# Effect of Hi-Pro Corn Distiller's Dried Grains with Solubles (DDGS) and Commonly Used Vegetable Protein Sources on Growth Performance, Amino Acid Digestibility and Intestinal Health in Broilers

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# ABSTRACT

The trial was carried out to compare the effect of Hi-Pro Distiller's dried grains with solubles (DDGS) with conventionally available vegetable protein ingredients such as soybean meal (SBM), sunflower meal (SFM), canola meal (CM) and rapeseed meal (RSM) on growth, ileal amino acid digestibility and apparent metabolisable energy (AME) in chicken. Six iso-caloric (ME 2900 Kcal/kg) and iso-nitrogenous (CP 20%) diets were formulated. The experimental diets were: a corn-SBM-based diet, 15% dietary protein replacement of SBM with CM, 15% dietary protein replacement of SBM with RSM, 15% dietary protein replacement of SBM with SFM and 15% dietary protein replacement of SBM with Hi-Pro corn DDGS. A positive control diet in which dietary protein was shared by SBM, Hi-Pro corn DDGS, CM, SFM and RSM was also formulated to check the associative effect of ingredients. Each treatment was replicated 5 times and contained 10 birds in each replicate. During day 1-35, growth performance and FCR was depressed (P<0.05) in birds fed RSM and positive control diet. Trial was lasted on 35 day of age and two birds/ replicate were killed and non-significant effect was observed on carcass, organ weight and intestinal integrity. Similar results were also observed on apparent ileal amino acids digestibility except for methionine, iso leucine and phenylalanine. The apparent ileal digestibility (AID) of methionine improved (P>0.05) in birds fed Hi-Pro corn DDGS based over the SFM-based diet. In conclusion, our study substantiates that Hi-Pro corn DDGS can effectively replace up to 15% of dietary protein from soybean and canola meals without compromising growth performance. Furthermore, Hi-Pro corn DDGS emerges as a superior protein source compared to sunflower and RMs, offering enhanced amino acid digestibility and supporting intestinal health.

# **INTRODUCTION**

Protein is one of the most critical and costly nutrients in broiler diets. Industrial processing of soybean seeds

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involves high temperature and pressure for making soybean meal (SBM) and full-fat soybean which either destroy or inactivate most of the heat sensitive anti-nutritional factors (ANF) i.e., trypsin inhibitors, antigenic proteins and lectins (Huang *et al.*, 2023). Thus, well-prepared SBM is generally free from these anti-nutritional factors.

Canola meal (CM), sunflower meal (SFM) and rapeseed meal (RSM) are commonly utilized as alternative protein sources (Arrutia *et al.*, 2020). Over the past few years, there has been a notable rise in canola cultivation. Canola is advantageous as it provides three times more oil content per acre compared to soybean. After extraction of oil from canola seeds, the resultant meal has comparable nutritional profile with SBM (Spragg and Mailer, 2007).

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MH and MS drafted the content, analyzed the subjects, and wrote the manuscript. MAM, FA critical insights on the subjects and AAK reviewed and improved the manuscript. All authors have read and approved the manuscript.

Key words

Hi-Pro corn DDGS, SBM, Performance, Intestinal integrity, Amino acids digestibility



Solvent extracted CM has little residual oil and thus less energy, high fiber (Bell, 1993; Slominski et al., 2012; Wickramasuriya et al., 2015) and non-starch polysaccharide content (17.9% in CM vs. 14.5% in SBM) that limits its inclusion in broiler diets (Lannuzel et al., 2022). SFM has variable composition that depends upon oil extraction and de-hulling of the oilseed. High crude fiber (CF), lower energy and low lysine contents are major factors that may limit the inclusion of SFM in broiler diets (Heuzé et al., 2020a, b). Crude protein (CP) contents of SFM range from 29 to 45%. It is available in 3 products with variable CP and CF contents i.e., corticated low protein (CP 23-30%) (Vidosavljević et al., 2022), partially decorticated moderated protein (CP 34-40%) (Ahmed et al., 2013) and decorticated high protein (CP 46%) (Levic et al., 2009). RSM has CP 36 to 37% and it is also an important protein ingredient. Higher inclusion of SFM and RSM depressed the growth performance and FCR in broilers (Amerah et al., 2015), which might be due to high NSP content and anti-nutritional factors (Khajali and Slominski, 2012; Lannuzel et al., 2022; Rama-Rao et al., 2006). To address rising feed prices, it's crucial to explore innovative animal feed options, such as using atypical ingredients. Replace traditional feed with alternative resources or byproducts in animal diets (Saeed et al., 2018, 2019, 2023, 2024; Attia et al., 2003, 2024, Aggoor et al., 2004).

The distiller's dried grains with solubles (DDGS) is a by-product of ethanol industry, obtained during dry milling process of different cereals like corn, wheat, sorghum, barley and rice. Use of corn DDGS as a protein substitute in broiler diets is well documented (Damasceno *et al.*, 2020a; Hristakieva *et al.*, 2023). The positive effects of DDGS on the intestinal villi and meat pH are well documented without harming other health and meat quality parameters (Damasceno *et al.*, 2020b). Similarly, Kim *et al.* (2018) reported that DDGS can be utilized in the broilers ration up to 20% without any negative consequences on health and performance. There are two types of corn DDGS, i.e., Hi-Pro corn DDGS and Low-Pro corn DDGS. Hi-Pro corn DDGS contains high CP and amino acids (AA) with low fiber (Buenavista *et al.*, 2021).

This study pioneers the comprehensive evaluation of Hi-Pro Corn DDGS alongside conventional vegetable protein sources such as soybean meal, SFM, canola meal, and RSM in poultry diets. Our findings introduce a new ingredient and its optimal replacement level, showcasing a superior alternative to traditional protein sources in enhancing growth performance, amino acid digestibility, and intestinal health in broilers.

The main objective of the present study was to compare the effects of Hi Pro corn DDGS with conventionally available vegetable protein sources, such as SFM, CM and RSM on growth performance, AA digestibility and apparent metabolisable energy (AME) in broilers.

## **MATERIALS AND METHODS**

### *Experimental design and grouping*

All the feed raw materials used in diets except Hi Pro corn DDGS were arrange by Sadiq Feeds Pvt. Ltd. Pakistan. Three hundred (300) day old broiler chicks (Ross 308) were procured from SB Chicks Rawalpindi- Pakistan.

A basal corn-SBM-based diet was formulated using feed formulation software WinFeed 2.8 and the nutritional matrix data for the ingredients were sourced from AminoDat® 5.0." of ingredient was refered from Amino dat which was further differentiated with replacing 15% dietary protein with CM, SFM (CP 36%), RSM and Hi-Pro corn DDGS (Table I). A positive control was also run to check the associative effect of ingredients. In positive control, dietary protein was contributed by SBM, Hi-Pro corn DDGS, CM, SFM (CP 36%) and RSM (on protein equivalent basis). In the preparation of the experimental diets, all feeds were subjected to a pelleting process at 80°C to ensure uniform texture and composition. Postpelleting, the diets were further processed into crumbles of appropriate sizes to cater to the different developmental stages of the birds

The experimental chicks (mixed male and female) were randomly allotted to 30 experimental units and each experimental diet was offered to 5 experimental units (10 birds per unit). The experimental chicks were raised on cemented floor and rice husk was used as a bedding material. Experimental chicks were vaccinated against ND, IB and IBD by following Ross 308 standard vaccination schedule. Feed was fed to the birds *ad libitum* for the period of trial. Availability of clean and fresh water was offered 24 h in a day. The experimental house temperature was achieved at 33°C on first day of experiment and reduced 3°C every week till 3<sup>rd</sup> week of age and later was maintained at 25°C. During the trial all birds were raised under identical management practices.

# Growth performance, digestibility assay and intestinal health

Starter (d 1-21) and finisher (d 22-35) phase data on BWG and FI were collected to compute FCR. The trial lasted until the end of 35 day. On 35 days, two (2) birds per experiment pen were arbitrarily picked and killed by cervical dislocation for determining carcass percentage, breast, thigh, length of duodenum, jejunum, ileum and soft organ weights. To measure the villus morphology, villus height (VH), crypt depth (CD), crypt width (CW) and VH:CD Table I. Ingredients and nutrients composition ofexperiment diets.

Ingredients	SBM	PC	DDGS	СМ	SFM	RSM
High pro DDGS	0.00	6.19	6.41	0.00	0.00	0.00
Maize	64.27				61.28	61.38
Rice polishing	0.00	0.53	0.00	0.00	0.00	0.00
Wheat bran	0.00	0.00	0.44	0.67	0.00	0.34
Canola meal	0.00	7.85	0.00	8.12	0.00	0.00
RSM	0.00	8.01	0.00	0.00	0.00	8.29
SFM 36%	0.00	7.98	0.00	0.00	8.25	0.00
Soybean meal 46%	30.50		23.50	23.25	23.25	23.25
Fish meal	0.00	0.00	0.00	1.00	1.00	1.00
Calcium carbonate	1.90	1.75	1.92	1.75	1.80	1.75
Monocalcium phos- phate	1.07	0.86	1.04	0.86	0.94	0.85
Sodium chloride	0.15	0.15	0.15	0.15	0.15	0.15
Sodium bicarbonate	0.21	0.21	0.21	0.21	0.21	0.21
Lysine sulphate 70%	0.25	0.83	0.52	0.32	0.42	0.36
DL-Methionine	0.19	0.11	0.17	0.16	0.17	0.16
L-Threonine	0.15	0.20	0.18	0.15	0.17	0.16
L- Valine	0.10	0.11	0.10	0.10	0.09	0.10
L- Tryptophan	0.00	0.02	0.02	0.00	0.00	0.00
L Isoleucine	0.07	0.15	0.10	0.09	0.09	0.11
Sunflower oil	0.50	3.32	0.00	1.25	1.54	1.25
Molasses	0.00	1.29	0.00	0.00	0.00	0.00
Vit. Min premix <sup>1</sup>	0.50	0.50	0.50	0.50	0.50	0.50
Microtech 1000 FTU	0.01	0.01	0.01	0.01	0.01	0.01
Betaine	0.13	0.13	0.13	0.13	0.13	0.13
Total	100	100	100	100	100	100
Nutrient composition	n (%)					
Crude protein	20.0	20.0	20.0	20.0	20.0	20.0
Dig. Lys	1.10	1.10	1.10	1.10	1.10	1.10
Dig. Met	0.46	0.46	0.46	0.46	0.46	0.46
Dig. Met + Cys	0.75	0.75	0.75	0.75	0.75	0.75
Dig. Trp	0.21	0.21	0.21	0.21	0.21	0.21
Dig. Thr	0.78	0.78	0.78	0.78	0.78	0.78
Dig. Ile	0.78	0.78	0.78	0.78	0.78	0.78
Dig. Val	0.89	0.89	0.89	0.89	0.89	0.89
ME Kcal/kg	2900	2900	2900	2900	2900	2900
Calcium	0.90	0.90	0.90	0.90	0.90	0.90
Avail. phosphorus	0.33	0.33	0.33	0.33	0.33	0.33

SBM, soybean meal; PC, positive control; DDGS, distiller's dried grains with solubles; CM, canola meal; SFM, sunflower meal; RSM, rapeseed meal. <sup>1</sup>. Vitamin A 10 mg/kg, vitamin D<sub>3</sub> 9 mg/ kg vitamin E 50 mg/ Kg, vitamin K<sub>3</sub> 3.6 mg/Kg, vitamin B<sub>1</sub> 1.7 mg/Kg, vitamin B<sub>2</sub> 10 mg/ Kg, vitamin B<sub>3</sub> 35 mg/Kg, vitamin B<sub>5</sub> 11.1 mg/Kg, vitamin B<sub>6</sub> 3.1mg/ Kg vitamin B<sub>9</sub> 1.1mg/Kg, vitamin B<sub>12</sub> 1.2 mg/Kg, vitamin H5 mg/Kg, Magnesium sulphate 18 mg/Kg and Zinc sulphate 2 mg/Kg.

were collected from the midpoint of jejunum by following Wang *et al.* (2015). Villus morphology was studied under light microscope by following Khan *et al.* (2022).

A digestibility bioassay was performed during last week (22-35 day) of trial to measure the AID of amino acids and AME of experimental diets. External acid insoluble ash (Celite®) was used as a digestibility marker and mixed 1% in all experimental diets. These diets containing Celite<sup>®</sup> were fed to the experimental birds from day 29 till the end of the trial. At the end of 35 day of trial 4 birds per replicate were randomly picked and ileal digesta was collected from the ileum (Meckel's diverticulum to 40 mm cranial to ileao-cecal junction) to measure amino acids digestibility and AME. Plastic cups (200 mL) were used to recover ileal digesta and a few drops of formalin solution were added to stop bacterial growth and immediacy shifted to an ice box. The collected digesta samples were dried in hot air oven at 65°C and finely ground by using a 0.5 mm sieve for further lab analysis.

#### Lab analysis of experimental diets

Experimental diets and ileal digesta were analyzed for total nitrogen (AOAC, 000) and acid insoluble ash (AIA) (Vogtmann *et al.*, 1975). For amino acid content analysis of the experimental diet and digesta, samples were prepared and analyzed using a Biochrom 30 Plus Amino Acid Analyzer (Biochrom Ltd., Cambridge, UK), following standard oxidation and hydrolysis procedures. The gross energy values of experimental diet and ileal digesta were measured by using Bomb calorimeter (Parr Instrument Co., Moline, IL).

#### Calculation and statistical analysis

N

Nutrient digestibility was determined following Ravindran *et al.* (1999).

utrient Digestibility (%) = 
$$\frac{\left(\frac{\text{nutrient}}{\text{AIA}}\right) \text{diet} - \left(\frac{\text{nutrient}}{\text{AIA}}\right) \text{digesta}}{\left(\frac{\text{nutrient}}{\text{AIA}}\right) \text{diet}}$$

The AME value of experimental diets was computed as,

$$AME_{(kcal/kg)} = GE_{diet} - GE_{di} \left[ esta \times \left( \frac{AIA \text{ In diet}}{AIA \text{ in digesta}} \right) \right]$$

AME was corrected to zero  $N_{ret}$  by using 8.22 kcal/g. This is a factor which was described by Hill and Anderson (1958).

 $AME_n = AME - (8.22 \times N_{ret})$ 

Where AMEn and N<sub>ret</sub> represents AME corrected for nitrogen and nitrogen retention respectively (Bolarinwa and Adeola, 2012).

All statistical analyses were conducted using the general linear model (GLM) procedure of the SAS statistical software (version 9.4, SAS Institute Inc., Cary, NC, USA). The model included treatment as the fixed effect and

Age Parameters days	Treatments						SEM	P-value	
	SBM <sup>1</sup>	PC <sup>2</sup>	DDGS <sup>3</sup>	CM <sup>4</sup>	SFM <sup>5</sup>	RSM <sup>6</sup>			
1-21	FI (g)	1357 <sup>ab</sup>	1264°	1376ª	1322 <sup>abc</sup>	1371ª	1292 <sup>bc</sup>	17.6	0.001
	BWG (g)	949 <sup>ab</sup>	858°	978ª	942 <sup>ab</sup>	962 <sup>ab</sup>	896 <sup>bc</sup>	18.7	0.001
	FCR (g/g)	1.430 <sup>ab</sup>	1.473ª	1.406 <sup>b</sup>	1.404 <sup>b</sup>	1.428 <sup>ab</sup>	1.442 <sup>ab</sup>	0.01	0.030
22-35	FI (g)	1867 <sup>b</sup>	1932 <sup>ab</sup>	1985 <sup>ab</sup>	2013 <sup>ab</sup>	2045 <sup>a</sup>	1936 <sup>ab</sup>	35.1	0.020
	BWG (g)	1173 <sup>ab</sup>	1175 <sup>ab</sup>	1268ª	1278ª	1234 <sup>ab</sup>	1153 <sup>b</sup>	26.0	0.006
	FCR (g/g)	1.593 <sup>ab</sup>	1.645 <sup>abc</sup>	1.567ª	1.575 <sup>ab</sup>	1.658 <sup>bc</sup>	1.680°	0.02	0.028
1-35	FI (g)	3225 <sup>b</sup>	3197 <sup>b</sup>	3361 <sup>ab</sup>	3335 <sup>ab</sup>	3417 <sup>a</sup>	3229 <sup>b</sup>	41.1	0.004
	BWG (g)	2122 <sup>ab</sup>	2033 <sup>b</sup>	2247ª	2221ª	2197ª	2050 <sup>b</sup>	30.2	0.001
	FCR (g/g)	1.520 <sup>abc</sup>	1.573ª	1.497°	1.501 <sup>bc</sup>	1.556 <sup>abc</sup>	1.575ª	0.01	0.005

Table II. Comparative effect of different vegetable protein sources on growth performance.

Mean values within a row having different subscripts are statistically significant (P < 0.05). Means of 5 replicates having 10 birds in each replicate. FI, feed intake; BWG, body weight gain; FCR, feed conversion rate. <sup>1</sup>Corn-SBM based diet. <sup>2</sup>Contained SBM, Hi-Pro DDGS, canola meal, RSM and SFM as a vegetable protein sources. <sup>3</sup>15% Dietary protein shared by Hi-Pro corn DDGS. <sup>4</sup>15% Dietary protein shared by canola meal. <sup>5</sup>15% Dietary protein shared by sunflower meal. <sup>6</sup>15% Dietary protein shared by rapeseed meal. For other abbreviations see Table I.

replicate (pen) as the random effect. Mean separation for significant effects was performed using the least significant difference (LSD) test at a significance level of P < 0.05. Results are presented as mean  $\pm$  standard error of the mean (SEM).

# RESULTS

## Growth performance and carcass response

During day 1-21 FI, BWG and FCR were depressed (P<0.05) in response to RSM and positive control-based diet, while FCR was only depressed (P<0.05) in birds fed positive control (PC) diets compared to Hi-Pro corn DDGS and CM diets (Table II). A similar trend was observed during d 22-35, wherein FCR and BWG were depressed (P<0.05) with the supplementation of RSM. During overall growth phase (day1-35), BWG was depressed (P<0.05) in response to PC and RSM-based diets. A similar trend was also observed in FCR value. Feed conversion was depressed (P<0.05) in response to PC and RSM diets (Table II).

Data on carcass, breast, thigh, liver, gizzard and heart percentage are shown in Table III. Carcass characteristics and organ weights were not different (*P*>0.05) in response to different vegetable protein sources.

#### Intestinal integrity

Intestinal integrity was measured in terms of duodenum, jejunum and ileum lengths, VH, CW, CD, VH:CD (Table IV). The small intestinal length was not different (P>0.05) due to different vegetable protein sources. However, numerically longest small intestine was observed for the PC group and shortest for Hi-Pro corn DDGS group. Similar trend (P>0.05) was observed on villus height, crypt depth and width in response to vegetable protein sources.

 Table III. Comparative effect of different vegetable

 protein source on carcass and organ characteristics.

Parame- ter (%)	$\sim$	SEM	Р					
	SBM	PC	DDGS	СМ	SFM	RSM		value
Carcass	61.6	61.6	61.54	61.5	61.86	61.59	1.06	0.96
Breast	26.2	26.3	26.83	27.1	27.93	26.45	0.67	0.47
Thigh	9.30	8.89	9.38	9.58	9.38	9.28	0.3	0.73
Liver	2.13	2.28	2.15	2.06	2.12	2.03	0.11	0.68
Gizzard	1.13	1.24	1.25	1.21	1.17	1.40	0.08	0.35
Heart	0.618	0.62	0.605	0.59	0.61	0.607	0.03	0.99

For abbreviations of treatment and statistical details see Table I and II.

 Table IV. Comparative effect of different vegetable

 protein sources on intestinal integrity.

Parame- ters		SEM	Р					
	SBM	PC	DDGS	СМ	SFM	RSM		value
Duodenum (cm)	32.2	34.5	29.9	31.3	32.1	33.5	1.26	0.17
Jejunum (cm)	88.0	95.3	83.7	96.1	93.1	95	3.02	0.06
Ileum (cm)	85.2	95.1	85.9	90.9	93.6	91.1	3.22	0.21
SI (cm)	205	225	199	218	219	220	6.33	0.06
VH (µm)	1049	913	1037	1011	975	1049	79.0	0.81
CD (µm)	126	157	132	151	155	134	20.4	0.82
CW (µm)	51.0	58.8	57.0	55.0	52.4	51.0	2.42	0.15
VH/ CD	8.37	6.21	8.34	7.49	6.55	7.85	0.79	0.29

SI, small intestine; VH, Villus height; CD, Crypt depth; CW, crypt width; VH/CD, villus height/crypt depth.For abbreviations of treatment and statistical details see Table I and II.

Parameters (%)	SBM	PC		SEM	P value			
			DDGS	СМ	SFM	RSM		
Crude protein	80.93	79.15	81.36	80.56	79.47	79.05	0.75	0.16
Cysteine	79.36	79.93	80.92	81.91	79.31	79.56	0.86	0.22
Methionine	88.53 <sup>ab</sup>	88.18 <sup>ab</sup>	89.26ª	88.56 <sup>ab</sup>	87.42 <sup>b</sup>	87.57 <sup>ab</sup>	0.41	0.04
Met + Cys	84.95	84.94	86.15	85.95	84.22	84.44	0.54	0.10
Threonine	83.28	82.09	83.99	83.88	81.97	82.17	0.76	0.23
Valine	81.97	80.26	83.86	82.94	81.54	80.86	0.83	0.05
Iso-leucine	80.48 <sup>ab</sup>	79.00 <sup>ab</sup>	80.84ª	78.97 <sup>ab</sup>	78.80 <sup>ab</sup>	77.11 <sup>b</sup>	0.78	0.03
Leucine	86.91	86.09	89.26	86.59	85.56	86.06	0.91	0.10
Phenylalanine	80.35 <sup>ab</sup>	79.02 <sup>ab</sup>	81.39ª	80.44 <sup>ab</sup>	77.99 <sup>b</sup>	77.60 <sup>b</sup>	0.74	0.009
Histidine	80.84	80.55	82.62	82.78	80.51	80.66	0.68	0.06
Lysine	80.04	78.91	80.46	79.82	79.08	79.00	0.66	0.48
Arginine	90.18	89.53	90.45	89.51	89.69	89.50	0.60	0.80
N <sub>ret</sub>	2.71 <sup>abc</sup>	2.73 <sup>abc</sup>	2.82ª	2.78 <sup>ab</sup>	2.62 <sup>d</sup>	2.64 <sup>cd</sup>	0.02	0.001
AME Kcal/kg	2951	2906	2966	2929	2920	2909	37.9	0.84
AMEn Kcal/kg	2929	2884	2943	2906	2899	2888	37.8	0.85

Table V. Comparative effect of different vegetable protein sources on amino acids digestibility and apparent metabolisable energy.

AME, apparent metabolisable energy. For abbreviations of treatment and statistical details see Table I and II.

#### Nutrient digestibility

Crude protein (CP) and apparent amino acids (AAs) digestibility was not different (P>0.05) with different vegetable protein sources (Table V), except methionine, iso-leucine and phenylalanine. Methionine digestibility was improved (P<0.05) in Hi-pro corn DDGS based diet over SFM based diet. Numerically (P>0.05) higher CP digestibility was recorded with group fed Hi-Pro corn DDGS based diet followed by SBM, CM, SM, positive control and RSM based diet.

Results of AME, AMEn and Nret are shown in Table V. Similar trends were observed on AME and AMEn. Numerically higher (P>0.05) AME value was observed in response to Hi-Pro corn DDGS followed by SBM, CM, SFM, RSM and PC. The nitrogen retention was higher (P<0.05) in response to Hi-Pro corn DDGS based diet over SFM and RSM based diet.

#### DISCUSSION

World-wide there is growing trend to use vegetable protein sources in commercial broiler diets rather than animal protein sources. This may be due to inconsistency in quality and high microbial contamination of animal protein sources (Van der Poel *et al.*, 2020). Protein is an expensive constituent of broiler diets, while exploring new vegetable protein sources could reduce cost and also increased the efficiency of feed (Ruelas *et al.*, 2023). However, growth performance of broilers in response to vegetable protein sources can be regulated by many factors as dietary composition, physical nature of diet and presence of anti-nutritional factors (Helmy *et al.*, 2022; Alshelmani *et al.*, 2021).

The finding of present study showed that different vegetable protein source had no effect on BWG and FCR except the RSM and PC diets. Growth performance was depressed by 8.7% and 9.48% with RSM and PC, respectively as compared to Hi-Pro corn DDGS. Numerically, FCR was also depressed with the supplementation of SFM than SBM, Hi-Pro corn DDGS and canola meal-based diets. Inclusion of SFM and RSM depressed growth performance. Similar findings were also published by Amerah et al. (2015) who used a combination of SFM and RSM and noted that higher inclusion level of RSM and SFM depressed growth performance and FCR. This depression might be due to presence of glucosinolate and non-starch polysaccharide, especially arabinoxylan that has anti-nutritional effects (Choct, 2006). In another study, similar findings were also observed by Min et al. (2009) who used graded levels (0, 5, 10, 15, 20 and 25%) of canola meal and corn DDGS and reported that FCR was not different. They also reported that higher inclusion of canola meal was more detrimental to BWG than corn DDGS. High percentage of fines in the diet may have contributed to reduction in FI and BWG. The relationship between percentage of fines and broiler growth performance was also explained by many studies (Greenwood *et al.*, 2004; Abd El-Wahab *et al.*, 2020; Novotný *et al.*, 2023).

In another study, growth performance and FCR were depressed beyond 25% HP DDGS protein replacement (Applegate *et al.*, 2009), but dietary protein replacement of Hi-Pro DDGS in current trial was relatively lower than used by Applegate *et al.* (2009), so depression of weight gain and FCR was not seen in present study. Jung and Batal (2009) also reported that HP DDGS could be an alternative to SBM and demonstrated that HP DDGS could be supplemented up to 12% in layer diets without compromising egg parameters i.e., production, weight and quality. Similar observation was observed by Abudabos *et al.* (2017) who reported that corn DDGS can partially replace corn and SBM without compromising growth performance and FE.

Elbaz *et al.* (2022) also reported that corn DDGS can be used in broiler diets up to 20%. Beyond this growth performance and FE were compromised. Higher inclusion of DDGS reduced bulk density and pellet quality. Recently, Kim *et al.* (2018) conducted a study and observed that growth performance of birds could be compromised with a 20% DDGS level; however, pellet durability index (PDI) was also compromised. The outcome of Loar *et al.* (2010) were also alike with the outcome of present study; they concluded that corn DDGS could be replaced up to 7.5% in broiler diets without compromising growth.

It is well documented that supplementation of corn DDGS did not improve carcass weight. Breast and thigh meat yields, fat contents were increased with corn DDGS inclusion (Shim *et al.*, 2011). Dozier and Hess (2015) also reported that inclusion of corn DDGS did not alter the partitioning of carcass in term of breast, thigh, leg and wing, although except fat pad percentage decreased with increasing the inclusion of corn DDGS. It was also seen in present trial, inclusion of different vegetable protein sources did not alter (P > 0.05) the carcass partitioning (carcass, thigh and breast weight) and soft organ weight (gizzard, heart and liver). It might be due all diets had similar caloric and digestible AA content.

Nutrient absorption in the small intestine depends on villus height; shortening of villus height reduced the absorption area and deeper crypt depth increased demand for energy and protein (Nassiri *et al.*, 2012). Changes in intestinal morphology is mainly associated with presence of the toxin (de Souza *et al.*, 2020). Findings of the present study showed that intestinal integrity (VH, CD, CW and VH: CD) was not affected (P > 0.05) by different vegetable protein sources. Numerically VH and VH/CD were observed to be improved in birds fed Hi-Pro corn DDGS based diet over SFM and the PC. Alizadeh *et al.* (2016) reported that histomorphology measurement was not different (*P*>0.05) among yeast cell wall supplemented diet and corn DDGS containing diet. Ruan *et al.* (2017) also reported that that intestinal integrity of the Chinese Yellow broiler was not compromised by the supplementation of graded level of corn DDGS. Similar observation was also observed by Wang *et al.* (2015) who concluded that different protein sources (Hi-Pro corn DDGS and meat and bone meal) had no effect on VH, CW and VH/CD. However, increasing level of SFM in broilers was reported to depress intestinal health (Nassiri *et al.*, 2012).

Protein, AA digestibilities and AME were not different (P>0.05) among different vegetable protein sources except methionine, phenylalanine, isoleucine and nitrogen retention (Table V). Though, protein digestibility was numerically improved (P>0.05) with Hi-Pro corn DDGS-based diet followed by SBM, CM, SFM, PC and RSM diets. Dry matter intake, feed ingredients processing, amount of CP, amino acid, crude fiber, ether extract and antinutritional content of diet are major factor that influenced the digestibility of diet (Gabert et al., 2001). Ullah et al. (2016) demonstrated that different cereal by-products and protein meals had different AA digestibility.in contrast, apparent ileal amino acids digestibility was not affected (P>0.05) with different vegetable protein sources. It might be due to feed formulation that based on digestible AAs. Also, synthetic AA were supplemented and supplemented proportion of tested ingredients were limited. Findings of Youssef et al. (2008), were also similar with the findings of present study who reported that protein digestibility was not reduced with increasing level of corn DDGS.

Kim *et al.* (2018) reported that CP digestibility was reduced with the supplementation of 20% corn DDGS than without corn DDGS. This contradiction might be due to higher inclusion of corn DDGS. In another study, Kim *et al.* (2010) concluded that newly developed DDGS (Elusieve DDGS and E-Mill DDGS) have higher AA digestibility and TME over conventionally available corn DDGS, which might be due to higher level of fiber and subjected to more heat; however, in our present study, different effects on ileal apparent AA digestibility and AME were not observed.

#### **CONCLUSION**

In conclusion, Hi-Pro corn DDGS (6.41% of diet) can effectively replace up to 15% dietary protein of SBM and CM protein without effecting growth performance. However, Hi-Pro corn DDGS appears to be a superior protein alternate compared to SFM and RSM in broiler

#### diets.

Future research could explore the potential of utilizing higher levels of Hi-Pro Corn DDGS in comparison to soybean meal and its substitution with other conventional protein sources commonly used in broiler diets. This area of study could reveal insights into optimizing broiler nutrition for enhanced growth performance and feed efficiency.

# DECLARATIONS

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### IRB approval

This research was approved by committee of Graduate Study, University of Agriculture, Faisalabad (IBR # DGS/56129-32).

## Ethical statement

This study was conducted in accordance with ethical standards and guidelines established by the University of Agriculture Faisalabad, Pakistan. Prior to commencing the re-search, all necessary ethical approvals were obtained from the Graduate Study, University of Agriculture, Faisalabad, Pakistan.

## Data availability

The current study is available from the corresponding author upon reasonable request. The authors declare full data transparency.

Statement of conflict of interest

The authors have declared no conflict of interest.

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