Opportunities in the feed milling industry by combining new generation xylanase and phytase enzymes

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AB Vista Feed Ingredients
Contents

- Introduction to AB Vista
- Phytase superdosing
- New insight in mode of action of NSP enzymes
- Using enzymes together
• AB Vista is an integrated supplier of new generation micro-ingredients for animal feeds.

• AB Vista was founded in 2004 and operates under the AB Agri division of Associated British Foods PLC. Its headquarters are in Marlborough, UK.
Global phytase market share

- Danisco, 32%
- DSM, 26%
- AB Vista, 15%
- BASF, 8%
- Chinese, 9%
- Enzyvia/ Optiphos, 5%
- Other, 5%
- Home mix

<table>
<thead>
<tr>
<th></th>
<th>Poultry</th>
<th>Swine</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Estimated market value</td>
<td>$250m</td>
<td>$150m</td>
<td>$400m</td>
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<td></td>
<td>€184m</td>
<td>€110m</td>
<td>€294m</td>
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Internal data, AB Vista 2012
What is phytate?

Phytate, which is present in many plant-based feedstuffs, is the main phosphorus (P) store in plants (Cosgrove, 1980).

Phytate is important as a possible source of P for poultry and swine. BUT monogastrics are less efficient at hydrolysing phytate which means that the phytate P is unavailable to them for absorption.

Phytate can bind with other minerals and proteins which makes them unavailable as well.

More information about phytate can be found at www.phytate.info
AvP release determination

Tibia Ash (mg)

Quantum Blue 2012 Broilers USA 111
Quantum® Blue delivers more phosphorus, more consistently

- 500 FTU/Kg Quantum Blue® gives you at least 0.15% av P*
- Quantum® Blue is markedly superior

Available P release calculated based on 18 days old broiler performance and bone parameters

*based on 90% confidence limits
Phytate

Anti-nutritional effects
Phytate can decrease feed digestibility by forming insoluble complexes with nutrients and digestive enzymes or reduce nutrient solubility.
Phytate reduces amylase activity in intestinal mucosa

Liu et al., 2008
IP6 is not the only problem
Phytase has to get rid of IP5→IP2 as well

Figure 6. Time course of phytic acid hydrolysis by E. coli phytase (Phyzyme XP, Danisco A/S, Brabrand, Denmark; 0.08 phytase unit/mL) and inhibition of porcine pepsin catalyzed azurine cross-linked casein hydrolysis by the hydrolyzates. Phytic acid hydrolysis was performed at 37°C; pepsin activity assay was carried out at 40°C. Each data point is an average of 2 separate experiments.
AB Vista definition of superdosing:

Supplementing high doses of phytase to maximize phytate degradation rather than P release.

‘Superdosing’ will depend on:

1. Species and category
2. Matrix and current feeding program

Caveats: What is a high dose?

Depends on biologically relevant characteristics of that enzyme

1. Enzyme activity at pH 3
2. Pepsin resistance
3. Low Km – high activity at low phytate concentrations
4. Rapid release
Equilibria to consider

Dietary Phytate $\rightarrow$ Insoluble Phytate $\rightarrow$ Soluble Phytate $\rightarrow$ IP3

Nuisance factor $\leftrightarrow$ ??? $\leftrightarrow$ kM $\leftrightarrow$ Substrate

Critical phytate concentration

High kM

Low kM

0.0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1.0

0 5 10 15 20 25

Phytate concentration

Phytate

Substrate
Quantum® Blue is optimised for phytate destruction

- Delivers high and consistent activity at gastric pH
- Reduces phytate even at low levels

Quantum® Blue reduces phytate even at low levels

Quantum® Blue gives 40% higher activity than Quantum® at lower phytate concentrations

Enhanced Quantum Blue
Quantum
Wild-type E.coli

In feed assay

Typical phytate level in avian gut

Soluble Phytate P Concentration (%)

Activity (FTU)
Quantum® Blue Superdosing – 4 point improvement in FCR

Composite analysis of 6 trials:
Body weight corrected FCR of broilers from d 0 to 35/42 (n = 35)

Body weight corrected FCR

PC  PC+DCP  PC+500 QB  NC  NC + 500 QB  NC + 1000 QB  NC + 1500 QB

y = -0.0255x + 1.638
R² = 0.9538

4 points in FCR currently worth €5 per tonne of feed
Extra-phosphoric effects of phytase

Broiler gizzard phytate, phytate ester and inositol concentration (d21)
Superdosing Quantum Blue decreased phytate and increased inositol concentration
Part of the superdosing response may be associated with inositol provision as well as phytate destruction

<table>
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<tr>
<th>Item, µmol/g</th>
<th>BW gain, kg</th>
<th>P =</th>
<th>FCR</th>
<th>P =</th>
<th>Tibia ash, g</th>
<th>P =</th>
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</thead>
<tbody>
<tr>
<td>Phytate</td>
<td>-0.37</td>
<td>***</td>
<td>0.46</td>
<td>***</td>
<td>-0.21</td>
<td>NS</td>
</tr>
<tr>
<td>Inositol</td>
<td>0.46</td>
<td>***</td>
<td>-0.57</td>
<td>***</td>
<td>0.19</td>
<td>NS</td>
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Walk et al., 2014
Inositol interacts with phytase. It is likely part of the superdosing effect.

FCR
Inositol* Phytase interaction p < 0.0143
LSD = 0.017
Optimised for maximum phytate destruction, Quantum Blue unlocks more value for your business than any other phytase.

- Greater phosphorus release
- Unrivalled intrinsic thermostability
- Proven additional feed efficiency value
Xylanase Mode of Action

1. Opens up feedstuff cell walls (insoluble fibre)
   - Corn-based diets
2. Reduces intestinal viscosity (soluble fibre)
   - Wheat and barley diets
3. Produces oligosaccharides (prebiotics)
Cell wall hypothesis

Samples taken from terminal ileum

no enzyme

xylanase added
Is cell wall hydrolysis realistic in the intestine?

- Time and pH

Fig. 2. Visualization of the degradation of aleurone arabinoxylan cages present in milled wheat after incubation with Ronozyme WX xylanase (1 g of enzyme/kg of diet) at 30°C. A, Milled maize as seen under a microscope; B–E, close-up of a cell wall structure containing ferulic acid, which fluoresces with an intense blue-green fluorescence. B and C present cell walls treated with a buffer solution lacking enzymes, and D and E present cell walls before and after addition of the xylanase, leading to breakdown and disappearance of the cell wall architecture.

Lei et al 2013
Xylanase Mode of Action

1. Opens up feedstuff cell walls (insoluble fibre)
   • Corn-based diets

2. Reduces intestinal viscosity (soluble fibre)
   • Wheat and barley diets

3. Produces oligosaccharides (prebiotics)
VFA production stimulates PYY release

1. Xylanases breakdown cell walls

2. Release prebiotic xylo-oligosaccharides

3. Fermented by bacterial species

4. Support growth of beneficial gut microflora and suppress growth of pathogenic bacteria

P-YY release → Gizzard → delays gastric emptying
Differences exist between xylanases in end products
Xylo-oligomer efficacy?

FCR 0-21

Red bars sig diff from control

Wheat

Con 0.3 AXOS 0.6 AXOS 1.2 AXOS 2.5 AXOS 5.0 AXOS 5.0 FOS 2 U Xyl 1.0 AXOS 2.5 AXOS

Corn

5% Turkeys LSD

Courtin et al 2008
Econase® XT increases Peptide-YY release

Plasma PYY pg/ml

Positive Control

0 8000 16000

0.00 5.00 10.00 15.00 20.00 25.00 30.00 35.00 40.00 45.00 50.00

cd c a

Negative Control – 100kcal

0 8000 16000

0

8000

16000

b

cd

Masey O’Neill, 2012
PYY release effects improved digestion?

Samples taken from terminal ileum

Improvement of digestion based on bigger gizzard activity and delay on gizzard emptying, not on enzyme directly opening cell walls!
Why should there be a benefit in combination

• Phytase improves Energy and amino acid digestibility
  – Leaves less for the xylanase to work on
  – Empirically the matrices are not additive
But ..... 

• Increased gastric residence time increases time available for phytase to work

• Better P matrix (more security) and better superdosing response

• SD = 4 points in FCR
• SD + Econase XT = 6 points

• Still value in the combination
SD combination with XT?

BWcFCR of broilers > 35 d fed QB with or without Econase XT
N = 70; 6 trials conducted in Mexico (1), Brazil (1), India (1), UK (2) and Germany (1)
Conclusions?

• Each enzyme has different substrate and hence activity in the animal

• BUT there is an overlap in the mechanisms by which they elicit improvements in digestibility

• Combination will result in better performance but reduced individual matrices

\[1 + 1 \not= 2, \quad \sim 1.5-1.6\]
Thank you!
For more information visit:
www.abvista.com or www.phytate.info