

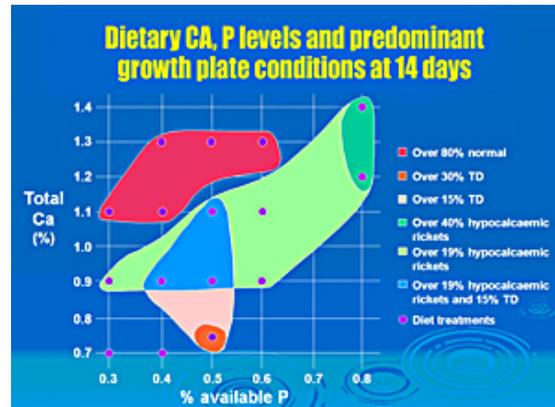
More work still needs to be done for phytases to achieve their maximum potential in animal feed.

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Impact of dietary total calcium and available phosphorous on bone quality in broilers.

With numerous phytase products on the market, but differing results when applied on farm, the need for the more effective testing and evaluation of phytase efficacy in animal feed was raised at the first [International Phytase Summit](#).

The difficulty was highlighted by Schothorst Feed Research's Dr Jan Dirk van der Klis who commented: "Results must be communicated to nutritionists so that they can choose the most promising products. But all companies are using the same arguments to sell their products, so without a reliable, standard assay, it is very hard for nutritionists to make that decision." He continued that, any in vitro methodology must be accurate, cheap and quick. However, he also highlighted the fact that assays can only really measure degradability, not actual digestibility, due to the large number of interactions and variability that exist within the bird, such as the differing response to phytase addition at various dietary calcium levels.

In clarifying the way forward, he stated: "It seems that you can develop in vitro methods to evaluate phytases as a tool to discriminate efficacy, but in order to predict what actually happens in the animal, it needs the measurements to be related to in vitro data."

According to Dr Ralph Greiner of the [Max Rubner Institute](#), the development of a reliable in vitro assay for phytase efficacy is further complicated by the fact that neither the phytase substrate nor the first degradation product can be measured by spectroscopy.

Changing environment

The main problem with the data is that phytases showing the same level of activity measured at pH 5.5 (the pH specified by the current standard [AOAC](#) assay methodology) can have dramatically differing activities at lower pH values.

Dr Mike Bedford of [AB Vista](#) commented that the variation in phytase activity with pH might not only help explain differences in bird performance, but also lead to the development of more effective in vitro assays for screening phytases for use in poultry diets. There was good evidence, he said, that the majority of phytase activity takes place in the stomach and gizzard, with measurements carried out at perhaps 2.5-3.0 likely to be more relevant to in vivo activity.

Bedford went on to present data from a broiler trial using five commercially available phytase enzymes at two different dose rates. In vitro phytase activity was measured for all diets at pH 2.5, 3.0, 3.5, 4.0 and 5.5, and then correlated to in vivo results. The best correlations were achieved when using in vitro activity measured at pH 3.0, both for weight gain and bone composition.

In comparison, phytase activity measured in vitro at pH 5.5 by the standard assay method had little correlation to actual phytase performance in broilers. As a result, Bedford suggested moving toward a new standard for bio-efficacy measurement of phytase in poultry diets, at pH 3.0, keeping the current AOAC pH 5.5 as a consistent and reliable tool for quality control purposes.

"The current assay measured at pH 5.5 is giving value in indicating the performance of the animal," Bedford told delegates. "It's quite clear that pH 3.0 gives the best results in broilers – the bird is telling you that if you want to predict bio-efficacy, use pH 3.0."

Accurate determination of phosphorous, calcium

Another message to come out of the summit was that the extensive use of phytase enzymes in pig and poultry diets has raised the need for more accurate determination of phosphorous and calcium requirements.

Pig and poultry requirements in the US are predominantly stated as "available" phosphorous, yet in Europe the dominant terminology is "digestible" phosphorous. Taking into account the number of factors affecting phosphorous digestibility, such as feedstuff variation, genetics, age, weight and calcium-to-phosphorous ratio, comparison between different studies is difficult.

This sentiment was echoed by professor Roselina Angel, [University of Maryland](#), who highlighted some important discrepancies in current [National Research Council](#) recommendations for poultry. Measures of available phosphorous and non-phytate phosphorous appeared to be (incorrectly) used interchangeably,

and calcium-to-phosphorous ratios were based on comparing total calcium, not available calcium, to available phosphorous.

“If we do not know the requirements, we do not know how to properly use phytase,” she said. “The current recommendation using total calcium is for a ratio of 2:1, but for available-calcium-to-available-phosphorous in commercial, low-calcium diets the value is more likely to be around 2.4:1.”

Professor Angel also called for a move toward predictive modeling of requirements to keep ahead of the rapid rate of genetic progress being made in poultry.

Assumptions

The inadequacy of existing requirements was confirmed by professor Colin Whitehead of the [Roslin Institute](#) who presented trial data showing that calcium-to-phosphorous ratios producing highest bone quality were not the same as current NRC recommendations. He also discussed the impact of vitamin D in phosphorous and calcium homeostasis, suggesting that limits for vitamin D inclusion should be revised upward.

“We currently make a lot of assumptions,” said associate professor Todd Applegate of [Purdue University](#), highlighting the large number of interactions that affect mineral nutrition that are not accounted for in current recommendations. He pointed to the significant decrease in overall efficiency of uptake in vivo when sufficiency of mineral supply is approached, yet requirement assays are typically carried out with animals in a deficient state.

In terms of finding a practical solution, associate professor Hans Stein of the [University of Illinois](#) presented results demonstrating the value of standard total tract digestibility as a measure of phosphorous requirements in pgs that was remarkably consistent across different studies. Subsequent trials showed that diets could be effectively formulated using an STTD value for phosphorous without affecting animal performance.

Broader recommendations needed

There was also general agreement that any new recommendations should go beyond phosphorous and calcium. Delegates were reminded that phytase use releases a range of bound cations, all of which should be taken into account during diet formulation.

In the Netherlands, the impact of phytase use on copper and zinc availability was already being investigated to help reduce pollution. Several regions of the country have copper and zinc concentrations in the water that exceed permitted levels.

The consensus among delegates was that some sort of working group was needed to help achieve standardization in terminologies and methodologies. The importance of quantifying interactions – particularly between calcium, phosphorous and vitamin D – was also noted, along with the possible need to adjust requirements based on breed, age, weight and feed intake.

The summit was held in September 2010 and hosted by AB Vista, Massey University, and the universities of Maryland and Sydney.