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Heat-processed bovine blood–rumen digesta meal and vegetable oil concentrate as partial replacement for soybean meal in broiler finisher diet

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ABSTRACT

Context. Blood and rumen contents, which are currently considered wastes in many abattoirs, could be valuable ingredients in poultry feeding; however, several factors including high fibre and low energy may limit their dietary inclusion. There is need for more research on feed processing and diet formulation to maximise utilisation of these by-products in poultry feeding. **Aim.** We investigated the effect on broiler finisher performance of feeding bovine blood, rumen digesta and vegetable oil concentrate as a partial replacement for soybean meal. **Methods.** Three isonitrogenous and isocaloric diets were formulated: a control diet based on maize and soybean meal, and two test diets. In the test diets, heat-processed blood–rumen digesta meal and vegetable oil concentrate replaced 50% of the soybean meal, with and without exogenous enzymes. Diets were allocated to five replicate pens containing six Ross broilers aged 21 days in a completely randomised design for a period of 21 days. **Key results.** Results showed no treatment effect ($P > 0.05$) on average daily feed intake but birds fed the test diets gained more weight ($P < 0.05$) and converted their feed more efficiently ($P < 0.05$) than the control group. Enzyme supplementation had no additional effect ($P > 0.05$) on growth performance. There were no dietary effects ($P > 0.05$) on relative weights of carcass and gut segments. Feed cost of liveweight gain and carcass weight was reduced ($P < 0.05$) on the test diets. **Conclusion.** Heat-processed blood–rumen digesta meal and vegetable oil concentrate can replace up to 50% of soybean meal in broiler fishier diets. At this level of replacement, enzyme supplementation is not required. **Implications.** The use of blood and rumen content in poultry diets has cost and environmental benefits.

Keywords: abattoir by-products, alternative ingredients, antinutritional factors, broiler performance, feed analysis, feed cost, feed formulation, feed processing.

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Introduction

Rumen digesta (RD, partially digested feed in the rumen) and blood are two important by-products of ruminant slaughter, which remain underutilised in many parts of the world, posing disposal concerns (Al Mamun *et al.* 2018). RD is an average source of protein but is high in fibre and low in energy (Mishra *et al.* 2015; Bekele *et al.* 2020) and may contain antinutritional factors associated with fodders consumed by the animal (Elfaki and Abdelatti 2016). Blood is a rich protein source high in lysine, methionine and cysteine (Prata and Sgarbieri 2008; Adeniji 2013). Poor palatability is the main limitation to using blood meal in animal feed; this can be overcome by feeding it with other protein sources (Dafwang *et al.* 1986). There are reports on the inclusion of blood–RD meal in broiler diets, but many of these studies did not formulate isonitrogenous and isocaloric diets (Adeniji and Balogun 2001; Oyedeji *et al.* 2015; Efrem *et al.* 2016; Yitbarek *et al.* 2016). In addition, several factors including diet composition, feed processing and class of birds may affect utilisation by poultry. Studies with broilers have reported beneficial effects of supplementation of blood–RD meal-based diets with yeast (Oyedeji *et al.* 2015) and cellulase (Mandey *et al.* 2015).

However, information on feeding of blood–RD meal-based diets with complex enzyme products is limited.

This study investigated the effect of replacing a portion of dietary soybean meal with heat-processed bovine blood–RD meal and vegetable oil concentrate in isocaloric and isonitrogenous broiler finisher diets, and the effect of complex enzyme supplementation. We tested the following hypotheses: (1) heat-processed bovine blood–RD meal and vegetable oil concentrate can replace half of dietary soybean meal in protein and energy balanced diets for finishing broilers; and (2) complex enzyme supplementation will improve the performance of birds fed the test diet.

Table 1. Analysed proximate composition of the blood meal, rumen digesta (RD) meal and soybean meal (SBM), and apparent metabolisable energy of RD meal.

Constituent	Blood meal	RD meal	Soybean meal
Dry matter (g/kg)	926	891	892
Crude protein (g/kg DM)	892	187	392
Either extract (g/kg DM)	29	26	63
Crude fibre (g/kg DM)	8	212	40
Ash (g/kg DM)	154	42	7.3
Apparent metabolisable energy (MJ/kg)	–	5.6	–

Materials and methods

The study was conducted at the University of the South Pacific (USP) Livestock Farm, Samoa Campus. The research protocol was approved by the Animal Ethics Committee of the USP.

Experimental site, source and processing of blood and rumen digesta

Blood and RD obtained from slaughtered cattle by the Mobile Slaughter Unit of the Animal Production and Health Department, Ministry of Agriculture and Fisheries, Samoa, were processed for the experiment. The materials were collected immediately after cattle slaughter, into plastic containers with lids, and delivered to the USP, Samoa Campus Farm. The RD and blood were processed via modified methods of Yitbarek *et al.* (2016). Fresh RD was spread on a Hessian sac, covered with aluminium sheet and placed on wire mesh over boiling water in a petroleum drum for 40 min to allow uniform distribution of moist heat through the sample. Fresh blood was mixed with an equal volume of boiling water in a petroleum drum and allowed to boil for another 30 min with constant stirring. Heated RD and blood were then sun-dried for 3 days to a moisture content ~11% and ground in a hammer mill through a 3-mm sieve to obtain RD and blood meals. Samples of soybean meal, blood meal and RD meal were analysed for proximate composition at the USP Samoa Campus Central Laboratory (Table 1).

Determination of apparent metabolisable energy (AME)

Owing to unexpected early mortality of the broiler chicks, an AME study to determine the metabolisable energy of the RD was conducted on old layers. For the AME study, 25% of a commercial layer mash was replaced with RD. Diatomaceous earth was added to each of the two diets (with or without 25% replacement with RD) at 5 g/kg as an insoluble marker.

Sixteen laying hens of similar weight (1850 ± 47 g) were placed in standard two-bird laying cages (30 cm by 42 cm) fitted with feeders, drinkers and excreta collection trays. Care was taken to avoid excreta contamination between cages. Each diet was made available, on *ad libitum* basis, to four replicate cages in a completely randomised design. The birds were adapted to the cages for 4 days and excreta collected from each cage daily for 3 days. Feathers, feed particles and other contaminants were removed and samples frozen immediately until analysis. Samples from replicate cages were pooled, mixed thoroughly and subsampled prior to gross energy (GE) and acid-insoluble ash (AIA) analyses. AME was calculated using the formula of Robbins and Firman (2006) as:

$$\mathrm{AME} = \mathrm{GE}_{\mathrm{diet}} - \left(\mathrm{GE}_{\mathrm{excreta}} \times \left(\frac{\mathrm{AIA}_{\mathrm{diet}}}{\mathrm{AIA}_{\mathrm{excreta}}}\right)\right)$$

Experimental diets and birds

Three diets were formulated: a basal diet based on soybean meal; and two test diets containing blood–RD and vegetable oil (BRDO) concentrate replacing soybean meal at 50%, with and without Challenzyme 1309A (Beijing Challenge International Trade Co., Beijing, China) (Table 2). The proportion of BRDO in the concentrate was calculated to contain protein and ME contents similar to soybean meal, in order to separate any effects of possible antinutritional factors in RD from those of dietary energy and protein. The diets met or exceeded the requirements of Ross broilers (Aviagen 2019).

In total, 90 male Ross 308 broiler chicks aged 21 days were assigned to 15 open-sided floor pens (238 cm by 109 cm) for a 21-day experiment. Diets were allocated to five replicate pens containing six birds in a completely randomised design. Feed and water were provided *ad libitum* throughout the experimental period. The lighting program was 16 h light and 8 h dark.

Data collection

Data were collected on average daily feed intake (ADFI), average daily weight gain (ADWG), feed-to-gain ratio (F:G),

Table 2. Ingredient composition (g/kg as fed) and nutrient content of the control diet and experimental diets, in which soybean meal was replaced at 50% with blood–rumen digesta meal and oil concentrate (BRDO), with or without enzyme.

	Control	BRDO	BRDO + enzyme
Ingredients			
Maize	640	645	644.7
Wheat bran	25	25.1	25
Soybean meal	293.8	147	147
Rumen digesta	0.0	69	69
Blood meal	0.0	58.8	58.8
Vegetable oil	5.2	19.7	19.7
Limestone	11.0	11.0	11.0
Coral sand	18	18	18
Calcium hydrogen phosphate	10.0	10.0	10.0
NaCl	3.0	3.0	3.0
HCl-Lysine	5.0	4.4	4.4
DL-Methionine	4.0	4.0	4.0
Threonine	1.0	1.0	1.0
Challenzyme[A]	0.0	0.0	4.0
Premix[B]	2.0	2.0	2.0
Analysed values			
Crude protein	195	194	194
Crude fibre	45	64	64
Calculated values			
Calcium	8.2	7.8	7.8
Available phosphorus	3.6	3.7	3.7
Calcium:phosphorus	2.3	2.1	2.1
Lysine	10.1	10.3	10.3
Methionine	4.2	4.2	4.2
Metabolisable energy (MJ/kg)	12.9	12.7	12.7

[A]Challenzyme 1309A, with eight enzyme activities (U/g): β-glucanase 800, xylanase 15 000, β-mannanase 100, α-galactosidase 100, proteases 800, amylase 500, pectinase 500, and cellulase 300; and matrix values crude protein 3000%, lysine 270%, methionine 64%, tryptophan 90%, threonine 120%; metabolisable energy 1000 Mcal (4184 MJ)/kg.

[B]Contains (mg supplied per kg diet): retinol 6.71, cholecalciferol 0.134, α-tocopherol 23, niacin 27.5, thiamine 1.8, riboflavin 5, pyridoxine 3, cyanocobalamin 0.015, menadione 2, pantothenic acid 7.5, biotin 0.06, folic acid 0.75, choline chloride 300, cobalt 0.2, copper 3, iodine 1, iron 20, manganese 40, selenium 0.2, zinc 30, anti-oxidant 1.25.

carcass and gut weight and length, pH of gut content, and feed cost of meat production. Birds were weighed at the start and end of the experiment and weight gain calculated by difference. The quantities of feed supplied and left were weighed per pen and ADFI calculated by difference. F:G was derived as the ratio of ADFI to ADWG.

At the end of the experiment (at age 42 days), two birds weighing closest to the mean of each pen were selected for carcass and gut measurements (weight, length and pH). The birds were fasted overnight and killed the next morning by decapitation. Birds were scalded in hot water at 50°C for 30 s, plucked and eviscerated. The carcass and cuts (breast, thighs and drumsticks) were weighed and expressed as percentage of the liveweight of the bird. The cost of 1 kg of each diet was calculated based on the market price of the ingredients and feed cost of meat production (US$/kg carcass) derived as:

$$\text{Feed cost per kg liveweight} = \text{Cost per kg feed} \times \text{feed conversion ratio.}$$

$$\text{Feed cost per kg carcass} = \text{Feed cost per kg liveweight} \div \text{dressing percentage}$$

Gut segments (gizzard, liver, proventriculus and intestine) were weighed on a digital scale (Jadever JKH-500 series, Smartfox, Auckland, New Zealand), and intestine length was determined by using a measuring tape and expressed in relation to liveweight. The pH of the contents of the gizzard and caeca was taken with a digital pH meter (model IQ120, 2075E; Corte Del Nogal, Carlsbard, CA, USA).

Chemical analysis

Blood meal, DR meal, soybean meal and the diets were analysed for proximate composition following standard procedures (AOAC 1995). Dry matter (DM) was determined in a forced-air oven (103°C) for 24 h. Nitrogen was analysed by the Kjeldahl method (method 954.01; AOAC 1995), and crude protein calculated as nitrogen × 6.25 (feed factor). Total fat, ash and crude fibre content were analysed according to AOAC (1995) methods 942.05, 920.39, and 962.09, respectively. Gross energy was determined by adiabatic bomb calorimetry (Autobomb; Gallenkamp, London, UK). The AIA content of feeds and excreta was determined according to the method of Van Keulen and Young (1977).

Statistical analyses

One-way analysis of variance (Steel and Torrie 1980) was performed on data using the GLM function in SPSS (SPSS for Windows 2013, ver. 22.0; IBM, Armonk, NY, USA). The pen was the experimental unit for ADFI, ADWG and F:G, whereas the individual bird was the unit for carcass traits, gut weight and pH. Treatment means were compared by using the least significant difference test, and the significance level was set at $P = 0.05$.

Results

Growth performance

Growth performance data of the broilers are presented in Table 3. There was no treatment effect ($P > 0.05$) on ADFI.

Table 3. Growth performance of broilers fed a control diet or diets containing blood–rumen digesta meal and vegetable oil concentrate (BRDO) with or without enzyme from 22 to 42 days of age.

	Diets			s.e.m.	*P*-value
	Control	BRDO	BRDO + enzyme		
ADFI (g/bird)	175.8	177.0	177.8	2.4	0.483
ADWG (g/bird)	80.0b	94.5a	98.0a	2.9	0.004
Feed:gain	2.20b	1.89a	1.82a	0.10	0.001
Final bodyweight (g/bird)	2183b	2438a	2555a	66	0.009

Within a row, means followed by the same letter are not significantly different ($P > 0.05$). s.e.m., standard error of the mean.

Broilers fed the test diets gained more weight, converted their feed better and weighed more (all $P < 0.05$) than the control group. Enzyme supplementation of the test diet had no additional effect ($P > 0.05$).

Carcass measurements

None of the carcass traits measured (relative weight of the carcass, breast muscle, thigh and drumstick) was affected ($P > 0.05$) by dietary treatment (Table 4).

Gut measurements and pH

The relative weights of gizzard, liver, proventriculus and intestine were not affected ($P > 0.05$) by dietary treatment (Table 5). A longer ($P < 0.05$) intestine was recorded in the control than the BRDO groups. Enzyme supplementation further reduced ($P < 0.05$) intestine length. There were no treatment effects ($P > 0.05$) on pH of the gizzard and caeca.

Feed cost of meat production

The cost of feed was reduced by approximately US$0.10/kg on the test diets compared with the control (soybean-based) diet (Table 6). There was a significant ($P < 0.05$) reduction in feed cost per kg liveweight and per kg carcass on the test diets.

Discussion

We tested the hypotheses that broilers will utilise BRDO concentrate in isonitrogenous and isocaloric diets in a comparable way to the control soybean meal-based diet, and that enzyme supplementation of the BRDO concentrate diet will be beneficial.

Chemical composition of rumen digesta meal

There are several published reports on the composition of blood–RD meal mixture but only few studies have analysed RD meal separately. The protein content of the RD meal used in this study (187 g/kg) is within the range (185–196 g/kg) reported by Elfaki and Abdelatti (2016) but higher than the 66 and 111 g/kg reported by Mandey *et al.* (2015) and Efrem *et al.* (2016), respectively. The crude fibre of the experimental RD meal (212 g/kg) is similar to the value (229 g/kg) observed by Efrem *et al.* (2016), whereas Mandey *et al.* (2015) reported a higher value (400 g/kg). RD is a variable product and several factors including species, age and diet of the ruminant, method of slaughter, and collection affect its composition (Agbabiaka *et al.* 2011; Garcia *et al.* 2021). At the time of this study, we did not come across any published reports on AME determination of RD meal for poultry.

Growth performance

In the present study, dietary inclusion of blood and RD did not affect ADFI but significantly increased ADWG and reduced F:G of the broilers. The reason for the improved ADWG and F:G despite similarities in ADFI is not well understood, but several properties of RD and blood meal might be involved. RD, which is a mixture of feed, fermentation products and dead microbial cells, is rich in microbial protein, essential amino

Table 4. Relative weights (% of liveweight) of carcass components of broilers fed a control diet or diets containing blood–rumen digesta meal and vegetable oil concentrate (BRDO) with or without enzyme from 22 to 42 days of age.

	Diets			s.e.m.	*P*-value
	Control	BRDO	BRDO + enzyme		
Carcass	77.2	78.5	78.8	1.0	0.491
Breast muscle	24.8	21.6	26.3	2.5	0.369
Thigh	10.8	11.9	10.9	0.6	0.389
Drumstick	9.4	11.2	11.1	1.5	0.353

s.e.m., standard error of the mean.

Table 5. Gut measurements of broilers fed a control diet or diets containing blood–rumen digesta meal and vegetable oil concentrate (BRDO) with or without enzyme from 22 to 42 days of age.

	Diets			s.e.m.	*P*-value
	Control	BRDO	BRDO + enzyme		
Relative weight (% of liveweight)					
Gizzard	18.1	18.6	20.0	1.4	0.654
Liver	14.0	12.8	12.7	1.0	0.589
Proventriculus	2.8	2.8	3.5	0.5	0.521
Intestine	27.0	25.6	27.0	2.4	0.899
Intestine length (cm/kg liveweight)	107.3a	94.4b	88.5c	1.5	0.001
pH of gizzard content	4.7	4.0	4.6	0.2	0.143
pH of caeca content	6.1	5.5	5.5	0.2	0.159

Within a row, means followed by the same letter are not significantly different ($P > 0.05$). s.e.m., standard error of the mean.

Table 6. Effect of replacing soybean meal with blood–rumen digesta meal and vegetable oil concentrate (BRDO) and enzyme supplementation in broiler chickens (22–42 days of age) on feed cost of production (US$).

	Diets			s.e.m.	*P*-value
	Control	BRDO	BRDO + enzyme		
Feed cost per kg	0.81	0.70	0.71	n.a.	n.a.
Feed cost per kg liveweight	1.77a	1.32b	1.28b	0.04	0.000
Feed cost per kg carcass	2.30a	1.70b	1.62b	0.06	0.000

Within a row, means followed by the same letter are not significantly different ($P > 0.05$).
s.e.m., standard error of the mean; n.a., not analysed.

acids, B vitamins and volatile fatty acids (Rao and Fontenot 1987; Sok *et al.* 2017). The beneficial effects of volatile fatty acids on intestinal health, liver functions and tissue synthesis (Lee and Gemmell 1972; van der Wielen *et al.* 2000) and possible functional properties of the micronutrients in blood and RD may be reasons for the pattern of growth performance observed in our study. Dietary vitamin B has been reported to improve the performance of poultry (Lindstrom *et al.* 1949; Dryden and Hartman 1971; Patel and McGinnis 1977; Coelho and McNaughton 1995; Suckeveris *et al.* 2020). The effect of B vitamins is mainly linked to their metabolic functions including cofactors of enzymatic reactions, metabolism of carbohydrates and amino acids, and synthesis of methyl groups and nucleic acids (Suckeveris *et al.* 2020). Although diets were supplemented with vitamin premix, possible additional vitamin B coming from RD might have been favourable for birds fed the test diets. Blood meal has a high digestibility coefficient of tryptophan, the third limiting amino acid in broilers (Ravindran *et al.* 2006). Tryptophan stimulates growth and protein accretion through regulation of metabolic pathways (Wu 2013; Qaid and Al-Garadi 2021). Inclusion of the test ingredient increased the fibre content of the diet. Possible beneficial effects of dietary fibre on nutrient utilisation (Li *et al.* 2017) and caecal microbiota (Li *et al.* 2018) may also be implicated in the growth pattern. Li *et al.* (2017) observed that increasing dietary fibre improved protein and energy utilisation by poultry. Lower caecal microbiota diversity and abundance were also reported in broilers fed low-fibre diets (Li *et al.* 2018; Jha and Mishra 2021). These effects (better nutrient utilisation and caecal health) may explain the growth improvement observed in terms of ADWG and F:G of the broilers on the test diets in this study.

Results from published works on the feeding of blood–RD meal to poultry have not been consistent. Up to 10% of the diet as sun-dried bovine blood–RD maintained the growth of pullet chicks (Adeniji and Balogun 2001) and broilers (Makinde *et al.* 2008). Oyedeji *et al.* (2015) recommended 10.5% of the diet as sun-dried bovine blood–RD for broiler growth. Odunsi *et al.* (2004) observed that replacing 15% of dietary soybean meal with sun-dried blood–RD meal was the safest level for broiler growth. Up to 18% sun-dried bovine blood–RD meal in the diet did not adversely affect growth performance parameters of broilers grown to 56 days of age (Yitbarek *et al.* 2016). Mishra *et al.* (2015) observed that replacing 30% of dietary soybean meal with sun-dried blood–RD maintained the performance of growing quails. Most studies on the feeding of blood and RD to poultry have employed sun-drying rather than heat processing as used in the present study.

Although previous researchers collected the blood–RD from the abattoir environment, the materials used in this study were collected directly from slaughtered animals into airtight plastic containers (to avoid contamination with other materials) and heat-processed. In addition, the BRDO concentrate was prepared so as to have protein and ME contents similar to soybean meal, and diets were formulated to be isocaloric and isonitrogenous. The source and processing of RD, diet formulation, and class and age of birds may all affect its utilisation by poultry. Contrary to previous reports on the beneficial effect of yeast (Oyedeji *et al.* 2015) and xylanase (Mandey *et al.* 2015) for supplementation of sun-dried RD meal, in this study enzyme supplementation provided no additional advantage. As earlier suggested, differences in the method of collection, processing, diet formulation and age of birds are all possible sources of variation in the performance of broilers fed RD-based diets.

Carcass measurements

There were no effects of RD inclusion or enzyme supplementation on the carcass traits measured. Nutrients absorbed at the end of digestion are used for tissue synthesis. This pattern of carcass performance of the broilers suggests that the birds obtained sufficient nutrients from the test diets, further supporting the growth pattern observed. In an earlier study, Odunsi *et al.* (2004) recommended 15% of the diet as sun-dried bovine blood–RD meal for carcass performance in broilers. Yitbarek *et al.* (2016) found no effect of dietary dried blood–RD inclusion at up to 24% on carcass development in broilers. Several factors including the source and processing of blood–RD meal, diet composition and age of birds may influence its utilisation by broilers.

Gut measurements

There were no dietary effects on the relative weights of gizzard, liver, proventriculus and intestine or on the pH of the gizzard and caeca. The reduced intestinal length of birds on the test diet, and further reduction with enzyme supplementation, are not understood, but possible effects of dietary fat and the enzyme on digesta transit time may be postulated. Danicke *et al.* (1999) reported faster digesta transit time in all gut segments of broilers fed rye-based diets supplemented with xylanase. Increasing poultry fat in the diet was reported to increase digesta transit time in broilers (Golian and Maurice 1992).

Feed cost of meat production

Birds fed the test diets performed better in terms of weight gain and feed utilisation. This improved performance, coupled with the lower cost of the test diets, was the main reason for the reduction in feed cost of meat production on the test diets compared with the control. The few studies we came across on the feeding of blood–RD meal in broiler diets did not assess the cost, but based on our results, with proper processing and adequate feed formulation the material could be used in broiler diets with a cost advantage.

Conclusions

Adequately processed blood and RD supplemented with fat in the right proportions can replace up to 50% of soybean meal in isocaloric and isonitrogenous broiler diets without affecting growth performance and carcass weight. At this level of inclusion, exogenous enzyme supplementation is not required. These findings provide useful information, especially for small- to medium-scale broiler production. There is need for more research into processing methods of the test ingredients and higher levels of inclusion. The AME of RD meal in this study was determined in layer hens. Considering the variability of AME of feed ingredients among classes of birds, there is need for more accurate estimates in broilers. Organoleptic studies of meat from broilers fed blood–RD-based diets are not documented and this is an area worthy of evaluation.

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