seeds contain phytase activity, except for wheat and rye and their hybrid, triticale (table 7).

Table 7. Phytate phosphorus and phytase activity in the major components of animal feeds

Ingredients	Phytic P, g/kg	Phytic P, % of Total P	Phytic Activity, U/Kg
Wheat	1.7-2.5	60-77	700 ± 100
Maize	1.7-2.2	66-85	n
Oats	1.9-2.3	55-63	n
Barley	1.9-2.5	51-66	400 ± 200
Triticale	2.5-2.6	65-68	1,500 ± 170
Rye	2.2-2.5	61-73	4,900 ± 620
Sorghum	1.8-2.2	60-74	-
Peas	1.2-1.7	40-50	n
Wheat bran	8.1-9.7	70-90	1,200 ± 150
Wheat shorts	4.7-5.8	66-85	1,900 ± 140
Rye bran	7.6	71	6,300 ± 1,100
Soybean meal	3.2-3.8	51-61	n
Rapeseed meal	6.0-7.3	60-73	n
Sunflower meal	6.2-9.2	73-80	n
Peanut meal	3.2-4.3	47-69	n

(Pointillart, 1993)

To date, experiments on the effects of phytase have been carried out using a microbial form of phytase supplied by either *Aspergillus ficuum* (Simmons et al., 1990; Nasi, 1990) or *Aspergillus niger* (Beers and Jongbloed, 1992). A problem with this preparation has been the sensitivity of the organism and the enzyme to temperature and hence the loss of activity above 80C, a temperature often experienced during the process of feed pelleting. An interesting development which may herald a new era in the use of enzymes in animal feed was recently reported when the gene coding for phytase activity in *Aspergillus niger* was engineered into tobacco seeds. The enzyme was expressed as 1% of the soluble protein in mature seeds. Supplementation of broiler diets with transgenic seeds resulted in an improved growth rate, comparable to diets supplemented with fungal phytase or P (Pen et al., 1993).

(1) Pigs

Cereals that contain phytase such as wheat, triticale, rye or their by-products, result in better phytate utilization when included in pig diets. Pointillart et al. (1993) designed diets which contained decreasing amounts of inorganic P and increasing dietary phytase activity by altering the dietary proportions of wheat, wheat by-products (bran and shorts), and rye bran. The proportion of inorganic P in the diet ranged from 0 to 0.3%. There was no effect on pig performance in the period to slaughter and no effect on bone density or strength at slaughter in pigs given the diet with low inorganic P supplemented with cereals containing phytase compared with the pigs given a standard diet supplemented with inorganic P. Pigs fed diets with and without added phytase commencing at 25 kg live weight showed that supplementation with phytase resulted in significant improvements in growth performance only at the lower levels of available P (Williams and Kelly, 1994). In an experiment to establish the P equivalency of *Aspergillusniger*-phytase in pigs, digestibility of P increased up to approximately 1,000 U/kg, with no further significant response beyond 1,000 U/kg. The mean P-equivalency calculated from various sources, using treatments with phytase activity up to 500 U/kg, was 432±169 U = 1 g P (Hoppe and Schwarz, 1993). In several experiments conducted with growing pigs of 11-70 kg, microbial phytase supplementation at a level of 800-1,550 U/ kg diet improved bioavailability of P and Ca by 22-56% and 10-16%, respectively (Soares and Hughes, 1995).

With phytase supplementation of the diets, growing-finishing pigs and pregnant sows may need little or no supplementary inorganic phosphate. By using phytase supplementation researchers have achieved the same performance as with a control feed while reducing P excretion by 30%. The use of phytase in liquid feeding systems for pigs appears to be particularly interesting, because phytase may already be liberating phytate P as soon as the feed has been mixed. As with N, phase feeding can result in a reduction in P excretion. The use of a grower diet from 45-70 kg and a finishing diet from 70-106 kg instead of one fattening diet from 45-106 kg results in a 6% reduction in excretion of P. Table 8 shows an example of the reduction of P excretion through different options.

Table 8. Options in feeding program and P level of pig diets and P excretion

Feeds (%)	Situation in 1990	Phosphorus restriction	Phytase supple-mentation	Phytase+P restr.+selected feedstuffs
Starter feed	0.60	0.58	0.47	0.40
Grower feed	0.50	0.46	0.44	0.40
Finishing feed	0.46	0.42	0.40	0.35
Fattening feed	0.50	-	-	-
P excretion, kg/pig	0.83	0.70	0.61	0.49

(Jongbloed and Lenis, 1992)

Although the use of phytase is targeted specifically at the availability of P, the presence of the enzyme can have effects on the digestibility of other dietary components. Nitrogen digestibility was significantly higher in pigs receiving diets supplemented with phytase but without inorganic P supplementation (Nasi, 1990). A number of other investigations have indicated that there is often strong binding between phytic acid and protein. The effects of phytase on protein and amino acid digestibility may indicate that phytate-protein binding is to some extent cleaved by phytase activity or that the inhibitory activity of phytic acid on trypsin and pepsin is diminished. It was also suggested that the digestibility of a number of other mineral components, such as Ca and Mg, may be improved by the addition of phytase.

Ca and vitamin D in the diet strongly interact with P utilization and may affect phytate-P utilization in weanling pigs (Lei et al., 1994) as well as chicks (Edwards, 1993). A normal level of Ca in the pig diet greatly reduced the efficacy of supplemental phytase. Raising vitamin D in the diet partially offset this effect but did not produce a further improvement when the Ca level was low. The introduction of high levels of vitamin D into the diet may enhance Ca absorption, thereby removing the possible Ca-phytate interaction in the gut and resulting in improved phytate P availability.

The interaction between a phytase and a carbohydrase in relation to the digestibility of nutrients and the growth performance of individually kept growing pigs were investigated. The results showed that the combination of the carbohydrase with the phytase was not favorable for growth performance and digestibility of nutrients. Possibly a negative interaction between the two enzyme mixtures existed. It seemed that the carbohydrase reduced the analyzed phytase activity in the diets (Wenk et al., 1993).

(2) *Poultry*

In broiler chickens, phytase supplementation at a level of 1,000 U/kg diet increased the bioavailability of P and Ca by 60% and 26%, respectively (Simons et al., 1990). The beneficial effects of phytase supplementation were illustrated by Zyla and Korelski (1993). The performance of birds fed available P deficient diets was improved by the addition of phytase to the diets. The *in vitro* activity (i.e., ability to dephosphorylate phytate) was also demonstrated, confirming the proposed mode of action of this enzyme. The direct benefits of dietary phytase supplementation on bone mineralization have been shown by Farrell and Martin (cited by Annison and Choct, 1993) who reported that tibial ash deposition was enhanced in birds fed phytase supplemented diets. Simons and Versteegh (1993) summarized the results of several experiments conducted in Netherlands. A microbial phytase product from *Aspergillus niger* was added to broiler feed with a low inorganic P level. The availability of total P could be increased up to 70%. In comparison with feed with increased levels of inorganic feed phosphates, a significantly larger amount of the P consumed was absorbed. Improved utilization of P decreased its excretion by 40% or more. Growth and feed conversion ratios were comparable with feed to which inorganic feed phosphate was added. In layers the degradation of phytate and the absorption of P was slightly decreased by higher amounts of Ca in the diets (4.0% vs. 3.0% Ca in feed), nevertheless at both levels the efficacy of phytase addition was satisfactory. In broilers up to 500 units of phytase per kg feed, 250

Table 9. Effects of supplemental phytases on the productivity and P excretion of laying birds

Experiment	Level of NPP[1], %	Supplemental phytase, units	Performance index		
			Egg production	Feed/egg mass	P excretion
1	0.37	0	100	100	100
	0.37	500	102.2	99.6	88.5
	0.24	500	100.4	100.4	70.5
	0.12	500	100.4	100.4	59.0
2	0.27	0	100	100	100
	0.22	250	100.3	100.5	88.5
	0.16	250	101.4	98.6	67.3
	0.11	250	99.1	100.3	57.7
3	0.25 (Ca 4%)	0	100	100	-
	0.25 (Ca 4%)	300	103.1	99.8	-
	0.25 (Ca 3%)	0	102.1	96.1	-
	0.25 (Ca 3%)	300	104.5	91.6	-
	0.15 (Ca 4%)	0	97.6	130.4	-
	0.15 (Ca 4%)	300	97.2	101.6	-
	0.15 (Ca 3%)	0	100.6	89.9	-
	0.15 (Ca 3%)	300	101.5	95.0	-

[1] Nonphytate phosphorus

Table 10. Effects of supplemental phytase on the productivity and P excretion of broilers

Experiment	Level of NPP1, %		Supplemental	Performance index		
	Starter	Finisher	phytase, units	Gain	Feed/gain	P excretion
1	0.45	0.40	0	100	100	100
	0.34	0.31	600	101.0	98.7	76.2
	0.23	0.21	600	99.3	101.3	54.8
	0.13	0.12	600	96.7	103.3	40.5
2	0.45	0.35	0	100	100	100
	0.35	0.25	0	89.4	100.6	84.8
	0.25	0.15	0	60.5	108.7	51.5
	0.25	0.15	600, Phyt-A[2]	82.2	109.3	39.4
	0.25	0.15	600, Phyt-B[3]	78.9	109.3	45.5
3	0.45	0.35	0	100	100	-
	0.45	0.35	500	101.7	100	-
	0.35	0.25	0	90.5	102.8	-
	0.35	0.25	500	95.2	100.7	-
	0.25	0.15	0	63.1	109.7	-
	0.25	0.15	500	77.6	107.6	-

[1] Nonphytate phosphorus, [2] Crude phytase A (soup + cell), [3] Crude phytase B (soup)

units phytase was equivalent for P absorption with 0.5 g monocalcium phosphorus (MCP) P per kg feed. Addition of up to 300 units phytase per kg feed for laying hens resulted in a minimal equivalency of 0.3 g MCP P per 100 units phytase.

In a feeding trial with laying hens the effectiveness of microbial phytase in diets based on corn-soya and wheat-soya was tested (Peter and Jeroch, 1993). The supplement of phytase (500 U/kg diet) or inorganic P (0.1% of diet) had a positive effect on the performance of the corn-soya group but no effect on that of the wheat-soya group. The highest breaking strength of the egg shell was recorded with hens that received the phytase supplement in the corn-soya group. Mineralization of the tibia bone was also improved with phytase addition. Provided phytate P content and plant phytase activity are taken into account, it should be possible to mix layer diets which require a minimum amount of supplementary inorganic P with a 250 U phytase supplement (Um et al., 1999), or do not require supplementary inorganic P sources with a 500 U phytase supplement (Um and Paik, 1999).

Although microbial phytases have shown an ability to enhance P availability in many varied situations, several technical problems still exist that prevent the universal application of such enzymes. These include instability to gastric pH and a severe loss in activity at elevated, present day processing temperatures. Such difficulties present a challenge to producers of microbial phytases, which the tools of biotechnology can address (Power, 1993).

Tables 9 and 10 show a summary of the feeding trials conducted with microbial phytases in author's laboratory.

3. Protease

Proteases of bacterial origin have been shown to be effective in increasing the digestibility in pigs of the energy and crude protein and amino acids of potatoes. A combination of a protease with α-amylase significantly improved the digestibility of lysine and of methionine (Jost et al., 1993).

A fungal protease was added to a broiler diet containing bitter lupin (*L. mutabilis tarwi*). The alkaloids in the lupin did not impair digestibility or metabolizability of energy and N in the feed. The addition of protease did not improve the performance but had a slightly positive effect on the utilization of the N in the diet (Pfirter et al., 1993). Feathers are composed almost exclusively of a protein termed keratin. Keratin is particularly resistant to the digestive process of the alimentary canal, largely due to the presence in the protein of covalent chemical bonds termed disulphide bonds, which stabilize the molecular structure. Pathogenic fungi have been the source of most of the enzymes that break down keratin. Regulatory agencies have prohibited the large-scale propagation of these fungi. However, a recent breakthrough in feather digestion has been the development of a pre-treatment that breaks the disulphide bond thereby exposing the protein. A neutral protease is then added at a relatively low level (0.1% for 4 h) to bring about total solubilization of feathers (Lyons and Walsh, 1993).

Growth promoters

Growth-promoting compounds, such as antibiotics, β-agonists and recombinant somatotropin, increase the ability of the animal to utilize the available dietary protein. Some of the growth-promoting antibiotics have a protein-sparing effect which results in reduced excretion of total nitrogen.

The effects of different anabolic agents on N excretion are shown in table 11. The use of β-agonists and recombinant somatotropin induces an increase in muscle growth which increases both N retention and growth rate.

These two complementary responses have differing levels of effect on N excretion. Increased N retention by the animal results in approximately a 5% reduction in N excretion. The major response in pigs treated with porcine somatotropin (PST) is increased growth rate and reduced time to slaughter; both feed intake and maintenance N contribution are reduced, resulting in a reduction in N excretion of more than 30%. The use of bovine somatotropin (BST) to increase milk yield in the dairy cow has a similar beneficial environmental impact. Whilst overall N excretion is increased in BST-treated cows, as a result of the increased feed intake, the N excretion per liter of milk is reduced by 15%. Under a quota situation this would result in an overall decrease in N excretion.

Table 11. Effects of anabolic agents on nitrogen excretion of livestock

Anabolic Agents	Animal Species	Reduction of N Excretion,%
β-agonist (Clenbuterol)	Pigs, 60-105 kg	14
rPST	Pigs, 48-84 kg	32
rPST	Pigs, 65-102 kg	35
β-agonist	Cattle (bull)	7.4
β-agonist	Calves	7.0
rBST	Dairy cows	15

(Williams, 1995)

Chelated minerals

Some micromineral supplements are produced in the form of protected forms. Metal-amino acid chelate (Ashmead, 1992), metal-proteinate and metal-polysaccharide complex are protected minerals. The protected minerals may be more available and not react with digesta due to both their chemical (electrically neutral, ligand and metal make up) and physical structures (size and ligand source). If this is the case, we could use less to achieve the same result. This would be excellent as potentially it would save world resources and reduce pollution (Lowe, 1993).

High levels of copper sulphate have been widely used as growth promotants in pigs and broilers. Copper polysaccharide complex (sequestered Cu) at a level of 62.5 ppm of Cu was as effective as 200 ppm Cu in the form of copper sulphate in weanling pigs and broilers (Paik and Kim, 1993). The performance enhancing effect of methionine-Cu complex at a level of 100 ppm Cu was greater than that of copper sulphate at a level of 200 ppm Cu in broilers. The excretion of copper was significantly less in the methionine-Cu treatment than in the copper sulphate treatment (Min et al., 1993, 1994).

Tables 12 and 13 show a summary of feeding trials conducted in the author's laboratory to test effects of chelated copper and zinc products.

Deodorases

Ammonia release from animal manure should be controlled to avoid air pollution and conserve N in the manure for use as fertilizer. The smell of pig slurry has four times the intensity of cattle, broiler and poultry manure (Pain, 1990). In terms of odour control, ammonia reduction may only play a contributory role since Schaefer (1977) correlated odour intensity with the concentrations of volatile fatty acids (C2-C5), phenol, *p*-cresol, indole, skatole and ammonia; the highest correlations were obtained with *p*-cresol. Conservation of N in manure is important because P or K usually limit use of poultry manure for crop production and other sources of N are needed when the manure application is limited to needs for fertilizer elements. Ammonia release from manure can be limited by using additives, by

Table 12. Effects of supplementary copper chelates

Experiment	Animals	Source and level of Cu, ppm	Difference from the control, % Gain	Feed intake	Feed/gain
1	Broilers	$CuSO_4$, 200	3.8	-0.2	-4.0
		SQM-Cu, 63.5	2.6	0.6	-3.5
		SQM-Cu, 127	3.5	-0.2	-4.0
		SQM-Cu, 191	3.8	1.7	-2.0
2	Pigs	$CuSO_4$, 200	6.4	4.0	-3.1
		SQM-Cu, 63.5	4.1	0.9	-3.5
		SQM-Cu, 127	7.5	3.1	-4.8
3	Broilers	$CuSO_4$, 200	-0.9	-0.6	0.0
		Met-Cu, 200	2.5	-0.5	-3.3
		SA-Cu, 200	1.1	-0.3	-1.6
4	Broilers	$CuSO_4$, 200	0.3	-2.0	-2.2
		Met-Cu, 200	2.1	-0.5	-2.2
		FM-Cu, 200	0.2	-1.4	-1.7
	Rats	$CuSO_4$, 200	-7.0	-5.3	1.9
		Met-Cu, 200	-7.5	-3.7	3.8
		FM-Cu, 200	-6.3	-3.0	3.4
5	Broilers	$CuSO_4$, 200	4.3	-1.4	-5.7
		Met-Cu, 200	6.3	3.1	-3.2
		Met-Cu-Zn-Fe, 200	2.2	0.0	-1.9
		FM-Cu, 200	2.0	1.2	-0.6
6	Pigs	$CuSO_4$, 200	7.5	-5.9	-3.8
		Met-Cu, 100	10.1	0.4	-1.9
		Met-Cu, 200	8.2	9.6	0.0
7	Pigs	$CuSO_4$, 200	5.5	2.6	-4.6
		Met-Cu, 200	17.8	-7.2	-9.8
		FM-Cu, 200	0.0	-16.4	-4.6

Footnotes: $CuSO_4$; $CuSO_4 \cdot 5H_2O$, SQM-Cu; sequestered mineral copper, Met-Cu; methionine-Cu chelate, SA-Cu; sodium alginate-Cu complex, FM-Cu; fish meal digest-Cu complex, Met-Cu-Zn-Fe; methionine-Cu-Zn-Fe complex.

Table 13. Effects of supplementary zinc oxide and chelates in weanling pigs

Exp.	Source and level of Zn, ppm	Difference from the control (ZnO, 100 ppm), % Gain	Feed intake	Feed/ gain
1	ZnO, 200	8.3	9.6	0.5
	Met-Zn, 100	3.0	3.0	0.5
	Met-Zn, 200	18.8	15.1	-3.8
2	ZnO, 1000	10.0	9.2	-1.4
	ZnO, 2000	11.0	12.8	1.4
	Met-Zn, 100	15.6	15.9	0.0
	Met-Zn, 1000	6.5	12.2	7.6
	Met-Zn, 2000	-2.0	9.4	11.7

Footnotes: Met-Zn; methionine-Zn chelate.

drying and by acidic conditions.

Research into minimizing air pollution from animal wastes is continuing and taking many different paths. In the Netherlands, for example, they have identified a microorganism (aerobic denitrifier) which, under aerobic conditions, converts the nitrogen of ammonia and other nitrogen containing compounds into nitrogen gas. Nitrogen gas can be released into the atmosphere without causing pollution problems. Adding such bacteria to manure would reduce the emission of ammonia and reduce the nitrogen content of the manure. They are looking at the possibility of adding these bacteria to the feed (Holthuijzen, 1993).

The ammonia-binding properties of the *Yucca* extract have been widely studied. The earlier reports on the action of a *Yucca* extract to prevent the accumulation of ammonia erroneously attributed its action to an inhibition of urease by its component three steroid saponins, i.e. sarsapogenin, smilagenin and hecogenin. But Headon et al. (1991) reported that the *Yucca* extract does not inhibit urease activity and that saponin-free De-Odorase had an ammonia-binding capacity similar to that of the unfractionated De-Odorase. Recent work by Headon and Power (unpublished, cited by Leek, 1993) demonstrated that the binding agents in the *Yucca* extract are glycocomponents. Because the ammonia-binding action starts to decline slowly from the fourth day onwards, atmospheric ammonia levels within houses can be significantly reduced by including this product in the diet.

Zeolite products have been used at a level of 1 to 2% of the diet to improve pelleting quality. It is also believed that zeolite may improve the litter condition and environment of the barn. Due to a high ion-exchange capacity, it is expected that zeolite may bind ammonium ion in the litter (Moon et al., 1991).

However, dietary supplementation of zeolite or top dressing of zeolite on the broiler litter did not significantly influence level of ammonia produced from the broiler litter (Blair and Jacob, unpublished).

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