



Original Research Article

The flow of non-starch polysaccharides along the gastrointestinal tract of broiler chickens fed either a wheat- or maize-based diet



Eunjoo Kim ^a, Natalie K. Morgan ^{a,*}, Amy F. Moss ^a, Lily Li ^b, Peter Ader ^c, Mingan Choct ^a

^a School of Environmental and Rural Science, University of New England, Armidale, NSW 2351, Australia

^b BASF South East Asia, 038987, Singapore

^c BASF SE, 68623, Lampertheim, Germany

ARTICLE INFO

Article history:

Received 23 June 2021

Received in revised form

7 October 2021

Accepted 9 November 2021

Available online 14 December 2021

Keywords:

Broiler chickens

Soluble fibre

Insoluble fibre

Non-starch polysaccharides

ABSTRACT

The present study characterised the types and amounts of non-starch polysaccharides (NSP) remaining undigested along the gastrointestinal tract (GIT) of broiler chickens offered a typical wheat- or maize-based diet. One-day old Cobb 500 mixed-sex chicks were assigned to 24 pens, with 10 birds/pen and 12 pens/treatment. Birds were offered the experimental diets in 3 phases (starter, day 0 to 10; grower, day 11 to 24 and finisher, day 25 to 35). Excreta and digesta samples from the crop, gizzard, duodenum, jejunum, ileum and caeca were collected at 12 and 35 days of age, and analysed for the NSP flow. The wheat-based diet contained higher levels of soluble NSP than the maize-based diet, whereas insoluble NSP levels were similar between the 2 diets. Detailed analysis of NSP constituents revealed that arabinoxylans were the primary NSP in the wheat-based diet, mostly in insoluble form. Pectins were the predominant NSP in the maize-based diet, followed by arabinoxylans. Overall, birds offered the wheat-based diet presented higher levels of soluble NSP remaining in all gut sections compared to birds offered the maize-based diet, at both 12 and 35 days of age ($P < 0.050$). Accumulation of insoluble NSP in the gizzard was noted in birds fed both diets, but was more pronounced in birds offered the maize-based diet compared to the wheat-based diet, at both 12 and 35 days of age ($P < 0.001$). The present study highlights marked differences in the amounts and types of NSP delivered to the different gut sections when feeding wheat-compared to maize-based diets, particularly in the gizzard and the lower GIT of birds.

© 2022 Chinese Association of Animal Science and Veterinary Medicine. Publishing services by Elsevier B.V. on behalf of KeAi Communications Co. Ltd. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

1. Introduction

Non-starch polysaccharides (NSP) and lignin are the primary constituents of plant cell walls, accounting for up to 20% of plant-based poultry diets (Choct, 2015). However, NSP are the least digestible constituent of poultry feed, as birds lack endogenous NSP-degrading enzymes. Microbial fermentation in the hindgut of birds can partially hydrolyse NSP, but only to a very small extent. Although NSP utilisation is limited in poultry, there is strong

evidence that intestinal microbiota, gut physiology and nutrient digestibility are directly influenced by dietary NSP, depending on their water solubility. Soluble NSP often increase digesta viscosity and hinder nutrient absorption (Choct and Annison, 1992; Kiarie et al., 2014), whereas insoluble NSP are mostly inert in the digestive tract, bulking up the total fibre levels of feed and excreta dry matter (Choct, 1997; Hetland et al., 2004).

To minimise the anti-nutritive impacts of NSP and improve their utilisation, exogenous feed enzymes active against specific NSP are routinely applied into modern broiler diet formulations. For instance, xylanase can be included for arabinoxylans hydrolysis, whereas β -glucanase for β -glucans degradation. Nonetheless, the use of these feed enzymes is not always successful, largely a consequence of poor understanding about dietary fibre, due to lack of consistency and clarification regarding definitions and analytical methods used for assessing dietary fibre. It is well established that crude fibre and detergent fibre systems do not capture all the NSP present in the feedstuff (Choct, 2015), yet poultry diets are still

* Corresponding author.

E-mail address: nmorga20@une.edu.au (N.K. Morgan).

Peer review under responsibility of Chinese Association of Animal Science and Veterinary Medicine.



Production and Hosting by Elsevier on behalf of KeAi

formulated based on these classical concepts. This means that the actual dietary fibre levels of a diet are overlooked, thereby exacerbating the anti-nutritive impacts of NSP and/or inconsistent responses to supplemental enzymes during broiler production.

NSP consist of different structural features in terms of linkage patterns, monomer composition, degree of branching and side-chain compositions, leading to variable nutritional properties in the digestive tract of animals (Hamaker and Tuncil, 2014). Thus, a detailed understanding of NSP quantity and composition, and thus its ability to be utilised by broilers, is vital for nutritionists to be able to precisely predict true fibre levels in feed formulations, and the influence of NSP in the chicken gastrointestinal tract (GIT).

In the present study, common wheat-soybean meal and maize-soybean meal-based diets, without the presence of exogenous enzymes, were used as NSP substrates for characterisation of the amount and types of NSP remaining undigested along the GIT of broiler chickens, at 12 and 35 days of age. The aim of this study was to provide insight into the flow of dietary NSP and their behaviour in the digestive tract of broilers.

2. Materials and methods

2.1. Animal ethics

The experimental procedures used in the present study were approved by the Animal Ethics Committee of the University of New England (AEC 18-089).

2.2. Experimental design, housing and diets

Two hundred and forty Cobb 500 mixed-sex broiler chicks were obtained at 1 day old from a commercial hatchery (Baiada, Australia). The chicks were individually weighed and assigned to 24 equal-sized floor pens (120 cm × 77 cm), with 10 birds per pen. There were two dietary treatments and 12 replicate pens per treatment. Birds were raised on clean wood shavings as a bedding material. The temperature was maintained at 32 °C during the first three days and then gradually reduced to 22 °C by 21 days of age. Birds received 24 h of light for the first three days post-hatch and then 20 h of light for the remainder of the study.

The two dietary treatments were either a wheat- or maize-based diet, and both diets were formulated to meet the nutrient specifications for Cobb 500 broilers (Cobb-Vantress, 2018). The composition of the experimental diets is shown in Table 1. Birds were offered the experimental diets in 3 phases (starter, day 0 to 10; grower, day 11 to 24 and finisher, day 25 to 35). Feed and water were provided *ad libitum*. The nutrient profile of ingredients was determined using near-infrared reflectance spectrometry (Foss NIR 6500, Denmark) standardised to Evonik AMINONIR Advanced calibration. The quantities of total NSP in the ingredients used in the present study were analysed according to a method described by Englyst et al. (1994) with some modifications (Theander et al., 1995; Morgan et al., 2019) to confirm that the values are in the range reported previously for wheat (Annison, 1993), maize (Bach Knudsen, 1997; Choct, 1997) and soybean meal (Langhout and Schutte, 1996; Bach Knudsen, 1997). The diets were cold-pelleted at 65 °C and the starter diets were crumbled. Antimicrobial agents, animal protein sources and exogenous enzymes were not included in any diet. TiO₂ (5 g/kg) was added to the diets as an indigestible marker.

2.3. Sample collection

Excreta samples were collected and pooled per pen on day 12 and 35. Four birds per pen on day 12 and three birds per pen on day

Table 1

The composition of experimental diets (% as fed).

Item	Starter, d 0 to 12		Grower, d 13 to 24		Finisher, d 25 to 35	
	Wheat	Maize	Wheat	Maize	Wheat	Maize
Ingredient						
Wheat	64.1	—	69.5	—	71.6	—
Maize	—	60.6	—	66.3	—	67.5
Soybean meal, 47%	29.0	33.4	23.4	27.5	19.9	25.0
Canola oil	2.17	1.15	2.87	1.72	4.31	3.19
Limestone	1.17	1.13	1.12	1.08	1.06	1.02
Dicalcium Phosphate	1.63	1.74	1.53	1.65	1.40	1.52
Salt	0.20	0.21	0.22	0.23	0.20	0.17
Sodium bicarbonate	0.22	0.26	0.11	0.16	0.18	0.31
TiO ₂	0.50	0.50	0.50	0.50	0.50	0.50
Vitamin-mineral premix ¹	0.18	0.18	0.18	0.18	0.18	0.18
Choline chloride, 70%	0.07	0.13	0.07	0.13	0.06	0.11
L-lysine HCl	0.31	0.28	0.24	0.22	0.26	0.22
DL-methionine	0.31	0.34	0.24	0.28	0.22	0.25
L-threonine	0.13	0.10	0.10	0.08	0.08	0.06
Calculated value						
ME, kcal/kg	2,950	2,950	3,050	3,050	3,150	3,150
Crude protein	23.0	22.0	20.8	19.5	19.5	18.4
Calcium	0.90	0.90	0.85	0.85	0.79	0.79
Available P	0.45	0.45	0.43	0.43	0.40	0.40
Digestible Lys	1.24	1.24	1.05	1.05	0.98	0.98
Digestible Met	0.58	0.63	0.49	0.55	0.45	0.51
Digestible Met + Cys	0.90	0.90	0.80	0.80	0.75	0.75
Digestible Thr	0.80	0.80	0.70	0.70	0.63	0.64
Digestible Trp	0.24	0.21	0.22	0.18	0.20	0.17
DEB, meq/kg	247.9	243.8	213.7	206.5	203.9	213.7

¹ Vitamin-mineral concentrate supplied per kilogram of diet: retinol 12,000 IU; cholecalciferol, 5,000 IU; tocopherol acetate, 75 mg; menadione, 3 mg; thiamine, 3 mg; riboflavin, 8 mg; niacin, 55 mg; pantothenate, 13 mg; pyridoxine, 5 mg; folate, 2 mg; cyanocobalamin, 16 µg; biotin, 200 µg; cereal-based carrier, 277 mg; mineral oil, 6.25 mg; Cu (sulphate), 16 mg; Fe (sulphate), 40 mg; I (iodide), 1.25 mg; Se (selenate), 0.3 mg; Mn (sulphate and oxide), 120 mg; Zn (sulphate and oxide), 100 mg.

35 of average weight were selected and euthanised by cervical dislocation. Digesta were obtained from the crop, gizzard, duodenum, jejunum, ileum and caeca. Excreta and digesta samples were immediately frozen at −20 °C for laboratory analysis.

2.4. Chemical analysis

All digesta and excreta samples were freeze-dried and finely ground using a centrifuge mill with a 0.5 mm screen (Model ZM 200, Retsch, Hann, Germany). Samples were analysed for dry matter and NSP, uronic acids and TiO₂. Dry matter was determined according to the standard method of AOAC (2012) (Method 930.15). The constituent sugar components of NSP were determined as alditol acetates using gas–liquid chromatography (Model CP3800, Varian Inc., Palo Alto, CA), according to the method described by Englyst et al. (1994) with some modifications (Theander et al., 1995; Morgan et al., 2019). Uronic acids were determined by spectrophotometry, at 400 and 450 nm (UV-1600PC, VWR, Darmstadt, Germany), using the method illustrated by Scott (1979). Quantification of TiO₂ was conducted using UV-spectroscopy at 410 nm (Cary 50 Bio UV–Visible spectrophotometer equipped with a Cary 50 MPR microplate reader, Varian Inc., Palo Alto, CA), using the method of Short et al. (1996). All the analysed values were corrected for dry matter percentages.

2.5. Calculation and statistical analysis

The flow of NSP was calculated using the concentrations of NSP constituent sugars and the TiO₂ marker ratio in the diets and digesta or excreta, based on the following formula:

NSP flow (g/kg marker) = NSP constituent sugar (g/kg) $\times$ (TiO₂ diet g/kg / TiO₂ digesta or excreta g/kg).

The lower values for NSP flow is an indication of NSP disappeared in the particular gut section.

All data were analysed using the GLM model procedure of IBM SPSS Statistics 25, with pen as the experimental unit. Data were tested for normality using Shapiro–Wilk test prior to statistical analysis. Significantly different means were separated by an independent *t*-test and accepted when *P* < 0.050.

3. Results

Insufficient quantities of digesta samples were collected from the crop and duodenum at 12 days of age, meaning not all the proposed chemical analyses were able to be conducted. Thus, these values are not presented herein.

3.1. Chemical composition of diets

The analysed chemical composition of the experimental diets is shown in Table 2. The wheat-based diet contained much higher levels of soluble NSP compared with the maize-based diet. The levels of insoluble NSP were similar between the two diets. Detailed NSP constituent sugar analysis confirmed that the main sugars present in the soluble NSP fraction were arabinose, xylose and uronic acid in the wheat-based diet, indicating the presence of arabinoxylans as the major soluble and insoluble NSP fractions. Pectic polysaccharides, followed by arabinoxylans, were the main

Table 2
Analysed chemical composition of experimental diets (% DM).¹

Item	Starter, d 0 to 12		Grower, d 13 to 24		Finisher, d 25 to 35	
	Wheat	Maize	Wheat	Maize	Wheat	Maize
Dry matter	88.4	88.0	88.8	89.6	89.3	88.7
Gross energy, kcal/kg	3893.4	3907.7	4348.7	4404.2	4524.9	4543.5
Crude protein ²	24.8	25.0	21.6	22.3	21.2	21.3
Ash	6.8	7.7	6.4	5.9	5.7	6.0
Crude fat	4.2	3.5	4.9	4.7	5.6	5.1
(hexane extract)						
Total starch	47.2	44.4	49.6	48.1	48.9	49.2
Klason lignin	1.6	1.2	1.5	1.3	1.6	1.4
Oligosaccharides	4.6	4.8	4.5	4.3	4.1	4.2
Total NSP ³	13.75	12.00	11.33	10.90	9.81	10.40
Soluble						
Rhamnose	0.004	0.003	0.005	0.003	0.004	0.005
Fucose	0.006	0.006	0.005	0.005	0.003	0.005
Ribose	0.04	0.03	0.04	0.03	0.04	0.03
Arabinose	0.38	0.13	0.34	0.10	0.51	0.11
Xylose	0.46	0.04	0.41	0.04	0.64	0.06
Mannose	0.14	0.13	0.23	0.16	0.19	0.27
Galactose	0.22	0.17	0.23	0.15	0.22	0.16
Glucose	0.10	0.06	0.16	0.06	0.18	0.13
Uronic acid	0.25	0.31	0.15	0.22	0.15	0.18
Total soluble	1.42	0.79	1.39	0.68	1.72	0.86
Insoluble						
Rhamnose	0.06	0.08	0.05	0.09	0.06	0.06
Fucose	0.11	0.13	0.10	0.12	0.08	0.10
Ribose	0.04	0.03	0.04	0.03	0.04	0.03
Arabinose	2.89	2.19	2.18	2.00	1.84	1.96
Xylose	3.06	2.09	2.54	2.03	1.91	1.90
Mannose	0.25	0.24	0.22	0.17	0.17	0.20
Galactose	1.78	2.09	1.42	1.80	1.17	1.75
Glucose	2.68	2.54	2.48	2.45	2.48	2.32
Uronic acid	2.38	3.13	2.11	2.74	1.53	2.34
Total insoluble	12.33	11.21	9.94	10.22	8.11	9.53

¹ Samples were analysed in duplicates.

² Crude Protein = N $\times$ 6.25.

³ NSP, non-starch polysaccharides.

NSP in the maize-based diet, as reflected by the predominant presence of uronic acid, arabinose and xylose. The glucose levels in the insoluble fraction were also relatively high in both diets, which partly originated from cellulose.

3.2. NSP flow along the GIT

The flow of total soluble and insoluble NSP and oligosaccharides along the GIT of birds fed the wheat- or maize-based diet at 12 and 35 days of age is shown in Table 3. Birds offered the wheat-based diet presented higher concentrations of soluble NSP in all gut sections (*P* < 0.001), except in the gizzard, at 12 days of age compared to those fed the maize-based diet. Similarly, at 35 days of age, soluble NSP determined in all gut sections (*P* < 0.001), except the caeca, were greater in birds fed the wheat-based diet compared to those offered the maize-based diet. The relative concentrations of soluble NSP in the different sections of the gut remained similar between the two diets, with peak accumulation occurring in the caeca at day 12, and in the gizzard and duodenum at day 35.

At 12 days of age, the wheat-based diet resulted in lower concentrations of insoluble NSP in the gizzard (*P* < 0.001), ileum (*P* = 0.016) and excreta (*P* = 0.001) compared to the maize-based diet, but the reverse was true for the caeca (*P* < 0.001). The relative concentrations of insoluble NSP in the different sections of the gut remained similar between the two diets, with accumulation beginning in the crop and peaking in the gizzard at both 12 and 35 days of age. In general, birds offered the maize-based diet presented lower concentrations of oligosaccharides from the jejunum to the total tract compared to those offered the wheat-based diet, at both 12 and 35 days of age.

4. Discussion

The present study aimed to provide detailed knowledge about the type and amount of NSP remaining undigested along the GIT of broilers offered a commercial-type wheat- or maize-based diet, devoid of enzymes, at different age. Although the physicochemical properties of NSP vary greatly within batches of the same ingredient, the results of this study could be used to predict the target substrates in various parts of the GIT. This is essential for developing nutritional strategies, such as more tailored enzyme selections, to enhance poultry production performance.

Although birds in the present study performed well, more than 30% of the nutrients in both the wheat- and maize-based diets used in the present study, which did not contain exogenous enzymes, remained undigested and were excreted by the broilers (data now shown). It appeared that NSP, mostly the insoluble fraction, were quantitatively the least digestible nutrient in both diets, accounting for 29% of the total undigested component. This again emphasises that nutritional approaches are required to minimise the prevalence of undigested nutrients, based on a clear understanding of target NSP substrates along the chicken GIT.

Distinct patterns of NSP flow between the two diets along the GIT were noted in the present study. The amount of soluble arabinose and xylose were much higher in the jejunum, ileum and excreta of birds fed the wheat-based diet compared with those fed the maize-based diet, at both 12 and 35 days of age. This higher flow of soluble NSP in wheat-fed birds is not surprising, as wheat is rich in soluble arabinoxylans with complex structures, which slows down digesta transit rate throughout the GIT, and are unlikely utilised by young birds (Choct and Annison, 1992; Annison, 1993). Nonetheless, birds offered the wheat-based diet displayed an age-related improvement in soluble NSP utilisation from the jejunum to the caeca, as illustrated by improved soluble NSP digestibility and reduced recoveries of soluble arabinose and xylose at 35 days

Table 3The flow of non-starch polysaccharides along the gastrointestinal tract of broiler chickens fed a wheat- or maize-based diet at 12 and 35 days of age (g/kg marker DM).¹

Item	Soluble NSP ²				Insoluble NSP				Oligosaccharides			
	Wheat	Maize	SEM ³	P-value	Wheat	Maize	SEM	P-value	Wheat	Maize	SEM	P-value
12 days of age												
Diet	14.2	7.9			123.3	112.1			45.5	47.8		
Crop	—	—			—	—			—	—		
Gizzard	16.2	13.3	0.89	0.101	377.8	556.1	24.67	<0.001	16.1	27.9	1.51	<0.001
Duodenum	—	—			—	—			—	—		
Jejunum	12.5	5.9	1.04	<0.001	102.9	93.7	2.94	0.124	68.9	47.6	4.50	0.009
Ileum	12.2	4.6	1.17	<0.001	82.6	90.7	1.79	0.016	23.0	20.8	1.73	0.557
Caeca	38.7	13.6	4.21	<0.001	67.8	17.7	7.98	<0.001	13.4	7.3	1.12	0.001
Excreta	12.0	5.8	0.70	<0.001	76.4	86.3	1.68	0.001	13.5	10.6	0.78	0.061
35 days of age												
Diet	17.2	8.6			81.1	95.3			41.4	41.9		
Crop	14.2	5.1	1.12	<0.001	106.6	110.0	1.53	0.292	16.5	22.1	1.19	0.024
Gizzard	27.7	15.4	2.07	<0.001	314.4	528.4	35.61	<0.001	31.3	38.6	2.23	0.102
Duodenum	25.0	13.2	1.84	<0.001	90.3	83.3	4.14	0.422	131.9	121.9	7.73	0.546
Jejunum	12.1	5.3	0.77	<0.001	75.6	81.1	1.81	0.128	89.4	73.5	3.15	0.017
Ileum	13.4	6.2	0.79	<0.001	68.8	74.8	1.51	0.045	31.7	22.9	1.34	<0.001
Caeca	13.5	9.6	1.95	0.279	34.1	22.6	4.35	0.083	8.2	5.4	1.13	0.130
Excreta	10.4	6.9	0.47	<0.001	72.9	77.4	1.51	0.068	10.1	7.4	0.65	0.045

¹ Each mean represents values from 12 replicates and each sample was analysed in duplicates.² NSP, non-starch polysaccharides.³ SEM, standard error of the mean.

of age, compared to at 12 days of age, similar to the outcome from Bautil et al. (2019) (Appendix Table 2). Although accumulation of soluble xylose and arabinose in the caeca of birds fed the wheat-based diet was noted at 12 days of age, no extensive NSP degradation occurred. It is believed that early access to fibre substrates can prime the bird to better utilise NSP later in life (Lee et al., 2017), through increasing fermentative capacity of the hindgut microbiota with advancing age even in the absence of exogenous enzyme supplementation (Svihus et al., 2013). Thus, this caecal accumulation of soluble sugars in young birds fed the wheat-based diet possibly trained intestinal microbiota to utilise dietary NSP in older birds. Moreover, in older birds offered the wheat-based diet, soluble NSP were likely already partially disappeared in the small intestine. The pre-caecal digestion presumably rendered wheat arabinoxylans less substituted, making them more favourably fermentable for the caecal microbiota, as previously evidenced by Choct et al. (1996) and Nguyen et al. (2021). As a result, birds fed the wheat-based diet utilised nearly 40% of the dietary soluble NSP after the caeca at 35 days of age. The negligible soluble NSP disappearance in birds offered the maize-based diet is most likely due to lower levels of dietary soluble NSP, which had an insignificant impact on GIT adaptation to fibre.

The superior total tract digestibility of insoluble NSP in younger birds compared to older ones was noted, regardless of the diet type. A possible explanation for this is litter intake in young birds, which might have influenced the intake of dry matter and insoluble fibre, subsequently diluting nutrient contents in the excreta and thereby affecting the digestibility values at a young age. There is evidence that chicks consume litter (up to 6% of feed intake) up to two weeks of age, and then their litter intake gradually declines with advancing age and developed appetite (Malone et al., 1983). Nonetheless, the results from the present study infer that degradation of insoluble NSP fraction is limited, but possible, even at a young age, particularly when wheat-based diets are fed.

Although wheat contains a lot more soluble NSP than maize, both cereals contain similar levels of insoluble NSP (Bach Knudsen, 2014). The main insoluble NSP in both cereals are arabinoxylans, but these polysaccharides are known to vary widely in chemical structure and 3-dimensional arrangement in the cell walls (Marcotuli et al., 2016; Bender et al., 2017). Such differences may manifest in a number of ways. In the current study, birds fed the wheat-based diet exhibited

lower levels of insoluble NSP flow in the gizzard, ileum and excreta compared with those fed the maize-based diet, but the reverse was true for the caeca. It is speculated that birds fed the wheat-based diet were exposed to more soluble NSP, which, as mentioned earlier, perhaps primed the gut microbiota to partially solubilise the insoluble NSP fraction, contrasting those fed the maize-based diet, where the insoluble NSP present in maize likely remained intact at the ileal level. Due to the solubilisation of insoluble NSP, much of the insoluble NSP in wheat were probably degraded into low molecular weight carbohydrates in the jejunum of birds offered the wheat-based diet, correspondingly increasing jejunal levels of oligosaccharides. Subsequently, young birds fed the wheat-based diet accommodated more NSP in their caeca compared to those offered the maize-based diet; there is evidence that only small soluble particles can enter the caeca (Svihus et al., 2013). On the other hand, insoluble arabinoxylans in the maize-based diet appeared to be more substituted, with a higher arabinose/xylose ratio than those in the wheat-based diet (1.04 vs. 0.94 in the starter diets), meaning that the arabinoxylans in maize are likely much less susceptible to fermentation than those in wheat.

Although the separation of TiO₂ and digesta is possible in the gizzard due to the fine particle size of the marker, it suffices to say that the gizzard of birds fed the maize-based diet held much more insoluble NSP, particularly uronic acid, galactose and glucose, compared that of birds fed the wheat-based diet at both 12 and 35 days of age (Appendix Tables 4 and 6). These results probably indicate two things: a) the hardness of the maize particles was conducive for gizzard holding, and b) there was enough microbial alternation of the NSP in the crop for the wheat-based diet as it had 'the fuel', i.e., more soluble NSP, to kick start microbial activity early in the foregut, potentially leading to significant modification of the soluble pectins present in soybean meal, where the uronic acid predominantly originates from. Interestingly, this difference in the uronic acid flow between the two diets largely disappears as the digesta moves down the GIT (Appendix Table 4). In the current study, maize and wheat were milled to the same particle size, although the pellet quality was slightly superior for the wheat-based diet compared with the maize-based diet.

This set of findings suggests that the type of grain used in broiler diets affects gizzard holding by mechanisms that are not limited to particle size. It is possible that the ratio between soluble and insoluble NSP fractions, as well as the in situ production of functional

oligosaccharides, such as xylo-oligosaccharides (XOS), may play a role in regulating digesta transit rates along the GIT. In the present study, birds fed the wheat-based diet exhibited elevated duodenal levels of arabinose and xylose as part of the oligosaccharide fraction (Appendix Table 8), suggesting the generation of XOS due to a breakdown of arabinoxylans in the small intestine. Bautil et al. (2019) demonstrated that the jejunal microbiota likely establishes with advancing age and subsequently solubilises insoluble arabinoxylans, to a small extent. Again, this emphasises that the early exposure of the bird to the appropriate type of soluble NSP substrates that sufficiently prime the microbiota to modulate digesta flow, starting from the foregut region. The gut health or nutritional outcome of such a change is difficult to speculate, but this suggests that future studies will need to pay more attention to the dynamics of digesta movement (crop and gizzard holding) and microbial changes (pH, fermentation etc.) in the foregut.

In conclusion, the present study demonstrates that different NSP fractions present in the wheat- or maize-based diet behaved differently along the chicken GIT. The higher soluble NSP contents in the wheat-based diet appeared to improve the ability of birds to utilise soluble NSP with advancing age; however, such age-related improvement was not observed for insoluble NSP. The lower soluble NSP level in the maize-based diet unlikely affected the fermentative capacity of birds. These, possibly with the physical properties of the insoluble NSP in the cell wall matrix of the diets, led to very different patterns of individual sugars in different parts of the GIT. Further research will need to explore digesta flow in the foregut of broilers fed diets that are nutritionally similar but differ in ingredient composition, e.g., different cereals. Such research will lay a solid foundation for devising highly tailored enzymes, not only to minimise undigested NSP fractions but also to maximise their effects on growth performance and gut health of birds.

Author contributions

Eunjoo Kim: Data curation, Formal analysis, Methodology, Investigation, Writing-Original draft preparation. **Natalie K. Morgan:** Conceptualisation, Investigation, Methodology, Writing-Review and Editing. **Amy F. Moss:** Writing-Review and Editing. **Lily Li:** Resources, Writing-Review and Editing. **Peter Ader:** Resources, Writing-Review and Editing. **Mingan Choct:** Conceptualisation, Data curation, Project administration, Writing-Reviewing and Editing, Supervision, funding acquisition.

Declaration of competing interest

We declare that we have no financial and personal relationships with other people or organizations that can inappropriately influence our work, and there is no professional or other personal interest of any nature or kind in any product, service and/or company that could be construed as influencing the content of this paper.

Acknowledgements

Authors gratefully acknowledge BASF SE for supporting this study.

Appendix

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.aninu.2021.11.004>.

References

- Annisson G. The role of wheat non-starch polysaccharides in broiler nutrition. *Aust J Agric Res* 1993;44:405–22.
- Bach Knudsen KE. Carbohydrate and lignin contents of plant materials used in animal feeding. *Anim Feed Sci Technol* 1997;67:319–38.
- Bach Knudsen KE. Fiber and nonstarch polysaccharide content and variation in common crops used in broiler diets. *Poult Sci* 2014;93:2380–93.
- Bautil A, Verspreet J, Buyse J, Goos P, Bedford M, Courtin C. Age-related arabinoxylan hydrolysis and fermentation in the gastrointestinal tract of broilers fed wheat-based diets. *Poult Sci* 2019;98:4606–21.
- Bender D, Schmatz M, Novalin S, Nemeth R, Chrysanthopoulou F, Tömösközi S, Török K, Schoenlechner R, D'amico S. Chemical and rheological characterization of arabinoxylan isolates from rye bran. *Chem Biol Technol Agric* 2017;4:1–8.
- Choct M. Feed non-starch polysaccharides: chemical structures and nutritional significance. *Feed Milling International* 1997;191:13–26.
- Choct M. Fibre-Chemistry and functions in poultry nutrition. In: *Proc. III Simposio Científico de Avicultura*, Málaga; 2015.
- Choct M, Annison G. Anti-nutritive effect of wheat pentosans in broiler chickens: roles of viscosity and gut microflora. *Br Poult Sci* 1992;33:821–34.
- Choct M, Hughes RJ, Wang J, Bedford M, Morgan A, Annison G. Increased small intestinal fermentation is partly responsible for the anti-nutritive activity of non-starch polysaccharides in chickens. *Br Poult Sci* 1996;37:609–21.
- Cobb-Vantress. Cobb 500 Broiler performance and nutrition supplement. 2018.
- Englyst HN, Quigley ME, Hudson GJ. Determination of dietary fibre as non-starch polysaccharides with gas-liquid chromatographic, high-performance liquid chromatographic or spectrophotometric measurement of constituent sugars. *Analyst* 1994;119:1497–509.
- Hamaker BR, Tuncil YE. A perspective on the complexity of dietary fiber structures and their potential effect on the gut microbiota. *J Mol Biol* 2014;426:3838–50.
- Hetland H, Choct M, Svihus B. Role of insoluble non-starch polysaccharides in poultry nutrition. *World Poult Sci J* 2004;60:415–22.
- Kiarie E, Romero L, Ravindran V. Growth performance, nutrient utilization, and digesta characteristics in broiler chickens fed corn or wheat diets without or with supplemental xylanase. *Poult Sci* 2014;93:1186–96.
- Langhout D, Schutte J. Nutritional implications of pectins in chicks in relation to esterification and origin of pectins. *Poult Sci* 1996;75:1236–42.
- Lee S, Apajalahti J, Vienola K, González-Ortiz G, Fontes C, Bedford M. Age and dietary xylanase supplementation affects ileal sugar residues and short chain fatty acid concentration in the ileum and caecum of broiler chickens. *Anim Feed Sci Technol* 2017;234:29–42.
- Malone G, Chaloupka G, Saylor W. Influence of litter type and size on broiler performance: 1. Factors affecting litter consumption. *Poult Sci* 1983;62:1741–6.
- Marcotuli I, Hsieh YS-Y, Lahnstein J, Yap K, Burton RA, Blanco A, Fincher GB, Gadaleta A. Structural variation and content of arabinoxylans in endosperm and bran of durum wheat (*Triticum turgidum* L.). *J Agric Food Chem* 2016;64:2883–92.
- Morgan NK, Keerqin C, Wallace A, Wu S-B, Choct M. Effect of arabinoxyloligosaccharides and arabinoxylans on net energy and nutrient utilization in broilers. *Anim Nutr* 2019;5:56–62.
- Nguyen HT, Bedford MR, Wu S-B, Morgan NK. Soluble non-starch polysaccharide modulates broiler gastrointestinal tract environment. *Poult Sci* 2021;101:183.
- Scott RW. Colorimetric determination of hexuronic acids in plant materials. *Anal Chem* 1979;51:936–41.
- Short F, Gorton P, Wiseman J, Boorman K. Determination of titanium dioxide added as an inert marker in chicken digestibility studies. *Anim Feed Sci Technol* 1996;59:215–21.
- Svihus B, Choct M, Classen HL. Function and nutritional roles of the avian caeca: a review. *World Poult Sci J* 2013;69:249–64.
- Theander O, Aman P, Westerlund E, Andersson R, Pettersson D. Total dietary fiber determined as neutral sugar residues, uronic acid residues, and Klason lignin (the Uppsala method): collaborative study. *J AOAC Int* 1995;78:1030–44.