Determination of metabolizable energy and amino acid digestibility in various hatchery byproducts for broiler chickens

Jong Hyuk Kim, Hyeon Seok Choi, Won Jun Choi, Hyun Woo Kim, and Dong Yong Kil [1]

Department of Animal Science and Technology, Chung-Ang University, Anseong-si, Gyeonggi-do 17546, Republic of Korea

ABSTRACT The objective of the present experiment was to determine ME concentrations and amino acid (**AA**) digestibility in various hatchery byproducts (**HBPs**) for broiler chickens. In experiment 1, a total of forty 60-day-old female broiler chickens were allotted to 1 of 5 dietary treatments with 8 replicates and used to measure ME concentrations in HBPs. The basal diet was prepared to contain corn, soybean meal, corn oil, and other non-energy ingredients. Additional 4 experimental diets were prepared to contain 10% of infertile eggs (**IFE**), unhatched eggs (**UHE**), low-grade or dead chicks (**LDC**), and mixture (**MIX**; 55% IFE, 10% UHE, 10% LDC, and 25% hatched eggshells). In experiment 2, a total of seven hundred and sixteen 1-day-old mixed-sex broiler chickens (1:1 ratio of males and females) were allotted to 1 of 5 dietary treatments with 7 replicates per treatment and used to determine apparent ileal digestibility (**AID**) and standardized ileal digestibility (**SID**) of AA in HBPs. The experimental diets consisted of a nitrogen-free diet and 4 diets containing IFE, UHE, LDC, or MIX as a sole source of AA. Results indicated that AME and AME_n values were greater ($P < 0.05$) for LDC than for IFE, which had greater ($P < 0.05$) AME and AME_n values for UHE and MIX. The AID and SID of most AA in LDC were greater ($P < 0.05$) than those in MIX, whereas IFE and UHE had intermediate AID and SID of those AA as compared to LDC and MIX. Average SID of essential AA in LDC was greater ($P < 0.05$) than in UHE and MIX, but the average SID of nonessential AA did not differ among 4 HBPs. In conclusion, LDC has the greatest ME concentrations and AA digestibility among 3 individual HBPs (IFE, UHE, and LDC). The mixture of HBPs has the least ME concentrations and AA digestibility in broiler chickens. The ME and AA digestibility of HBPs are likely affected by inclusion amounts of hatched eggshells. However, high concentrations of ME and available AA demonstrate that individual HBPs and their mixture are potential protein ingredients for broiler diets.

Key words: amino acid, broiler chicken, digestibility, hatchery byproduct, metabolizable energy

2022 Poultry Science 101:101544
https://doi.org/10.1016/j.psj.2021.101544

INTRODUCTION

The production of poultry byproducts steadily increases with increasing production of poultry meats or eggs (Das et al., 2002; Glatz et al., 2011). Various types of poultry byproducts are currently produced in the poultry industry. One of the most common poultry byproducts is hatchery byproducts (**HBPs**), which originate from a hatching process in the hatchery. The HBPs are generally classified as infertile eggs (**IFE**), unhatched eggs (**UHE**), low-grade or dead chicks (**LDC**), or hatched eggshells (**HE**; Rasool et al., 1999; Shahriar et al., 2008; AAFCO, 2016). In the commercial hatchery, IFE is collected after examination of fertilization, whereas UHE, LDC, and HE are typically collected together at the end of a hatching system (Choi et al., 2021). However, IFE, UHE, LDC, and HE are normally composted together into a mixture (**MIX**) as a final product in the hatchery (Choi et al., 2021). Currently, HBPs are mostly disposed in landfills or utilized as a fertilizer, which increases disposal costs and environmental pollutions, consequently motivating researchers to find a solution for the proper use of HBPs (Das et al., 2002; Sung et al., 2020).

The HBPs contain high amounts of energy, protein, and calcium (**Ca**) although variations in nutritional compositions are relatively high, indicating that HBPs can be a potential ingredient in animal diets (Abiola et al., 2012; Sung et al., 2020; Choi et al., 2021). However, the detailed information regarding nutritional values for HBPs in animal diets is largely lacking. In

Received July 19, 2021.
Accepted October 10, 2021.

[1]Corresponding author: dongyong@cau.ac.kr

particular, few studies have performed to determine ME concentrations and apparent ileal digestibility (**AID**) and standardized ileal digestibility (**SID**) of AA in individual HBPs and their mixture for broiler chickens, which currently limits the use of HBPs in broiler diets. Therefore, in order to increase the use of the HBPs in animal diets, the accurate estimation of available energy and amino acid (**AA**) concentrations in HBPs is required.

Therefore, the objective of the present experiment was to determine the values for ME concentrations and AA digestibility in various HBPs (i.e., IFE, UHE, LDC, and MIX) for broiler chickens.

MATERIALS AND METHODS

All experimental procedures were reviewed and approved by the Institutional Animal Care and the Use Committee at Chung-Ang University (approval No. 2018-00136).

Preparation of HBPs

The collection and preparation of HBP samples were conducted based on the methodology described in our previous experiment (Choi et al., 2021). Briefly, the IFE, UHE, LDC, and HE were individually collected from a local broiler hatchery (Dongsan Broiler Hatchery, Cheonan-si, Republic of Korea). The mixture of 4 ingredients (MIX; 55% IFE, 10% UHE, 10% LDC, and 25% HE) was also prepared according to the typical production proportion of each HBP in the commercial broiler hatchery. Each of collected HBPs (i.e., IFE, UHE, LDC, or MIX) was ground using a meat chopper (MN-225, Hankook Fujee, Hwaseong-si, Republic of Korea) and dried immediately at 50°C for 24 h in a vertical convention oven (LDO-630F, Daihan Labtech, Namyangju-si, Republic of Korea) to achieve optimal DM concentrations (i.e., close to 96% DM). All dried HBPs were finely ground again using a screen grinder (CM 290 Cemotec, FOSS, Hilleroed, Denmark) and stored in the refrigerator at −20°C before further analysis. The analyzed energy and nutrient concentrations in the 4 HBPs are presented in Table 1.

Birds, Diets, and Experimental Design

In experiment 1, a total of forty 60-day-old Ross 308 female broiler chickens were used to determine AME and AME_n of 4 HBPs. All birds were randomly allotted to 1 of 5 dietary treatments with 8 replicates per treatment. Each bird was placed in a metabolic cage (35.2 cm × 45.0 cm × 55.3 cm, width × length × height). Room temperature was set at 20°C and light was provided for 24 h throughout the experiment. The basal diet was prepared to contain corn, soybean meal, corn oil (99%), and other non-energy ingredients (1%). Diets were then prepared to include 10% of each of HBP samples at the expense of energy ingredients in the basal diet (Table 2).

Table 1. Analyzed energy and nutrient concentrations of hatchery byproducts, as-fed basis.

	Hatchery byproducts[1]			
Items[2]	IFE	UHE	LDC	MIX
Gross energy, kcal/kg	5,133	4,753	5,945	3,764
DM, %	98.3	97.8	96.6	98.0
CP, %	34.2	39.4	60.0	31.2
Crude ash, %	27.6	27.6	6.6	40.6
AEE, %	23.3	23.8	24.6	16.7
Calcium, %	11.7	11.7	1.2	16.8
Phosphorus, %	0.7	0.7	1.0	0.6
Essential AA, %				
Arg	2.00	2.80	3.95	1.50
His	0.79	1.01	1.32	0.58
Ile	1.73	1.90	2.34	1.02
Leu	3.15	3.64	4.71	1.91
Lys	2.46	2.65	3.51	1.50
Met	1.29	1.30	2.31	0.73
Phe	1.96	2.24	2.78	1.14
Thr	1.85	2.22	2.84	1.27
Trp	0.47	0.43	0.50	0.28
Val	2.21	2.69	3.25	1.50
Nonessential AA, %				
Ala	2.10	2.73	3.98	1.40
Asp	3.78	4.31	5.47	2.38
Cys	0.97	1.35	1.58	0.84
Glu	4.80	4.17	8.54	3.30
Gly	1.31	2.69	4.97	1.22
Pro	1.38	2.38	3.83	1.29
Ser	2.61	3.02	3.65	1.72
Tyr	1.03	1.22	1.52	0.66

[1]IFE, infertile egg; LDC, low-grade or dead chick; MIX, mixture (55% IFE, 10% UHE, 10% LDC, and 25% hatched eggshells); UHE, unhatched egg.
[2]AA, amino acid; AEE, acid-hydrolyzed ether extract.

In experiment 2, a total of seven hundred and sixteen 1-day-old mixed-sex broiler chickens (1:1 ratio of males and females) were used to determine the AID and SID of AA in 4 HBPs. All birds were raised in an

Table 2. Ingredients and chemical composition of experimental diets, as-fed basis (experiment 1).[1]

Items	Basal	IFE	UHE	LDC	MIX
Ingredients (%)					
Corn	70.0	62.9	62.9	62.9	62.9
Soybean meal (46% CP)	25.0	22.5	22.5	22.5	22.5
Corn oil	4.0	3.6	3.6	3.6	3.6
HBPs	0.0	10.0	10.0	10.0	10.0
NaCl	0.3	0.3	0.3	0.3	0.3
$NaCO_3$	0.3	0.3	0.3	0.3	0.3
Vitamin premix[2]	0.2	0.2	0.2	0.2	0.2
Mineral premix[3]	0.2	0.2	0.2	0.2	0.2
Analyzed energy and nutrient concentrations					
Gross energy, kcal/kg	4,097	4,139	4,016	4,258	3,976
CP, %	17.7	19.2	21.0	19.9	17.4
DM, %	90.0	90.9	90.6	90.7	90.9

[1]HBPs, hatchery by-products; IFE, infertile egg; LDC, low-grade or dead chick; MIX, mixture (55% IFE, 10% UHE, 10% LDC, and 25% hatched eggshells); UHE, unhatched egg.
[2]Provided per kilogram of the complete diet: vitamin A (from vitamin A acetate), 13,000 IU; vitamin D_3, 5,000 IU; vitamin E (from DL-α-tocopheryl acetate), 80 IU; vitamin K_3, 4 mg; vitamin B_1, 4 mg; vitamin B_2, 10 mg; vitamin B_6, 6 mg; vitamin B_{12}, 20 μg; calcium pantothenate, 20 mg; folic acid, 2 mg; biotin, 200 μg; niacin, 60 mg.
[3]Provided per kilogram of the complete diet: Zn (as ZnO), 100 mg; Mn (as $MnSO_2 \cdot H_2O$), 120 mg; Fe (as $FeSO_4 \cdot 7H_2O$), 60 mg; Cu (as $CuSO_4 \cdot 5H_2O$), 16 mg; Co (as $CoCO_3$), 1,000 μg; I (as $Ca(IO_3)_2 \cdot H_2O$), 1.25 mg; Se (as Na_2SeO_3), 300 μg.

environmentally controlled room. Room temperature was set at 30°C during the first week and reduced by 2°C during the subsequent week. Birds were fed a commercial diet for 20 d. Feed and water were supplied ad libitum and birds received the continuous lighting throughout the experiment. On d 21, all birds were weighed, and 275 birds with extremely high and low BW were discarded. The remaining 441 birds were allotted to 35 battery cages with a similar average BW (initial BW = 642 ± 12.9 g). Battery cages were allocated to 1 of 5 dietary treatments with 7 replicates per treatment in a completely randomized design. Each battery cage, except for those assigned to the nitrogen-free diet (**NF**), had 12 birds. In the NF treatment, each battery cage had 15 birds. The birds were fed the experimental diets for 7 d. The experimental diets consisted of NF, IFE, UHE, LDC, and MIX (Table 3). Each HBP served as the sole source of AA in dietary treatments. The NF diets were formulated to measure the endogenous losses of AA (Adedokun et al., 2014). All experimental diets included 0.5% of chromic oxide as an indigestible marker to calculate ileal AA digestibility.

Sample Collection

In experiment 1, the AME and AME_n values for the experimental diets were determined as Bourdillon et al. (1990) described but with a minor modification (Kim et al., 2021). At the start of the experiment, average feed intake (**FI**) were measured for 2 d. Afterward, all birds were adapted to experimental diets for 72 h with feeding 80% of the average FI to minimize ingredient selection in feeders for 55 h and fasting for 17 h. The fasting period was required to empty the gastrointestinal tract before the start of the collection period. The collection period lasted for 96 h with feeding 180 g of each diet every day for 79 h and fasting 17 h. The excreta were collected daily and immediately stored at −20°C. Collected excreta samples were dried in a forced-air drying oven at 60°C for 48 h and finely ground using a 1-mm screen grinder (CM 290 Cemotec, FOSS).

In experiment 2, sample collection and digesta processing were conducted as described by Ravindran et al. (2017). All birds were euthanized using CO_2 asphyxiation and immediately dissected. Digesta was collected from the entire ileum. The ileum was defined as the portion of the small intestine extending from Meckel's diverticulum to a point 40 mm proximal to the ileo-cecal-colonic junction. Ileal digesta were collected by gently flushing with distilled water into a plastic container. Ileal samples from birds within a replicate cage were pooled, frozen immediately after collection and subsequently freeze-dried. The HBP samples, experimental diets, and ileal digesta samples were ground through a 1-mm screen grinder and stored in airtight containers at −20°C for chemical analyses.

Table 3. Composition of experimental diets, as-fed basis (experiment 2).[1]

Items	N-free	IFE	UHE	LDC	MIX
Cornstarch	17.76	17.76	17.76	17.76	17.76
Dextrose	63.16	12.62	17.72	32.92	3.80
Soybean oil	5.00	3.00	5.00	2.00	8.70
Infertile eggs (IFE)	0.00	55.60	0.00	0.00	0.00
Unhatched eggs (UHE)	0.00	0.00	48.30	0.00	0.00
Low grade chicks (LDC)	0.00	0.00	0.00	34.20	0.00
Mixture (MIX)	0.00	0.00	0.00	0.00	60.60
Cellulose	5.00	5.00	5.00	5.00	3.00
Limestone	1.32	0.00	0.00	1.90	0.00
MDCP	1.94	0.20	0.40	0.40	0.32
Chromic oxide mixture[2]	2.50	2.50	2.50	2.50	2.50
Celite	1.00	1.00	1.00	1.00	1.00
NaCl	0.20	0.20	0.20	0.20	0.20
Potassium chloride	0.28	0.28	0.28	0.28	0.28
Magnesium oxide	0.20	0.20	0.20	0.20	0.20
$NaHCO_3$	0.80	0.80	0.80	0.80	0.80
Choline chloride	0.24	0.24	0.24	0.24	0.24
Vitamin premix[3]	0.30	0.30	0.30	0.30	0.30
Mineral premix[4]	0.30	0.30	0.30	0.30	0.30
Total	100.00	100.00	100.00	100.00	100.00

[1]IFE, infertile egg; LDC, low-grade or dead chick; MIX, mixture (55% IFE, 10% UHE, 10% LDC, and 25% hatched eggshells); N-free, nitrogen free diet; UHE, unhatched egg.

[2]Chromic oxide mixture = 20% of chromic oxide mixed with 80% of corn starch.

[3]Provided per kilogram of the complete diet: vitamin A (from vitamin A acetate), 13,000 IU; vitamin D_3, 5,000 IU; vitamin E (from DL-α-tocopheryl acetate), 80 IU; vitamin K_3, 4 mg; vitamin B_1, 4 mg; vitamin B_2, 10 mg; vitamin B_6, 6 mg; vitamin B_{12}, 20 μg; calcium pantothenate, 20 mg; folic acid, 2 mg; biotin, 200 μg; niacin, 60 mg.

[4]Provided per kilogram of the complete diet: Zn (as ZnO), 100 mg; Mn (as $MnSO_2 \cdot H_2O$), 120 mg; Fe (as $FeSO_4 \cdot 7H_2O$), 60 mg; Cu (as $CuSO_4 \cdot 5H_2O$), 16 mg; Co (as $CoCO_3$), 1,000 μg; I (as $Ca(IO_3)_2 \cdot H_2O$), 1.25 mg; Se (as Na_2SeO_3), 300 μg.

Chemical Analysis

The HBP samples, experimental diets, and excreta samples were measured for DM (method 930.15; AOAC, 2005), CP (method 990.03; AOAC, 2005), and GE using bomb calorimetry (Model 6400, Parr instruments Co., Moline, IL). Benzoic acid was used as the standard for calibration of GE analysis. Additionally, the HBP samples were analyzed for crude ash (method 942.05; AOAC, 2005) and acid-hydrolyzed ether extract (**AEE**, method 996.01; AOAC, 2005). The HBP samples were weighed and digested to measure Ca and phosphorus (**P**) concentrations using an inductively coupled plasma spectrometer (Optima 5300 DV, Perkin Elmer Inc., Shelton, CT) as demonstrated by Kim et al. (2016). The concentrations of AA in experimental diets, ileal digesta, and HBP samples were determined using a high performance liquid chromatography (Ultimate 3000, Thermo Dionex, Sunnyvale, CA) according to the method as described by AOAC (2005; method 982.30).

Calculation

In experiment 1, the AME and AME_n values for the experimental diets were calculated as followed (Wolynetz and Sibbald, 1984; Lee et al., 2018):

$$\text{AME (kcal/kg)} = (\text{GEi} - \text{GEo})/\text{FI},$$

$$\text{AME}_n \text{ (kcal/kg)} = [\text{GEi} - \{\text{GEo} + (\text{Ni} - \text{No}) \times 8.22\}]/\text{FI},$$

where GEi indicates the GE intake; GEo indicates the GE output; Ni − No indicates the gram N balance; GEe indicates endogenous loss of energy; 8.22 equals the N retained value (Hill and Anderson, 1958). The values for AME and AME_n of 4 HBP samples as an ingredient were calculated by difference procedure (Barzegar et al., 2019).

In experiment 2, the AID of AA was calculated as follows using the Cr marker as described by Ravindran et al. (2005) and Kong and Adeola (2013).

$$\text{AID, \%} = \left[(\text{AA/Cr})_d - -(\text{AA/Cr})_i\right]/(\text{AA/Cr})_d \times 100$$

Where AID is the apparent ileal AA digestibility (%), $(AA/Cr)_d$ = ratio of AA to Cr in diet; and $(AA/Cr)_i$ = ratio of AA to Cr in ileal digesta.

The endogenous losses of AA were calculated using following equation as described by Kong and Adeola (2013).

$$EAL = (Cr_d/Cr_i) \times AA_i$$

Where EAL is the endogenous losses of each AA measured in ileal digesta after feeding the NF, Cr_d is the Cr concentrations in NF, Cr_i is the Cr concentrations in ileal digesta, and AA_i is the concentration of each AA in ileal digesta.

The AID data were converted to SID values, using the endogenous AA losses values as described by Kong and Adeola (2013).

$$\text{SID, \%} = \text{AID} + (EAL/AA_d) \times 100$$

Where SID represents the standardized ileal AA digestibility (%), AA_d is the dietary concentrations of the AA.

Statistical Analysis

All data were analyzed by ANOVA as a completely randomized design using the GLM procedure of SAS (SAS Institute., Cary, NC). The replicate was used as the experimental unit for all analyses. Outlier data were checked using the UNIVARIATE procedure of SAS (Steel et al., 1997). The means of AME and AME_n values for experimental diets and various HBPs and those of AID and SID of AA in various HBPs were compared using the Duncan multiple range test. Significance for statistical tests was set at $P < 0.05$.

RESULTS

Energy and Nutrient Concentrations in HBPs

The concentrations of GE, CP, and AA were the greatest for LDC but the least for MIX among 4 HBPs. However, the concentrations of ash and Ca were the greatest for MIX but the least for LDC. The primary reason for these differences in energy and nutrients is related to different inclusion amounts of HE in HBPs because LDC had no additional inclusion of HE, MIX had the greatest inclusion of HE, and IFE and UHE contained the relatively small inclusion of HE.

Table 4. Metabolizable energy of experimental diets (experiment 1).[1]

	Experimental diets[2]						
Items	Basal	IFE	UHE	LDC	MIX	SEM	*P*-value
ME (kcal/kg)							
AME	3,317[c]	3,450[b]	3,342[c]	3,519[a]	3,347[c]	22.2	<0.01
AME_n	3,168[c]	3,279[b]	3,155[c]	3,368[a]	3,192[c]	20.4	<0.01

[a-c]Means within a variable with no common superscript differ significantly ($P < 0.05$).

[1]Data are least squares means of 8 observations per treatment.

[2]IFE, infertile egg; LDC, low-grade or dead chick; MIX, mixture (55% IFE, 10% UHE, 10% LDC, and 25% hatched eggshells); UHE, unhatched egg.

Table 5. Metabolizable energy of hatchery byproducts (experiment 1).[1]

	Hatchery byproducts[2]					
Items[3]	IFE	UHE	LDC	MIX	SEM	*P*-value
ME (kcal/kg)						
AME	4,680[b]	3,608[c]	5,375[a]	3,654[c]	230.2	<0.01
AME_n	4,314[b]	3,074[c]	5,198[a]	3,443[c]	197.8	<0.01

[a-c]Means within a variable with no common superscript differ significantly ($P < 0.05$).

[1]Data are least squares means of 8 observations per treatment.

[2]IFE, infertile egg; LDC, low-grade or dead chick; MIX, mixture (55% IFE, 10% UHE, 10% LDC, and 25% hatched eggshells); UHE, unhatched egg.

ME Concentrations and AA Digestibility

The ME (i.e., AME and AME_n) values for diets containing LDC were greater ($P < 0.05$) than those of diets containing IFE, which were greater ($P < 0.05$) than those of basal diets and diets containing UHE or MIX (Table 4). Likewise, the ME (i.e., AME and AME_n) values for LDC was greater ($P < 0.05$) than those for IFE, which were greater ($P < 0.05$) than for UHE or MIX (Table 5).

The average AID of essential AA was greater ($P < 0.05$) in IFE and LDC than in MIX with UHE having intermediate values for the average AID of essential AA (Table 6). The IFE, UHE, and LDC had greater ($P < 0.05$) AID of Met and Thr compare with MIX. The AID of Arg and Trp was greatest ($P < 0.05$) for LDC, intermediate for IFE and UHE, and least for MIX. There were no differences in the AID of Ile, Leu, Lys, and Phe among 4 HBPs.

The average AID of nonessential AA in IFE, UHE, and LDC was greater ($P < 0.05$) than those in MIX. Similar results were observed for the AID of Pro. As seen for the AID of essential AA, LDC had the greatest ($P < 0.05$) AID of Ala, Asp, Glu, and Gly, MIX had the least ($P < 0.05$) AID of those 5 nonessential AA, and IFE and UHE had intermediate AID values. The AID of Ser and Tyr did not differ among 4 HBPs.

The results for the SID of AA in HBPs were close to those for the AID of AA (Table 7). The average SID of essential AA was greater ($P < 0.05$) for LDC than for UHE and MIX with intermediate values being observed for IFE. The SID of 6 essential AA (Arg, His, Met, Thr, Trp, and Val) was greater ($P < 0.05$) for LDC than for MIX. The SID of His and Trp was greater ($P < 0.05$) for

Table 6. Apparent ileal amino acid digestibility (AID) of hatchery byproducts (experiment 2).[1]

Items	Hatchery byproducts[2]				SEM	*P*-value
	IFE	UHE	LDC	MIX		
AID, %						
Essential amino acid						
Arg	84.7[ab]	82.9[bc]	87.0[a]	80.7[c]	1.13	<0.01
His	80.9[b]	81.5[b]	85.3[a]	78.7[b]	1.09	<0.01
Ile	87.3	84.3	87.9	86.2	0.97	0.07
Leu	88.3	85.7	88.2	86.1	1.04	0.19
Lys	85.7	85.2	88.5	83.1	1.43	0.09
Met	88.1[a]	89.5[a]	91.0[a]	84.5[b]	0.95	<0.01
Phe	88.9	85.1	86.7	87.1	1.07	0.12
Thr	82.1[a]	80.5[a]	83.7[a]	76.5[b]	1.34	<0.01
Trp	86.5[b]	86.2[b]	97.8[a]	79.8[c]	1.24	<0.01
Val	86.8[ab]	84.1[b]	87.4[a]	84.2[b]	0.91	0.03
Mean	85.9[ab]	84.5[bc]	88.4[a]	82.7[c]	1.04	<0.01
Nonessential amino acid						
Ala	85.5[b]	85.2[b]	89.0[a]	82.6[b]	1.20	<0.01
Asp	83.3[ab]	81.5[b]	86.0[a]	81.5[b]	1.21	<0.05
Cys	71.5[bc]	77.4[a]	74.7[ab]	69.9[c]	1.44	<0.01
Glu	86.2[ab]	85.5[ab]	88.7[a]	83.3[b]	1.10	0.02
Gly	80.1[bc]	83.7[b]	87.5[a]	77.5[c]	1.24	<0.01
Pro	86.1[a]	80.2[a]	80.1[a]	66.2[b]	2.73	<0.01
Ser	79.1	78.7	80.4	78.4	1.21	0.68
Tyr	86.9	82.3	83.8	84.1	1.15	0.07
Mean	82.3[a]	81.8[a]	83.8[a]	77.9[b]	1.23	0.02

[a-c]Means within a variable with no common superscript differ significantly ($P < 0.05$).

[1]Data are least squares means of 7 observations per treatment.

[2]IFE, infertile egg; LDC, low-grade or dead chick; MIX, mixture (55% IFE, 10% UHE, 10% LDC, and 25% hatched eggshells); UHE, unhatched egg.

LDC than for IFE and UHE. The SID of Thr and Trp was greater ($P < 0.05$) for IFE than for MIX. The SID of Met and Trp was greater ($P < 0.05$) for UHE than for MIX.

Table 7. Standardized ileal amino acid digestibility (SID) of hatchery byproducts (experiment 2).[1]

Items	Hatchery byproducts[2]				SEM	*P*-value
	IFE	UHE	LDC	MIX		
SID, %						
Essential amino acid						
Arg	88.9[ab]	87.0[b]	91.3[a]	85.6[b]	1.13	<0.01
His	85.7[b]	85.9[b]	90.0[a]	83.7[b]	1.09	<0.01
Ile	90.7[ab]	87.8[b]	91.8[a]	90.6[ab]	0.97	<0.05
Leu	92.0	89.4	92.2	90.8	1.04	0.23
Lys	90.0	90.0	93.6	88.8	1.43	0.13
Met	90.6[bc]	92.1[ab]	94.5[a]	87.4[c]	0.95	<0.01
Phe	92.4	88.6	90.6	91.7	1.07	0.09
Thr	88.8[a]	87.2[ab]	90.7[a]	84.6[b]	1.34	0.02
Trp	89.1[b]	89.8[b]	100.7[a]	83.7[c]	1.24	<0.01
Val	90.3[ab]	87.7[b]	91.5[a]	88.6[b]	0.91	0.03
Mean	89.9[ab]	88.5[b]	92.7[a]	87.5[b]	1.04	0.01
Nonessential amino acid						
Ala	90.4[ab]	89.7[b]	93.5[a]	88.6[b]	1.20	<0.05
Asp	87.0[ab]	85.3[b]	90.1[a]	86.1[b]	1.21	<0.05
Cys	79.8[ab]	83.4[a]	81.2[a]	76.4[b]	1.44	0.02
Glu	89.9	88.8	92.1	87.7	1.10	0.06
Gly	86.5[ab]	87.4[a]	90.2[a]	83.0[b]	1.24	<0.01
Pro	90.9[a]	85.4[a]	84.1[a]	73.3[b]	2.73	<0.01
Ser	83.3	83.1	85.2	83.6	1.21	0.59
Tyr	91.0	86.8	89.7	88.9	1.15	0.10
Mean	87.4	86.2	88.3	83.5	1.23	0.06

[a-c]Means within a variable with no common superscript differ significantly ($P < 0.05$).

[1]Data are least squares means of 7 observations per treatment.

[2]IFE, infertile egg; LDC, low-grade or dead chick; MIX, mixture (55% IFE, 10% UHE, 10% LDC, and 25% hatched eggshells); UHE, unhatched egg.

The average SID of nonessential AA did not differ among 4 HBPs. However, the SID of Pro was greater ($P < 0.05$) for IFE, UHE, and LDC than for MIX. The SID of Ala and Asp was the greatest ($P < 0.05$) for LDC, intermediate for IFE, and the least ($P < 0.05$) for MIX and UHE. There were no differences in the SID of Glu, Ser, and Tyr among 4 HBPs.

DISCUSSION

The accurate determination of energy values for HBPs is mandatory if the HBPs are properly used for broiler diets. However, there have been scarce data for energy concentrations of HBPs, especially for individual sources of HBPs such as IFE, UHE, and LDC, because HBPs are typically collected and used as a mixed form (i.e., MIX) for broiler diets. The GE concentrations of MIX used in the current experiment were similar or lower than those reported in previous studies (Rasool et al., 1999; Mehdipour et al., 2009; Sung et al., 2019). The variable GE concentrations in MIX result from different sources and inclusion amounts of individual HBPs, which are largely dependent of the hatchability in the commercial hatchery (Rasool et al., 1999; Thaler and Holden, 2010; Sung and Kim, 2020). The MIX used in the previous studies contained relatively small amounts of HE, whereas the MIX used in this experiment contained 25% HE. This difference in the inclusion amount of HE in the MIX may be the reason why the GE concentrations of MIX used in the current experiment were similar or lower than those reported in previous experiments.

The values for AME_n of 4 HBPs measured in this experiment ranged from 3,074 to 5,198 kcal/kg, falling within the range (2,706−5,712 kcal/kg) reported in the literature (Ilian and Salman, 1986; Sharara et al., 1992; Abiola and Onunkwor, 2004). The values for AME and AME_n of LDC were greater than those of IFE. The main reason for this difference is likely due to the different GE concentrations but similar energy metabolizability between LDC and IFE. This reason also explains why the values for AME and AME_n of IFE were greater than those of UHE. However, the values for AME and AME_n of UHE did not differ from those for MIX although the GE concentrations of MIX (3,764 kcal/kg) was lower than those of UHE (4,753 kcal/kg). Based on GE intake and GE excretion, the GE metabolizability was calculated and the values were less for UHE (64.7%) than for LDC (87.4%), IFE (84.0%), and MIX (91.5%). It is not clear why such a trend was identified because UHE is likely an intermediate form during a hatching process and no experiments have reported that the specific chemical and physical components in UHE decreased energy and nutrient utilization in UHE.

In the current experiment, the CP concentrations in 4 HBPs ranged from 31.2 to 60.0%, which were closed to the values (32.2−66.3%) reported in previous experiments (Sung et al., 2019; Sung et al., 2020; Sung and Kim, 2020). These results indicate that HBPs used in

the current experiment are similar to those used in previous experiments. Interestingly, the LDC had the greatest CP concentrations among 3 individual HBPs, probably due to few eggshells present in LDC. The CP concentrations in LDC were similar or lower than those values reported in previous experiment (Sung et al., 2019; Sung et al., 2020).

Among the essential AA, 4 HBPs contained high concentrations of total Arg, Leu, Lys, and Val, but low concentrations of total His and Trp. These AA compositions are likely similar to those in eggs (Norberg et al., 2004; Donadelli et al., 2019) and other animal byproducts (Ravindran et al., 2005; Adedokun et al., 2014; Sung et al., 2020). Among 4 HBPs, LDC had the highest concentrations of essential AA, followed by IFE and UHE, whereas MIX had the least concentrations of essential AA, which is likely a consequence of different amounts of eggshells included in each HBP.

The HBPs can be used in broiler diets as an alternative source to conventional animal protein ingredients such as meat and bone meal and fish meal. Thus, AA digestibility of HBPs is worthy of comparison with other protein ingredients. In the previous studies, the average AID of AA in meat and bone meal ranged from 61 to 80%, and that in fish meal ranged from 77 to 87% (Angkanaporn et al., 1996; Ravindran et al., 1999, 2005; Kadim et al., 2002; Huang et al., 2005). In addition, the average SID of AA in meat and bone meal ranged from 64 to 78%, and that in fish meal ranged from 82 to 91% (Angkanaporn et al., 1996; Kadim et al., 2002; Lemme et al., 2004; Adedokun et al., 2014). Our values for the average AID of AA in HBPs ranged from 81 to 86%, and the SID of AA ranged from 86 to 91%. Therefore, the values for both AID and SID of AA in HBPs used in the current experiment were similar or greater than those values measured in meat and bone meal and fish meal. This result indicates that the individual HBPs and their mixture (i.e., IFE, UHE, LDC, and MIX) can be potential animal protein ingredients for broiler diets.

The LDC had the greatest AID of most of the essential AA, whereas the MIX had the least AID of those AA with IFE and UHE showing the intermediate values. This result appeared to be associated with different amounts of total AA among 4 HBPs because the AID of AA increases with increasing AA concentrations in HBPs due to a concomitant decrease in the relative contribution of endogenous losses of AA in the excreta (Stein et al., 2007). However, the SID of most of the essential AA was also greater for LDC than for UHE and MIX, indicating that essential AA in LDC can be more digestible than in UHE and MIX. To our knowledge, this is the first experiment reporting the AID and SID of AA in various HBPs for broiler chickens. Thus, the direct comparison of our measured values with previous ones in poultry experiments was difficult. However, Sung et al. (2020) reported that the AID and SID of most of the essential AA were greater for IFE than for LDC when fed to nursery pigs. In addition, there were no differences in the AID and SID of most of the essential AA between LDC and MIX. The reason for this observation is likely related to differences in the drying process of HBPs. In the present study, the HBPs were dried at 50°C for 24 h in a vertical convention oven, whereas the HBPs were dried at 130°C for 20 h in a dryer in the other experiment (Sung et al., 2020). The possible reason for the difference in AA digestibility may be associated with a decrease in AA availability when ingredients were dried at the high temperature (Navarro et al., 2018). Another possible explanation for differences in AA digestibility is the different proportions of individual HBPs in the MIX. The HBPs used in our experiment contained 55% IFE, 10% UHE, 10% LDC, and 25% HE, whereas the HBPs used by Sung et al. (2020) contained 20% IFE, 20% UFE, and 60% LDC. These results may indicate that the AID and SID of AA in various HBPs may be affected by the drying process and inclusion proportions of HBPs in the MIX.

CONCLUSIONS

The LDC has the greatest ME concentrations and AA digestibility among 3 individual HBPs (IFE, UHE, and LDC), whereas the MIX has relatively less ME concentrations and AA digestibility for broiler chickens. The ME and AA digestibility of HBPs are likely affected by inclusion amounts of HE. However, high concentrations of ME and available AA indicate that individual HBPs and their mixture are potential protein ingredients for broiler diets.

ACKNOWLEDGMENTS

This research was carried out with the support of the Cooperative Research Program for Agriculture Science and Technology Development (Project No. PJ01252804), Rural Development Administration, Republic of Korea. This research was also supported by the Chung-Ang University Graduate Research Scholarship (Academic scholarship for College of Biotechnology and Natural Resources) in 2021.

DISCLOSURES

The authors declare no conflict of interest for the data presented in this experiment.

REFERENCES

Abiola, S. S., N. E. Radebe, C. V. D. Westhuizen, and D. O. Umesiobi. 2012. Whole hatchery waste meal as alternative protein and calcium sources in broiler diets. Archiv. Zootec. 61:229–234.

Abiola, S. S., and E. K. Onunkwor. 2004. Replacement value of the hatchery waste meal for fish meal in layer diets. Bioresour. Technol. 95:103–106.

Adedokun, S. A., P. Jaynes, M. E. Abd El-Hack, R. L. Payne, and T. J. Applegate. 2014. Standardized ileal amino acid digestibility of meat and bone meal and soybean meal in laying hens and broilers. Poult. Sci. 93:420–428.

Angkanaporn, K., V. Ravindran, and W. L. Bryden. 1996. Additivity of apparent and true ileal amino acid digestibilities in soybean meal, sunflower meal, and meat and bone meal for broilers. Poult. Sci. 75:1098–1103.

Association of Official Analytical Chemists (AOAC). 2005. Official Methods of Analysis of AOAC International. 18th ed. AOAC International, Gaithersburg, MD.

Association of American Feed Control Officials (AAFCO). 2016. Official Publication. AAFCO, Atlanta, GA.

Barzegar, S., S.-B. Wu, J. Noblet, and R. A. Swick. 2019. Metabolizable energy of corn, soybean meal and wheat for laying hens. Poult. Sci. 98:5876–5882.

Bourdillon, A., B. Carré, L. Conan, J. Duperray, G. Huyghebaert, B. Leclercq, M. Lessire, J. McNab, and J. Wiseman. 1990. European reference method for the in vivo determination of metabolisable energy with adult cockerels: reproducibility, effect of food intake and comparison with individual laboratory methods. Br. Poult. Sci. 31:557–565.

Choi, W. J., J. H. Kim, G. P. Han, C. H. Kwon, and D. Y. Kil. 2021. Effects of dietary hatchery by-products on growth performance, relative organ weight, plasma measurements, immune organ index, meat quality, and tibia characteristics of broiler chickens. Anim. Biosci. 34:1181–1192.

Das, K. C., M. Y. Minkara, N. D. Melear, and E. W. Tollner. 2002. Effect of poultry litter amendment on hatchery waste composting. J. Appl. Poult. Res. 11:282–290.

Donadelli, R. A., C. G. Aldrich, C. K. Jones, and R. S. Beyer. 2019. The amino acid composition and protein quality of various egg, poultry meal by-products, and vegetable proteins used in the production of dog and cat diets. Poult. Sci. 98:1371–1378.

Glatz, P., Z. Miao, and B. Rodda. 2011. Handling and treatment of poultry hatchery waster: a review. Sustainability 3:216–237.

Hill, F. W., and D. L. Anderson. 1958. Comparison of metabolizable energy and productive energy determinations with growing chicks. J. Nutr. 64:587–603.

Huang, K. H., V. Ravindran, X. Li, and W. L. Bryden. 2005. Influence of age on the apparent ileal amino acid digestibility of feed ingredients for broiler chickens. Br. Poult. Sci. 46:236–245.

Ilian, M. A., and A. J. Salman. 1986. Feeding processed hatchery wastes to poultry. Agric. wastes 15:179–186.

Kadim, I. T., P. J. Moughan, and V. Ravindran. 2002. Ileal amino acid digestibility assay for the growing meat chicken−comparison of ileal and excreta amino acid digestibility in the chicken. Br. Poult. Sci. 43:588–597.

Kim, J. H., G. H. Park, G. P. Han, and D. Y. Kil. 2021. Effect of feeding corn distillers dried grains with solubles naturally contaminated with deoxynivalenol on growth performance, meat quality, intestinal permeability, and utilization of energy and nutrients in broiler chickens. Poult. Sci. 100:101215.

Kim, J. W., J. H. Kim, J. E. Shin, and D. Y. Kil. 2016. Relative bioavailability of copper in tribasic copper chloride to copper in copper sulfate for laying hens based on egg yolk and feather copper concentrations. Poult. Sci. 95:1591–1597.

Kong, C., and O. Adeola. 2013. Additivity of amino acid digestibility in corn and soybean meal for broiler chickens and white pekin ducks. Poult. Sci. 92:2381–2388.

Lee, B. B., T. S. Yang, D. Goo, H. S. Choi, F. M. Pitargue, H. Jung, and D. Y. Kil. 2018. Effects of dietary β-mannanase supplementation on the additivity of true metabolizable energy values for broiler diets. Asian-Australas. J. Anim. Sci. 31:564–568.

Lemme, A., V. Ravindran, and W. L. Bryden. 2004. Ileal digestibility of amino acids in feed ingredients for broilers. Worlds Poult. Sci. J. 60:423–438.

Mehdipour, M., M. S. Shargh, B. Dastar, and S. Hassani. 2009. Effects of different levels of hatchery wastes on the performance, carcass and tibia ash and some blood parameters in broiler chicks. Pak. J. Biol. Sci. 12:1272–1276.

Navarro, D. M. D. L., J. K. Mathai, N. W. Jaworski, and H. H. Stein. 2018. Amino acid digestibility in six sources of meat and bone meal, blood meal, and soybean meal fed to growing pigs. Can. J. Anim. Sci. 98:860–867.

Norberg, S. E., R. N. Dilger, H. Dong, B. G. Harmon, O. Adeola, and M. A. Latour. 2004. Utilization of energy and amino acids of spray-dried egg, plasma protein, and soybean meal by ducks. Poult. Sci. 83:939–945.

Rasool, S., M. Rehan, A. Haq, and M. Z. Alam. 1999. Preparation and nutritional evaluation of hatchery waste meal for broilers. Asian-Australas. J. Anim. Sci. 12:554–557.

Ravindran, V., L. I. Hew, G. Ravindran, and W. L. Bryden. 1999. A comparison of ileal digesta and excreta analysis for the determination of amino acid digestibility in food ingredients for poultry. Br. Poult. Sci. 40:266–274.

Ravindran, V., L. I. Hew, G. Ravindran, and W. L. Bryden. 2005. Apparent ileal digestibility of amino acids in dietary ingredients for broiler chickens. Anim. Sci. 81:85–97.

Ravindran, V., O. Adeola, M. Rodehutscord, H. Kluth, J. D. van der Klis, E. van Eerden, and A. Helmbrecht. 2017. Determination of ileal digestibility of amino acids in raw materials for broiler chickens − results of collaborative studies and assay recommendations. Anim. Feed Sci. Technol. 225:62–72.

Shahriar, H. A., K. Nazer-Adl, J. Doolgarisharaf, and H. Monirifar. 2008. Effects of dietary different levels of hatchery wastes in broiler. J. Anim. Vet. Adv. 7:100–105.

Sharara, H. H., H. Y. El-Hammady, and H. Abd-El-Fattah. 1992. Nutritive value of some non-conventional by-products as poultry feed ingredients. 2. Biological determination of metabolizable energy. Assiut J. Agric. Sci. 23:349–363.

Steel, R. G. D., J. H. Torrie, and D. A. Dickey. 1997. Principles and Procedures of Statistics: A Biometrical Approach. 3rd ed. McGraw Hill Book Co., New York, NY.

Stein, H. H., B. Seve, M. F. Fuller, P. J. Moughan, and C. F. M. de Lange. 2007. Invited review: amino acid bioavailability and digestibility in pig feed ingredients: terminology and application. J. Anim. Sci. 85:172–180.

Sung, J. Y., S. Y. Ji, and B. G. Kim. 2020. Amino acid and calcium digestibility in hatchery byproducts fed to nursery pigs. Anim. Feed Sci. Technol. 270:114703.

Sung, J. Y., and B. G. Kim. 2020. Effects of a hatchery byproduct mixture on growth performance and digestible energy of various hatchery byproduct mixtures in nursery pigs. Animals 10:174.

Sung, J. Y., S. Y. Ji, A. R. Son, and B. G. Kim. 2019. Energy concentration and phosphorus digestibility in hatchery byproducts fed to nursery pigs. Animals 9:255.

Thaler, B., and P. Holden. 2010. By-product feed ingredients for use in swine diets. National Swine Nutrition Guide. US Pork excellence Center, Iowa State University Extension Service, Ames, IA.

Wolynetz, M. S., and I. R. Sibbald. 1984. Relationships between apparent and true metabolizable energy and the effects of a nitrogen correction. Poult. Sci. 63:1386–1399.