Efficiency of a prebiotic and a plant extract on broiler performance and intestinal physiology

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The objective of this experiment was to evaluate the effects of a short chain fructo-oligosaccharide (scFOS) from sugar beet (PROFEED®), and a plant extract (XTRACT™), alone or mixed, on broiler performance up to 36d. Ileal histomorphometry, anaerobic sulphite-reducing bacteria count, intestinal and caecal pH, caecal volatile fatty acids production and litter score were also evaluated. Five supplemented diets were tested: scFOS 600 ppm (P); plant extract 100 ppm (X); P/X 600/100 ppm (XPH); P/X 450/75 ppm (XPM); P/X 300/50 ppm (XPL). These experimental diets were compared to a negative control (C) and a positive control with 10ppm avilamycin (AV).

Compared to C, avilamycin improved broiler performance throughout the experiment while the effects of plant extract and prebiotic employed alone were noticeable during the finisher period (from 22 to 36d). At 36d of age, broilers fed AV, P or X were heavier than those fed C. Throughout the trial, the same broilers tended to have a better feed conversion ratio (FCR). When the two additives were mixed, from 1 to 36d of age, XPM or XPL improved the body weight gain as AV did, but only XPL improved the FCR compared to C. With the higher mixture dose (XPH), no broiler performance improvement was observed suggesting a negative associative effect of the two additives. No statistical effect was observed in any treatment for the other parameters measured.

Based on these results, prebiotic and plant extract could be considered as potential alternatives to antibiotic growth promoters in broilers up to 36d of age, though further research is needed to understand the basis of their growth promoter effect.

Keywords: broiler; performance; feed additives; prebiotic; plant extract.

Introduction

Though the possible relationship between resistant microorganisms selected during the use of antibiotic growth promoters (AGP) in poultry and antibiotic-resistant infections in humans is still under debate (Dibner and Richards, 2005), European Union has preventively banned the use of antibiotics as growth promoters since 1st January 2006. Therefore, alternatives to AGP need to be proposed to livestock producers in order to maintain animal health, productivity and microbial food safety.

Plant extracts and prebiotics that have shown some capacity to modify gut microbiota could be considered as potential alternatives to AGP. Antimicrobial activity from some active principles of the first ones has been quoted, as could be cinnamaldehyde and carvacrol (Hernández et al., 2004; Dahiya et al., 2006). The second ones, fructo-oligosaccharides (FOS), are non-digestible and non-absorbable oligosaccharides available as substrate for beneficial intestinal bacteria while controlling populations of potential pathogens by competitive exclusion (Xu et al., 2003). Nevertheless, though both potential alternatives to AGP act on intestinal microbiota (plant extract by its antimicrobial activity; prebiotic by

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its promoting effect on beneficial intestinal bacteria), there is no report on broiler evaluating the response to supplementation with combinations of prebiotics and plant extracts.

Therefore, the objective of this work was to evaluate, as potential alternatives to AGP, the effect of a prebiotic (PROFEED®) and a plant extract (XTRACT $^{\text{TM}}$), alone or in combination, on animal performance and a large set of variables related to the gastrointestinal tract: ileal morphometry, anaerobic sulphite-reducing bacteria (ASR) count, intestinal and caecal pH, caecal volatile fatty acids (VFA) production and litter score (LS) in male broilers.

Materials and methods

The experiment was conducted on 3780 Ross PM3 male broilers from the age of one-day-old to 36d. Broilers were randomly distributed in 84 floor pens located in a poultry house environmentally controlled by heating, ventilation and lighting systems. 45 one-day-old broiler chickens were placed in each 3x1 m floor pen. All broilers were individually identified at 7-8d by numbered wing bands.

A three-phase feeding program was used with corn-wheat-soya based diets: starter (from 1 to 10d), grower (from 11 to 21d) and finisher (from 22 to 36d). The diets were steam pelleted using a 2.5 mm die. Seven treatments were involved: negative control (C); positive control 10 ppm avilamycin (AV); 600 ppm PROFEED® (P); 100 ppm XTRACT™ (X); high level combination PROFEED®/XTRACT™ 600/100 ppm (XPH); medium level PROFEED®/XTRACT™ 450/75 ppm (XPM) and low level PROFEED®/XTRACT™ 300/50 ppm (XPL). 540 broilers were assigned to each treatment, so 12 replicates per treatment were tested in a randomized design. XTRACT™ was a mixture of plant extract provided by Pancosma¹ which chromatogram analysis showed the following composition: 5.44% carvacrol, 3.25% cinnamaldehyde and 1.93% standardized *Capsicum* oleoresin. PROFEED® was a sugar beet short-chain FOS obtained by enzymatic synthesis provided by Beghin-Meiji². Feed and water were provided *ad libitum*.

Body weight (BW) of each broiler was measured on 1d, 11d, 21d and 36d for body weight gain (BWG) calculation. Feed intake was measured per floor pen throughout the experiment and the feed conversion ratio (FCR) was calculated from 1 to 11d, 12 to 21d, 22 to 36d and 1 to 36d on a pen weight basis. Mortality and BW of dead chicks were recorded daily. The weights of dead and eliminated birds were taken into account for the FCR calculation.

At 28d of age, 12 broiler chickens per treatment (one per floor pen) were sacrificed and the content of the intestine (pool of duodenum, jejunum, ileum and colon) and the two caeca were collected for immediate pH measurement. Caeca content was stored (-20°C) for later VFA determination by gas liquid chromatography. For ileal morphology, a segment (1.5 cm) was cut in the mid part of the ileum. At least 10 villi and 20 crypts from each subsample (so 120 villi and 240 crypts per treatment) were dissected under a stereo microscope and photographed. Villi height and width, as well as crypts depth and width, were measured using image analysis software. The surfaces of villi and crypts were calculated from the height or depth, and the width.

At 26 and 34d of age, faecal samples were taken from the cloacae for ASR count. For each treatment, six pools of faeces were collected on 10 broilers from two floor pens (five chickens per pen). 1/10 dilutions of the samples were immediately done with peptone solution and 1 mL aliquots were pipetted and sowed into plates with Triptose Cycloserin Agar medium. The plates were incubated at 37°C for 20h in anaerobic conditions using BBL[™] GasPak[™] system³. Floor pen ASR counts were log 10 transformed prior to statistical analysis.

At 33d of age, based upon a visual assessment of the floor pen by three independent persons, a percentage of litter surface was assigned to five score levels according to humidity degree and a litter score calculated from 1(dry and friable) to 5 (wet and compacted).

Diets effect on BW, BWG, FCR, intestinal and caecal pH, ASR count, caecal VFA, villus and crypt parameters and LS were analyzed statistically by one-way ANOVA with STATVIEW 5.0 software. Differences among means with 0.05<P<0.10 were accepted as representing tendencies to differences.

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Results and discussion

On average, BW of the one-day-old broilers was 40.6 g and mortality was 4.44%, and both parameters were not different among treatments. Also, there were no differences for parameters related to the gastrointestinal physiology. Thus, the mean value for villus height, crypt depth, villus height and crypt depth ratio, and villus surface were 0.5102 mm, 0.1286 mm, 4.008, and 0.6382 mm², respectively. When AGV were evaluated, the mean value of total AGV production and the proportion of acetic, propionic and butyric acids were 26.2 mg, 61.3%, 24.9% and 9.7%, respectively. The average intestinal and caecal pH were 6.09 and 6.18. For microbial counts, 26 and 34d ASR log10 UFC counts were 4.44 and 4.05. Finally, the litter score was a medium score of on average, 2.1 corresponding to quite good litters.

Performance parameters were affected by treatment diets. AV improved by 2% BWG and FCR throughout the experiment in comparison to C.

During starter period (1-11d of age), BWG of all experimental groups was lower than C or AV. During grower period (12-21d of age), the best BWG was observed with AV. BWG of all experimental groups was not different from C, except for XPH, which gained less than C. During finisher period (22-36d of age), AV, X, P, XPM and XPL gained more weight than C. In addition, BWG of P was higher than that of AV, and BWG of XPH was not different from C.

During starter period, FCR was improved by AV compared to the other treatment groups. This improvement was similar during grower period, but XPH group was not different from AV group. During finisher period, there was no difference between treatments for FCR. Considering the whole experimental period (1-36d of age), FCR of AV, X, P and XPL groups tended to be improved compared to C group. FCR of broilers fed XPH and XPM were not different from C.

Environmental conditions (e.g. density) and stress status of the animals are important to detect growth performance responses to feed additives. Growth promoters, especially antibiotics, were more effective when used in unhealthy rather than in healthy birds and the expected response level for performance should be lower for animals in production systems with high standards of hygiene (Thomke and Elwinger, 1998). Consequently a well known additive (positive control treatment) and a negative control treatment ought to be compared in order to validate the experimental model. The positive control should improve broiler performance in order to rank the responses of the animals to the other tested additives.

In the present experiment, from 1 to 11d of age, BWG of birds fed AV were not different from those of C group, indicating that stress status was low at the beginning of the experiment. Later, while the animals were aging and certainly more stressed due to crowding, AV effect was in the same range as in field conditions with an improvement of broiler performance by 2% in comparison to C group. Consequently, the experimental scheme should be considered as relevant for the comparison of the alternatives to AGP tested.

Jamroz and Kamel (2002) and Jamroz et al. (2003) reported improvements in broiler performance when feeding 150-300 ppm of the same plant extract used in the present study. In contrast, Hernández et al. (2004) did not detect any growth promoting effect of this extract used at 200 ppm in male broilers diets. It is important to note that this last trial was done under high hygienic conditions which could have alleviated the animal's response, so the results could no to be applicable to real farm conditions. In the present experiment, X treatment improved BWG in the same way as AV treatment at the end of the experiment, when the broilers were probably in heavy environmental conditions. In fact, total weight of broiler reached 29 kg/m² at the end of the experiment and consequently the litter quality was impaired. This plant extract could also enhance the digestibility of nutrients, thus improving growth or/and FCR (Jamroz et al., 2003; Catalá et al., 2004).

P treatment was a sugar beet FOS used for its prebiotic effect. Tianxing et al. (1999) reported an improvement of BW and FCR in broilers fed 0.25 to 0.50% FOS, in comparison to unsuplemented group, and Xu et al. (2003) reported similar improvements with the incorporation of 0.4% of FOS in the diet. However, no positive control group with AGP was included in these studies. In our experiment we observed a significant effect on broiler performance with a very low FOS dosage

(0.06%). As for the plant extracts, the FOS were more efficient at the end of the experiment when overcrowding induced poor hygienic conditions.

At the end of the experiment, performance of birds fed the highest mixture of X and P (XPH) was not different from C group. It seemed that 600 ppm of prebiotic and 100 ppm of plant extract exerted an antagonist effect that mutually inhibits their beneficial effects on broiler performance. As the main effect of these compounds is related to controlling gut microbiota, further research is needed to elucidate the targeted microorganisms in order to understand this antagonist effect. For the lower dosages of X and P (XPM and XPL), a positive effect was shown, and results of performance were similar to feeding each additive X or P alone.

Based on these results, prebiotic and plant extract could be considered as potential alternatives to antibiotic growth promoters in broilers up to 36d of age, though further research is needed to understand the basis of their growth promoter effect.

Table 1 Effect of treatments on broiler performance

	Diets								
	C	AV	P	X	XPH	XPM	XPL	SEM ¹	p^2
BWG (g)									
1-11d	261 ^a	264 ^a	256 ^b	254 ^b	254 ^b	255 ^b	254 ^b	0.6	***
12-21d	583 ^b	594 ^a	577 ^{bc}	581 ^b	572°	578 ^{bc}	582 ^b	1.1	***
22-36d	1237 ^c	1261 ^b	1284 ^a	1278 ^{ab}	1242°	1271 ^{ab}	1261 ^b	2.5	***
1-36d	2084 ^{bc}	2121 ^a	2121 ^a	2115 ^a	2071 ^c	2105 ^{ab}	2097^{ab}	3.2	***
FCR (g/g)									
1-11d	1.15 ^b	1.10^{a}	1.15 ^b	1.14 ^b	1.16 ^b	1.15 ^b	1.15 ^b	0.004	**
12-21d	1.45 ^b	1.41^{a}	1.44 ^b	1.44 ^b	1.43 ^{ab}	1.44 ^b	1.43 ^b	0.003	**
22-36d	1.81	1.79	1.77	1.77	1.79	1.78	1.78	0.004	NS
1-36d	1.62 ^c	1.59 ^a	1.60 ^{ab}	1.60^{ab}	1.61 ^{bc}	1.60 ^{abc}	1.60^{ab}	0.003	†

^{a, b, c}: Mean values on a same row with different superscripts differ significantly ($p \le 0.05$).

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¹ Standard error of the mean.

² Signification level: NS, non significant P > 0.10; †, tendency 0.05 < P < 0.10; ** 0.001 < P < 0.01; ***P < 0.001