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Effect of different dietary fibre raw material sources on production and gut development in fast-growing broilers

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Abstract

Many studies have been published recently about the beneficial effect of different fibre sources in a broiler diet. To assess the effects of raw material sources available in southern Africa, a trial was done with four treatments: control diet, 2% sunflower hulls, 2% malt culms, and 0.8% of a commercial lignocellulose product. Using a completely randomised block design, each treatment had 24 repetitions (96 pens in total), with 48 birds per pen. The effects were measured weekly by assessing production parameters, gut development, and the humoral immune response. Production parameters were measured per pen, gut measurements were done on 12 birds per treatment each week, and humoral immune response on 24 birds per treatment at 32 days. The promising responses seen in previous studies on fibre sources such as sugar beet pulp and oat hulls were not fully repeated here with the local fibre sources. There were no statistical differences from the control group based on production parameters. Sunflower hulls produced a substantial improvement over the control group with regards to caecal and overall intestinal lengths at 7 d. There was a numerical improvement in gizzard weights at 7 d for the malt culms group. No marked differences were detected in serology. The results indicate that there could be merit in including sunflower hulls in the pre-starter period (days 0–7), although it did not translate to production advantages here. Different inclusion levels should be investigated.

Keywords: gut length, immunity, intestinal health, sunflower hulls

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Introduction

Dietary fibre has been recognized as an essential ingredient with beneficial effects on gut health and a balanced gut flora (Neufeld, 2010). Dietary fibre is a form of carbohydrate made up of the largely indigestible parts of plants, passing through the small intestine relatively unchanged (Stribling and Ibrahim, 2023). Chemically, it is considered as the sum of non-starch polysaccharides (NSP) and lignin (Theander *et al.*, 1994). The NSP can be divided into two sub-groups, i.e., water-soluble and insoluble fibre (Raninen *et al.*, 2011). The water-soluble fibre increases gut viscosity, resulting in a slower feed passage rate and is mostly fermented in the large intestine and caeca. The insoluble fibre increases the rate of feed passage and is non-fermentable (Sozcu, 2019; Rohe *et al.*, 2020). Lignocellulose largely consists of insoluble cellulose, hemicellulose, and phenolic lignin (Rohe *et al.*, 2020).

The non-fermentable fibre which is inert to digestive enzymes in the upper gastrointestinal tract (e.g., cellulose and lignin), plays an important role in intestinal transit. This fibre induces contractions, which facilitates peristalsis, and has a laxative effect by mechanical stimulation of the intestines, promoting mucous secretion (Stribling and Ibrahim, 2023). It also tends to stay in the upper gastrointestinal tract for longer, mechanically stimulating the gizzard muscles to develop (Jiménez-Moreno *et al.*, 2019).

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Fermentable fibre is fermented in the large intestine by bacteria, stabilizing the natural gut flora and assisting in the control of certain pathogens (Neufeld, 2010). Some of these fermentation by-products, such as butyric acid, can be an important energy source for colonocytes. Butyrate also has anti-inflammatory effects and can strengthen the mucosal barrier integrity by upregulating the expression of tight junction proteins (Van Immerseel *et al.*, 2010).

Every year an increasing number of studies are published in which the effect of different fibre sources is measured in broilers. Most of these studies have been done on fibre sources not readily available in sufficient quantity in Southern Africa, such as oat hulls and sugar beet pulp. For this study, the focus was thus on fibre more readily-available locally, i.e., sunflower hulls, malt culms, and a commercial lignocellulose product. Table 1 summarises different fibre sources tested in broilers, with the effect that these fibre sources had on different parameters. This indicated that sunflower hulls were a good option to trial to determine whether the results could be replicated locally with this readily-available product. Lignocellulose was chosen for its lower health risk and the uniformity of the commercial product, even though there is not an abundance of published data. Malt culms were chosen due to availability, although its lower crude fibre content, compared to other fibre sources, probably excluded it from other trials. Considering the data available as well as local experiences, an inclusion of 2% (weight) was used for sunflower hulls and malt culms. The commercial lignocellulose product was included at the manufacturer's recommended level of 0.8%.

Table 1 Summary of the effect of different fibre sources on different parameters

Product	Gizzard weight	Gizzard pH	Body weight	FCR	Humoral immunity
Oat hulls	+[d]+[i]+[j]+[l]+[m]+[n]+[r]	+[d]+[l]+[m]+[n]+[o]	+[i]+[k]+[m]+[n]+[r]	+[i]+[k]+[m]+[n]	
Sunflower hulls	+[b]+[c]+[d]	+[b]+[d]	+[b]+[c]+[k]	+[b]+[k]	
Sugar beet pulp	+[m]+[n]	+[l]+[m]+[n]+[p]	+[m]+[n] -[f]-[h]	+[m]+[n] -[f]-[h]	+[f]+[h]
Soybean hulls	+[e]+[i]+[q]	+[o]	+[i]+[q]	+[i]	
Rice hulls	+[d]	+[d]	+[k] -[f]-[h]	+[h]+[k] -[h]	+[f]+[g]
Lignocellulose	+[b]		+[b]		
Wheat bran	+[a]				
Malt culms					

A "**+**" sign represents a study found to positively influence the parameter, while a "**-**" sign represents a negative influence. Empty blocks are indicative of a lack of available data to assess the parameter.

References are indicated by the following superscripts in the table: [a] (Shang *et al.*, 2020); [b] (Moradi *et al.*, 2020); [c] (Jamshidi and Moradi, 2020); [d] (Jiménez-Moreno *et al.*, 2019); [e] (Tejeda and Kim, 2020); [f] (Sadeghi *et al.*, 2015); [g] (Sabour *et al.*, 2019); [h] (Saadatmand *et al.*, 2019); [i] (Gonzalez-Alvarado *et al.*, 2007); [j] (González-Alvarado *et al.*, 2008); [k] (Jimenez-Moreno *et al.*, 2016); [l] (Jimenez-Moreno *et al.*, 2009a); [m] (Jimenez-Moreno *et al.*, 2010); [n] (Jimenez-Moreno *et al.*, 2009b); [o] (Jimenez-Moreno *et al.*, 2009c); [p] (Aziz-Aliabadi *et al.*, 2023); [q] (Tejeda Martinez, 2021); [r] (Rasool *et al.*, 2023)

There are many studies that indicate that fibre sources can improve production and gut development in fast-growing broilers, despite their low nutritive value (Table 1). These fibre sources improve the development of the upper gastrointestinal tract in particular and can also have an immunomodulatory effect by altering the gut microflora and its interaction with the gut-associated lymphoid tissues.

In addition to the parameters in Table 1, other parameters were also included in the current trial. Mortality rate was included due to its ease in measurement, together with the production parameters. Gizzard erosion scores were added to assess whether altered gizzard pH would influence the gizzard's integrity. Intestinal length was measured to macroscopically assess whether there are differences in development of the intestinal tract. Finally, bursal weights were taken to ensure that differing serological titres weren't due to differences in bursal health.

The objectives of this research were to measure the effect of no additional fibre (control), the inclusion of 2% sunflower hulls, 2% malt culms, and 0.8% of a commercial lignocellulose product on:

1. Production parameters: live body weight, average daily gain (ADG), feed conversion ratio (FCR) and mortality;

2. Intestinal health: gizzard pH, gizzard erosions, gizzard weights, total intestinal and caecal lengths;
3. The immune system: Bursa of Fabricius weights and enzyme-linked immunosorbent assay (ELISA) titres in response to Newcastle Disease Virus (NDV) vaccination.
4.

The overarching and main objective is to determine whether locally-available fibre sources could achieve the outcomes as mentioned above, since all the fibre sources listed in Table 1 are not readily-available on a commercial scale in this country.

Materials and Methods

Ethical clearance for this research was granted by the Research Ethics Committee of the University of Pretoria's Faculty of Veterinary Science (reference number REC206-21).

The trial pens were arranged in a line along the middle of a commercial broiler house, extending for approximately 70% of the length of the house. On either side of this line, extending along the whole house, were two separate camps also stocked with broilers of the same age, to mimic commercial full-house conditions in the industry. Litter for this trial consisted of pine shavings. The trial house consists of 96 pens, each able to house 48 birds. This provided a total of 4 608 birds. Day-old chicks (n = 4 608) were sexed with only males used in the trial to limit variability. The parents were a 46-week old, Ross 308 flock. All birds received the same vaccination program:

- Hatchery administration
  - Live NDV vaccine, sprayed
  - HVT-vectored Newcastle Disease vaccine, subcutaneous injection
  - Live Infectious Bursal Disease (IBD) vaccine, subcutaneous injection
  - Live Mass-type and 793B-type Infectious Bronchitis vaccines, sprayed
- Day 10 on-farm administration
  - Live 793B-type Infectious Bronchitis vaccine, sprayed

All feed was manufactured by Simple Grow in Centurion, South Africa. The control diet had no additional fibre sources included. In the sunflower hull diet, the sunflower hulls were unground at a 2% inclusion, with a crude fibre value of 49.8%. For the malt culm diet, the malt culms were also unground at a 2% inclusion, with a crude fibre value of 15%. The latter is a by-product of the beer fermentation process. In the fourth group, a commercial lignocellulose product was used, which was homogenous, of consistent quality, with a consistent supply throughout all seasons. The lignocellulose product had a standard combination of fermentable and non-fermentable fibres. The non-fermentable fibre had a physical action on the gastrointestinal tract, whereas the fermentable fibre served as a food source for bacteria that produce short chain fatty acids, especially butyrate. This product was included at the recommended 0.8%, with a crude fibre value of ~59%. It was a natural lignocellulose product made from fresh wood, with the crumbles of 0.4–1.6 mm.

All four rations were formulated to be isocaloric, with each fibre product included on their full matrix values, not added on top. These inclusions applied to all five phases within a ration: pre-starter (0–7 d), starter (7–14 d), grower (14–21 d), finisher (21–28 d), and post-finisher (28–32 d). The formulated dietary values for all diets are indicated in Table 2. The crude fibre values were the only major differences (Table 3).

Table 2 Formulated percentage values for each major dietary parameter

	Pre-starter	Starter	Grower	Finisher	Post-Finisher
Moisture	10.94	11.15	11.08	10.94	10.89
Protein	22.86	21.25	20.23	19.23	19.30
Fat (EE)	5.09	5.17	5.65	6.68	7.20
Fat (AH)	5.77	5.73	6.23	7.26	7.78
Calcium	0.94	0.87	0.76	0.68	0.68
Phosphorus	0.64	0.51	0.43	0.35	0.35
Sodium	0.17	0.15	0.14	0.14	0.14
Chloride	0.30	0.30	0.25	0.25	0.25
Potassium	0.86	0.87	0.83	0.78	0.79

EE – Ether Extract; AH – Acid Hydrolysis

Table 3 Percentage crude fibre values for each broiler phase-fed ration

	Pre-starter	Starter	Grower	Finisher	Post-Finisher
Control	3.85	3.91	3.75	3.57	3.58
Sunflower hulls	4.82	4.88	4.72	4.54	4.54
Malt culms	4.05	4.11	3.94	3.77	3.77
Lignocellulose	4.31	4.37	4.21	4.03	4.04

All 4 rations had Flavophospholipol (Biesterfeld, Hamburg, Germany) included at 12 ppm in all five phases for clostridial control, as well as Lasalocid (Zoetis, NJ, USA) at 75 ppm for coccidiosis control.

The trial was divided into four groups with each of the 96 pens representing one treatment. Each group thus had 24 repetitions, totalling 1 152 birds per group. Each pen was stocked with 51 chicks at placement, reduced to 48 chicks at 7 d. This was done to improve uniformity by removing the non-starter or weakest birds. The final stocking density was thus 24 birds per m^2, mimicking the highest stocking densities experienced under commercial conditions in southern Africa. A completely randomised block design was used to assign treatments to different pens, generated in Microsoft Excel.

Production parameters were measured every seven days (7, 14, 21, and 28 d of age) and again on the day before slaughter (32 d of age). All parameters were measured per pen, which equates to 24 repetitions per group. The birds' growth was assessed by totalling the live weight of all birds in a pen weekly. Average daily gain (ADG) was calculated with the total weight per pen being divided by the number of birds, and then the number of days in the specified period. Weights were also recorded for each mortality. This data was not used in the calculations for live body weight or ADG but was used for the calculation of mortality-corrected FCR (described below).

The total weight of the feed used per period was measured for each pen. The mortality-corrected feed conversion ratio (FCR) was calculated by dividing the total feed used per week by the total weekly weight (live & mortalities) gain.

The total amount of birds that died per week was stated as a percentage of the initial number of birds placed per pen, which was 51 birds. The mortality rates in the trial were artificially elevated due to the initial selection in week 1 and weekly culling of birds for gut measurements. This was equivalent to one bird being removed from every second pen each week (12 birds per treatment, each week).

Birds were sacrificed every 7 d, with the parameters discussed below being measured on the gastrointestinal tract during post-mortem evaluation. One bird per pen was sacrificed weekly from every second block. The blocks sampled were alternated each week.

The gizzard pH was measured using a Hanna HI 98190 pH waterproof portable meter. Upon culling, the gizzard was the first organ to be opened, with the meter's tip put directly into the gizzard and its contents. Elanco Animal Health's 2010 Broiler Disease Guide was used to assess the gizzard erosions (Elanco, 2010).

The scoring system is described as:

- Score 0 indicates a normal smooth lining
- Score 1 indicates roughening of the gizzard lining
- Score 2 indicates erosions of the gizzard lining
- Score 3 indicates erosions extending into the mucosal area.

Gizzards were removed *in toto*, rinsed free from contents, drip dried, and then weighed individually.

The intestine was measured, stretched from the proximal duodenum to the distal colon. The entire length of the intestine was straightened in order to simplify measuring. This included breaking the duodenal loop's attachments to the pancreas. In order to compare the intestinal lengths of different-sized birds, the lengths in cm (centimetres) were expressed as ratios of body weight. The lengths of both caeca were measured in cm, and the average per bird used for statistical analysis. Bursa of Fabricius weights were recorded weekly on the same birds sacrificed for the gut measurements. In order to compare the bursas of different-sized birds, the bursa weights were expressed as ratios of body weight.

Serology was done by means of BioChek's ELISA kit for NDV (Reeuwijk, Netherlands), performed at CAL (Central Analytical Laboratories) in Roodepoort, South Africa. Blood samples (2 mL) were collected by venipuncture of the wing vein on the day before slaughter from 1 bird per pen. This was equivalent to 2% of the birds being sampled in total, or 24 repetitions per treatment.

Data were recorded manually and transferred to Excel spreadsheets. Each outcome (e.g., live body weight, ADG) was compared for each of the treatment groups against the control group at time points (age) using linear mixed models with fixed effects for group, age, block, and a group × age interaction, a random effect for pen, and a Bonferroni adjustment for multiple comparisons. The $\log_{10}$-transformed NDV titres were compared between groups using analysis of variance. The significance level was set at 0.05. Statistical analyses were done using Stata 17.0 (StataCorp, College Station, TX, USA). Comparisons were thus only done between each group and the control, with no between-group comparisons.

Results and Discussion

Mean values are indicated in the figures. ‘Sunflower hulls’ is abbreviated as ‘sun hulls’ in all figures. The commercial lignocellulose group is referred to as “Opticell” in all figures.

For live body weights per week, no statistically significant differences were observed, but the malt culms group showed numerically lower weights at 21 d and 32 d.

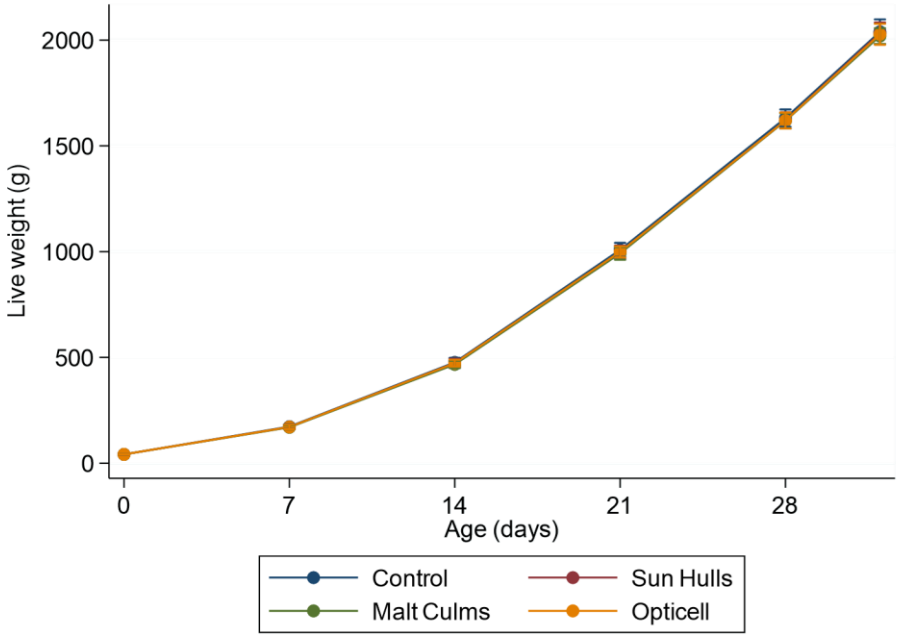

Figure 1 Live body weights per week in broilers fed diets containing different fibre sources

For average daily gains per week, no statistically significant differences were observed, but the malt culms group showed a numerically lower ADG for the fifth week.

For feed conversion ratio per week, no statistically significant differences were observed, but the lignocellulose group showed a numerically higher FCR for the first week.

No statistically significant differences were observed for weekly mortality percentages.

The lignocellulose group had a gizzard pH (3.07) that was higher than the control group at 28 d (*P* = 0.0053). The sun hulls group showed a numerically higher pH at 28 d.

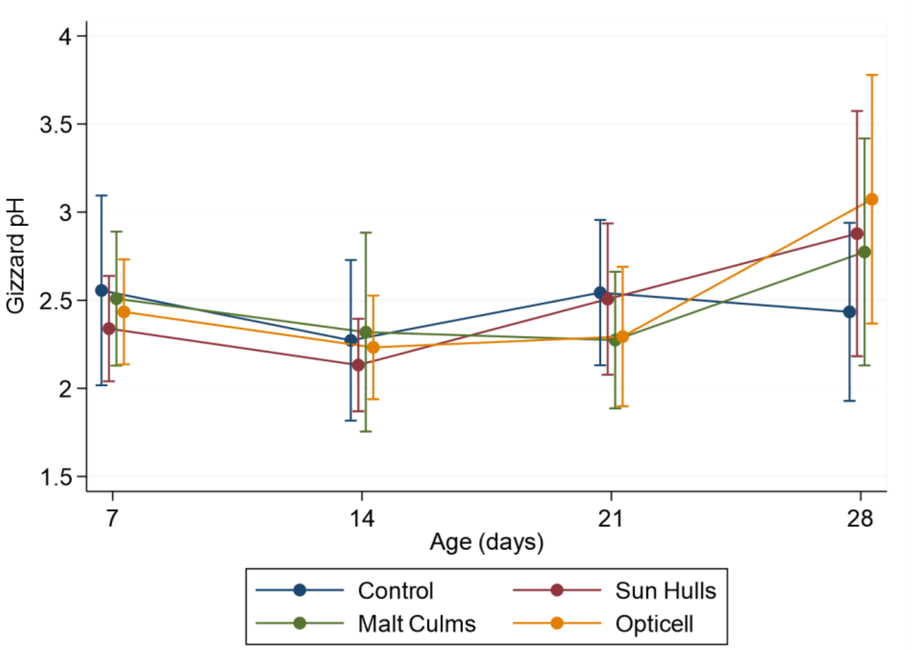

Figure 2 Gizzard pH per week in in broilers fed diets containing different fibre sources

No statistically significant differences were observed for gizzard erosion scores.

For gizzard weight to body weight ratios, no statistically significant differences were observed, but the malt culms group showed a numerically higher ratio at 7 d.

The sun hulls group had a total intestinal length to body weight ratio that was higher than the control group at 7 days ($P = 0.02$). The lengths over all treatments for different ages:

- d7, 98–132 cm
- d14, 117–184 cm
- d21, 155–200 cm
- d28, 163–219 cm

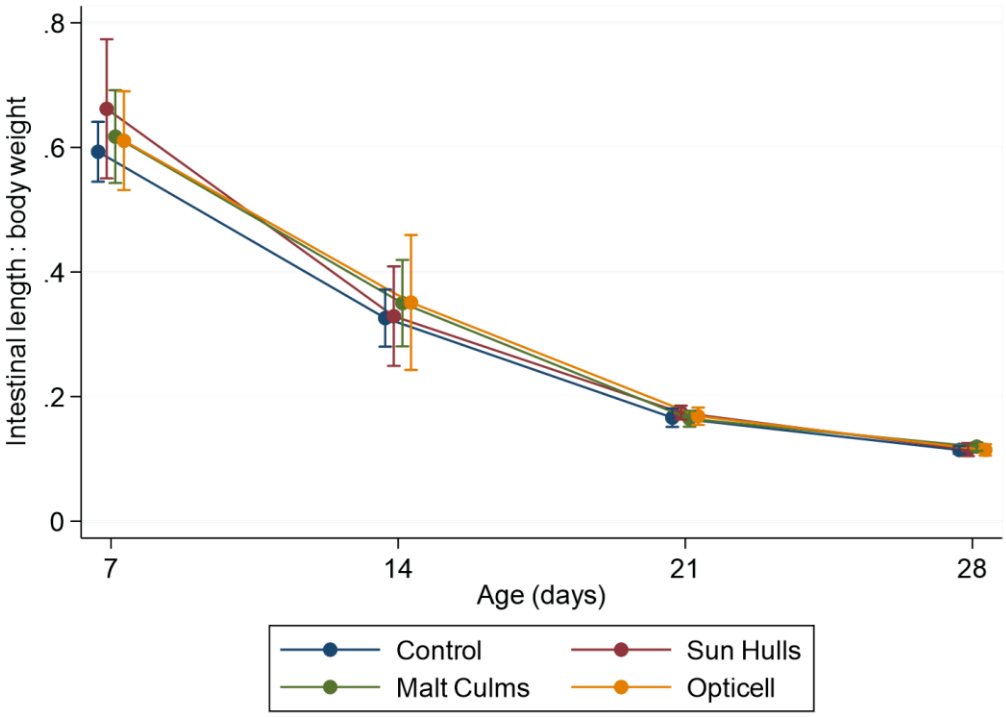

Figure 3 Intestinal length:body weight ratios per week in in broilers fed diets containing different fibre sources

Similar to the total intestinal length data, the sun hulls group also had a caecal length ratio that was higher than the control group at 7 d ($P = 0.03$). The malt culms group had a numerically higher ratio at 14 d. The caecal lengths over all treatments for different ages:

- d7, 8–11 cm
- d14, 9–14 cm
- d21, 12–18 cm
- d28, 14–20 cm

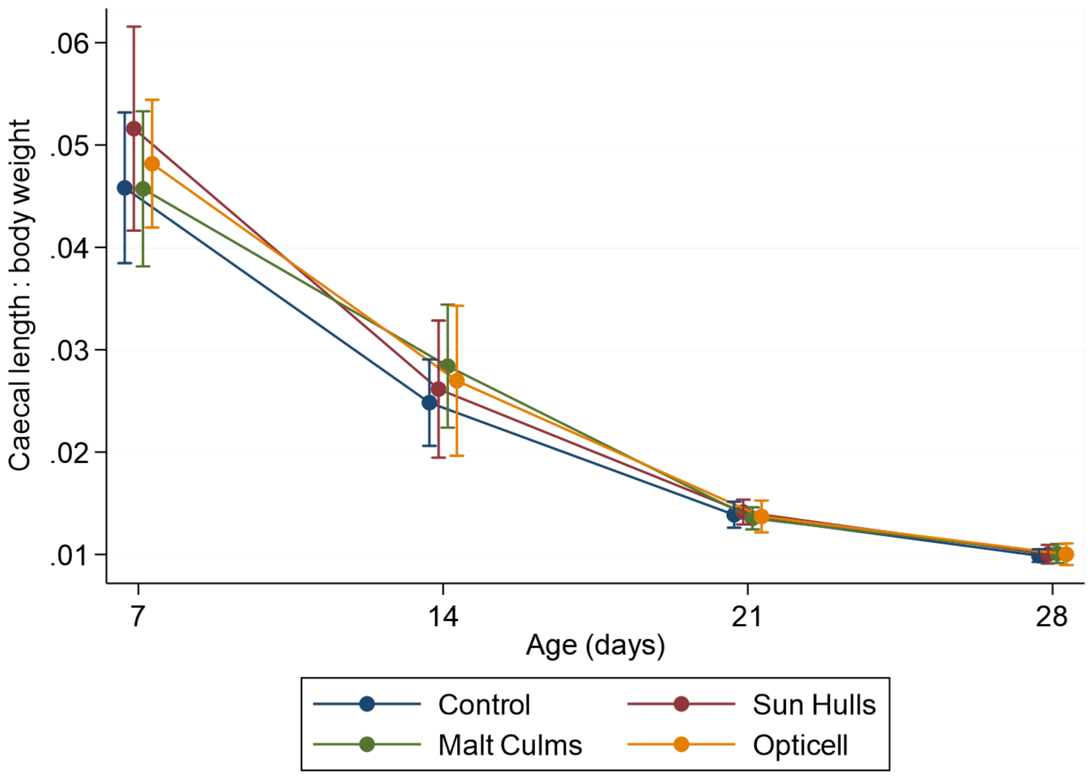

Figure 4 Caecal length:body weight ratios per week in in broilers fed diets containing different fibre sources

For bursa weight to body weight ratios and Newcastle Disease Virus (NDV) titres, no statistically significant differences were observed. The median titre for the control group was 873.

Only two parameters showed statistical differences compared to the control group. Firstly, the sun hull group had proportionately longer intestines (including the separately-measured caecal parameter) at 7 d. Secondly, the lignocellulose group had a higher gizzard pH at 28 d. The sun hull group's longer intestines at 7 d indicates improved early development of the intestinal tract. It is expected that a better-developed intestinal tract could lead to improved nutrient absorption and thus feed efficiency. It was therefore surprising that growth and FCR showed no improvement in this group. Tejeda & Kim's comments (2021) could explain this as they reported that increased villi height, and thus absorptive surface, does not necessarily lead to the expected production advantage. It was proposed that the increased villi height could represent an increased burden on nutrient requirements due to a higher cell turnover, which negates any possible advantage.

The range of intestinal lengths measured at different ages could serve as a baseline and reference. The lengths seen at 7, 14, 21, and 28 d compares well with Ross 308 data in 2017, which showed total intestinal lengths of ~210 cm at 42 d of age and a 2 101 g body weight (Kokoszynski *et al.*, 2017). The individual caecal lengths compared well with Kokoszynski's data, which reported ~20.5 cm at 42 d of age.

The lignocellulose group had a statistically higher gizzard pH at 28 d, with the sun hulls group showing a numerically higher pH at the same age. When considering the other trials' results (Table 1), a higher pH did not promote performance. There were wide differences between ages and even within groups, which suggests that statistical differences would be hard to achieve. Only the control group's gizzard pH remained relatively constant with means of 2.27–2.56 over the four measurement periods. This compares with Lee's pH measurements in broilers of 1.99–2.29 (Lee *et al.*, 2021). Nishi (2016), however, reported a pH of 3.51 at 28 d. These different ranges could well be attributed to differences in dietary composition and physiological differences between individual birds.

Although not statistically significant, the malt culms group showed a numerically higher gizzard to body weight ratio at 7 d, numerically lower body weights at 21 and 32 d, and a lower ADG for the 28–32-d period. The crude fibre value of malt culms was 15%, compared with 50% and 59% for sunflower hulls & lignocellulose, respectively. This may explain the slight differences observed and, in retrospect, would exclude malt culms from future trials for the purpose of adding dietary fibre.

Considering the bursa weight and NDV titres, no immunological effect could be observed. As different levels of dietary fibre supplementation could lead to microbiota changes (Qiu *et al.*, 2022), it was expected that a positive immunomodulatory effect could be seen if the microbiota changes led to improved health of the birds. Effects on gut health related to the microbiota could have been limited due to the insolubility of a fibre source such as sunflower hulls, which should have limited effects on the intestinal microflora.

This trial did not take into account the interaction of dietary fibre with different inclusions of fat, enzymes, and dietary organic acids, as have been done in other trials (Table 1). Furthermore, the potential need for additional amino acids and energy to compensate for increased intestinal growth due to fibre addition (Tejeda and Kim, 2021) was also not considered in formulation of these diets. The main objective was to determine whether local fibre sources could achieve similar positive effects and which fibre source would be best suited for further trials.

Conclusion

Previous trials with sunflower hulls and other fibre sources, such as oat hulls, sugar beet pulp, and rice hulls, showed higher gizzard weights, lower gizzard pH, lower FCR, and improved humoral immunity. These promising responses seen in other trials were not observed in the current trial.

Proportional intestinal length was the only parameter which showed potential, specifically in the sunflower hulls group. Owing to improved early development of the intestinal tract in this group at a 2% inclusion, there may be merit in including sunflower hulls in the pre-starter period (days 0–7). Different inclusion levels could be investigated and may show improved production performance which wasn't observed at a 2% inclusion. An inclusion of 1% may be sufficient to show some advantage without changing the base diet much but could be too low to show significant effects. An inclusion level of 3% may be cost-prohibitive on a commercial scale, due to costly ingredients required to keep the diets isocaloric. The commercial lignocellulose product could be investigated at similar inclusion levels to the sunflower hulls. Its recommended inclusion level of 0.8% may be too low to show significant effects.

The main objective of investigating locally-available dietary fibre sources was achieved, with sunflower hulls showing the most potential for future trials, wherein different inclusion levels should be investigated in the pre-starter period.

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Authors' contributions

ABF collaborated in the design of the experiment, collected the data for this study, collaborated in the interpretation of the results, and wrote the initial draft of this manuscript. DBRW collaborated in the design of the experiment, collaborated in the interpretation of the results, and revised the manuscript.

Conflict of interest declaration

The authors declare they have no conflicts of interest that are directly or indirectly related to the research.

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