Animal Nutrition xxx (2016) 1–9

Contents lists available at ScienceDirect

Animal Nutrition

journal homepage: http://www.keaipublishing.com/en/journals/aninu/

Original Research Article

Reducing agent and exogenous protease additions, individually and in combination, to wheat- and sorghum-based diets interactively influence parameters of nutrient utilisation and digestive dynamics in broiler chickens

Peter H. Selle [a,*], Ha H. Truong [a,b], Leon R. McQuade [c], Amy F. Moss [a], Sonia Yun Liu [a]

[a] Poultry Research Foundation, The University of Sydney, Camden 2570, NSW, Australia
[b] Poultry CRC, University of New England, Armidale, 2351, NSW, Australia
[c] Australian Proteome Analysis Facility, Macquarie University, Sydney 2109, NSW, Australia

ARTICLE INFO

Article history:
Received 22 June 2016
Received in revised form
26 July 2016
Accepted 1 August 2016
Available online xxx

Keywords:
Amino acids
Broiler chickens
Protease
Sodium metabisulphite

ABSTRACT

The objective of the study was to investigate the possibility that tandem inclusions of a reducing agent and a protease may advantage chicken-meat production and to ascertain if the established benefits of including sodium metabisulphite in sorghum-based diets extend to wheat-based diets. The study comprised a 2 × 2 × 2 factorial array of treatments in which either nutritionally iso-nitrogenous and iso-energetic wheat- or sorghum-based diets, without and with sodium metabisulphite (2.75 g/kg), without and with protease (1,000 units/kg) were offered to broiler chickens from 7 to 28 days post−hatch. The effects of dietary treatments on growth performance, nutrient utilisation, protein (N) and starch digestibility coefficients and digestive dynamics were determined. A preliminary investigation into the effects of two treatments on concentrations of free amino acids and glucose in the portal circulation was conducted. There was significant feed grain by sodium metabisulphite interactions ($P = 0.03$ to 0.005) for parameters of nutrient utilisation (AME, ME:GE ratios, N retention, AMEn). For example, sodium metabisulphite inclusions in sorghum-based diets enhanced AME by 0.18 MJ (12.47 versus 12.29 MJ/kg) but depressed AME by 0.43 MJ (11.88 versus 12.31 MJ/kg) in wheat-based diets. There was a linear relationship between starch:protein disappearance rate ratios in the distal ileum with weight gain ($r = -0.484$; $P = 0.0012$) indicating that condensed ratios (or absorption of more protein relative to starch) advantaged growth performance. Concentrations of free amino acids in the portal circulation or the post-enteral availability of certain amino acids, including the branched-chain amino acids, methionine, phenylalanine and threonine, were significantly correlated to FCR. For example, threonine concentrations were negatively correlated to FCR ($r = -0.773$; $P = 0.005$). Finally, tandem inclusions of sodium metabisulphite and protease in sorghum-based diets may hold merit but it appears that the established 'energy sparing' effects of sodium metabisulphite inclusions in sorghum-based diets are not duplicated in wheat-based diets.

1. Introduction

The inclusion of the reducing agent sodium metabisulphite in sorghum-based broiler diets has been shown to enhance energy utilisation in broiler chickens (Liu et al., 2014; Selle et al., 2013a; 2014; Truong et al., 2016). For example, Truong et al. (2016) reported that 3.50 g/kg sodium metabisulphite increased N-corrected apparent metabolisable energy (AMEn) by 0.42 MJ from 11.43 to 11.85 MJ/kg in broiler diets based on five different sorghum

* Corresponding author.
E-mail address: peter.selle@sydney.edu.au (P.H. Selle).

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

Production and Hosting by Elsevier on behalf of KeAi

http://dx.doi.org/10.1016/j.aninu.2016.08.001

varieties. This 'energy sparing' effect could be attributed to the sulphite reducing agent via either the depolymerisation of starch polysaccharides by oxidative–reductive reactions (Paterson et al., 1996, 1997) and/or the reduction of disulphide cross-linkages in protein. In respect of the latter, the reduction of disulphide bonds in the periphery of kafirin protein bodies and the mitigation of starch–protein interactions in sorghum endosperm may hold particular relevance (Taylor and Emmambux, 2010).

In one feeding study completed at this institution, supplementation of a sorghum-based broiler diet with another exogenous protease displayed promise. This protease increased protein (N) and, to a lesser extent, starch digestibility coefficients (Selle et al., 2013b) and increased amino acid digestibility coefficients and the digestion rates of the majority of amino acids assessed (Liu et al., 2013). However, exogenous proteases hydrolyse proteins and almost certainly lack the capacity to reduce disulphide bonds. Thus, the assumption was made that inclusions of a protease and a reducing agent in tandem may be advantageous in sorghum-based broiler diets because of the distinct modes of action in their digestion of protein.

In Australia, wheat is more commonly used than sorghum as a feed grain in chicken-meat production. However, it has not been demonstrated if the advantages observed pursuant to the inclusion of sodium metabisulphite in sorghum-based diets extend to wheat-based diets. Thus the prime objectives of this study were two-fold. One was to investigate the possibility that inclusions of a reducing agent in wheat-based diets would be of benefit. The other was to explore the possibility that tandem inclusions of a reducing agent and an exogenous protease in wheat- and sorghum-based diets would be advantageous. Additionally, concentrations of free amino acids and glucose in the portal circulation for 2 treatment groups (wheat-based diets without and with sodium metabisulphite plus protease) were determined as a preliminary investigation.

2. Materials and methods

Wheat, sorghum and soybean meal were characterised and iso-nitrogenous and iso-energetic wheat- and sorghum-based diets were formulated to recommended nutrient specifications for Ross 308 grower (11 to 24 days) as shown in Table 1. Sodium metabisulphite was substituted for sodium bicarbonate in the 2 basal diets in order to maintain dietary sodium levels at 1.80 g/kg and adjustments for this and protease addition was made at the expense of Celite. The experimental diets were steam-pelleted at a temperature of 80 °C with a 14 s residence time in the conditioner, and crumbled after passing through a vertical cooler. The steam-pellet press (Palmer PP330 pellet press; Palmer Milling Engineering, Griffith, NSW, Australia) contained a 4.0 mm die. Wheat, sorghum and soybean meal had been ground through a 3.2 mm hammer-mill screen prior to being mixed into the diets. The trial design consisted of a 2 × 2 × 2 factorial arrangement of dietary treatments: wheat or sorghum-based diets, without or with 2.75 g/kg sodium metabisulphite, without or with exogenous protease. The selected enzyme was a serine protease [Ronozyme ProAct (CT); DSM] produced by a genetically modified strain of *Bacillus licheniformis*. The feed enzyme addition rate of 500 g/kg provided 1,000 units of protease activity per kg of feed.

A total of 288 feather-sexed, male Ross 308 chicks were housed in an environmentally-controlled facility with unlimited access to feed and water under a "18 h on-6 h off" lighting schedule. There were 6 replicate cages (6 birds per cage) for each of the 8 dietary treatments and birds were initially offered a proprietary wheat-based starter diet prior to being allocated to dietary treatments from 7 to 28 days post–hatch. At 7 days post–hatch birds were weighed and allocated into 48 cages so that the mean and standard deviation of body-weight for each cage was nearly identical. Body weights were again determined at 28-days post hatch and caged feed intakes recorded over the entire 21-day period to calculate FCR with adjustments made from the weight of any dead or culled birds, which were monitored on a daily basis. At 28-days post hatch the birds were euthanised (intravenous injection of sodium pentobarbitone) and digesta samples were collected in their entirety from the proximal jejunum, distal jejunum, proximal ileum and distal ileum and pooled for each cage. This was in order to determine nutrient (starch and crude protein) digestibility coefficients and nutrient disappearance rates [g/(bird·day)].

Total excreta collection over a 48 h period at 25 days post–hatch was used to determine apparent metabolisable energy (AME) on a dry matter basis, metabolisable energy to gross energy (ME:GE) ratios, nitrogen (N) retention and AMEn. Total excreta were quantitatively collected from each cage and feed intakes recorded for the 48 h collection period. Excreta were dried in a forced-air oven at 80 °C for 24 h and the GE of excreta and diets were determined using an adiabatic bomb calorimeter. The AME values of the diets on a dry matter basis were calculated from the following equation:

Table 1
Composition and nutrient specifications of the basal wheat- and sorghum-based diets (as is basis).

Item	Wheat-based diet[4]	Sorghum-based diet[5]
Ingredient, g/kg		
Wheat	638.2	–
Sorghum	–	682.3
Soybean meal	255.0	235.0
Canola oil	45.0	20.0
Limestone	13.0	13.0
Dicalcium phosphate	12.0	12.0
Sodium bicarbonate[1]	6.0	5.7
Lysine HCl	3.4	4.5
Methionine	2.6	3.0
Threonine	1.6	1.6
Arginine	5.6	5.0
Choline chloride	0.6	0.9
Vitamin-trace mineral premix[2]	2.0	2.0
Celite[3]	15.0	15.0
Nutrient level, g/kg		
Metabolisable energy, MJ/kg	13.06	12.92
Protein	215.1	215.7
Calcium	8.05	7.81
Total phosphorus	6.54	6.22
Available phosphorus	4.05	3.85
Sodium	1.79	1.80
Digestible amino acids		
Lysine	11.40	11.50
Methionine	5.40	5.78
Methionine + cystine	8.61	8.66
Tryptophan	2.13	2.13
Arginine	17.2	15.6
Threonine	7.68	7.68
Leucine	12.6	17.5
Isoleucine	8.18	8.31

[1] Sodium metabisulphite (2.75 g/kg) replaced 2.34 g/kg sodium bicarbonate to maintain Na levels, difference corrected with Celite.
[2] Vitamin-trace mineral premix supplied per tonne of feed; [million international units, MIU] retinol 12, cholecalciferol 5, [g] tocopherol 50, menadione 3, thiamine 3, riboflavin 9, pyridoxine 5, cobalamin 0.025, niacin 50, pantothenate 18, folate 2, biotin 0.2, copper 20, iron 40 manganese 110, cobalt 0.25, iodine 1, molybdenum 2, zinc 90, selenium 0.3.
[3] Protease added at the expense of Celite.
[4] On analysis, wheat-based diets contained 456 g/kg starch and 205 g/kg protein (N).
[5] On analysis, sorghum-based diets contained 501 g/kg starch and 196 g/kg protein (N).