Endogenous Nitrogen Flow in Chickens as Influenced by Dietary Levels of Viscous and Non-Viscous

Roy M. PADILLA¹ ¹Instructor, AFTD, DOSCST, Mati, Davao Oriental

Abstract

This study was undertaken to examine the antinutritional influence of -soluble non-starch polysaccharides (NSP) in monogastric specie. Specifically, the influence of NSP on ileal nitrogen digestibility and flow were determined in broiler chickens. Different levels (3% or 6%) of purified maize arabinoxylan (AX) and barley β-glucan extract Glucagel TM (BG) were substituted to wheat starch in enzymatically hydrolysed casein-based (EHC) diets. Five experimental diets consisting of EHC, cellulose, wheat starch, dextrose and vegetable oil were formulated. These diets contained titanium oxide as an indigestible marker. Each experimental diet (control, 3% and 6% of BG or AX) was fed for 7 days to 27-day old birds in cages, with 4-5 birds/cage. Inclusion of AX and BG did not significantly influence feed intake (P>0.05). AND was numerically depressed at 90.37% and 90.4% for 6% AX and BG as compared to 91.1% for control diet. Ilea' nitrogen content and endogenous nitrogen flow were numerically increased with increased levels of AX and BG, though statistically significant differences were not observed due to high variations among the replicates. Inclusion of 6% BG significantly depressed dry matter digestibility (P<0.05), suggesting preservation of hydration property of gelling BG. It is then concluded that the anti-nutritional effect of soluble NSP was evident in chicken as indicated by decreased dry matter digestibility (P<0.05) and the extent of increase in ileal flow of nitrogen in chicken. The cause of increased nitrogen flow with increased levels of NSP is not clear, but could be due to increased secretion of endogenous protein, decreased reabsorption, or combination of both.

Keywords: Antinutritional, Non-Starch Polysaccharides, Ileal Nitrogen Digestibility, Broiler Chickens, Purified Maize Arabinoxylan, Barley β-Glucan Extract,

Introduction

An estimate of 140 million tons of fibrous materials is produced from cereals and other sources, and such values are increasing dramatically every year. In most grains or cereals, the fibre component is primarily consisting of non-starch polysaccharides that form part of the cell wall structure (Choct, 1997).

Non-starch polysaccharides (NSP) played a crucial role in poultry nutrition due to the anti-nutritive effect elicited by soluble pentosans components (arabinoxylan and beta-glucan) and their effects on bird performance (Choct, 1997). The detrimental effects of NSP are believed to be associated to its viscous nature that has physiological, morphological and microbiological effects on the digestive tract. These events negatively impact nutrient digestion and also cause wet and sticky droppings (Bedford and Schulze, 1998).

It has been established that NSP in wheat influence the apparent ileal digestibility of nutrients, including protein (Choct and Annison, 1992). The reduction in protein digestibility can be attributed to impaired digestion, inhibition of amino acid absorption or increased secretion of endogenous protein derived from gut secretion and sloughed off epithelial cells to the digesta.

In the present study, the influence of different types and levels of NSP on endogenous ileal nitrogen losses in broiler chickens was examined using the peptide alimentation method. (Moughan et al., 1990). Two types of soluble NSP, namely, viscous (barley ß-glucan) and non-viscous (maize arabinoxylan) were compared.

Materials and Methods

Five diets were prepared. including a control diet and test diets that contained 3.0 and 6.0% purified maize arabinoxylan or barley beta-glucan extract GlucagelTM). The diet composition is shown in Table 1. All diets contained 0.5% titanium oxide as an indigestible marker.

Ingredients	D Control	1 3% Ax ²	E 6% Ax ²	T 3% BG ³	S 6% BG ³
Starch	569.7	539.7	509.7	539.7	509.7
Dextrose	100.0	100.0	100.0	100.0	100.0
EHC ¹	180.0	180.0	180.0	180.0	180.0
Arabinoxylan	Contraction of the	30.0	60.0		100.0
Beta-glucan	-	- 1		30.0	60.0
Vegetable oil	50.0	50.0	50.0	50.0	50.0
Cellulose	35.0	35.0	35.0	35.0	35.0
Titanium oxide	0.5	0.5	0.5	0.5	0.5
Common Ingredients ⁴	60.3	60.3	60.3	60.3	60.3

Table1. Ingredient composition (g/kg) of the experimental diets.

-Enzymatically Hydrolysed Casein; ²-Arabinoxylan; ³-Beta-glucan; ⁴- Dicalcium phosphate, 24.0; Dipotassium hydrogen phosphate, 14.3; Sodium bicarbonate, 12.0;

The treatments diets were assigned as follows:

Control=	0% AX / BG
3% AX=	3% Arabinoxylan
6% AX =	6% Arabinoxylan
3% BG=	3% Beta-glucan
6% BG=	6% Beta-glucan

Three-week old male broilers (Ross) were selected based on body weight (1.3 to 1.7 kg) and allocated for 25 colony cages (4 — 5 birds per cage) so that all cages had a similar average weight. The 25 pens were assigned at random to the five dietary treatments so that there were five replicates per group.

The cages were housed in an electrically heated grower shed (22 - 24 oc)during the trial. All birds had ad libitum access to feed and water. The birds were given a mash commercial-type diet till Day 27. Following overnight fasting on Day 27, a casein-based diet was introduced and fed for the next three days. The casein-based diet was similar to the control diet (Table 1) except that casein was used in place of EHC. The casein-based diet was withdrawn on the evening of Day 30 and the test diets (Table I) were introduced on the morning of Day 31. The test diets were offered for 36 hours and records of feed intake during this period was maintained. The birds were then euthanased by an intracardial injection of sodium pentabarbitone and the contents of the lower half of the ileum were collected by gently flushing with distilled water into plastic containers. The ileum was defined as that portion of the small intestine extending from Meckel's diverticulum to a point 40 mm proximal to the ileo-cecal junction. The digesta were pooled from birds within a pen, frozen immediately after collection and subsequently freeze-dried. Diet and dried ileal digesta samples were ground to pass through a 0.5 mm sieve and subsequently analysed for titanium and nitrogen. Ileal nitrogen flow, as described by Moughan et al., 1990 and 1993, related to the ingestion of lg of food dry matter was calculated using the following equation:

Nitrogen flow = Nitrogen in the digesta x (TiO₂ diet/TiO₂ digesta) Apparent digestibility of nitrogen (ADN) was calculated using the formula: ADN = [(N diet/TiO₂ diet) - N digesta/TiO₂ digesta)] / (N diet/TiO₂ diet) The dry matter digestibility (DMD) was then calculated using the formula: DMD= [(TiO₂ in ileal sample/TiO₂ in diet) -1]/ (TiO₂ in ileal sample/TiO₂ in diet)

In this study, the endogenous nitrogen (N) flow was calculated assuming a digestibility value of 100% for EHC. This assumption was based on previous trial where EHC had been shown to be completely digested by broiler chickens (Ravindran, 2000 unpublished data). Thus, based on the above assumption, endogenous N losses is, therefore, equal to nitrogen flow.

Results

The experiment involved feeding of birds with casein-based and enzymatically hydrolyzed casein diets. The endogenous nitrogen flow, dry matter and nitrogen digestibility's, and dry matter and total feed intake (5 groups with 5 birds/group) were detailed in Table 2. Total feed intake for 36 hours was similar (P>0.05) in broiler fed different types and levels of no starch polysaccharides. Though no significant differences in feed consumed

over the specified period, a common trend was evident wherein total feed

intake is quantitatively decreased with addition of soluble NSP in the diet. Inclusion of soluble NSP did not influence nitrogen digestibility (P>O.05), at least in this trial, having nitrogen digestibility values in excess of 90% for different levels (3% and 6%) and types (soluble- β -glucan, arabinoxyian; insoluble- cellulose) of dietary NSP. However, it is interesting to note that the lowest values for nitrogen digestibility (90.4%) were observed in birds fed by the highest inclusion level (6%) of arabinoxylan (AX) and β -glucan (BG), both were soluble NSP.

DIETS	DMD ¹	lleal N (g/gTiO2)	ENF ² (ug/gDMI)	ADN 3	TFI ⁴ kg(36h)
Control	0.878ª	0.457	2383	0.911	0.99
3.0% AX	0.882ª	0.552	2549	0.906	1.07
6.0% AX	0.879ª	0.605	2625	0.904	1.04
3.0% β-G	0.884ª	0.506	2372	0.914	1.10
6.0% β-G	0.8565	0.535	2700	0.904	1.03
Pooled SEM	0.0055	0.055	269	0.010	0.055

Table 2. Influence of type and level of NSP on feed nitrogen flow in broilers.	intake and endogenous
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^{3-b} Values in the same column with different superscripts are significantly different at (P< 0.05), LSD; ¹-Dry matter digestibility; ²-ENF Endogenous nitrogen flow; ³- Apparent digestibility of nitrogen; ⁴- Total feed intake for 36 hours; AX-Arabinoxylan; B-G-Beta-glucan.

Ileal nitrogen contents, expressed as g/g titanium (g/gT), were numerically increased with the addition of soluble NSP (0.605 g/gT against 0.552 g/gT for 6% and 3% arabinoxylan; and 0.535 g/gT against 0.506 g/gT for 6% and 3% β -glucan), but the influences were not statistically significant due to high variation among replicates. The same trend was also observed for endogenous nitrogen flow, where increase in the inclusion levels of soluble NSP in the diets resulted to quantitative increase of ilea! nitrogen flow. Highest endogenous nitrogen flow was observed in 6% level of β -glucan (2700 ug/g DMI), followed by 6% level of arabinoxylan (2625 ug/g DMI), however, the said differences did not sustain statistical significance (P>0.05) due to greater variation among replicates.

On the other hand, the dry matter digestibility was not influenced by inclusion of arabinoxylan or 3% β -glucan (with values ranging from 0.879, 0.882 and 0.884 for 6% and 3% arabinoxylan and 3% β -glucan level of inclusion, respectively) but was significantly lowered (P<0.05) with 6% inclusion of β -glucan, with the dry matter digestibility value reduced to 0.856.

Discussions and Conclusion

In the peptide alimentation method (Moughan et al., 1990), the animal is fed a purified diet containing enzymatically hydrolyzed casein (EHC) (composed of free amino acids and peptides with a molecular weight of less than 10,000 Da) as sole source of nitrogen. The digesta are then collected from the animal and the endogenous protein (molecular weight, Da) is separated from unabsorbed free amino acids and peptides by centrifugation and ultrafiltration. In the present study, difficulties were experienced with the ultrafiltration step. The endogenous flow was therefore calculated assuming a digestibility value of 100% for EHC. This assumption was based on the previous work where EHC has been shown to be completely digested by broiler chickens (Ravindran, 2000 unpublished data).

The influence of soluble NSP in bird performance can be manifested in the amount of feed consumed per unit time. Several studies in broilers showed nonsignificant effect of the dietary inclusion of soluble NSP on feed intake. Maisonnier et al. (2001) showed similar feed intake in male broiler chickens given diet based on wheat, and maize added with different levels of guar gum, however, other parameters like feed conversion ratio (FCR) and growth rate were significantly influenced. Moreover, the experiment of Ouhida et al. (2000a) showed no difference in feed intake when broilers were fed NSP-rich diet (maize-barley-wheat based diets) with or without enzyme supplementation. The absence of differences in feed intake in this trial suggests that most of the differences in animal performance were related to impairment of the digestibility factors in the dietary constituents such as the like of NSP.

Apparent nitrogen digestibility was numerically depressed with increasing content of arabinoxylan and β -glucan in the diet, in which, greater depression at 90.3% and 90.4% were observed in 6% inclusion arabinoxylan and β -glucan, as compared to control diet (91.1%). However, such numerical depressions were not statistically significant due to high variations among

replicates. This result is unexpected, and in contrast to common notion that soluble NSP negatively influenced protein digestibility in broilers, as shown by previously completed studies (e.g., Choct and Annison, 1990, 1992; Maqueda, 1999; Angkanaporn et al., 1994; Hesselman and Aman, 1986). The absence of depressant effects of soluble NSP in this trial can be explained, in part, by the high variation among replicates. It can also be explained by the manner the β-glucan extract reacts in water, forming a soft gel and not a viscous solution, thereby making the extract behave differently than native β-glucan in solutions of the same concentration and thereby neutralizing some anti-nutritive effects (G.D. Coles pers comm., cited in Maqueda, 1999). Moreover, for nitrogen digestibility to be depressed, the concentration of soluble NSP should reach threshold levels, which were probably not achieved in this experiment. Also, Choct and Annison (1992) concluded that birds can tolerate minimal increases in gut viscosity brought by presence of soluble NSP, and birds' performance was not depressed once viscosity is reduced to certain level. Dry matter digestibility was significantly lowered with inclusion of 6% betaglucan to broiler diet. This said result is in concordance to the fact that dry matter digestibility (DMD) was negatively related to intake of soluble NSP. The result of the current trial is in agreement with the experiment of Jorgensen et al. (1996) that increased fiber levels for pea, wheat bran and oat bran significantly depressed ileal digestibility of dry matter in diminishing sequence, as presented. Similar result was also indicated by Refstie et al. (1999), who concluded that non-starch polysaccharide content of different soya products significantly depressed organic matter digestibility.

The inclusion of increasing levels of NSP numerically increased the ilea! nitrogen content (g/g Titanium) and calculated endogenous nitrogen flow (as ug/g DMI), though differences were not statistically significant due to high variation among replicates. Authors such as Angkanaporn et al. (1994) and Beames and Eggum (1981) demonstrated that wheat pentosans and some fiber sources depressed overall digestibility of amino acid and increased secretion of endogenous amino acid flow in chicken and rat, respectively. The said flow is consistent with the numerical increase of this study though no statistical significance was achieved. In general, the above results showed that increased levels of soluble NSP depressed dry matter digestibility. The present data also suggest that soluble NSP may influence the extent of the increase in the ileal flow of nitrogen in chicken.

It is a common knowledge that soluble dietary fibers possess anti nutritive factors, particularly influencing digestion process in poultry. There are proposed mechanisms that help to explain the anti-nutritive factors of NSP. Soluble NSP, like arabinoxylan and ß-glucan, from endosperm wall hinders access of digestive enzymes to nutrients found in the cereal endosperm (Edwards et al., 1988). However, several evidences like depressed growth performance and digestibility in broiler fed carboxymethylcellulose (CMC) (Smits et al., 1997), wheat pentosans (Choct et al., 1996) and barley ß-glucan (White et al, 1981), and improvements noticed in fat digestibility, a non-endosperm-stored nutrients, raises doubt about this theory (Ouhida et al., 2000b). The topic of increased digesta viscosity in chicken associated with soluble NSP is very well discussed and documented (Kocher et al. 2000: Ouhida et al. 2000ab: lii, 1999: Choct, 1997; Choct et al., 1996; Annison, 1993; Choct and Annison, 1992 and 1990). Increased digesta viscosity, caused byaräbinoxylan from wheat and β-glucan from barley, influenced intestinal tract digestion by limiting the diffusion of nutrients and digestive enzymes (Choct et al., 1996) restricting access of these enzymes to their dietary substrate, and through primary and secondary increase of microbial activity of the small intestine. (White et al., 1988). Moreover, viscous digesta had been implicated with longer retention time in gastro-intestinal tract that increases microbial activity (Annison, 1993), which in part, inhibits digestibility of nutrients. Further, the increased digesta resistance to peristalsis, due to high viscosity, may cause a stimulation of endogenous secretions by a feedback mechanism (Angkanaporn et al., 1994). These factors suggest increased nitrogen recovery and flow, as indicated in the current trial. Investigation done by Angkanaporn et al. (1994) on the influence

of soluble fibre on endogenous nitrogen losses in chicken suggested that increased rate of dietary pentosans inclusion inhibited direct protein breakdown and amino acid absorption that leads to decreased nitrogen digestibilities and conseauently caused an increase in endogenous amino acid losses.

In conclusion, increased level of soluble NSP significantly depressed dry matter digestibility. The current results also suggest that soluble NSP may increase ilea] flow of nitrogen in chickens. The exact causes of the increased nitrogen flow with increased NSP levels are not known. It could be due to increased secretion of endogenous proteins into the gut, decreased reabsorption of endogenous protein or a combination of both effects. As suggested by Angkanaporn et al. (1994), it is possible that soluble NSP interacts with the gut wall modifying the actions of the peptide hormones, which regulate gut functions including stimulation of secretion of endogenous protein.

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