

An innovative phytase for efficient phytate degradation

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In grain based feedingstuffs, around 70-80% of the phosphorus (P) is bound in phytate (inositol hexaphosphate = IP6), which is not efficiently degraded within the intestinal tract of monogastric animals. To make phytate-bound P available for the animal, phytase enzymes, produced by microbial fermentation, are commonly used in feeds.

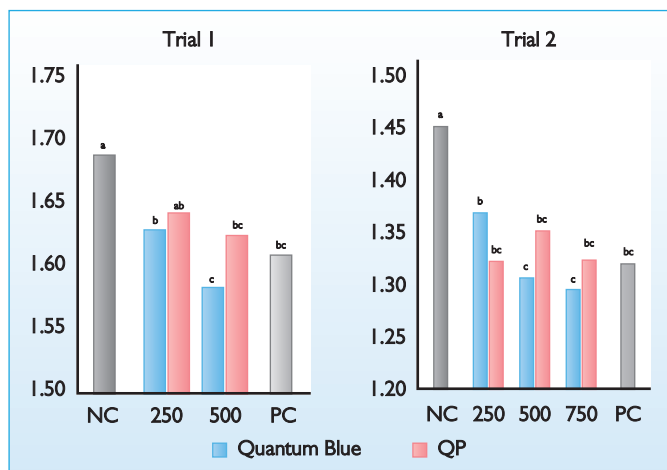
The phytases on the market differ in their characteristics and, as such, also differ in their efficacy within the animal intestinal tract.

Effect on digestibility

The first approach to evaluating the efficiency of a phytase is to study its effect on P digestibility in low P diets when compared to other phytase products with well known levels of efficacy.

For the phytase product Quantum (QP), an *E. coli* derived, enhanced phytase with improved thermo- and gastric-stability and efficacy, there is a huge database of published trials which acts as a good reference against which to test new phytases.

Fig. 1. Effect of two *E. coli* derived phytases (Quantum (QP) and Quantum Blue) on the feed conversion ratio (FCR) of broilers fed positive control (PC), negative control (NC, with reduced P and Ca) and NC with 250, 500 or 750U/kg phytase diets.



In January 2012, Quantum Blue, a more advanced version of Quantum, was launched on to the international phytase market. Quantum Blue has further improvements in several enzymological characteristics compared with QP, including thermo-stability and protease stability. Several trials have been carried out to compare the biological efficacy of these two products.

Two trials have been completed where broilers were fed negative control (NC) diets with reduced P (4.7g/kg in trial 1; 4.5g/kg in trial 2) and calcium (Ca) levels (5.7g/kg in trial 1 and 7.0g/kg in trial 2). Ileal P digestibility measured after 37 (trial 1) and 22 (trial 2) days was improved on addition of Quantum Blue, and the effects were more distinct than those measured for QP.

In trial 1 ileal P digestibility of the control birds was 48.4% which was increased by 11.1 and 12.2 percentage points when diets were supplemented with Quantum Blue at 250 and 500FTU/kg, respectively, whereas QP at the same application rates led to an improvement of only 6.7 and 6.2 percentage points.

In trial 2, a similar superior effect of Quantum Blue compared to QP was noted. Ileal P digestibility of the

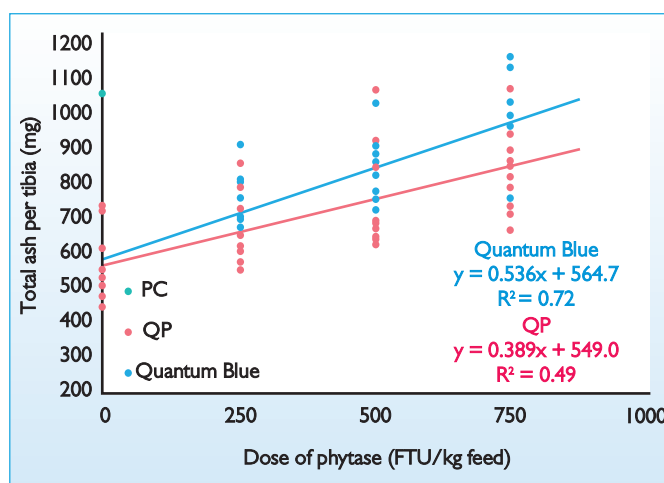


Fig. 2. Bone mineralisation (mg total ash per tibia) in broilers as influenced by dietary P and Ca levels or phytase addition.

control birds in this trial was 49.0% and increased by 18.4 to 18.5 percentage points when diets were supplemented with 500 and 750FTU/kg Quantum Blue, respectively, whereas the improvement with QP at the same dose rates were 9.0 and 16.3 percentage points.

There are clear limitations in evaluating phytase efficiency by digestibility measurements alone, even though ileal digestibility compares well with P utilisation in poultry as it excludes the error of renal excretion.

However, in digestibility experiments there is not enough time for adequate adaptation in P absorption based on the P level of the diet, and the relevance for bone mineralisation and growth are not measured.

In particular, comparison with a positive control (PC) diet, which ordinarily has high P and Ca levels, is not really possible due to significant adaptations to the diet which influence the digestibility of Ca and P markedly compared with the NC.

Due to this, it is not possible to draw a firm conclusion on the optimal feed mineral content or phytase dosage at different P, Ca and sodium (Na) levels based on digestibility measurements alone. Therefore, growth parameters as well as bone mineralisation should be evaluated.

For both broiler trials mentioned

above, results in Fig. 1 show the feed conversion ratio in broilers fed positive control (PC), negative control (NC) and the NC diet containing either QP or Quantum Blue.

While 500FTU/kg feed of QP in trial 1 and 2 improved feed conversion ratio (FCR) by 7.0 and 10.0 points, respectively, broilers fed the same level of Quantum Blue achieved an improvement in FCR of 11.0 and 14.0 points, respectively.

Improved daily gain

Both phytases also improved daily gain, and the differences between the phytase products were similar to those noted for FCR.

Compared to those broilers fed the PC (P level 2.0-2.9g/kg higher and Ca level 2.0-4.0g higher than in the NC diets), in trial 1 the 500FTU/kg and in trial 2 the 750FTU/kg feed Quantum Blue additions achieved similar daily gain, whereas QP fed birds did not achieve the PC performance level.

In trial 2, bone mineralisation also reflected the performance effect of both phytases, with Quantum Blue again being superior to QP (Fig. 2).

Although animals in the phytase treatments received diets with lower P and Ca levels than the PC,

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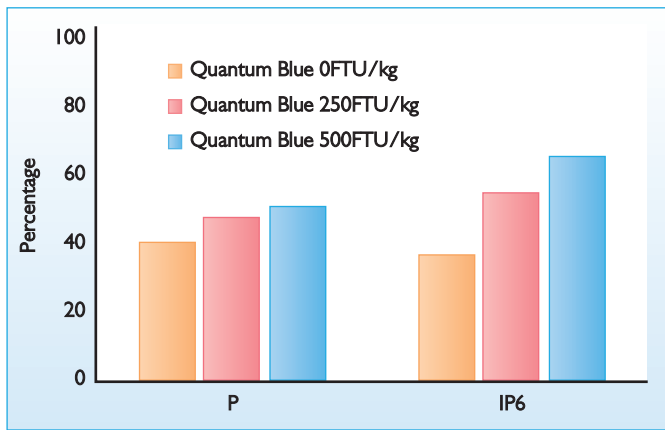


Fig. 3. Effect of Quantum Blue inclusion rate on P digestibility and phytate (IP6) degradation at the end of the small intestine in broilers.

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 tibia ash and P contents of birds fed Quantum Blue at 750FTU/kg feed were not significantly different to those of the PC birds, whereas QP fed broilers failed to achieve the mineralisation levels of the PC birds. In this trial, ileal digestibility alone suggested that there is no improvement with an increase in Quantum Blue dosage from 500 to 750FTU/kg. However, growth and bone mineralisation data differ in this regard as there was a clear improvement with an increase of the phytase dosage to 750FTU/kg.

to P release but additionally improves availability of those minerals (cations) and proteins associated with the phytate. This effect of binding cations is mainly relevant for IP6 and IP5 molecules, whereas less phosphorylated phytate degradation products such as IP4-IP1 no longer have significant chelating effects. Therefore, further degradation of IP4 to IP1 lead to a further P release but does not substantially change mineral or protein availability. Phytases that preferentially

degrade IP6 and IP5 are thus better at releasing bound minerals or proteins. Evaluation of the differences in the rate of IP6 degradation within the upper intestinal tract therefore could be a good tool in the select of next-generation phytases. The effect of Quantum Blue on IP6 degradation was analysed in a trial with broilers fed low P and Ca diets (starter: 4.6g/kg P, 5.9g/kg Ca; grower 4.2g/kg P, 6.0g Ca).

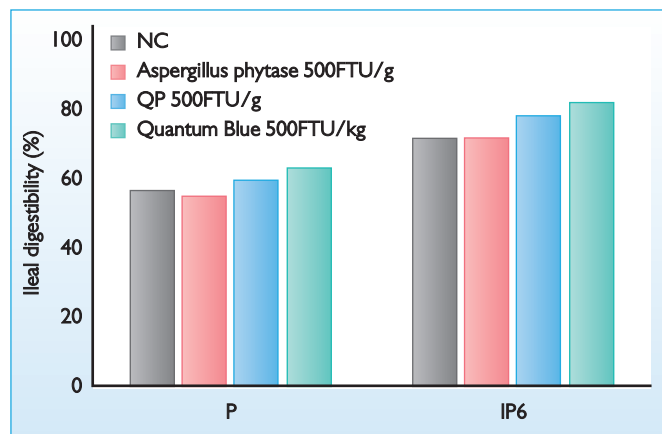
Results shown in Fig. 3 demonstrate that Quantum Blue supplemented at 250 and 500FTU/kg significantly improved ileal P digestibility by 7.0 and 11.1 percentage points, respectively, compared to the control group. Interestingly, the effects on IP6 degradation were more pronounced, with 18.9 and 27.9 percentage points improvement at the end of the small intestine. In another trial, several different phytases were evaluated for their effects on IP6 degradation. The aim of this trial was to test if an *Aspergillus* derived 3-phytase could degrade phytate as efficiently as 6-phytases of *E. coli* origin. Additionally, it was of interest to determine if slight changes in the characteristics of the phytase, as is evident between Quantum Blue compared to QP, have any impact

on IP6 degradation in broilers. In this trial, 600 16-day-old broilers were allocated to 40 floor pens (10 pens per diet). The low P corn and soybean meal-based diets, without any mineral P source added (4.5g/kg P, 7.0g/kg Ca), were fed without or with a phytases (*Aspergillus* phytase, QP or Quantum Blue), all at a level of 500FTU/kg. At 25 days of age, digesta was collected from the bird intestine and analysed for ileal P digestibility and IP6 degradation (Fig. 4). IP6 degradation tended to be higher with the *E.coli* derived 6-phytases (QP, 79.0%; Quantum Blue, 82.0%) compared to the *Aspergillus* derived 3-phytase (74.0%) or the control (74.0%). Accordingly, P net absorption at the end of the ileum tended to be higher with the 6-phytases (QP: 60.0%; QP2: 64.0%) compared to the 3-phytase (56.0%) or the control (57.0%). Even though there were no significant differences compared with the control, there was interestingly no numerical effect of the 3-phytase on IP6 degradation at the end of the small intestine. In contrast, QP and Quantum Blue increased IP6 degradation by 5.0 and 8.0 percentage points.

Influence animal growth

It is clear that improvements in phytases can lead to more efficient phytate degradation, which can positively influence animal growth performance and bone mineralisation. The degradation path and rate of phytate destruction seems to have an important influence on the overall efficacy of phytases and should be taken into account in phytase development and comparison. Phytate (IP6, with a total of six attached phosphate units) complexes with minerals and proteins in the animal intestinal tract. Phytate hydrolysis therefore leads not only

Fig. 4. Effect of different phytases at 500FTU/kg on P digestibility and phytate (IP6) degradation at the end of the small intestine in broilers.



Conclusion

It is clear that phytases have different phytate degradation patterns in the intestinal tract, and this influences their efficiency in poultry. Hence, phytases clearly should not be compared on the basis of enzyme activity level determined in the laboratory. Even small changes in the characteristics of a phytase can improve the overall efficiency in the animal, as demonstrated by Quantum Blue phytase compared to QP. Recently published results suggest that greater emphasis should be placed on rapid IP6-phytate destruction as this may be a key attribute of a potent feed phytase. ■