Standardized amino acid digestibility in cecectomized roosters and lysine bioavailability in chicks fed distillers dried grains with solubles

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ABSTRACT This study was conducted to compare the concentration of standardized digestible (SDD) Lys and relative bioavailable Lys in 7 sources of corn distillers dried grains with solubles (DDGS). A second objective was to evaluate 2 in vitro methods, reactive Lys and color score, to predict the concentration of SDD Lys and bioavailable Lys in DDGS. Seven sources of DDGS were fed to cecectomized roosters, and digestibility of amino acids was measured using the total excreta collection method. To measure the relative bioavailable Lys in DDGS, a standard curve ($r^2 = 0.96$, $P < 0.01$) was constructed from 9-d weight gain of young chicks fed a Lys-deficient basal diet or diets containing increasing concentrations of L-Lys-HCl. Seven additional diets were formulated by adding each of the 7 sources of DDGS to the basal diet, and total weight gain of chicks was measured. Weight gain of chicks fed each DDGS-containing diet was then compared with the standard curve to calculate the bioavailable Lys and bioavailability of Lys in each source of DDGS. All DDGS sources were analyzed for reactive Lys using the guanidination procedure, and a Hunterlab color score was used to measure the degree of lightness (L), redness (a), and yellowness (b). Results showed that the mean SDD Lys values and the mean relative bioavailability of Lys were 61.4 and 69.0%, respectively. Differences between the concentration of SDD Lys and the concentration of bioavailable Lys were not observed in 5 of 7 sources of DDGS. The concentration of SDD Lys was correlated ($r^2 = 0.84$, $P < 0.05$) with the concentration of reactive Lys in DDGS. Greater Hunterlab L scores were associated with a greater ($r^2 = 0.90$, $P < 0.05$) concentration of bioavailable Lys in DDGS. In conclusion, the concentration of SDD Lys in DDGS does not overestimate the concentration of bioavailable Lys for poultry. Values for reactive Lys may be used to estimate the concentration of SDD Lys, whereas Hunterlab L may be used to estimate the concentration of bioavailable Lys in DDGS.

Key words: amino acid, availability, distillers dried grains with solubles, standardized digestibility, rooster

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INTRODUCTION

Distillers dried grains with solubles (**DDGS**) is a coproduct from the dry milling of grains, which is the residual component of the grain kernel after the starch has been fermented by yeast to produce ethanol. Heat processing is needed to reduce the moisture concentration of wet distillers grains, but it may reduce the utilization of heat-sensitive amino acids (**AA**) such as Lys (Cromwell et al., 1993). Lysine and reducing sugars in DDGS can interact, which can lead to the initiation of the Maillard reaction (Maillard, 1912). When Lys is complexed with reducing sugars, it becomes unreactive Lys (Hurrell and Carpenter, 1981; Friedman, 1982). Because unreactive Lys is biologically unavailable but may be partially absorbed in the intestine (Finot and Magnenat, 1981), it is hypothesized that the conventional digestibility measurement may overestimate the amount of bioavailable Lys in DDGS.

Measurement of digestibility and relative bioavailability of AA are relatively expensive and tedious. Among the in vitro methods that may be used to estimate bioavailable Lys is the reactive Lys procedure that measures the amount of free ε-NH_2 groups of Lys in heated proteins (Hurrell and Carpenter, 1981). There is, however, no information on the correlation between the measured quantity of reactive Lys in DDGS, the quantity of standardized digestible (**SDD**) Lys, and the amount of relative bioavailable Lys in DDGS fed to poultry. It may also be possible to evaluate the quality of DDGS based on color, because darker DDGS results in lower average daily gain of broiler chicks (Cromwell et al., 1993). However, there is no information about the correlation between bioavailable Lys and color of DDGS.

The objective of this study was to compare the concentration of relative bioavailable Lys and SDD Lys in

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Table 1. Composition of 7 sources of distillers dried grains with solubles (DDGS), as-fed basis[1]

	DDGS source							
Item	1	2	3	4	5	6	7	Mean
DM, %	87.5	86.9	89.4	87.9	86.8	88.3	83.3	87.2
CP, %	26.9	25.8	23.9	28.0	24.6	26.5	28.9	26.4
Acid detergent fiber, %	12.5	9.2	8.1	8.7	8.3	9.2	12.8	9.8
Neutral detergent fiber, %	41.5	33.8	33.8	36.6	36.0	38.1	41.1	37.3
Crude fat, %	13.0	9.6	9.3	10.3	10.2	11.0	9.0	13.0
Indispensable amino acids, %								
Arg	1.35	1.12	1.03	1.24	1.04	1.19	1.15	1.16
His	0.80	0.70	0.65	0.76	0.62	0.71	0.69	0.70
Ile	1.08	1.00	0.92	1.04	0.84	1.02	1.02	0.99
Leu	3.32	3.09	2.83	3.45	2.67	3.17	3.21	3.11
Lys	0.94	0.78	0.65	0.84	0.71	0.74	0.72	0.77
Met	0.57	0.51	0.46	0.57	0.45	0.52	0.49	0.51
Phe	1.40	1.29	1.19	1.43	1.14	1.35	1.34	1.31
Thr	1.05	0.93	0.91	1.10	0.88	1.05	1.04	0.99
Trp	0.18	0.17	0.16	0.18	0.16	0.18	0.18	0.17
Val	1.49	1.36	1.26	1.41	1.14	1.39	1.39	1.35
Dispensable amino acids, %								
Ala	2.01	1.84	1.69	2.00	1.61	1.86	1.91	1.85
Asp	1.81	1.63	1.52	1.77	1.45	1.70	1.73	1.66
Cys	0.54	0.49	0.45	0.55	0.43	0.49	0.48	0.49
Glu	4.20	3.79	3.36	3.91	3.22	3.49	3.79	3.68
Gly	1.19	1.02	0.96	1.09	0.93	1.06	1.04	1.04
Pro	1.99	1.81	1.68	2.00	1.61	1.82	0.19	1.59
Ser	1.23	1.05	1.05	1.33	1.06	1.25	1.23	1.17
Tyr	1.03	0.98	0.92	1.16	0.90	1.07	1.06	1.02

[1]Values are the mean of duplicate analysis.

DDGS fed to poultry. The second objective was to determine if the concentration of reactive Lys or the color of DDGS can be used to predict the concentration of SDD Lys and bioavailable Lys.

MATERIALS AND METHODS

Samples of DDGS

Seven sources of DDGS from dry-grind ethanol plants in Minnesota, Michigan, Missouri, Illinois, and South Dakota were used in the experiment (Table 1). The SDD of AA in each source of DDGS was measured using cecectomized roosters, whereas the relative bioavailability of Lys was measured by chick growth assay (see below). The DDGS sources were analyzed for reactive Lys, and the degree of lightness (**L**), redness (**a**), and yellowness (**b**) was measured using the Hunterlab colorimeter (Hunter Associates Laboratory, Reston, VA). All experimental protocols involving use of animals were approved by the University of Illinois Animal Care and Use Committee.

Birds, Housing, and Experimental Design

Digestibility Study. Cecectomized Single Comb White Leghorn roosters (45 wk old) were used in the experiment. Roosters were cecectomized using the procedure of Parsons (1985) and housed in 22.5- × 36-cm individual cages with raised wire floors in an environmentally controlled room. A 16-h light and 8-h dark cycle was provided, and water was accessible at all times. Five roosters were allotted to each of the 7 DDGS sources in a completely randomized design. The roosters were deprived of feed for 24 h and then fed 30 g of DDGS via crop intubation. The basal endogenous losses of AA were measured from 5 additional roosters that were deprived of feed for 48 h. The excreta were collected quantitatively for 48 h starting immediately after crop intubation with collection via a plastic tray that was placed under each rooster. The SDD of AA were calculated using the method described by Sibbald (1979).

Lysine Bioavailability Study. The relative bioavailability of Lys in the 7 sources of DDGS was measured using the standard curve method (de Muelenaere, 1967a,b; Robbins and Baker, 1980). Crystalline L-Lys-HCl was used as the reference AA. New Hampshire × Columbian Plymouth Rock male chicks (total of 220 chicks; mean BW of 97.8 g) were fed a corn and soybean meal-based pretest starter diet for 7 d. This diet was formulated to contain nutrients according to NRC (1994) requirements. Chicks were housed in battery cages and raised wire floors in an environmentally controlled room. Water and artificial light was provided at all times. On d 8 posthatch, chicks were randomly allotted to 11 diets (a basal diet, 3 diets with increasing Lys supplementation from L-Lys-HCl to the basal diet, and 7 diets with Lys from DDGS) in a completely randomized design with 5 chicks per pen and 4 replicate pens per diet.

To construct the growth standard curve, 3 diets were mixed by adding feed-grade L-Lys-HCl at 0.094, 0.1875, and 0.281%, respectively, to the basal diet at the expense of cornstarch (Table 2). Thus, the calculated supplemental (bioavailable) Lys from L-Lys-HCl (78% L-Lys) was 0, 0.075, 0.15, and 0.225% for the basal plus Lys-supplemented diets, respectively. A preliminary experiment conducted before this experiment and with the same source of chicks had confirmed that, at these levels of Lys supplementation to the basal diet, a linear response in growth of chicks is obtained in response to increasing Lys intake. The 7 DDGS-containing diets were formulated by adding 20% of each source of DDGS to the basal diet at the expense of cornstarch. Chicks were fed the experimental diets from d 8 to 17 posthatch, and the total weight gain and feed consumption during this period were recorded.

Reactive Lys Analysis. The quantity of reactive Lys was analyzed in the 7 DDGS sources using the homoarginine procedure that involves guanidination of samples with O-methylisourea (Rutherfurd and Moughan, 1990). A 0.2-g sample of each source of DDGS was placed in a 125-mL flask, and 6 mL of 0.6 *M* O-methylisourea solution (pH 11.4) was added to the flask. The samples were stirred for 12 h at 20°C using a magnetic stirrer (MultiMagnestir 1278, Lab-Line Instruments, Melrose Park, IL) followed by a 60-h incubation at 20°C. The guanidinated samples were air-dried and analyzed for homoarginine (method 982.30E, step a; AOAC International, 2006). The reactive Lys was calculated based on the amount of homoarginine in the samples.

Colorimetry. The 7 DDGS sources were analyzed for L, a, and b using a Hunterlab Miniscan XE colorimeter (Hunter Associates Laboratory). Each DDGS sample was placed in a 1-cm-deep clear Petri dish with a transparent cap. Ten color measurements were obtained for each sample. Based on the Hunterlab scale (Hunterlab, 2001), a lower L value represents a darker color (0 = black), whereas a greater L value represents a lighter color, with white having an L value of 100. Positive, negative, or zero values of "a" indicate that the sample is predominantly red, green, or neutral, respectively. Positive, negative, or zero values of "b" indicate that the sample is predominantly yellow, blue, or neutral, respectively.

Other Analyses. All diets and the dried excreta from cecectomized roosters were analyzed for all AA except Met, Cys, and Trp following 22-h acid hydrolysis (method 982.30E, step a; AOAC International, 2006). The DDGS samples were analyzed for CP by the combustion method (method 990.03; AOAC International, 2006), and all AA. Methionine and Cys were analyzed after oxidation hydrolysis with performic acid (method 982.30E, step b; AOAC International, 2006). Tryptophan was analyzed after alkali oxidation using 4.2 *M* NaOH and boiling at 110°C for 24 h (method 982.30E, step c; AOAC International, 2006). The AA concentration of samples was quantified using HPLC. The DDGS samples were also analyzed for acid detergent fiber (**ADF**; method 973.18, AOAC International, 2006), neutral detergent fiber (**NDF**; Holst, 1973), and fat (method 954.02; AOAC International, 2006).

Table 2. Composition of the Lys-deficient basal diet and the diets containing distillers dried grains with solubles (DDGS) used in the Lys bioavailability study, as-fed basis

	Diet	
Item	Basal	DDGS
Cornstarch	20.00	—
Ground corn	40.00	40.00
Corn gluten meal	25.00	25.00
Soybean meal	8.00	8.00
DDGS	—	20.00
Soybean oil	2.00	2.00
Dicalcium phosphate	2.00	2.00
Limestone	1.40	1.40
NaCl	0.40	0.40
Vitamin premix[1]	0.20	0.20
Trace mineral premix[2]	0.15	0.15
Choline chloride (60%)	0.13	0.13
L-Trp	0.10	0.10
L-Thr	0.12	0.12
L-Lys	—	—
L-Arg	0.45	0.45
DL-Met	0.05	0.05
Bacitracin premix[3]	0.025	0.025
Total	100.0	100.0
Calculated content		
CP, %	22.78	28.38
TME_n, kcal/kg	3,386	3,326
Digestible Lys, %	0.52	0.64

[1]Provided per kilogram of diet: retinyl acetate, 4,400 IU; cholecalciferol, 25 μg; DL-α-tocopheryl acetate, 11 IU; vitamin B_{12}, 0.01 mg; riboflavin, 4.41 mg; D-pantothenic acid, 10 mg; niacin, 22 mg; menadione sodium bisulfite, 2.33 mg.

[2]Provided per kilogram of diet: Fe, 75 mg ($FeSO_4 \cdot H_2O$); Zn, 75 mg (ZnO); Mn, 75 mg (MnO); Cu, 5 mg ($CuSO_4 \cdot 5H_2O$); I, 0.75 mg (ethylenediamine dihydroiodide); Se, 0.1 mg (Na_2SeO_3).

[3]Contributed 25 mg of bacitracin per kilogram of diet as bacitracin methylene disalicylate (Alpharma Inc., Fort Lee, NJ).

Calculations

The concentration (i.e., % per unit weight) and digestibility values (i.e., SDD coefficient × 100) of AA were calculated using the following equation (Sibbald, 1979):

$$\text{SDD AA (\%)} = 100 \times \{\text{AA intake (mg)} - [\text{AA in excreta (mg)} - \text{endogenous AA loss (mg)}]\}/\text{AA intake (mg)}$$

where SDD AA is the standardized digestibility, and endogenous AA is the AA from excreta of roosters that were deprived of feed. The concentration of SDD Lys in each DDGS source was subsequently calculated:

$$\text{Concentration of SDD Lys (\%)} = \text{analyzed Lys in DDGS (\%)} \times [\text{SDD Lys (\%)}/100].$$

For Lys bioavailability in the chick assay, the standard curve for the relative bioavailability assay was constructed from the total gain of chicks fed the basal diet and the 3 diets containing increasing concentration of L-Lys-HCl. A best-fit regression equation was then derived. Bioavailable Lys was estimated by substituting weight gain of chicks (y) fed each source of DDGS into the standard-curve linear regression, after which the intake of bioavailable Lys (x) was calculated (Sasse and Baker, 1973). Intake of bioavailable Lys divided by total observed DDGS intake resulted in an estimate of the bioavailable Lys concentration in each sample of DDGS.

Relative bioavailability of Lys (%) was calculated using the following equation:

$$\text{Relative bioavailability} = 100 \times \text{calculated bioavailable Lys (g/kg of DDGS)/analyzed Lys (g/kg of DDGS)}.$$

The concentration of reactive Lys in DDGS was calculated based on the concentration of homoarginine in the samples after guanidination. The concentration of homoarginine was then converted to Lys (reactive Lys) on a molar basis (Rutherfurd et al., 1997):

$$\text{Reactive Lys (\%)} = [\text{homoarginine (\%)/MW of homoarginine}] \times \text{MW of Lys}.$$

Statistical Analysis

Digestibility data were analyzed as a completely randomized design using PROC MIXED of SAS (SAS Institute, 2004). The experimental unit was the rooster, the fixed effect was source of DDGS, and the random effect was the replicate. Mean digestibility values were calculated as least squares means and compared using the PDIFF option of SAS. In the bioavailability study, the experimental unit was the pen of 5 chicks, the random effect in the model was the replicate, and the fixed effect was the source of DDGS. Means were separated using the PDIFF option of SAS. The concentrations of relative bioavailable Lys and SDD Lys in the 7 sources of DDGS were compared by the *t*-test procedure (Rao, 1997) using SAS.

Concentrations of SDD Lys and relative bioavailable Lys in each source of DDGS were predicted from the concentrations of NDF, ADF, reactive Lys, and Hunterlab color scores using PROC CORR of SAS. In all comparisons, a difference of $P < 0.05$ was considered significant.

RESULTS

The mean CP, ADF, NDF, and crude fat concentrations in the 7 sources of DDGS were 26.4, 9.8, 37.3, and 13.0%, respectively. Lysine ranged from 0.65 to 0.94% with a mean of 0.77% (Table 1). The DDGS sources differed ($P < 0.05$) in SDD for Leu, Lys, Met ($P = 0.054$), Glu, and Pro, whereas the SDD for all other AA was similar among sources (Table 3). The mean SDD for Lys was 61.4 ± 3.16%. Source 6 (52.7%) had the lowest ($P < 0.05$) SDD for Lys, whereas source 1 (70.4%) had the greatest ($P < 0.05$) SDD for Lys.

The best-fit regression equation in the chick bioavailability assay was y = 77.29 + 118.88x (where y is chick gain, g, and x is supplemental Lys intake, g); an r^2 of 0.97 was obtained (Figure 1). The calculated mean relative bioavailability of Lys as percentage of analyzed Lys in each source of DDGS was 69.0% (Table 4). Differences were observed in gain:feed of chicks fed different sources of DDGS. Also, the bioavailable Lys determined by the regression was lower ($P < 0.05$) for source 7 than for source 2. However, differences were

Table 3. Standardized amino acid (AA) digestibility of 7 sources of distillers dried grains with solubles (DDGS)[1]

	DDGS source									
Item	1	2	3	4	5	6	7	Mean	SEM	*P*-value
Indispensable AA, %										
Arg	89.3	88.9	88.2	87.3	87.6	87.9	90.0	88.5	1.18	0.649
His	88.0	87.6	86.3	87.6	86.7	84.7	85.8	86.7	1.28	0.555
Ile	84.1	85.0	83.2	82.8	82.6	82.0	85.2	83.5	1.41	0.593
Leu	91.0[b]	92.6[b]	91.1[b]	91.8[b]	91.2[b]	91.0[b]	88.2[c]	91.0	0.79	0.024
Lys	70.4[a]	63.1[ab]	57.7[bc]	63.5[ab]	62.3[ab]	52.7[c]	59.8[bc]	61.4	3.16	0.021
Met	87.9	88.7	87.2	87.4	84.9	83.6	88.5	86.9	1.24	0.054
Phe	87.9	88.7	87.2	87.9	87.3	86.8	88.8	87.8	0.98	0.743
Thr	78.8	78.5	75.6	78.7	77.5	76.1	77.8	77.6	1.74	0.781
Val	84.2	85.9	83.6	83.4	82.5	82.4	84.4	83.8	1.50	0.691
Dispensable AA, %										
Ala	86.1	87.4	84.7	86.5	85.8	84.7	84.8	85.7	1.14	0.551
Asp	79.0	78.7	76.2	77.4	76.8	75.6	76.3	77.1	1.64	0.709
Cys	85.4	85.1	84.6	83.8	81.7	80.4	81.9	83.3	1.88	0.409
Glu	89.3[a]	89.9[a]	87.2[abc]	88.2[ab]	87.7[ab]	85.7[bc]	84.4[c]	87.5	1.01	0.008
Pro	89.8[b]	89.9[b]	87.7[b]	89.6[b]	88.7[b]	87.4[b]	−29.6[c]	72.0	5.54	<0.010
Ser	85.6	85.4	83.0	85.6	84.2	84.0	83.5	84.5	1.51	0.788
Tyr	87.8	90.1	88.6	89.2	88.3	89.3	89.1	88.9	1.07	0.801

[a–c]Digestibility values within a row with different superscript letters differ ($P < 0.05$).

[1]Each digestibility value is the mean of 5 observations.

Table 4. Relative bioavailability of Lys in 7 sources of distillers dried grains with solubles (DDGS) fed to chicks, as-fed basis[1]

Item	Chick total gain, g	G:F, g/kg	Relative bioavailable Lys,[2] %	Relative bioavailability of Lys,[3] %
Source of DDGS				
1	102.9	0.527[a]	0.546[ab]	58.1
2	107.6	0.522[a]	0.616[a]	79.0
3	99.6	0.494[b]	0.456[ab]	70.2
4	105.3	0.515[a]	0.573[ab]	68.2
5	106.1	0.519[a]	0.587[ab]	82.6
6	100.1	0.515[a]	0.481[ab]	65.0
7	96.8	0.515[a]	0.431[b]	59.9
Mean	102.6	0.5	0.527	69.0
SEM	3.3	0.01	0.056	7.72
P-value	0.238	0.010	0.050	0.264

[a,b]Values within a column with different superscript letters differ ($P < 0.05$).

[1]Values are means of 4 replicate pens of 5 chicks during a 9-d feeding period.

[2]Calculated from the best-fit regression equation using standard curve methodology: chick gain (g) = 77.29 + 118.88 (supplemental Lys intake, g); $r^2 = 0.96$.

[3]100 × (bioavailable Lys/total analyzed Lys). See Table 1 for total analyzed Lys in each DDGS source.

not observed in either total weight gain of chicks or Lys bioavailability among DDGS sources.

The mean concentrations of SDD Lys and bioavailable Lys were 0.47 and 0.53%, respectively (Table 5), but only DDGS sources 2 and 5 had a greater ($P <$ 0.05) concentration of bioavailable Lys than SDD Lys.

Hunterlab L, a, and b scores had mean values of 52.81, 12.48, and 39.51, respectively, for the 7 sources of DDGS (Table 6). The CV for the Hunterlab L score was 5.16% compared with 11.37% for Hunterlab a score and 7.46% for Hunterlab b score. The concentration of reactive Lys was correlated with the concentration of SDD Lys in DDGS ($r^2 = 0.84$, $P < 0.05$; Table 7). However, the concentration of SDD Lys was not correlated with the concentration of ADF and NDF. Likewise, there was no correlation between any of the color scores and SDD Lys. The concentration of reactive Lys was poorly correlated with the concentration of relative bioavailable Lys ($r^2 = 0.46$). No correlation was observed between the concentration of ADF and NDF

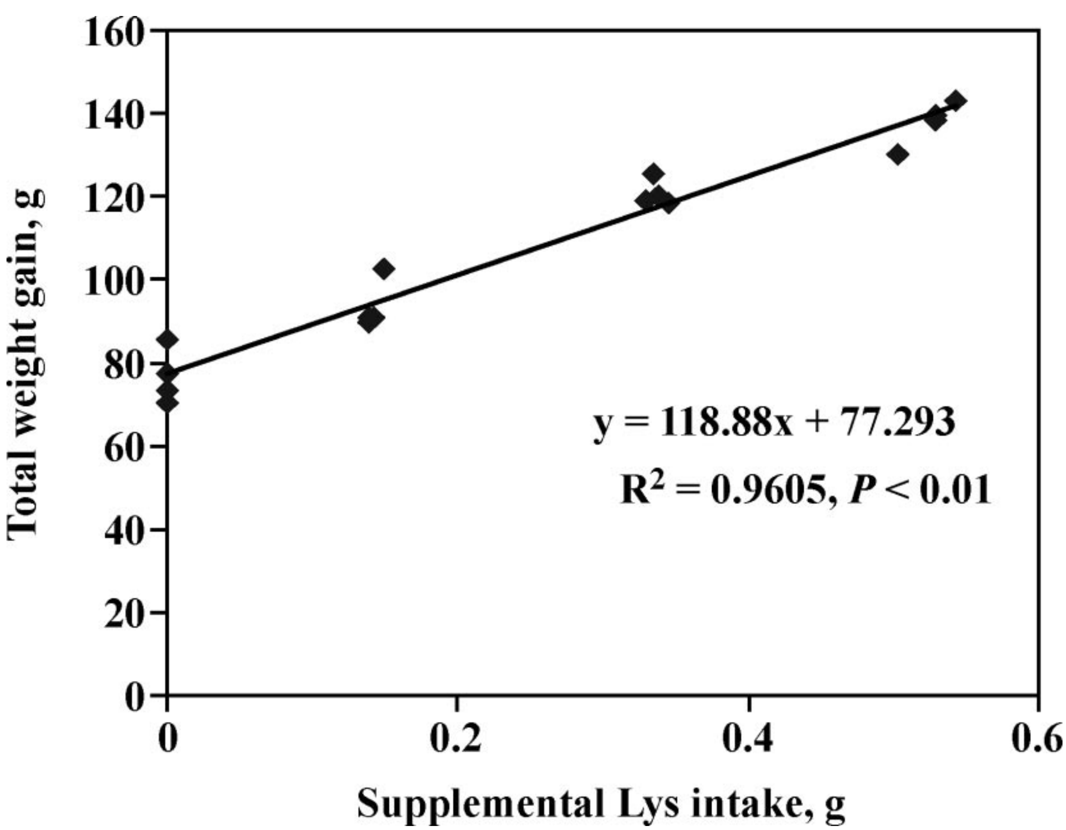

Figure 1. Total weight gain of chicks (y) in response to increasing intake of supplemental Lys (x).

Table 5. Comparison of rooster standardized digestible (SDD) Lys and relative chick bioavailable Lys in 7 sources of distillers dried grains with solubles (DDGS), as-fed basis

DDGS source	Concentration of SDD Lys,[1] %	Relative bioavailable Lys,[2] %	SED[3]	*P*-value
1	0.66	0.55	0.05	0.054
2	0.49	0.62	0.02	<0.010
3	0.38	0.46	0.06	0.220
4	0.53	0.57	0.03	0.286
5	0.44	0.59	0.06	0.038
6	0.39	0.48	0.08	0.303
7	0.43	0.43	0.07	0.990
Mean	0.47	0.53	—	—

[1]Measured by the cecectomized rooster assay. Calculated by multiplying the concentration of analyzed Lys (see Table 1) by SDD Lys (%; see Table 4) and divided by 100. Each value is the mean of 5 observations.

[2]Measured by the chick growth assay. Each value is the mean of 4 observations.

[3]Standard error of difference.

and the concentration of relative bioavailable Lys. The concentration of bioavailable Lys was highly correlated with Hunterlab L score ($r^2 = 0.90$, $P < 0.01$), but bioavailable Lys was poorly correlated with Hunterlab b score ($r^2 = 0.47$) and not correlated with Hunterlab a score.

DISCUSSION

The composition of DDGS used in this study is similar to previously published values (NRC, 1994; Spiehs et al., 2002; Stein et al., 2006). The range of SDD values of AA in the 7 sources of DDGS also concur with previously reported values (NRC, 1994; Batal and Dale, 2006; Fastinger et al., 2006).

The greater CV for the SDD of Lys compared with the SDD for other indispensable AA (9 vs. <3%; data not shown) is in agreement with previous observations showing that the variability in digestibility of Lys is greater than for most other AA in DDGS (Fiene et al., 2006; Parsons, 2006; Stein et al., 2006). This is likely because of the negative effect of heat processing on digestibility. Several steps in the starch extraction process used in the ethanol plant (jet cooking, liquefaction, saccharification) involve application of heat, and drying of wet distillers grains and condensed distillers solubles to produce DDGS also involves heat (Rausch and Belyea, 2006). Although drying of wet distillers grains and condensed solubles is the stage in which heat application is most aggressive, some Lys in the wet distillers grains and condensed solubles appears to have already been damaged by heat before drying (Pahm, 2008).

The mean bioavailability of Lys in DDGS obtained in this study (69%) is lower than the 80% value reported by Lumpkins and Batal (2005). However, the range in the concentration of relative bioavailable Lys (0.43 to 0.62%) is in agreement with previously reported values of 0.47 to 0.71% (Combs and Bossard 1969; Parsons et al., 1983). The lack of a difference between the concentration of SDD Lys and the concentration of bioavailable Lys suggests that when SDD of Lys is measured using the cecectomized rooster assay, the concentration of bioavailable Lys is not overestimated. This result is not in agreement with observations in pigs (Wiseman et al., 1991; van Barneveld et al., 1994) and rats (Craig and Broderick, 1981), where it was shown that heat application to feedstuffs lowered the efficiency of utilization of digested Lys. The fact that only 75% (0.58% reactive Lys vs. 0.77% analyzed Lys) of the Lys is reactive suggests that part of the Lys in DDGS is bound to reducing sugars. This agrees with previous results showing that approximately 24% of the Lys in DDGS is unreactive (Pahm, 2008). It appears that when fed to chicks, these unreactive Lys residues do not cause an overestimation of the concentration of digestible Lys in relation to the concentration of bioavailable Lys. The SDD procedure appears to correct for the reduction in the efficiency of digested Lys by taking into account the unavailable Lys.

The strong correlation between SDD Lys and reactive Lys is an indication that greater amounts of Lys are digested by roosters when the ε-NH_2 group of Lys is not bound to sugars. A similar relationship between the amount of reactive Lys in DDGS and ileal digestibility was obtained in pigs fed DDGS-containing diets (Pahm et al., 2006). Maillard products can reduce the digestibility of Lys by competing with absorption of

Table 6. Color scores of 7 sources of distillers dried grains with solubles (DDGS)[1]

DDGS source	Color score L	Color score a	Color score b
1	53.38	10.37	36.81
2	56.43	13.38	42.02
3	50.00	11.78	36.70
4	55.66	13.20	43.93
5	53.57	12.95	41.49
6	51.34	11.20	36.95
7	49.28	14.47	38.68
Mean	52.81	12.48	39.51
CV	5.16	11.37	7.46

[1]Measured by using the Hunterlab color scale; L, a, and b scores are measures of degree of lightness, redness, and yellowness, respectively. Each value is a mean of 10 measurements.

Table 7. Correlation of chemical composition and color score with concentration (% of sample) of standardized digestible (SDD) Lys and relative bioavailable Lys in 7 sources of distillers dried grains with solubles

	SDD Lys[1]		Relative bioavailable Lys[2]	
Predictor	r^2	*P*-value	r^2	*P*-value
Chemical composition				
Reactive Lys[3]	0.84	0.003	0.46	0.093
NDF	0.23	0.272	0.11	0.463
ADF	0.21	0.301	0.14	0.403
Color score[4]				
L	0.29	0.215	0.90	0.001
a	0.10	0.500	0.00	0.973
b	0.02	0.749	0.47	0.088

[1]Measured the using adult cecectomized roosters.

[2]Measured by the chick growth assay.

[3]Measured by the homoarginine procedure (Rutherfurd and Moughan, 1990). The concentration of reactive Lys in DDGS sources 1, 2, 3, 4, 5, 6, and 7 was 0.72, 0.59, 0.43, 0.69, 0.59, 0.51, and 0.50%, respectively (mean = 0.58%).

[4]Measured by the Hunterlab color scale; L, a, and b scores are measures of degree of lightness, redness, and yellowness, respectively.

Lys (Sherr et al., 1989) or inhibit the release of protein-bound Lys by inhibition of carboxypeptidases (Hansen and Millington, 1979).

The low correlation between the concentration of SDD Lys and Hunterlab L scores was due, in part, to DDGS from source 1 that had a dark color but a high concentration of SDD Lys. Excluding this sample, the r^2 between the concentration of SDD Lys and Hunterlab L score was 0.70 (data not shown). Heat processing (such as roasting of corn) is accompanied by brown discoloration (Costa et al., 1976), which may indicate nonenzymatic browning (Moran and Summers, 1968). A moderate correlation (r^2 = 0.52) of SDD Lys and Minolta L* scores in DDGS has also been observed (Fastinger et al., 2006), whereas a high correlation (r = 0.87) between the concentration of SDD Lys and Minolta L* score was reported by Batal and Dale (2006). This suggests that the relationship between color score and SDD Lys in DDGS can vary and may be influenced by the type of colorimeter and the procedure used to measure color (Pedersen et al., 2005).

The strong correlation between Hunterlab L score and the concentration of bioavailable Lys indicates that darker colored DDGS may have undergone considerable binding of Lys with reducing sugars, which initiated a browning reaction. Results of this experiment agree with previous data (Cromwell et al., 1993) showing a positive relationship between Hunterlab L and a (but not b) scores and chick performance (weight gain, gain:feed). A similar relationship between color and performance has been reported in diets containing heated soybean meal fed to broilers (McNaughton et al., 1981).

The guanidination procedure (Mauron and Bujard, 1964) can be used to measure reactive Lys, and it appears that values obtained with this procedure correlate well with Lys utilization in vivo (e.g., Hurrell and Carpenter, 1974; Nair et al., 1978). However, the relatively low correlation between reactive Lys and the concentration of bioavailable Lys obtained in this study may be due to poor absorption of some of the reactive Lys in DDGS because some Lys may be trapped in indigestible peptides (Desrosiers et al., 1989; Moughan et al., 1996).

In conclusion, the concentration of SDD Lys in DDGS fed to poultry does not appear to overestimate the concentration of bioavailable Lys. The concentration of reactive Lys and Hunterlab L values are alternative parameters to evaluate DDGS quality in addition to AA digestibility and the chick growth assay.

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