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# Concentrations of analyzed or reactive lysine, but not crude protein, may predict the concentration of digestible lysine in distillers dried grains with solubles fed to pigs<sup>1</sup>

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**ABSTRACT:** The objectives of this experiment were to evaluate procedures that may be used to predict the concentration of standardized ileal digestible (SID) Lys in distillers dried grains with solubles (DDGS) fed to pigs and to evaluate the accuracy of a published equation to predict SID Lys in DDGS. Twenty-one sources of DDGS were analyzed (as-fed basis) for CP (23.8% to 33.6%; CV = 8.3%), Lys (0.69% to 1.17%; CV = 12.4%), and furosine (0.02% to 0.22%; CV = 91.4%). The concentration of reactive Lys (%; as-fed basis) was calculated as analyzed Lys (%) – furosine (%) ÷ 0.32 × 0.40 and ranged from 0.47% to 1.15% (CV = 20.7%) in the 21 sources of DDGS. Twenty-one diets that each contained 60.0% of 1 source of DDGS as the sole source of CP and AA were formulated. An N-free diet was also formulated and was used to determine basal endogenous losses of CP and AA. Twenty-two barrows with an initial BW of 45.2 kg (SD = 3.1 kg) were fitted with a T-cannula in the distal

ileum and allotted to a 22 × 10 Youden square design with the 22 diets and 10 periods. The SID of CP and AA were calculated for each source of DDGS. The SID of CP ranged from 69.8% to 79.6%, and the SID of Lys ranged from 45.3% to 74.1%. The concentration of SID Lys in the 21 samples of DDGS was highly related to the concentration of analyzed Lys ( $P < 0.001$ ;  $r^2 = 0.849$ ) and with the concentration of reactive Lys in the samples ( $P < 0.001$ ;  $r^2 = 0.898$ ). In contrast, the concentration of SID Lys in the 21 sources of DDGS was not related to the concentration of CP in the samples ( $P = 0.558$ ;  $r^2 = 0.021$ ). However, values for SID Lys were in good agreement with values predicted using a published prediction equation. In conclusion, analyzed Lys in DDGS, but not CP, may be used to predict the concentration of SID Lys in DDGS fed to pigs. However, analysis of furosine in addition to Lys and subsequent calculation of reactive Lys improve the prediction accuracy of digestible Lys concentration in DDGS.

**Key words:** amino acids, digestible lysine, distillers dried grains with solubles, furosine, lysine, reactive lysine

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## INTRODUCTION

The concentration and digestibility of Lys in distillers dried grains with solubles (DDGS) are more variable than that of most other AA (Stein et al., 2006; Pahl et al., 2008a). If the concentration of digestible

Lys in DDGS can be predicted, mixed diets that contain DDGS can be more accurately formulated. Procedures to predict the concentration of digestible Lys in DDGS are therefore needed.

Some of the variability in the digestibility of Lys in DDGS is likely caused by the Maillard reaction, which may occur during production and drying of DDGS (Cromwell et al., 1993; Stein et al., 2006; Pahl et al., 2008a). As a result of the Maillard reaction, Lys is bound to reducing sugars, and the bound Lys is called “unreactive” Lys, which is biologically unavailable to animals because it cannot be used for protein synthesis (Finot and Magnenat, 1981). In contrast, the Lys that is not bound to a reducing sugar is called “reactive” Lys, which may be absorbed from the intestinal tract and used for protein synthesis. Reactive Lys can be determined

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using the furosine procedure (Pahm et al., 2008b). Furosine is an AA produced during acid hydrolysis of unreactive Lys, also called Amadori products. Of the Lys that is changed to Amadori products, 32% is typically converted to furosine, 28% to pyridosine and 40% to regenerated Lys after undergoing acid hydrolysis, and the regenerated Lys is included in the analyzed Lys concentration (Bujard and Finot, 1978; Pahm et al., 2008b; Figure 1). Thus, the concentration of regenerated Lys can be calculated from the concentration of furosine. By subtracting the regenerated Lys from the analyzed Lys, the concentration of reactive Lys is calculated (Pahm et al., 2008b).

The objective of this experiment was to test the hypothesis that the concentration of digestible Lys and other AA in DDGS can be estimated from the concentration of CP or the concentration of Lys or other AA. A second objective was to validate a previously published equation to predict the concentration of digestible Lys in DDGS.

## MATERIALS AND METHODS

The protocol for the experiment was reviewed and approved by the Institutional Animal Care and Use Committee at the University of Illinois. All pigs used in this experiment were the offspring of Landrace boars that were mated to Duroc × Large White females (Pig Improvement Company, Hendersonville, TN).

### Animals and Diets

Twenty-one sources of corn DDGS were procured, with 19 sources originating from dry grind ethanol plants in the Midwest and 2 sources originating from ethanol plants in the European Union (Table 1). The ileal digestibility of CP and AA in each source of DDGS was determined using 22 barrows (initial and final BW,  $45.2 \pm 3.1$  and  $68.2 \pm 7.3$  kg, respectively). Pigs were surgically fitted with a T-cannula in the distal ileum using procedures adapted from Stein et al. (1998). After the surgery, pigs were housed individually in pens ( $1.2 \times 1.5$  m) that were equipped with a feeder and a nipple drinker and had fully slatted stainless steel T-bar floors.

Twenty-one diets were formulated by mixing 60% DDGS with cornstarch, sucrose, vitamins, and minerals (Tables 2 and 3). The 21 sources of DDGS were used in 1 diet each, and DDGS was the sole source of AA in the diets. An N-free diet was also prepared to determine basal endogenous losses of CP and AA. All diets contained 0.5% chromic oxide as an indigestible marker. Vitamins and minerals were included in all diets to meet or exceed requirement estimates for growing pigs (NRC, 1998). Pigs were systematically allotted to a  $22 \times 10$  incomplete Latin square design with the 22 diets and 10 periods comprising the rows and the columns in the square, respectively. A

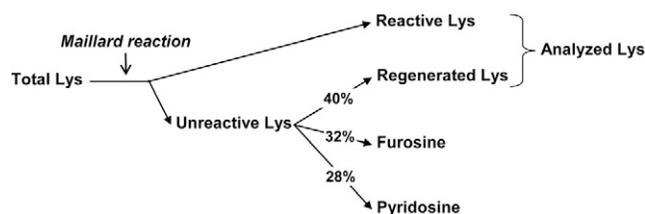
spreadsheet program was used during the allotment to balance potential residual effects (Kim and Stein, 2009).

### Feeding and Sample Collection

Feed was provided at daily levels of 3 times the estimated maintenance requirement for energy (i.e., 106 kcal ME/kg BW<sup>0.75</sup>; NRC, 1998), and equal meals were provided at 800 and 1700 h. The feed allowance for each pig was adjusted at the beginning of each period when the BW of the pigs was recorded. Animals had free access to water from a nipple drinker throughout the experiment. Each period lasted 7 d. The initial 5 d were an adaptation period to the diet, and ileal digesta samples were collected for 8 h on d 6 and 7. A plastic bag was attached to the cannula barrel using a cable tie, and digesta flowing into the bag were collected. Bags were removed whenever they were filled with digesta or at least once every 30 min. Collected samples were stored at  $-20^{\circ}\text{C}$  to prevent bacterial degradation of AA in the digesta.

### Chemical Analysis

At the conclusion of the experiment, frozen ileal samples were allowed to thaw at room temperature, mixed within animal and diet, and a subsample was collected for chemical analysis. Ileal digesta samples were lyophilized and finely ground before chemical analysis. All samples of ingredients, diets, and ileal digesta were analyzed for DM (method 930.15; AOAC International, 2007) and for CP by combustion (method 999.03; AOAC International, 2007) using a rapid N cube (Elementar Americas Inc., Mt. Laurel, NJ) with Asp as the internal standard. Amino acids were analyzed by an AA analyzer (Hitachi Amino Acid Analyzer Model L8800; Hitachi High Technologies America, Inc., Pleasanton, CA) using ninhydrin for postcolumn derivatization and norleucine as the internal standard [method 982.30 E (a, b, c); AOAC International, 2007]. Samples were hydrolyzed with 6N HCl for 24 h at  $110^{\circ}\text{C}$  before being analyzed. Methionine and Cys were analyzed as Met sulfone and cysteic acid after cold performic acid oxidation overnight before hydrolysis. Tryptophan was determined after NaOH hydrolysis for 22 h at  $110^{\circ}\text{C}$ . The 21 sources of DDGS were also analyzed for ash (method 975.03; AOAC International, 2007), ADF



**Figure 1.** Products of the Maillard reaction from Lys. During acid hydrolysis, 40% of the unreactive Lys is changed to regenerated Lys, 32% is changed to furosine, and 28% is changed to pyridosine.

**Table 1.** Analyzed nutrient composition (%) of 21 sources of distillers dried grains with solubles, as-fed basis

Item	Source of distillers dried grains with solubles																					Mean	SD
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21		
DM	89.5	89.3	89.3	89.1	87.6	90.7	89.3	88.2	87.0	88.6	86.2	89.7	89.6	88.0	86.9	88.2	88.5	88.7	88.7	85.3	88.0	88.4	1.3
GE, cal/g	4,797	5,010	4,814	4,717	4,912	4,907	4,765	4,793	4,753	4,865	4,756	4,751	4,955	4,876	4,849	4,946	4,768	4,861	4,716	4,665	4,704	4,821	95
CP	26.0	26.1	27.0	27.4	25.3	25.0	33.6	26.5	25.7	27.2	25.2	31.2	27.2	23.9	23.8	26.5	26.3	25.7	25.6	25.7	27.2	26.6	2.2
Ash	5.27	5.86	5.11	5.45	4.50	5.39	2.38	4.99	4.83	4.20	4.21	3.34	4.94	4.16	3.88	3.96	3.82	3.56	3.96	3.86	4.06	4.37	0.83
NDF	34.2	32.3	36.5	33.9	33.7	32.7	44.3	41.1	35.1	37.4	33.3	40.6	36.0	37.0	33.5	36.1	37.5	33.9	38.7	33.1	35.9	36.0	3.1
ADF	12.0	12.9	9.9	13.3	10.2	10.4	13.2	11.9	9.5	12.1	11.5	11.5	10.8	9.9	10.4	11.2	10.7	10.0	13.0	9.3	11.2	11.2	1.2
Furosine	0.030	0.199	0.030	0.039	0.067	0.037	0.071	0.059	0.039	0.055	0.037	0.018	0.017	0.038	0.073	0.037	0.050	0.198	0.027	0.220	0.050	0.066	0.060
Indispensable AA																							
Arg	1.12	1.11	1.31	1.24	1.12	1.16	1.34	1.18	1.18	1.24	1.14	1.38	1.32	1.12	1.10	1.23	1.23	1.13	1.22	1.05	1.27	1.20	0.09
His	0.74	0.74	0.82	0.78	0.73	0.74	1.00	0.80	0.78	0.84	0.77	0.93	0.85	0.74	0.73	0.81	0.75	0.75	0.73	0.72	0.80	0.79	0.07
Ile	0.97	0.96	1.03	0.96	0.89	0.90	1.35	0.99	0.98	1.05	0.94	1.23	1.02	0.91	0.87	0.98	1.00	0.91	0.97	0.87	0.96	0.99	0.11
Leu	3.17	3.30	3.18	3.01	2.79	2.74	4.59	3.09	2.94	3.25	2.85	3.82	3.23	2.74	2.63	3.01	3.02	2.89	2.90	2.89	2.99	3.10	0.43
Lys	0.69	0.74	1.01	0.90	0.84	0.93	0.96	0.96	0.99	1.00	0.98	1.17	1.05	0.97	0.91	0.98	0.96	0.78	0.99	0.74	0.98	0.93	0.11
Met	0.48	0.47	0.52	0.50	0.48	0.47	0.71	0.51	0.46	0.53	0.47	0.62	0.51	0.44	0.46	0.51	0.50	0.48	0.52	0.48	0.52	0.51	0.06
Phe	1.26	1.29	1.31	1.30	1.36	1.06	1.78	1.30	1.16	1.35	1.21	1.57	1.30	1.07	1.03	1.25	1.28	1.25	1.24	1.21	1.27	1.28	0.16
Thr	0.87	0.89	0.96	0.97	0.89	0.90	1.23	0.96	0.92	1.01	0.93	1.11	0.98	0.89	0.88	0.92	0.94	0.90	0.94	0.91	0.97	0.95	0.08
Trp	0.17	0.16	0.21	0.20	0.18	0.18	0.20	0.19	0.21	0.20	0.17	0.23	0.21	0.19	0.19	0.20	0.20	0.20	0.20	0.17	0.19	0.19	0.02
Val	1.38	1.38	1.47	1.40	1.26	1.32	1.82	1.43	1.41	1.51	1.36	1.70	1.49	1.32	1.28	1.43	1.43	1.31	1.39	1.26	1.39	1.42	0.14
Total	10.9	11.0	11.8	11.3	10.5	10.4	15.0	11.4	11.0	12.0	10.8	13.8	12.0	10.4	10.1	11.3	11.3	10.6	11.1	10.3	11.3	11.4	1.2
Dispensable AA																							
Ala	1.87	1.92	1.94	1.88	1.70	1.70	2.62	1.90	1.82	1.96	1.75	2.26	1.98	1.71	1.64	1.84	1.83	1.78	1.80	1.81	1.86	1.88	0.21
Asp	1.53	1.51	1.66	1.59	1.52	1.51	2.13	1.67	1.62	1.74	1.56	2.00	1.72	1.52	1.48	1.61	1.62	1.52	1.63	1.50	1.64	1.63	0.16
Cys	0.43	0.43	0.46	0.47	0.46	0.44	0.63	0.48	0.43	0.51	0.44	0.56	0.50	0.43	0.42	0.49	0.47	0.46	0.48	0.42	0.49	0.47	0.05
Glu	3.51	3.58	3.44	3.25	3.30	3.02	5.36	3.56	3.28	3.62	3.12	4.51	3.77	3.05	2.92	3.37	3.40	3.34	3.38	3.28	3.54	3.50	0.54
Gly	0.99	0.94	1.11	1.09	0.99	1.01	1.21	1.07	1.03	1.10	1.02	1.19	1.12	0.99	0.97	1.07	1.08	1.02	1.08	0.97	1.10	1.05	0.07
Pro	2.08	2.08	2.08	2.02	1.88	1.81	2.94	2.03	1.90	2.11	1.87	2.46	2.18	1.77	1.75	1.98	1.97	1.87	1.95	1.92	2.01	2.03	0.26
Ser	0.96	0.99	1.04	1.10	1.04	0.97	1.37	1.03	0.95	1.08	1.02	1.18	1.05	0.96	0.96	0.99	1.02	1.03	1.02	1.05	1.08	1.04	0.09
Tyr	0.89	0.89	0.93	0.89	0.90	0.78	1.38	0.96	0.84	0.98	0.87	1.15	0.88	0.71	0.70	0.79	0.86	0.80	0.82	0.85	0.92	0.89	0.15
Total	12.3	12.3	12.7	12.3	11.8	11.2	17.6	12.7	11.9	13.1	11.7	15.3	13.2	11.1	10.8	12.1	12.3	11.8	12.2	11.8	12.6	12.5	1.5

(method 973.18; AOAC International, 2007), and NDF (Holst, 1973), and GE was determined by bomb calorimetry (Model 6300, Parr Instruments, Moline, IL), with benzoic acid being the internal standard.

For furosine analysis, the 21 samples of DDGS were hydrolyzed with 6*N* HCl, and the hydrolysates were run on reversed phase HPLC with gradient mobile phases (0.1% trifluoroacetic acid in deionized water for mobile phase A and 0.1% trifluoroacetic acid in methanol for mobile phase B). The concentration of furosine in the sample was detected by tandem mass spectrometry in the multiple reaction monitoring operation mode. The quantification was performed on the basis of an external calibration with 5 standards made of furosine dihydrochloride (NeoMPS, Neosystems Laboratory, Strasbourg, France).

The concentration of Cr in diets and ileal digesta samples was determined using an inductive coupled plasma atomic emission spectrometric method (method 990.08; AOAC International, 2007) after nitric acid–perchloric acid wet ash sample preparation (method 968.088D; AOAC International, 2007).

### Calculation and Statistical Analysis

Values for apparent ileal digestibility (AID), basal endogenous ileal losses, and standardized ileal digestibility (SID) of CP and AA in each source of DDGS were calculated (Stein et al., 2007). Unreactive Lys in each source of DDGS was calculated by dividing the concentration of furosine in the sample by 0.32 (Pahm et al., 2008b). The quantity of regenerated Lys in each sample was calculated by multiplying unreactive Lys by 0.40, and the concentration of reactive Lys was then calculated by subtracting regenerated Lys from analyzed Lys (Pahm et al., 2008b).

Data were analyzed using the MIXED procedure (SAS Inst. Inc., Cary, NC). The model included diet as the fixed effect and period and animal as random effects. Means were calculated using the LSMeans statement. The animal was considered the experimental unit for calculations, and an alpha level of 0.05 was used to assess differences among means.

Regression analyses were conducted to estimate the concentration of SID Lys (%) in each source of DDGS from the concentration (%) of CP, analyzed Lys, analyzed Lys as percentage of CP, furosine, furosine as percentage of analyzed Lys, or reactive Lys. On the basis of the SID of CP and AA and analyzed CP and AA, the concentrations (%) of SID CP and SID of all AA were calculated for each source of DDGS. The REG procedure was used to establish regression equations with the concentration of SID Lys as the dependent variable and CP, analyzed Lys as percent of CP, analyzed Lys, and reactive Lys as independent variables. Regression equations were also developed to predict the concentration of the digestible

quantity of each AA from the concentration of CP in each source of DDGS or from the concentration of each AA in DDGS. The Cook's distance for the 21 sources of DDGS was calculated using the REG procedure in SAS to identify outliers (Neter et al., 1990). Two sources of DDGS were detected as outliers (>0.2 Cook's distance) in the regression to predict the SID of Lys and the concentration of SID Lys and thus were excluded from the final analysis. The accuracy of a previously published equation (Pahm et al., 2008b) to predict the concentration of digestible Lys in DDGS was assessed by regressing the determined minus the predicted values for SID of Lys on the predicted values centered to the mean (Seo et al., 2006).

## RESULTS AND DISCUSSION

### Composition of DDGS Samples

The concentration (as-fed basis) of GE in the 21 sources of DDGS (Table 1) ranged from 4,665 to 5,010 kcal/kg (mean = 4,821 kcal/kg; SD = 94.6 kcal/kg), and CP ranged from 23.8% to 33.6% (mean = 26.6%; SD = 2.2%). The average GE in DDGS, as well as the SD, observed in this experiment was very close to the average values previously reported (Stein and Shurson, 2009). The average CP obtained in this experiment was slightly less than the average of previous reports, but the average NDF (mean = 36.0%; SD = 3.1%) was much greater than the average value (25.3%) reported by Stein and Shurson (2009). Analyzed Lys ranged from 0.69% to 1.17% (as-fed

**Table 2.** Ingredient composition of experimental diets, as-fed basis

Ingredient, %	DDGS diets <sup>1</sup>	N-free diet
DDGS <sup>1</sup>	60.00	—
Cornstarch	17.40	68.20
Sucrose	20.00	20.00
Soybean oil	—	4.00
Solka floc <sup>2</sup>	—	4.00
Ground limestone	1.40	0.80
Dicalcium phosphate	—	1.30
Potassium carbonate	—	0.40
Magnesium oxide	—	0.10
Salt	0.40	0.40
Vitamin-mineral premix <sup>3</sup>	0.30	0.30
Chromic oxide	0.50	0.50

<sup>1</sup>DDGS = distillers dried grains with solubles. Twenty-one diets were formulated with a specific source of DDGS being the sole source of AA in each diet.

<sup>2</sup>Fiber Sales and Development Corp., Urbana, OH.

<sup>3</sup>Supplied per kilogram of complete diet: vitamin A as retinyl acetate, 11,128 IU; vitamin D<sub>3</sub> as cholecalciferol, 2,204 IU; vitamin E as *dl*- $\alpha$ -tocopheryl acetate, 66 IU; vitamin K as menadione nicotinamide bisulfite, 1.42 mg; thiamin as thiamine mononitrate, 0.24 mg; riboflavin, 6.58 mg; pyridoxine as pyridoxine hydrochloride, 0.24 mg; vitamin B<sub>12</sub>, 0.03 mg; D-pantothenic acid as D-calcium pantothenate, 23.5 mg; niacin as nicotinamide, 1.0 mg, and nicotinic acid, 43.0 mg; folic acid, 1.58 mg; biotin, 0.44 mg; Cu, 10 mg as copper sulfate; Fe, 125 mg as iron sulfate; I, 1.26 mg as potassium iodate; Mn, 60 mg as manganese sulfate; Se, 0.3 mg as sodium selenite; and Zn, 100 mg as zinc oxide.

**Table 3.** Analyzed nutrient composition (%) of experimental diets, as-fed basis

Item	Source of distillers dried grains with solubles																					N-free diet	
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21		
DM	91.8	90.6	91.9	89.5	90.7	91.7	92.2	89.9	91.5	90.5	91.0	89.6	90.4	91.6	90.9	90.0	90.7	90.5	92.9	92.0	91.0	91.9	
CP	16.3	16.4	16.5	17.2	16.2	15.4	21.1	16.5	15.5	17.4	16.1	19.4	15.6	15.4	14.8	16.7	15.9	17.0	17.6	16.4	17.4	0.8	
Indispensable AA																							
Arg	0.68	0.65	0.77	0.80	0.72	0.67	0.77	0.71	0.66	0.72	0.70	0.85	0.69	0.65	0.63	0.75	0.72	0.71	0.80	0.65	0.78	0.01	
His	0.46	0.46	0.46	0.48	0.45	0.41	0.56	0.49	0.46	0.49	0.48	0.58	0.45	0.43	0.42	0.50	0.47	0.46	0.52	0.45	0.51	0.00	
Ile	0.57	0.57	0.61	0.64	0.58	0.54	0.81	0.61	0.58	0.62	0.60	0.78	0.56	0.53	0.52	0.59	0.57	0.55	0.64	0.56	0.61	0.01	
Leu	1.96	2.03	1.92	2.00	1.82	1.64	2.73	1.91	1.76	1.94	1.82	2.43	1.78	1.60	1.57	1.87	1.79	1.81	1.95	1.81	1.91	0.03	
Lys	0.43	0.47	0.61	0.58	0.53	0.55	0.57	0.58	0.57	0.60	0.59	0.73	0.55	0.56	0.53	0.60	0.58	0.48	0.67	0.47	0.61	0.02	
Met	0.29	0.28	0.33	0.33	0.29	0.28	0.42	0.32	0.27	0.32	0.29	0.37	0.27	0.26	0.26	0.31	0.29	0.30	0.33	0.29	0.33	0.00	
Phe	0.77	0.79	0.80	0.84	0.76	0.68	1.06	0.77	0.72	0.80	0.75	1.00	0.73	0.66	0.65	0.76	0.73	0.76	0.80	0.74	0.78	0.01	
Thr	0.56	0.55	0.60	0.62	0.55	0.52	0.73	0.58	0.54	0.61	0.56	0.69	0.51	0.51	0.49	0.57	0.56	0.57	0.62	0.54	0.59	0.01	
Trp	0.13	0.14	0.15	0.14	0.15	0.14	0.16	0.15	0.15	0.15	0.15	0.16	0.14	0.14	0.14	0.15	0.15	0.15	0.15	0.14	0.14	<0.04	
Val	0.80	0.80	0.85	0.90	0.82	0.77	1.06	0.86	0.80	0.87	0.84	1.05	0.78	0.75	0.73	0.84	0.80	0.78	0.89	0.79	0.86	0.01	
Total	6.65	6.74	7.10	7.33	6.67	6.20	8.87	6.98	6.51	7.12	6.78	8.64	6.46	6.09	5.94	6.94	6.66	6.57	7.37	6.44	7.12	0.10	
Dispensable AA																							
Ala	1.16	1.20	1.19	1.27	1.13	1.05	1.58	1.20	1.10	1.19	1.14	1.45	1.08	1.03	1.00	1.16	1.11	1.13	1.22	1.16	1.21	0.02	
Asp	0.97	0.95	1.05	1.09	0.99	0.94	1.29	1.05	0.98	1.06	1.01	1.27	0.95	0.92	0.89	1.01	0.98	1.00	1.09	0.97	1.06	0.02	
Cys	0.31	0.30	0.33	0.35	0.31	0.26	0.38	0.30	0.25	0.32	0.27	0.34	0.27	0.25	0.24	0.29	0.29	0.29	0.31	0.28	0.32	0.01	
Glu	2.38	2.41	2.31	2.43	2.19	2.03	3.40	2.38	2.17	2.37	2.23	3.00	2.25	2.03	1.97	2.27	2.19	2.20	2.37	2.26	2.35	0.06	
Gly	0.60	0.58	0.68	0.73	0.66	0.63	0.73	0.67	0.61	0.68	0.66	0.76	0.59	0.60	0.58	0.67	0.65	0.64	0.72	0.62	0.71	0.01	
Pro	1.28	1.30	1.27	1.37	1.24	1.11	1.80	1.31	1.20	1.31	1.24	1.48	1.14	1.12	1.11	1.27	1.24	1.21	1.34	1.23	1.30	0.02	
Ser	0.65	0.63	0.67	0.67	0.60	0.54	0.81	0.61	0.58	0.67	0.58	0.73	0.55	0.55	0.52	0.63	0.63	0.65	0.67	0.61	0.63	0.01	
Tyr	0.51	0.51	0.55	0.56	0.51	0.45	0.74	0.51	0.46	0.54	0.49	0.67	0.47	0.43	0.43	0.50	0.51	0.51	0.53	0.48	0.54	0.00	
Total	7.86	7.88	8.05	8.47	7.63	7.01	10.73	8.03	7.35	8.14	7.62	9.70	7.30	6.93	6.74	7.80	7.60	7.63	8.25	7.61	8.12	0.15	

basis), and the average concentration of Lys (0.93%; SD = 0.11%) in the samples used in this experiment was greater than the values reported in many previous experiments (Fastinger and Mahan, 2006; Stein et al., 2006, 2009; Pahl et al., 2008a; Urriola et al., 2009). However, the concentration of other indispensable AA was in good agreement with previous publications. The furosine concentration (as-fed basis) in the 21 samples of DDGS ranged from 0.02% to 0.22% (mean = 0.07%; SD = 0.06%). The fact that furosine was analyzed in all the DDGS samples indicates that they had undergone Maillard reactions to varying degrees, which has also been observed in previous experiments (Cromwell et al., 1993; Pahl et al., 2008b; Cozannet et al., 2010).

### *Digestibility of AA in DDGS Samples*

The AID of CP in the 21 DDGS samples ranged from 57.3% to 68.3%, but the AID of Lys varied from 35.8% to 66.9% (Table 4). The SID of CP also varied less than that of Lys (69.8% to 79.6% vs. 45.3% to 74.1%; Table 5). However, the average SID for Lys obtained in this experiment (63.3%) was slightly greater than the average value (60.6%) obtained for corn DDGS in previous experiments (Stein and Shurson, 2009). The variation from the least to the greatest value for the SID of Lys observed in this experiment was, however, in good agreement with data from previous experiments (Stein, 2007). Likewise, for most indispensable AA, the average values for SID obtained in this experiment were in very good agreement with the average values reported in previous experiments (Stein and Shurson, 2009), with the exception that the SID of Trp obtained in this experiment is approximately 10 percentage units greater than the average from previous experiments, but it is not clear why the SID of Trp

**Table 4.** Apparent ileal digestibility (%) of CP and AA in diets containing 21 sources of distillers dried grains with solubles fed to pigs<sup>1</sup>

Item	Source of distillers dried grains with solubles																					SEM	Mean	CV, <sup>2</sup> %
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21			
CP	60.0	65.9	66.7	61.1	63.6	57.3	68.6	60.8	63.3	63.2	63.4	64.4	65.1	66.1	63.5	68.3	63.1	63.2	64.2	58.8	62.0	2.0	63.5	4.5
Indispensable AA																								
Arg	74.2	74.1	80.0	77.0	75.0	70.8	73.0	73.2	75.3	72.4	75.3	74.4	76.1	77.1	75.2	80.2	78.3	73.5	76.3	70.1	75.2	1.8	75.1	3.5
His	73.1	79.5	78.5	73.2	76.6	70.1	77.9	75.2	77.8	75.3	78.6	79.3	80.2	79.8	77.7	82.1	77.6	77.7	78.9	77.1	75.5	1.4	77.2	3.6
Ile	69.4	76.8	75.4	71.0	73.4	66.4	77.6	70.6	72.7	71.1	72.1	74.5	74.6	73.8	71.7	76.5	71.1	71.5	73.3	72.2	69.4	1.6	72.6	3.7
Leu	80.9	84.2	85.2	78.8	83.8	77.4	87.4	81.6	83.7	81.3	82.5	83.5	85.3	83.8	82.5	86.0	82.4	84.0	83.0	85.0	80.3	1.2	83.0	2.9
Lys	39.3	60.4	63.8	54.7	53.6	52.5	47.7	52.6	60.9	55.5	61.4	62.2	60.7	63.5	57.5	66.9	60.7	37.4	57.5	35.8	58.4	3.3	55.4	15.8
Met	79.5	84.8	81.4	81.4	80.8	73.2	86.2	78.0	77.0	76.7	78.3	81.3	80.8	80.5	78.3	82.2	74.9	82.3	80.1	82.7	78.6	1.3	80.0	3.8
Phe	76.7	82.8	80.8	77.9	79.5	72.1	83.3	76.0	77.1	77.3	76.9	79.5	79.9	78.7	77.4	81.2	76.3	79.3	78.8	78.3	75.4	1.4	78.3	3.3
Thr	60.2	66.1	67.5	60.0	63.1	54.6	68.8	59.3	61.1	62.2	61.2	65.7	63.0	64.0	61.2	68.0	62.9	64.8	64.2	61.1	59.3	2.0	62.8	5.4
Trp	71.5	78.1	75.8	68.7	75.2	69.5	75.5	71.5	74.3	70.0	74.3	72.8	75.1	75.0	73.8	78.9	75.1	76.8	73.8	73.9	68.9	1.9	73.7	3.9
Val	69.1	74.9	74.9	69.8	73.0	66.3	76.3	69.8	72.6	71.0	72.7	74.1	74.1	74.1	72.1	76.7	71.4	71.9	72.7	72.7	69.4	1.5	72.4	3.5
Total	72.1	77.7	77.9	72.8	75.2	68.8	78.5	72.6	75.0	73.1	74.8	76.4	76.8	76.4	74.3	79.2	74.6	74.1	75.2	73.6	72.6	1.4	74.8	3.3
Dispensable AA																								
Ala	71.9	77.4	78.0	74.7	75.6	69.6	79.4	73.7	75.9	74.0	76.2	76.3	75.5	76.6	75.3	78.7	74.5	74.5	75.2	73.7	73.8	1.5	75.3	3.0
Asp	59.1	65.1	67.2	57.8	63.6	55.2	69.1	60.5	65.0	62.0	63.0	69.1	66.0	65.5	63.8	68.5	61.6	63.5	62.9	63.0	60.1	2.0	63.4	5.7
Cys	66.4	72.5	75.1	66.3	71.6	62.7	71.9	65.6	66.5	68.9	69.5	72.2	72.4	71.4	68.6	73.4	69.1	69.7	70.8	67.4	68.1	1.9	69.5	4.4
Glu	75.4	80.5	81.1	74.2	79.0	72.7	83.5	77.2	79.7	76.6	79.3	80.8	81.6	80.5	79.0	82.0	78.3	78.6	78.8	79.9	76.0	1.4	78.8	3.4
Gly	31.6	34.0	45.6	45.1	39.4	32.4	38.7	35.6	34.4	40.8	43.6	38.6	34.3	43.2	39.1	47.1	40.0	35.2	41.4	33.7	42.5	4.3	38.9	11.9
Pro	21.8	10.9	32.4	40.7	21.3	11.3	27.1	23.1	25.3	21.5	27.7	5.6	12.7	25.4	23.1	34.3	27.3	9.2	23.1	12.7	35.1	9.6	22.5	41.5
Ser	71.8	74.0	75.7	68.3	71.3	62.2	76.1	68.0	70.5	70.7	68.1	72.3	70.4	71.6	69.1	75.8	71.8	73.1	72.5	72.0	68.0	1.7	71.1	4.5
Tyr	76.1	81.5	81.7	77.8	79.3	70.9	83.6	75.8	76.8	77.7	76.5	79.4	78.3	77.5	76.5	81.1	78.0	80.2	77.3	78.9	75.1	1.5	78.1	3.5
Total	60.1	62.5	67.5	63.6	62.8	55.3	67.7	61.1	63.2	61.7	63.9	62.9	62.8	64.6	62.7	68.2	63.2	60.6	62.9	61.0	63.1	2.5	62.9	4.4

<sup>1</sup>Each least squares mean represents 10 observations.<sup>2</sup>The CV is based on the mean values from each of the 21 sources of distillers dried grains with solubles.

was so much greater in the DDGS used in the present experiment.

The CV for the SID of Lys was much greater than the CV for the SID of CP or other indispensable AA, which indicates that there is more variability in Lys digestibility than the digestibility of other AA. A similar observation has been reported from other experiments with corn DDGS (Stein, 2007) and also for wheat DDGS (Cozannet et al., 2010). This is most likely a result of heat damage to some sources of DDGS because heat damage results in reduced SID of Lys (Pahm et al., 2008b). This observation, therefore, supports the hypothesis that some of the DDGS samples may have been heat damaged, as also indicated by the concentration of furosine in the samples. The need for identifying procedures that can predict the concentration of digestible Lys in a specific source of DDGS is emphasized by the variability in Lys digestibility observed.

### *Reactive and Unreactive Lys in DDGS Samples*

The concentration of Lys as a percentage of CP ranged from 2.66 to 4.05 with an average of 3.51 (Table 6). These values are considerably greater than values observed in previous experiments (Stein, 2007), and they are a consequence of the slightly reduced CP concentrations and the increased Lys concentrations observed in the DDGS samples used in this experiment compared with the samples used in previous experiments. The samples used in this experiment were produced in the spring of 2009, whereas the samples used in the summary by Stein (2007) were produced in 2004 and 2005. The fact that Lys as a percentage of CP is greater in the samples used in this experiment indicates that ethanol plants may have improved their production procedures and are destroying less

**Table 5.** Standardized ileal digestibility (SID; %) of CP and AA in 21 sources of distillers dried grains with solubles fed to pigs<sup>1,2</sup>

Item	Source of distillers dried grains with solubles																					SEM	Mean	CV, <sup>3</sup> %	
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21				
Indispensable AA																									
CP	71.8	77.5	78.3	71.9	75.3	69.8	77.7	72.1	75.7	74.1	75.2	74.0	77.2	78.5	76.4	79.6	75.0	74.3	75.3	70.5	73.0	2.0	74.9	3.6	
Arg	85.1	85.3	89.6	86.0	85.1	81.8	82.6	83.4	86.5	82.5	85.7	82.8	86.7	88.4	86.8	89.9	88.4	83.8	85.7	81.5	84.6	1.8	85.4	2.9	
His	77.3	83.6	82.7	77.1	80.8	74.8	81.3	79.1	82.0	79.2	82.6	82.5	84.4	84.3	82.2	85.9	81.7	81.9	82.6	81.5	79.3	1.4	81.3	3.3	
Ile	74.9	82.2	80.5	75.8	78.8	72.2	81.5	75.6	78.1	76.1	77.2	78.4	80.2	79.7	77.6	81.7	76.6	77.2	78.3	77.8	74.5	1.6	77.8	3.3	
Leu	83.7	86.9	88.0	81.5	86.8	80.8	89.5	84.4	86.8	84.1	85.5	85.7	88.3	87.2	86.0	88.9	85.4	87.0	85.9	88.0	83.2	1.2	85.9	2.7	
Lys	49.6	69.7	71.1	62.1	61.8	60.5	55.5	60.0	68.6	62.7	68.8	68.1	68.6	71.4	65.8	74.1	68.3	46.5	64.2	45.3	65.6	3.3	63.3	12.8	
Met	82.3	87.8	83.9	83.9	83.6	76.2	88.2	80.6	80.1	79.3	81.2	83.5	83.9	83.7	81.4	84.8	77.8	85.0	82.6	85.6	81.1	1.3	82.7	3.6	
Phe	80.9	86.8	84.8	81.6	83.7	76.8	86.3	80.1	81.5	81.2	81.1	82.6	84.2	83.5	82.2	85.3	80.6	83.4	82.8	82.6	79.4	1.4	82.5	2.9	
Thr	68.9	74.8	75.6	67.6	71.9	64.0	75.5	67.5	70.0	70.0	69.8	72.5	72.4	73.5	71.0	76.4	71.4	73.2	72.1	70.1	67.4	2.0	71.2	4.4	
Trp	79.7	85.5	82.8	76.0	82.1	77.0	82.1	78.3	81.3	76.9	81.3	79.2	82.5	82.5	81.2	85.8	82.0	83.7	80.9	81.5	76.4	1.9	80.9	3.4	
Val	74.7	80.3	80.1	74.6	78.3	72.0	80.5	74.8	78.1	76.0	78.0	78.2	79.7	80.0	78.1	81.9	76.8	77.5	77.7	78.3	74.5	1.5	77.6	3.2	
Total	77.6	83.1	83.1	77.7	80.7	74.8	82.6	77.8	80.6	78.2	80.1	80.5	82.4	82.4	80.4	84.4	80.1	79.6	80.3	79.3	77.7	1.4	80.2	2.9	
Dispensable AA																									
Ala	78.2	83.4	84.2	80.3	82.0	76.6	84.1	79.7	82.5	80.1	82.5	81.2	82.2	83.7	82.5	84.9	81.1	80.9	81.3	80.0	79.8	1.5	81.5	2.5	
Asp	66.8	72.9	74.4	64.6	71.1	63.3	75.0	67.6	72.6	69.0	70.4	74.9	73.8	73.7	72.2	75.8	69.2	70.9	70.0	70.9	67.1	2.0	70.8	4.9	
Cys	71.7	77.8	80.0	70.9	76.9	69.0	76.2	71.0	73.0	74.0	75.5	76.9	78.4	78.0	75.3	78.9	74.6	75.2	76.1	73.3	73.2	2.0	75.0	3.9	
Glu	79.5	84.5	85.4	78.1	83.5	77.6	86.4	81.2	84.2	80.7	83.7	84.0	85.9	85.3	83.9	86.2	82.7	83.0	83.0	84.3	80.1	1.4	83.0	3.1	
Gly	61.3	64.2	71.8	68.9	66.0	60.6	63.2	61.6	63.5	66.6	70.3	61.5	64.0	72.8	69.5	73.2	67.1	62.6	66.4	62.5	67.4	4.3	66.0	6.0	
Pro	76.8	64.3	87.9	90.8	77.4	74.6	66.4	75.8	83.8	74.5	83.9	52.0	73.5	88.1	85.9	88.6	83.4	66.6	70.1	88.8	9.6	77.6	12.9		
Ser	78.7	81.0	82.4	74.8	78.6	70.5	81.7	75.2	78.2	77.3	75.7	78.3	78.5	79.7	77.6	82.8	78.9	79.9	79.2	79.4	75.0	1.7	78.3	3.7	
Tyr	80.8	86.2	86.1	82.0	84.0	76.2	86.9	80.5	82.1	82.1	81.4	82.9	83.3	83.1	82.1	85.8	82.7	84.9	81.9	84.0	79.5	1.5	82.8	3.0	
Total	75.6	77.7	82.6	77.5	78.5	72.6	79.0	75.9	79.7	76.4	79.7	75.1	79.1	82.1	80.6	83.5	79.0	76.3	77.8	77.0	77.9	2.5	78.3	3.3	

<sup>1</sup>Each least squares mean represents 10 observations.<sup>2</sup>Values for SID were calculated by correcting apparent ileal digestibility values for basal endogenous losses. Basal endogenous losses were determined using pigs fed the N-free diet (g/g DMI): CP, 20.9; Arg, 0.80; His, 0.21; Ile, 0.34; Leu, 0.60; Lys, 0.48; Met, 0.09; Phe, 0.35; Thr, 0.53; Trp, 0.11; Val, 0.48; Ala, 0.80; Asp, 0.82; Cys, 0.18; Glu, 1.07; Gly, 1.94; Pro, 7.67; Ser, 0.49; and Tyr, 0.26.<sup>3</sup>The CV is based on the mean values from each of the 21 sources of distillers dried grains with solubles.

Lys via heat damage than they did previously. Nevertheless, the presence of furosine, and therefore also unreactive Lys, in the samples indicates that some of the samples used in this experiment were heat damaged. The concentration of unreactive Lys ranged from 0.05% to 0.69%, with a CV of 91.4%. This wide variation further indicates that some of the sources of DDGS used in this experiment were very little heat damaged, whereas other samples were severely heat damaged. This observation supports the hypothesis that the variability in the SID of Lys observed among samples was likely caused by heat damage to some of the samples. The conclusion, therefore, is that although the ethanol industry seems to have improved production processes in recent years, there are still some plants using production processes that result in heat damage to DDGS.

The concentration of regenerated Lys was calculated as 40% of the unreactive Lys (Finot and Magneat, 1981) and ranged from 0.02% to 0.28%. The regenerated Lys is the Lys that is analyzed as Lys but is biologically unavailable to the pig, and it is therefore necessary to quantify the regenerated Lys to estimate the quantity of Lys that can be utilized by the pig.

By subtracting the concentration of regenerated Lys from the concentration of analyzed Lys, the concentration of reactive Lys in the samples was calculated. These values ranged from 0.47% to 1.15% (mean = 0.85%; CV = 20.7%) and are greater than the values reported by Pahm et al. (2008b), which further

indicates that there is less heat damage in the samples used in this experiment compared with samples used in previous experiments.

The assumption that, as DDGS is acid hydrolyzed, 40% and 32% of the total Amadori compounds are regenerated as Lys and converted to furosine, respectively, is based on values that have been estimated in heat-damaged milk products (Bujard and Finot, 1978; Finot et al., 1981). These percentages have previously been used to estimate regenerated Lys and furosine in DDGS (Pahm et al., 2008b; Cozannet et al., 2010), but research to confirm that 40% of Amadori compounds in DDGS are regenerated as Lys and that 32% are converted to furosine when heat-damaged DDGS is acid hydrolyzed has not been conducted. Estimates for regenerated Lys in heat-damaged DDGS are, therefore, based on the untested assumption that acid hydrolysis of DDGS results in conversion of Amadori compounds in a way that is similar to acid hydrolysis of heat-damaged milk products. This is a weakness of the procedures used to calculate reactive and unreactive Lys in DDGS, and research to generate knowledge about the conversion of Amadori compounds after acid hydrolysis of heat damaged DDGS is needed.

**Prediction of Digestible Lys and AA in DDGS**

Regression analyses revealed that the analyzed Lys, analyzed Lys as percentage of CP, and reactive Lys were positively related to the SID of Lys ( $P < 0.001$ ;  $r^2 > 0.50$ ; Table 7), indicating that the SID of Lys increases if analyzed Lys, analyzed Lys as percentage of CP, or reactive Lys increases. In contrast, furosine and furosine as percentage of analyzed Lys were negatively related to the SID of Lys ( $P < 0.001$ ;  $r^2 > 0.70$ ), indicating that as the furosine concentration in DDGS increases, the SID of Lys is reduced. This observation supports the notion that heat damage of DDGS results in reduced digestibility of Lys and increased generation of furosine. However, the concentration of CP

**Table 6.** Concentration (%) of CP, analyzed Lys, Lys as percentage of CP, furosine, unreactive Lys, regenerated Lys, and reactive Lys in 21 samples of distillers dried grains with solubles, as-fed basis

Item	Mean	Minimum	Maximum	SD	CV, %
CP	26.6	23.8	33.6	2.2	8.3
Analyzed Lys	0.93	0.69	1.17	0.12	12.4
Lys as percentage of CP	3.51	2.66	4.05	0.42	11.9
Furosine	0.07	0.02	0.22	0.06	91.4
Unreactive Lys <sup>1</sup>	0.21	0.05	0.69	0.19	91.4
Regenerated Lys <sup>2</sup>	0.08	0.02	0.28	0.08	91.4
Reactive Lys <sup>3</sup>	0.85	0.47	1.15	0.18	20.7

<sup>1</sup>Unreactive Lys = furosine ÷ 0.32.

<sup>2</sup>Regenerated Lys = unreactive Lys × 0.40.

<sup>3</sup>Reactive Lys = analyzed Lys – regenerated Lys.

was not related to the SID of Lys ( $P = 0.685$ ;  $r^2 = 0.01$ ), indicating that the SID of Lys in DDGS cannot be predicted from the concentration of CP in the sample.

Further regression analyses indicated that the analyzed Lys and reactive Lys, but not CP, were highly related to the concentration of SID Lys in DDGS (Table 8). The  $r^2$  for analyzed Lys and reactive Lys and the concentration of SID Lys were 0.849 and 0.898, respectively ( $P < 0.001$ ). By using the analyzed Lys as percentage of CP as an additional independent variable in these regressions, the  $r^2$  improved to 0.921 and 0.930, respectively. However, CP concentrations in DDGS were not related to the concentration of SID Lys in DDGS ( $P = 0.558$ ;  $r^2 = 0.021$ ). These data indicate that the concentration of SID Lys in DDGS may be predicted from the concentration of analyzed Lys in the samples, but a slightly better prediction is observed if Lys as a percentage of CP is used as the second independent variable in the regression equation. The accuracy of the prediction equations is further improved if the concentration of reactive Lys is used rather than the concentration of analyzed Lys.

The present results are in agreement with the data reported by Pahm et al. (2008b), who observed

**Table 7.** Linear regression analysis of standardized ileal digestibility (SID) of Lys (%) using concentrations (%) of CP, analyzed Lys, analyzed Lys as percentage of CP, furosine, furosine as percentage of analyzed Lys, and reactive Lys as independent variables in distillers dried grains with solubles fed to pigs<sup>1,2</sup>

Intercept			Variable	Model statistics						
Estimate	SE	P-value		Estimate	SE	P-value	P-value	RMSE <sup>3</sup>	r <sup>2</sup>	
72.5	21.6	0.004	CP	-0.3	0.8	0.685	0.685	7.922	0.010	
5.9	12.8	0.651	Lys <sup>4</sup>	60.6	13.4	<0.001	<0.001	5.356	0.548	
-3.2	10.7	0.770	Lys as percentage of CP <sup>4</sup>	18.6	3.0	<0.001	<0.001	4.360	0.700	
71.2	1.4	<0.001	furosine	-123.0	17.0	<0.001	<0.001	3.946	0.754	
69.9	1.3	<0.001	furosine as percentage of Lys <sup>4</sup>	-0.9	0.1	<0.001	<0.001	4.030	0.744	
27.5	6.0	<0.001	reactive Lys <sup>5</sup>	41.2	6.7	<0.001	<0.001	4.444	0.688	

<sup>1</sup>n = 19.

<sup>2</sup>The dependent variable is SID of Lys (%).

<sup>3</sup> RMSE = root-mean-square error.

<sup>4</sup>Analyzed Lys (%).

<sup>5</sup>Reactive Lys (%) = analyzed Lys (%) – regenerated Lys (%) = analyzed Lys (%) – furosine (%) ÷ 0.32 × 0.40.

**Table 8.** Linear regression to predict the concentration (%) of standardized ileal digestible (SID) Lys from the concentrations (%) of CP, analyzed Lys as percentage of CP, analyzed Lys, and reactive Lys as independent variables in distillers dried grains with solubles fed to pigs<sup>1,2</sup>

Intercept			Independent variable									Model statistics	
Estimate	SE	P-value	Variable 1			Variable 2			P-value	RMSE <sup>3</sup>	r <sup>2</sup>		
			Variable	Estimate	SE	P-value	Variable	Estimate				SE	P-value
0.418	0.326	0.217	CP	0.007	0.012	0.558	—	—	—	—	0.558	0.120	0.021
-0.368	0.175	0.050	Lys as percentage of CP <sup>4</sup>	0.273	0.048	<0.001	—	—	—	—	<0.001	0.071	0.651
-0.482	0.112	<0.001	Lys <sup>4</sup>	1.148	0.117	<0.001	—	—	—	—	<0.001	0.047	0.849
-0.636	0.093	<0.001	Lys <sup>4</sup>	0.858	0.116	<0.001	Lys as percentage of CP <sup>4</sup>	0.120	0.032	0.002	<0.001	0.035	0.921
-0.016	0.052	0.762	reactive Lys <sup>5</sup>	0.716	0.058	<0.001	—	—	—	—	<0.001	0.039	0.898
-0.206	0.083	0.025	reactive Lys <sup>5</sup>	0.576	0.072	<0.001	Lys as percentage of CP <sup>4</sup>	0.087	0.032	0.016	<0.001	0.033	0.930

<sup>1</sup>n = 19.<sup>2</sup>The dependent variable is Lys concentrations based on SID.<sup>3</sup>RMSE = root-mean-square error.<sup>4</sup>Analyzed Lys concentration.<sup>5</sup>Reactive Lys (%) = analyzed Lys (%) - regenerated Lys (%) = analyzed Lys (%) - furosine (%) ÷ 0.32 × 0.40.

the greatest r<sup>2</sup> (0.66) between reactive Lys and the concentration of SID Lys in DDGS and the least r<sup>2</sup> (0.22) between CP and the concentration of SID Lys. Pahlm et al. (2008b) also reported that the relationship between the analyzed Lys and the concentration of SID Lys was much greater than that between CP and SID Lys, and the present results are in agreement with that observation.

Further regression analyses were conducted to predict the concentration of ileal digestible CP and AA from the concentration of CP in the samples of DDGS (Table 9). The concentration of the digestible quantities of most indispensable AA was predicted relatively well from the concentration of CP in DDGS (r<sup>2</sup> > 0.72; P < 0.001), but the concentration of digestible Arg, Lys,

**Table 9.** Linear regression analysis to predict the concentration (%) of standardized ileal digestible CP or AA from the concentration (%) of CP in distillers dried grains with solubles fed to pigs<sup>1</sup>

Dependent variable <sup>2</sup>	Intercept			Independent variable <sup>3</sup>				Model statistics		
	Estimate	SE	P-value	Variable	Estimate	SE	P-value	P-value	RMSE <sup>4</sup>	r <sup>2</sup>
CP	-1.187	1.982	0.557	CP	0.794	0.074	<0.001	<0.001	0.729	0.871
Indispensable AA										
Arg	0.461	0.190	0.027	CP	0.021	0.007	0.008	0.008	0.070	0.349
His	0.008	0.095	0.934	CP	0.024	0.004	<0.001	<0.001	0.035	0.729
Ile	-0.357	0.117	0.007	CP	0.042	0.004	<0.001	<0.001	0.043	0.846
Leu	-1.922	0.405	<0.001	CP	0.172	0.015	<0.001	<0.001	0.149	0.883
Lys	0.418	0.326	0.217	CP	0.007	0.012	0.558	0.558	0.120	0.021
Met	-0.242	0.055	<0.001	CP	0.025	0.002	<0.001	<0.001	0.020	0.897
Phe	-0.633	0.173	0.002	CP	0.063	0.006	<0.001	<0.001	0.064	0.850
Thr	-0.120	0.086	0.180	CP	0.030	0.003	<0.001	<0.001	0.032	0.838
Trp	0.093	0.034	0.013	CP	0.002	0.001	0.071	0.071	0.012	0.179
Val	-0.183	0.159	0.267	CP	0.048	0.006	<0.001	<0.001	0.059	0.794
Total	-2.445	1.139	0.047	CP	0.435	0.043	<0.001	<0.001	0.419	0.860
Dispensable AA										
Ala	-0.629	0.177	0.002	CP	0.081	0.007	<0.001	<0.001	0.065	0.899
Asp	-0.387	0.209	0.082	CP	0.058	0.008	<0.001	<0.001	0.077	0.766
Cys	-0.084	0.058	0.167	CP	0.017	0.002	<0.001	<0.001	0.021	0.775
Glu	-2.626	0.523	<0.001	CP	0.208	0.020	<0.001	<0.001	0.192	0.869
Gly	0.410	0.137	0.008	CP	0.011	0.005	0.045	0.045	0.050	0.215
Pro	0.739	0.506	0.163	CP	0.031	0.019	0.116	0.116	0.186	0.139
Ser	-0.091	0.108	0.411	CP	0.034	0.004	<0.001	<0.001	0.040	0.809
Tyr	-0.791	0.122	<0.001	CP	0.058	0.005	<0.001	<0.001	0.045	0.903
Total	-3.430	1.208	0.011	CP	0.498	0.045	<0.001	<0.001	0.444	0.877

<sup>1</sup>n = 19.<sup>2</sup>The dependent variables are concentrations (%) of standardized ileal digestible CP and AA.<sup>3</sup>The independent variable is the analyzed concentration of CP (%).<sup>4</sup>RMSE = root-mean-square error.

and Trp could not be predicted from the concentration of CP in the samples ( $r^2 < 0.35$ ). Given the importance of Lys and Trp in diet formulation, it is apparent that the nutritional value of DDGS cannot be adequately predicted from the concentration of CP in DDGS.

However, the concentration of each indispensable AA in DDGS was highly related to the concentration of digestible quantities of each AA ( $r^2 > 0.84$ ;  $P < 0.001$ ; Table 10). This observation indicates that by analyzing DDGS for total AA, the concentration of the digestible AA can be calculated. The fact that the  $r^2$  for Lys and Trp was greater than 0.84 indicates that acceptable predictions were also obtained for these 2 AA. It can thus be concluded that it is necessary to analyze samples of DDGS for the concentration of AA to obtain an acceptable prediction of the quantities of digestible AA.

### Accuracy of Published Prediction Equation

It has been suggested that the concentration of SID Lys in DDGS can be estimated using the following equation (Pahm et al., 2008b):

$$\text{SID Lys (\%)} = 0.023 + 0.637 \times \text{reactive Lys (\%)}. [1]$$

The concentrations of SID Lys that were obtained in this experiment were, therefore, plotted against the concentrations calculated from Eq. 1 (Figure 2). Results of this comparison indicate that the prediction equation by Pahm et al. (2008b) estimates the concentration of SID Lys in most of the sources of DDGS that were used in this experiment relatively well. The average determined concentration of SID Lys in the sources of DDGS used in this experiment was 0.611%, whereas the predicted concentration was 0.581%. The reliability of Eq. 1 was further tested by the regression analysis. The slope representing a linear bias was not different from 0, whereas the intercept representing a mean bias was greater than 0. These observations indicate that Eq. 1 slightly underestimates SID Lys in DDGS. However, given the variability in analyzing both chromium and AA in diet and digesta samples, the difference between observed and predicted values indicates that the accuracy of the prediction equation is acceptable. It appears therefore that the equation presented by Pahm et al. (2008b) may be used to predict the concentration of SID Lys in DDGS.

**Table 10.** Linear regression analysis to predict the concentration (%) of standardized ileal digestible CP or AA from the concentration (%) of CP or the corresponding AA in distillers dried grains with solubles fed to pigs<sup>1</sup>

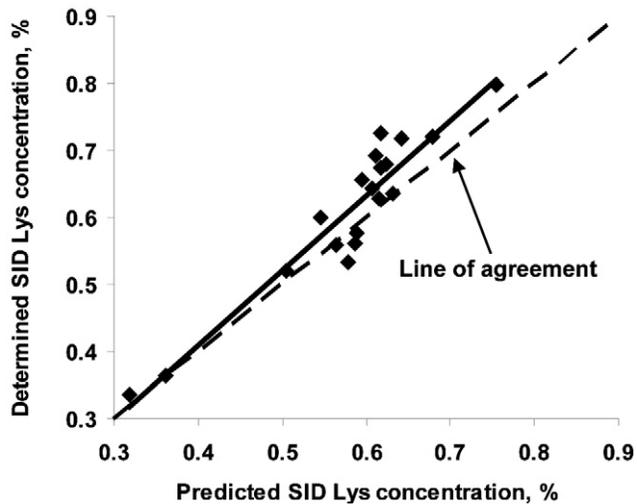
Dependent variable <sup>2</sup>	Intercept			Variable	Independent variable <sup>3</sup>			Model statistics		
	Estimate	SE	P-value		Estimate	SE	P-value	P-value	RMSE <sup>4</sup>	r <sup>2</sup>
CP	-1.187	1.982	0.557	CP	0.794	0.074	<0.001	<0.001	0.729	0.871
Indispensable AA										
Arg	-0.022	0.103	0.830	Arg	0.872	0.085	<0.001	<0.001	0.032	0.860
His	-0.026	0.053	0.626	His	0.847	0.067	<0.001	<0.001	0.021	0.904
Ile	-0.093	0.043	0.047	Ile	0.873	0.044	<0.001	<0.001	0.022	0.959
Leu	-0.225	0.114	0.064	Leu	0.934	0.037	<0.001	<0.001	0.070	0.975
Lys	-0.482	0.112	<0.001	Lys	1.148	0.117	<0.001	<0.001	0.047	0.849
Met	-0.070	0.027	0.018	Met	0.962	0.052	<0.001	<0.001	0.014	0.953
Phe	-0.117	0.045	0.019	Phe	0.916	0.035	<0.001	<0.001	0.026	0.976
Thr	-0.117	0.079	0.155	Thr	0.835	0.082	<0.001	<0.001	0.029	0.860
Trp	-0.005	0.017	0.785	Trp	0.832	0.087	<0.001	<0.001	0.005	0.843
Val	-0.108	0.080	0.195	Val	0.853	0.056	<0.001	<0.001	0.034	0.932
Total	-0.862	0.563	0.144	Total	0.878	0.049	<0.001	<0.001	0.251	0.949
Dispensable AA										
Ala	-0.101	0.072	0.180	Ala	0.870	0.038	<0.001	<0.001	0.036	0.969
Asp	-0.264	0.124	0.048	Asp	0.870	0.075	<0.001	<0.001	0.053	0.888
Cys	-0.030	0.030	0.335	Cys	0.814	0.063	<0.001	<0.001	0.014	0.908
Glu	-0.245	0.123	0.061	Glu	0.903	0.035	<0.001	<0.001	0.083	0.976
Gly	0.127	0.157	0.431	Gly	0.543	0.147	0.002	0.002	0.042	0.444
Pro	0.994	0.322	0.007	Pro	0.286	0.158	0.087	0.087	0.184	0.162
Ser	-0.094	0.079	0.251	Ser	0.871	0.075	<0.001	<0.001	0.030	0.888
Tyr	-0.060	0.029	0.057	Tyr	0.895	0.032	<0.001	<0.001	0.021	0.978
Total	0.343	0.633	0.595	Total	0.757	0.050	<0.001	<0.001	0.334	0.931

<sup>1</sup>n = 19.

<sup>2</sup>The dependent variables are concentrations (%) of standardized ileal digestible CP and AA.

<sup>3</sup>The independent variable is analyzed concentration (%) of CP or AA.

<sup>4</sup>RMSE = root-mean-square error.



**Figure 2.** Comparison of determined standardized ileal digestible (SID) Lys concentration (%) and predicted SID Lys concentration (%). The following equation (Pahm et al., 2008b) was used to generate values for predicted SID Lys:  $\text{SID Lys (\%)} = 0.023 + 0.637 \times \text{reactive Lys (\%)}$ . On the basis of a regression analysis of determined minus predicted SID Lys concentration (%) on the predicted SID Lys concentration (%) adjusted to the mean as 0, the slope (0.124; SE = 0.092;  $P = 0.194$ ) was not different from 0, whereas the intercept (0.030; SE = 0.009;  $P = 0.003$ ) was greater than 0.

### Conclusions

Results of this experiment confirmed that the digestibility of Lys in corn DDGS is more variable than the digestibility of all other indispensable AA and that the analyzed concentration of furosine in the sources of DDGS that were used confirmed that one of the reasons for this variability is that some of the samples likely had been heat damaged. It is, however, important to note that the concentration of Lys in the samples used in this experiment, on average, was considerably greater than what has been reported from previous experiments, which indicates that many ethanol plants now use production procedures that allow them to reduce heat damage in DDGS.

On the basis of the results from this experiment, it is concluded that the analyzed Lys, but not CP, may be used to predict the concentration of digestible Lys in DDGS when fed to pigs. The reason for that is most likely that the concentration of CP in DDGS is unaffected by heat damage, whereas the concentration of Lys is reduced if DDGS is heat damaged. Analysis of furosine in addition to Lys and subsequent calculation of the concentration of reactive Lys improve the accuracy of the prediction, but the relatively modest improvement in the prediction accuracy may not always be of practical importance.

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