The nutritional contribution of probiotics in broiler feed formulations

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One of the major advances achieved in poultry production is genetic development of birds. Genetic selection has produced birds with a tremendous growth potential. In a recent study the increase, from 1978 to 2005, in final body weight (56 days) of broilers has been calculated at 132%.

This huge growth potential increase has greatly modified nutritional needs; therefore, nutritionists nowadays must be much more careful when formulating feeds for these new breeds. It is not enough to look at the nutritional composition of the raw materials used in the formula, digestibility gains more importance. The interaction of the raw material with the gastrointestinal tract becomes crucial.

Raw material	EMA (kcal/kg)	Digestibility PB (%)	MAX* inclusion (%)
Corn	3280	85	Free
Wheat	3185	85	25
Lupins	1950	87	5
Oats	2500	76	5
Rapeseed meal	1700	80	5
DDGS (Corn)	2310	68	1

Table 1. Raw material nutritional content and limitations (FEDNA) (*for starter diets).

Sustainability challenge

The access to alternative raw materials is helping to achieve this century's sustainability challenge, but at the same time, some of those materials lack information about their utilisation by the birds' digestive system, or are known to have lower digestibilities or inclusion limitations.

Moreover, the nutrient content of these feedstuffs is lower compared to the traditionally used ingredients. In any case, the use of these alternatives is very attractive as it eases formulation, relaxes dependencies on certain raw materials and lowers costs. The supplementation with additives capable of unlocking the full potential of these ingredients is eventually necessary.

These products must work at intestinal level modifying viscosity, enhancing gut epithelium structure, increasing absorptive surface and improving digestibility, etc. In other words, the objective is to obtain better gut health. In this sense, a new range of natural growth promoters comes to occupy the niche created by the new challenge.

Traditionally this area has been occupied by antibiotic growth promoters (AGP). Currently this group of additives is under the spotlight as suspected of creating drug resistant bugs. In a 2014 report, FAO estimated in 25,000 human deaths per year due to these pathogens resistant to antibiotics in Europe, the cost to society generated by these conditions was estimated as €1,500 million per year.

For this reason and the suspicion of abusive use in some cases, these antibiotic growth promoters, are being phased out of the market on all continents.

Recently Wal-Mart US publically asked meat suppliers to reduce the use of antibiotics in animals. Tyson Foods said in April that it plans to eliminate the use of human antibiotics in its chicken flocks and McDonald's Corp also declared in March that its US restaurants would gradually stop buying chickens raised with human antibiotics.

Within the new range of natural growth promoters, we can find different categories: short and medium chain organic acids, prebiotics, probiotics and essential oils.

Importance of probiotics

Probiotics are gaining attention from animal nutrition professionals across the globe due to the combination of the various benefits they exhibit.

They can inhibit the growth of pathogens not generating resistance to antibiotics (through disruption of quorum sensing mechanisms and synthesis of bacteriocines, lactic acid, etc), promote beneficial flora, play an important role in the development and structure of the intestinal epithelium and positively modulate the immune response.

Probiotics, like most bacteria, use their own enzymes to metabolise nutrients found in the medium. For this purpose, they synthesise and excrete a variety of specialised enzymes capable of degrading complex compounds such as carbohydrates, proteins and fats.

The products of this metabolism are used as a source of energy, or as structural components in the bacteria growth and reproduction. The enzymatic activity of each species varies, having different genera and species varied efficiencies. This activity is also related to the growth rate of each species.

Bacillus amyloliquefaciens is a great example of a probiotic with a high enzyme activity. Industry has benefited from Bacillus amyloliquefaciens' elevated enzyme production capacity in various fields.

As examples, paper and sugar industrial processes, waste treatment, detergents and drugs production and its lipases excretion is key to the dairy products transformation industry. The industrial use of these enzymes is attractive due to its high

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Table 2. Effect of supplementing a probiotic on broiler digestibility (Sánchez, 2006).

	Digestibility (%)		
	Protein	Fat	Starch
Control	70.8	75.4	84.2
Ecobiol (10^6 UFC/g)	76.5	80.9	87.4
P	P<0.0001	P<0.02	P<0.05

Table 3. Experimental design ([a]reduced ME -60kcal/kg and digs. AA -2.3%; [b]reduced ME -120kcal/kg and digs. AA, -4.6%; [c]reduced ME -180kcal/kg and digs. AA -6.9%).

Treatments	Diets	Ecobiol (kg/t)
T1	PC: Positive control	-
T2	NC1: Negative control 1[a]	-
T3	NC2: Negative control 2[b]	-
T4	NC3: Negative control 3[c]	-
T5	PC: Positive control	1.00
T6	NC1: Negative control 1[a]	1.00
T7	NC2: Negative control 2[b]	1.00
T8	NC3: Negative control 3[c]	1.00

Treatment			Final body weight (g)	Body weight gain (g)	Feed intake (g)	Feed conversion rate (g)	Livability (%)
Group	Nutrient reduction	Ecobiol (kg/MT)					
T1		0.0	2658	2615	4021	1.538^{d}	96.7
T2	-60 kcal/kg; -2.3% aa	0.0	2617	2574	4015	1.560^{c}	96.7
T5		1.0	2659	2617	4001	1.529^{d}	100.0
T6	-60 kcal/kg; -2.3% aa	1.0	2671	2628	4036	1.536^{d}	98.3
SEM			34.350	49.963	0.008	0.010	1.790
Source		df	P values				
Treatment		7	0.3976	0.7986	0.0001	0.0003	0.5534
Diet (A)		3	0.1170	0.3321	0.0001	0.0001	0.3017
Ecobiol (B)		1	0.5297	0.8634	0.0350	0.0159	0.3302
A x B		3	0.8426	0.9752	0.5886	0.4666	0.7557

Table 4. Effect of Ecobiol in productive parameters (0-35 days of age). ^{a,b,c,d,e}***Means within column with no common superscript differ significantly ($p<0.05$).***

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rate of extracellular secretion and considerable thermostability.

Taking all this information into account, it could be hypothesised that this extracellular enzyme production can increase the digestibility of feed when this probiotic is administered in combination with the diet.

In this sense we can find examples in the scientific literature that confirm this hypothesis: in a trial conducted by Sanchez et al (2006) it was demonstrated how the use of B. amyloliquefaciens CECT5940 (Ecobiol) increased the digestibility of crude protein, ether extract and starch in broiler diets.

So the subsequent question would be if this digestibility increase could enable a reduction in the nutrient concentration of the diet, compen-

Fig. 1. Analysis for dry matter content.

$$\text{AME (kcal/kg as is)} = \frac{(\text{GE feed} \times \text{g feed consumed})-(\text{GE excreta} \times \text{g excreta})}{(\text{g feed consumed})}$$

$$\text{AMEn (kcal/kg as is)} = \frac{((\text{GE feed} \times \text{g feed consumed})-(\text{GE excreta} \times \text{g excreta}))-\text{NR} \times \text{k}}{(\text{g feed consumed})}$$

NR = nitrogen retention which is assumed to be (20% of body weight gain/loss)/6.25. K = constant which equals to 8.21kcal/g nitrogen retention. AME = apparent metabolisable energy. AMEn = apparent metabolisable energy corrected by nitrogen.

Fig. 2. Calculating apparent ileal protein digestibility.

$$AID_{CP} = \frac{CP_d/Cr_d - CP_i/Cr_i}{CP_d/Cr_d} \times 100$$

AID_{CP} = Apparent ileal digestibility of crude protein.
CP_d = Crude protein concentration in diet.
CP_i = CP concentration in ileal digesta.
Cr_d = Marker concentration in diet.
Cr_i = Marker concentration in ileal digesta.

sating this reduction with the addition of the probiotic.

If this is possible, we could relax the formulation requirements, thus lowering the final price of feed.

Trial in Thailand

To check this effect Norel's technical department designed the following trial. A study was conducted at BARC (Bangkok Research Center), Thailand. A total of 480 (Arbor Acres Plus) male broiler chicks were randomly allocated to eight different treatments with six replications using 10 birds in a pen as experimental unit. The test period covered starter (0-18 days of age) and grower (18-35 days of age).

Feeds were prepared in pellets (80°C conditioning temperature) and were provided to birds in crumble form to 12 days and in pellet form thereafter until finishing the 35 day test period. Feeds and water were provided freely. Birds were maintained on the lighting program, house temperature and management according to the Arbor Acres Plus broiler management recommendation.

Body weight of birds and leftover feed of each pen were measured on day 18 and 35. Dead and culled birds were recorded daily. At the end of each growing period, individual body weight was measured.

Body weight gain, feed intake, FCR, and livability were subjected to ANOVA as a 4 x 2 factorial arranged in a RCBD. Additionally, a digestibility study was conducted to verify if the compensatory effect of the probiotic could be attributed to the increased nutrient's digestibility.

240 male broiler chicks, aged 21 days, were randomly allocated to 48 metabolic cages, at five birds/cage. The same test diets prepared for grower broilers in the growth study were provided to birds (six cages/diet)

During the 96 hour collection period, the amount of feed given and the left over feed at the end of collection period were recorded for feed intake calculation.

All excreta from each cage were collected daily. All wet excreta were dried at 80°C for 24 hours. Test diet and excreta samples were collected and analysed for dry matter content (105°C, four hours) and gross energy (bomb calorimeter, Leco model AC-350, Isoperibol method) as shown in Fig. 1. All diets used in the AME and digestibility were added a marker. After finishing the 96-hour collection period, all birds in the AME test were sacrificed for digesta sample collection from the ileum.

The collected digesta samples were freeze dried. The test diet and dried ileal digesta samples were analysed for protein and marker contents. The apparent ileal protein digestibility was calculated based on dry matter by using the equation as shown in Fig. 2.

Treatment			AMEn as fed (cal/g)	AMEn as dry (cal/g)	Protein digestibility (g)
Group	**Nutrient reduction**	**Ecobiol (kg/MT)**			
T1		0.0	$2,793^{abc}$	$3,090^{bc}$	69.25
T2	-60 kcal/kg; -2.3% aa	0.0	$2,766^{cd}$	$3,071^{bcd}$	68.66
T5		1.0	$2,813^{ab}$	$3,117^{ab}$	69.66
T6	-60 kcal/kg; -2.3% aa	1.0	$2,830^{a}$	$3,141^{a}$	69.22
SEM			15.513	17.185	0.955
Source		**df**	**P values**		
Treatment		7	<0.0001	<0.0001	0.3365
Diet (A)		3	<0.0001	<0.0001	0.1040
Ecobiol (B)		1	0.0009	0.0006	0.2987
A x B		3	0.5813	0.6718	0.9087

Table 5. Effect of Ecobiol in apparent metabolisable energy ^{a,b,c,d,e} Means within column with no common superscript differ significantly ($p<0.05$) and protein digestibility.

Results

From the results of the first nutrient reduction step it can be concluded that Ecobiol was able to compensate a reduction of -60 kcal/kg and -2.3% in the lys and met content, being performance equal to the group receiving a higher nutrient concentration feed.

According to the complementary study, this effect can be explained by the higher values of metabolisable energy and protein digestibility observed in birds supplemented with Ecobiol. Once demonstrated this compensatory effect, it is plausible to use hypothetical nutritional values in the Ecobiol formulation matrix. This way, the formulation software will take into account the probiotic 'nutrient contribution', relaxing the formulation constraints and thus providing a cheaper solution. It is also important to remark that this probiotic nutrient contribution must not be fixed, as it depends on diet formulation.

The energy and amino acids released by the additive will be correlated to the amino acid profile and content, and also to the fat, protein and starch concentration of the feed.

Bacillus amyloliquefaciens CECT 5940 proves to be an extremely versatile tool, summing up a number of different capabilities: pathogen antagonist, immune system modulator, gut health enhancer and now a significant digestibility improver.

All these action mechanisms working together at gut level helps the new selected broiler breeds to express their full potential. ■

References are available from the author on request

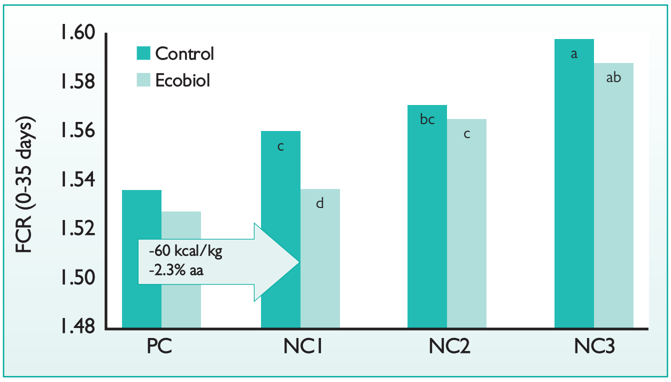

Fig. 3. FCR results (0-35 days of age).

Fig. 4. Apparent metabolisable energy corrected by nitrogen.

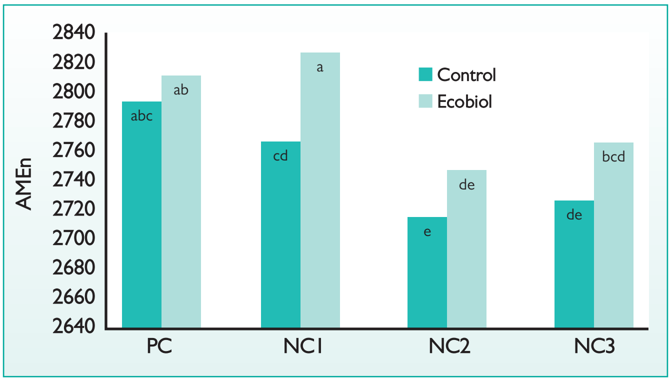

Fig. 5. Protein digestibility.