Determination and prediction of standardized ileal amino acid digestibility of wheat in broilers

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ABSTRACT This experiment evaluated the standard ileal digestibility (**SID**) of amino acids (**AA**) in 8 different sources of wheat fed to broilers and established prediction equations based on the chemical properties of wheat. A total of five hundred forty 1-day-old broilers were tested in 2 stages (from d 10 to 13 and from d 25 to 28). On d 13, 324 broilers were randomly assigned to 9 diets (6 replicate cages, 6 broilers per cage); on d 28, 216 broilers were randomly assigned to 9 diets (6 replicate cages, 4 broilers per cage). The 9 experimental diets included 8 test diets and 1 nitrogen-free diet. Titanium dioxide was added as an exogenous indicator at 0.5% of the diet. In 8 wheat samples, the mean values of total amino acids (**TAA**), dispensable amino acids (**DAA**), and indispensable amino acids (**IAA**) were 12.16% (CV 13.70%), 7.97% (CV 15.49%), and 4.20% (CV 11.47%). On d 13, the lowest SID of AA was Lys (86.71%), and the highest was Pro (97.98%). On d 28, the lowest SID of AA was His (81.31%), and the highest was Pro (96.83%). There was an effect of wheat source on the SID of AA except for Trp ($P < 0.05$); the broiler age had an effect on the SID of AA except for Tyr ($P < 0.05$); the SID of most AA were higher at d 13 compared to d 28. At d 13, the SID of AA was correlated with CP, NDF, and ST ($P < 0.05$). At d 28, the SID of AA was correlated with EE, Ash, ADF, and NDF ($P < 0.05$). The R^2 value of stepwise regression equations to predict the SID of AA at d 13 was highest for Leu ($R^2 = 0.972$), lowest for Asp ($R^2 = 0.785$); at d 28 was highest for Gly ($R^2 = 0.995$), lowest for His ($R^2 = 0.678$). In conclusion, this experiment showed that the chemical properties of wheat can be used to establish accurate equations for predicting the SID of AA. This made it more efficient to obtain the SID of AA for wheat.

Key words: broiler, standardized ileal amino acid digestibility, wheat, prediction equation

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INTRODUCTION

Corn and soybean meal are commonly used in the preparation of feed, but their prices have risen in the past two years and are unstable (An et al., 2020). As a result, other suitable raw materials should be used in place of corn, soybean meal, and other raw materials in the feed formula to reduce costs and improve economic benefits.

Wheat is increasingly being used in livestock production. However, different breeds and growing environments may cause differences in their chemical properties (Azhar et al., 2019; Olukosi and Bedford, 2019; Lasek et al., 2020), which affect the intake, digestion, and utilization of nutrients by poultry. To use wheat for precise diet configurations, it is vital to accurately determine the amino acid digestibility of wheat in poultry.

There are two commonly used methods for assessing the digestion and utilization of amino acids (**AA**) in poultry: apparent ileal digestible of amino acids (**AIDAA**) and standardized ileal digestible of amino acids (**SIDAA**). SIDAA accounts for the ileal endogenous amino acid (**IEAA**) flow (Ravindran, 2021), which is more appropriate for evaluating the utilization of AA in poultry (Sadighi et al., 2020). However, determining the SIDAA of the raw material is a very complicated procedure.

Many scholars have established equations that predict amino acid digestibility based on the chemical properties of raw materials (Ebadi et al., 2011; Wang et al., 2020), and the establishment of similar equations for wheat will make its use in poultry more efficient. Therefore, this study aims to determine the standardized ileal digestibility (**SID**) of AA of wheat in broilers at d 13 and 28, and to establish regression prediction equations based on chemical properties.

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Table 1. Analyzed chemical properties of wheat samples (%, DM).

	Wheat[1]											
Item	W-1	W-2	W-3	W-4	W-5	W-6	W-7	W-8	Mean	Min	Max	CV%
DM	87.81	86.20	87.69	87.37	86.30	87.74	87.29	87.54	87.24	86.20	87.81	0.73
CP	14.30	14.28	14.53	14.59	10.57	14.29	16.92	9.81	13.66	9.81	16.92	17.01
EE	2.02	2.43	1.38	1.52	1.35	1.34	2.40	2.10	1.82	1.34	2.43	25.97
Ash	1.54	2.18	1.68	2.03	1.76	2.26	1.86	1.82	1.89	1.54	2.26	13.07
CF	2.71	3.33	3.19	2.64	2.88	1.83	2.15	3.12	2.73	1.83	3.33	19.06
NDF	10.99	10.82	13.13	11.53	14.34	11.93	13.61	11.20	12.19	10.82	14.34	10.86
ADF	3.57	3.37	3.50	3.11	4.70	3.15	3.51	3.63	3.57	3.11	4.70	13.92
ST	67.00	74.80	67.96	66.75	76.50	68.84	65.70	74.85	70.30	65.70	76.50	6.17
Indispensable amino acids												
Arg	0.64	0.66	0.65	0.66	0.48	0.73	0.91	0.54	0.66	0.48	0.91	19.43
His	0.33	0.30	0.32	0.35	0.24	0.36	0.37	0.24	0.31	0.24	0.37	16.15
Ile	0.46	0.41	0.40	0.42	0.90	0.49	0.45	0.30	0.48	0.30	0.90	37.46
Leu	0.95	0.89	0.88	0.98	1.02	1.03	1.02	0.69	0.93	0.69	1.03	12.21
Lys	0.39	0.37	0.39	0.41	0.32	0.43	0.41	0.33	0.38	0.32	0.43	10.25
Met	0.31	0.37	0.38	0.41	0.23	0.40	0.36	0.25	0.34	0.23	0.41	20.11
Phe	0.66	0.59	0.62	0.53	0.58	0.66	0.55	0.34	0.57	0.34	0.66	18.15
Thr	0.39	0.37	0.39	0.38	0.31	0.43	0.36	0.27	0.36	0.27	0.43	13.89
Trp	0.18	0.22	0.15	0.19	0.19	0.22	0.22	0.21	0.20	0.15	0.22	12.62
Val	0.61	0.58	0.62	0.64	0.48	0.66	0.64	0.43	0.58	0.43	0.66	14.30
IAA	4.91	4.77	4.78	4.99	4.75	5.41	5.27	3.60	4.81	3.60	5.41	11.34
Dispensable amino acids												
Ala	0.51	0.51	0.58	0.60	0.42	0.59	0.60	0.47	0.54	0.42	0.60	12.68
Asp	0.73	0.68	0.73	0.80	0.59	0.81	0.78	0.61	0.72	0.59	0.81	11.65
Cys	0.38	0.37	0.40	0.41	0.29	0.44	0.39	0.29	0.37	0.29	0.44	14.64
Glu	4.66	4.58	4.56	4.91	3.45	5.11	5.09	3.16	4.44	3.16	5.11	16.58
Gly	0.63	0.63	0.64	0.64	0.50	0.70	0.68	0.43	0.61	0.43	0.70	15.27
Pro	1.41	1.64	1.25	1.61	0.89	1.70	1.79	1.01	1.41	0.89	1.79	23.59
Ser	0.68	0.68	0.66	0.72	0.53	0.73	0.73	0.49	0.65	0.49	0.73	14.13
Tyr	0.38	0.39	0.38	0.39	0.58	0.32	0.44	0.31	0.40	0.31	0.58	21.08
DAA	9.37	9.49	9.20	10.08	7.25	10.39	10.48	6.76	9.13	6.76	10.48	15.30
TAA	14.28	14.26	13.98	15.07	12.00	15.81	15.75	10.36	13.94	10.36	15.81	13.52

Abbreviations: ADF, Acid Detergent Fiber; Ala, Alanine; Arg, Arginine; Ash, coarse ash; Asp, Aspartic acid; CF, coarse fiber; CP, crude protein; CV, coefficient of variation; Cys, Cystine; DAA, dispensable amino acid; DM, dry matter; EE, ether extract; GE, gross energy; Glu, Glutamic acid; Gly, Glycine; His, Histidine; IAA, Dispensable amino acid; Ile, Isoleucine; Leu, Leucine; Lys, Lysine; Met, Methionine; Min, minimum; Max, maximum; NDF, neutral detergent fiber; Phe, Phenylalanine; Pro, Proline; Ser, Serine; ST, starch; Tyr, Tyrosine; TAA, Total amino acid; Thr, Threonine; Trp, Tryptophan; Val, Valine.

MATERIALS AND METHODS

The study was approved by the Institutional Animal Care and Use Committee of the China Agricultural University (permit number: AW11112202-1-2).

Wheat Samples

W-1 to W-8 were wheat samples from eight provinces in China (Heilongjiang, Jilin, Hebei, Henan, Liaoning, Yunnan, Anhui, and Neimenggu). Before chemical analysis and feeding broilers, all wheat samples were ground to pass through a sieve with 40 mesh size and stored at −4°C for further analysis. The chemical properties of wheat are shown in Table 1.

Experimental Procedures

A total of seven hundred twenty 1-day-old male broilers (Arbor Acres) were obtained from commercial hatcheries and were fed standard commercial diets during the non-experimental period. Broilers were provided ad libitum access to water and feed throughout the experiment. The experimental diets included 8 test diets and 1 nitrogen-free diet (NFD) (Table 2). In the test diets, wheat was the only source of CP and AA. The NFD was used to measure the IEAA. The analyzed properties of the test diets are shown in Table 3. Titanium dioxide was used as an exogenous indicator in each experimental diet; vitamins and minerals were supplemented to meet or exceed the estimated nutrient requirements for broilers as recommended by the NRC (1994). Each diet was fed for 3 D before ileal content collection on d 13 and 28. The broilers in 6 replicate cages (n = 6 broilers per cage) were euthanized, and the ileal

Table 2. Ingredient properties of experimental diet and nitrogen-free diet (%, air-dried basis).

Ingredients	Wheat diets	N-free diet
Wheat	91.4	-
Soybean oil	4.00	-
Corn starch	-	59.88
Glucose	-	31.50
Carboxymethylcellulose	-	4.20
Dicalcium phosphate	1.90	1.90
Limestone	1.30	1.30
Sodium chloride	0.30	0.30
Choline chloride (60%)	0.20	0.20
Vitamin and mineral premix[1]	0.40	0.22
Titanium dioxide	0.50	0.50
Total	100	100

[1]The premix provided the following per kg of diets: vitamin A 12500 IU, vitamin D3 3,500 IU, vitamin E 20 IU, vitamin K3 3 mg, vitamin B1 0.01 mg, vitamin B2 8.00 mg, vitamin B6 4.5 mg, vitamin B12 0.02 mg, nicotinic acid 34 mg, pantothenic acid 12 mg, folic acid 0.5 mg, biotin 0.2 mg, Fe 80 mg, Cu 8 mg, Zn 80 mg, Mn 80 mg, I 0.7 mg. Se 0.3 mg.

Table 3. Analyzed properties of the test diets (%, DM).

Item	Wheat							
	W-1	W-2	W-3	W-4	W-5	W-6	W-7	W-8
Indispensable amino acids								
Arg	0.62	0.55	0.57	0.62	0.46	0.68	0.65	0.46
His	0.31	0.28	0.29	0.32	0.23	0.32	0.32	0.22
Ile	0.47	0.41	0.45	0.48	0.35	0.48	0.50	0.34
Leu	0.87	0.79	0.83	0.90	0.66	0.90	0.92	0.63
Lys	0.37	0.34	0.36	0.39	0.30	0.38	0.40	0.31
Met	0.22	0.20	0.20	0.22	0.16	0.22	0.23	0.17
Phe	0.63	0.55	0.57	0.61	0.43	0.62	0.65	0.43
Thr	0.38	0.35	0.36	0.39	0.30	0.39	0.40	0.29
Trp	0.14	0.14	0.16	0.16	0.12	0.16	0.15	0.13
Val	0.61	0.55	0.58	0.61	0.45	0.61	0.63	0.45
IAA	4.62	4.12	4.37	4.69	3.46	4.77	4.86	3.43
Dispensable amino acids								
Ala	0.50	0.44	0.47	0.49	0.36	0.50	0.51	0.39
Asp	0.67	0.59	0.62	0.68	0.50	0.68	0.69	0.54
Cys	0.27	0.25	0.27	0.29	0.21	0.28	0.28	0.20
Glu	4.12	3.78	3.82	4.25	2.88	4.20	4.38	2.80
Gly	0.53	0.49	0.51	0.56	0.39	0.55	0.56	0.42
Pro	1.38	1.26	1.30	1.47	1.00	1.45	1.49	0.99
Ser	0.60	0.56	0.56	0.61	0.44	0.60	0.63	0.42
Tyr	0.37	0.26	0.32	0.33	0.23	0.36	0.37	0.24
DAA	8.43	7.62	7.87	8.66	6.03	8.61	8.91	6.00
TAA	13.05	11.74	12.24	13.36	9.48	13.39	13.78	9.43

Abbreviations: Ala, Alanine; Asp, Aspartic acid; Arg, Arginine; Cys, Cystine; DAA, dispensable amino acid; His, Histidine; IAA, Dispensable amino acid; Ile, Isoleucine; Leu, Leucine; Lys, Lysine; Met, Methionine; Phe, Phenylalanine; Pro, Proline; Ser, Serine; TAA, Total amino acid; Thr, Threonine; Trp, Tryptophan; Tyr, Tyrosine; Val, Valine.

contents were collected on day 13. The remaining broilers were fed the standard commercial diet until d 25 when 216 broilers were randomized to 6 replicate cages (n = 4 broilers per cage) for each experimental diet. Broilers were euthanized, and the ileal content were removed on d 28.

All euthanasia was performed by injecting pentobarbital, and all ileal digesta from the lower half of the ileum were collected by gently flushing with distilled water into aluminum containers. The ileum is defined as the part of the small intestine that extends from Meckel's diverticulum to the proximal ileocecal 40 mm. The ileal digesta from all broilers with a cage were pooled and frozen immediately after collection and subsequently freeze-dried. The dried ileal digesta were stored in a refrigerator at −4°C until required for chemical analysis.

Measurements

Dry matter (**DM**), crude protein (**CP**), crude ash (Ash), crude fiber (**CF**), and ether extract (**EE**) were analyzed according to AOAC International (2000) analytical methods (934.01, 990.03, 920.39, 978.10, 942.05, respectively). Neutral detergent fiber (**NDF**) and acid detergent fiber (**ADF**) were determined using an Ankom220 Fiber Analyzer (Ankom Technology, NY) following a modification of the procedures described by Van Soest et al. (1991).

The AA content of wheat samples, experimental diets, and ileal digesta were analyzed by the Ministry of Agriculture Feed Potency and Safety Supervision, Inspection and Testing Center (Beijing) (method 982.30; AOAC, 2000). The titanium dioxide content in experimental diets and ileal digesta was measured as described by Short et al. (1996).

Calculation

IEAA flow in broilers fed the NFD calculated as milligrams of AA content per kilogram of DM intake (**DMI**) using the formula proposed by Adedokun et al. (2008):

$$\text{IEAA, mg/kg of DMI} = [\text{amino acid in ileal digesta, mg/kg} \times (\text{TiO}_2 \text{in diet, mg/kg}/\text{TiO}_2 \text{ in digesta, mg/kg})].$$

The AIDAA values were calculated using the following equations by Lemme et al. (2004):

$$\text{AIDAA, \%} = [1 - (\text{TiO}_2 \text{ in diet}/\text{TiO}_2 \text{ in digesta}) \times (\text{amino acid in digesta}/\text{amino acid in diet})].$$

The SIDAA values were calculated using the following equations by Adewole et al. (2017):

$$\text{SIDAA, \%} = \text{AIDAA, \%} + [(\text{IEAA flow, g/kg of DMI})/(\text{amino acid content of the raw material, g/kg of DM})] \times 100.$$

Statistical Analysis

All analyses were performed using SPSS25.0 software. The data were analyzed using one-way ANOVA. The cage and wheat sample were the experimental units for analyzing the data. Means were compared using the Tukey's studentized range test. The relationship between chemical properties and SIDAA values was analyzed using bivariate correlation analysis by SPSS procedure. The forward stepwise regression method was used to establish linear regression equations for predicting the SIDAA values from the chemical properties of wheat. Differences were considered statistically significant at $P < 0.05$.

RESULTS

Chemical Properties of Wheat

The analyzed properties of the wheat samples are shown in Table 1. The content of CP, EE, Ash, CF, NDF, ADF, and ST were from 9.81 to 16.92%, 1.34 to 2.43%, 1.54 to 2.26%, 1.83 to 3.33%, 10.82 to 14.34%, 3.11 to 4.70%, 65.70 to 76.50%, respectively. The CV of all other properties except ST was more than 10%. Among the 8 sources of wheat, total amino acid (TAA) ranged from 10.36 to 15.81%, and CV was 13.52%. The content of indispensable AA (IAA) was from 3.60 to 5.41%, and the CV was 11.34%. The concentration of dispensable AA (DAA) was from 6.76 to 10.48%, and the CV was 15.30%.

Table 4. Coefficients of standardized ileal digestibility of amino acids in 8 sources of wheat fed to broilers at d 13 (%, DM).

	Wheat											
Item	W-1	W-2	W-3	W-4	W-5	W-6	W-7	W-8	Mean	CV %	SEM	*P*-value
Indispensable amino acids												
Arg	91.28^{ab}	92.89^{ab}	96.82^{b}	95.79^{b}	92.41^{ab}	89.98^{a}	95.73^{b}	87.88^{a}	92.83	3.39	0.56	<0.01
His	90.13^{ab}	92.86^{bc}	96.32^{c}	95.7^{c}	92.22^{bc}	89.98^{ab}	95.45^{c}	86.09^{a}	92.22	3.93	0.56	<0.01
Ile	91.06^{ab}	95.37^{bcd}	99.2^{d}	97.49^{cd}	95.75^{bcd}	89.98^{ab}	98.05^{d}	90.02^{a}	94.82	3.69	0.55	<0.01
Leu	90.06^{a}	95.27^{bcd}	98.51^{d}	96.53^{cd}	95.49^{bcd}	89.98^{ab}	97.85^{d}	90.27^{a}	94.45	3.55	0.51	<0.01
Lys	83.03^{ab}	87.2^{bc}	93.78^{c}	92.64^{c}	87.74^{bc}	89.98^{ab}	91.93^{c}	76.55^{a}	86.71	7.10	0.96	<0.01
Met	90.8^{bc}	92.03^{bcd}	96.31^{d}	94.82^{cd}	92.52^{bcd}	89.98^{abc}	95.03^{cd}	85.82^{a}	92.20	3.63	0.53	<0.01
Phe	92.96^{bcd}	95.66^{de}	97.86^{e}	95.62^{de}	94.12^{cde}	89.98^{ab}	97.35^{de}	86.97^{a}	93.67	4.20	0.58	<0.01
Thr	87.56^{ab}	92.79^{bcd}	99.16^{d}	97.57^{d}	91.17^{abcd}	89.98^{ab}	96.78^{cd}	84.01^{a}	91.74	6.39	0.94	<0.01
Trp	87.2^{a}	91.43^{abc}	95.83^{c}	94.83^{bc}	92.41^{abc}	89.98^{ab}	94.42^{bc}	86.21^{a}	91.08	4.00	0.67	<0.01
Val	90.24^{ab}	93.78^{bc}	97.87^{c}	96.43^{c}	93.06^{bc}	89.98^{ab}	96.48^{c}	87.05^{a}	93.01	4.18	0.61	<0.01
IAA	89.43^{ab}	92.93^{bc}	97.17^{c}	95.74^{c}	92.41^{bc}	89.98^{ab}	95.91^{c}	86.09^{a}	92.27	4.30	0.62	<0.01
Dispensable amino acids												
Ala	86.27^{ab}	90.75^{bc}	95.91^{c}	94.11^{c}	90.91^{bc}	85.22^{ab}	94.69^{c}	83.69^{a}	90.20	5.16	0.76	<0.01
Asp	88.51^{ab}	90.32^{abcd}	96.53^{d}	95.81^{cd}	90.01^{abcd}	85.75^{a}	94.4^{bcd}	85.55^{a}	90.86	4.74	0.70	<0.01
Cys	91.09^{abc}	93.02^{bcd}	96.59^{d}	95.53^{cd}	91.67^{bc}	89.98^{ab}	95.61^{cd}	86.55^{a}	92.50	3.66	0.53	<0.01
Glu	97.12^{abc}	98^{bcde}	99.17^{e}	98.99^{de}	97.67^{bcde}	96.51^{ab}	98.87^{cde}	95.6^{a}	97.74	1.31	0.19	<0.01
Gly	89.04^{ab}	91.97^{bc}	96.94^{c}	96.03^{c}	91.00^{abc}	89.98^{ab}	95.52^{c}	84.69^{a}	91.56	4.82	0.71	<0.01
Pro	97.28^{ab}	97.99^{ab}	99.42^{b}	99.4^{b}	97.98^{ab}	89.98^{a}	99.38^{b}	95.71^{a}	97.98	1.42	0.23	<0.01
Ser	93.05^{abc}	94.98^{bcd}	98.83^{d}	98.12^{d}	93.11^{abc}	90.76^{ab}	97.3^{cd}	89.58^{a}	94.47	3.63	0.54	<0.01
Tyr	95.02^{abc}	95.37^{bc}	99.52^{c}	96.85^{c}	93.21^{abc}	89.98^{a}	97.1^{c}	89.25^{ab}	94.35	4.09	0.68	<0.01
DAA	92.17^{abc}	94.05^{bcde}	97.86^{e}	96.86^{de}	93.19^{abcd}	89.98^{ab}	96.61^{cde}	88.83^{a}	93.71	3.50	0.52	<0.01
TAA	90.65^{ab}	92.93^{bc}	97.48^{c}	96.24^{c}	92.41^{bc}	89.98^{ab}	96.22^{c}	87.31^{a}	92.91	3.93	0.58	<0.01

Abbreviations: Ala, Alanine; Asp, Aspartic acid; Arg, Arginine; CV, coefficient of variation; Cys, Cystine; DAA, dispensable amino acid; Glu, Glutamic acid; Gly, Glycine; His, Histidine; IAA, Dispensable amino acid; Ile, Isoleucine; Leu, Leucine; Lys, Lysine; Max, maximum; Min, minimum; Met, Methionine; Phe, Phenylalanine; Pro, Proline; Ser, Serine; TAA, Total amino acid; Thr, Threonine; Trp, Tryptophan; Tyr, Tyrosine; Val, Valine.

$^{a-e}$ Means within a row lacking a common superscript differ ($p < 0.05$).

Digestibility of AA

The SID of AA in the wheat samples at days 13 and 28 are presented in Tables 4 and 5. At day 13, the SIDAA were different ($P < 0.01$) among the 8 sources of wheat. For the SID of IAA, the lowest was Lys (86.71%), the highest was Ile (94.82%), and the average was 92.27%. For the SID of DAA, the lowest was Ala (90.2%), the highest was Pro (97.98%), and the average was 93.71%. The average SID of TAA on d 13 was 92.91%. On d 28, the SID of Phe, Val, Cys, Pro, and Tyr were different ($P < 0.05$) among the 8 sources of wheat. For the SID of IAA, the lowest was His (81.31%), the highest was Ile

Table 5. Coefficients of standardized ileal digestibility of amino acids in 8 sources of wheat fed to broilers at d 28 (%, DM).

	Wheat											
Item	W-1	W-2	W-3	W-4	W-5	W-6	W-7	W-8	Mean	CV %	SEM	*P*-value
Indispensable amino acids												
Arg	90.99	89.78	88.36	87.06	91.77	88.55	92.71	89.92	89.89	2.09	0.49	0.056
His	90.43	89.1	87.92	87.68	90.53	88.22	92.47	88.44	89.35	1.86	0.47	0.150
Ile	95	91.75	92.14	91.16	96.51	91.06	96.13	93.88	93.45	2.39	0.54	0.123
Leu	92.73	91.18	89.89	90.65	94.43	90.15	94.98	91.92	91.99	2.09	0.47	0.068
Lys	81.75	78.97	80.79	77.83	84.07	79.77	86.61	80.73	81.31	3.49	0.86	0.175
Met	90.55	89.95	89.85	88.08	91.14	89.36	92.44	88.61	90	1.54	0.44	0.233
Phe	92.74^{ab}	91.63^{ab}	89.56^{a}	90.31^{ab}	93.7^{ab}	89.54^{a}	94.75^{b}	91.8^{ab}	91.75	2.08	0.46	0.015
Thr	91.23	89.28	87.44	82.9	94.48	84.57	92.41	88.67	88.87	4.38	1.00	0.051
Trp	88.94	87.71	87.76	83.59	89.95	87.22	87.26	89.2	87.7	2.21	0.70	0.443
Val	91.51^{ab}	89.22^{ab}	89.05^{ab}	86.32^{a}	92.62^{ab}	88.56^{ab}	92.85^{b}	89.03^{ab}	89.89	2.49	0.55	0.022
IAA	90.59	88.86	88.28	86.56	91.92	87.7	92.26	89.22	89.42	2.26	0.55	0.120
Dispensable amino acids												
Ala	85.57	83.72	83.54	80.96	87.09	84.06	89.25	84.83	84.88	2.95	0.74	0.142
Asp	88.04	86.27	85.07	82.52	89.81	84.99	90.1	87.59	86.8	2.98	0.78	0.223
Cys	92.9^{ab}	92.21^{ab}	91.66^{a}	88.4^{ab}	94.33^{b}	89.4^{ab}	92.89^{ab}	90.77^{ab}	91.57	2.14	0.50	0.043
Glu	96.77	96.32	94.93	95.03	96.71	95.35	97.52	95.48	96.01	0.99	0.23	0.059
Gly	90.72	88.1	87.05	85.91	90.69	87.03	91.93	88.61	88.76	2.41	0.57	0.107
Pro	97.77^{ab}	97.17^{ab}	96.5ab	94.86^{a}	98.05^{b}	96.18^{ab}	98.13^{b}	95.98^{ab}	96.83	1.19	0.27	0.012
Ser	94.92	94.06	91.96	90.33	96.34	91.18	95.17	93.18	93.39	2.25	0.53	0.061
Tyr	96.04^{ab}	94.17^{ab}	94.48^{ab}	93.6^{ab}	99.12^{b}	93.35^{a}	98.08^{ab}	95.93^{ab}	95.6	2.21	0.50	0.013
DAA	92.84	91.5	90.65	88.95	94.02	90.19	94.13	91.55	91.73	1.99	0.48	0.098
TAA	91.59	90.03	89.33	87.62	92.85	88.81	93.09	90.25	90.45	2.13	0.52	0.108

Abbreviations: Ala, Alanine; Asp, Aspartic acid; Arg, Arginine; CV, coefficient of variation; Cys, Cystine; DAA, dispensable amino acid; Glu, Glutamic acid; Gly, Glycine; His, Histidine; IAA, Dispensable amino acid; Ile, Isoleucine; Leu, Leucine; Lys, Lysine; Max, maximum; Min, minimum; Met, Methionine; Phe, Phenylalanine; Pro, Proline; Ser, Serine; TAA, Total amino acid; Thr, Threonine; Trp, Tryptophan; Tyr, Tyrosine; Val, Valine.

^{a,b} Means within a row lacking a common superscript differ ($p < 0.05$).

Table 6. Effect of age on standardized ileal digestibility of amino acids in wheat fed to broilers.

Item	Day 13	Day 28	SEM	*P*-value
Indispensable amino acids				
Arg	93.38	89.90	0.41	<0.01
His	92.89	89.37	0.41	<0.01
Ile	95.49	93.46	0.41	0.013
Leu	95.09	92.02	0.39	<0.01
Lys	87.88	81.35	0.75	<0.01
Met	92.79	90.03	0.38	<0.01
Phe	94.47	91.78	0.40	0.001
Thr	92.82	88.92	0.74	0.008
Trp	91.65	87.65	0.53	<0.01
Val	93.75	89.91	0.47	<0.01
IAA	93.02	89.44	0.46	<0.01
Dispensable amino acids				
Ala	91.09	84.88	0.64	<0.01
Asp	91.58	86.79	0.61	<0.01
Cys	93.09	91.61	0.38	0.055
Glu	97.98	96.02	0.19	<0.01
Gly	92.35	88.75	0.50	<0.01
Pro	98.22	96.84	0.20	<0.01
Ser	95.05	93.41	0.40	0.041
Tyr	95.04	95.63	0.41	0.474
DAA	94.30	91.74	0.39	0.001
TAA	93.59	90.46	0.43	<0.01

Abbreviations: Ala, Alanine; Asp, Aspartic acid; Arg, Arginine; CV, coefficient of variation; Cys, Cystine; DAA, dispensable amino acid; Glu, Glutamic acid; Gly, Glycine; His, Histidine; IAA, Dispensable amino acid; Ile, Isoleucine; Leu, Leucine; Lys, Lysine; Max, maximum; Min, minimum; Met, Methionine; Phe, Phenylalanine; Pro, Proline; Ser, Serine; TAA, Total amino acid; Thr, Threonine; Trp, Tryptophan; Tyr, Tyrosine; Val, Valine.

(93.45%), and the average was 89.42%. For the SID of DAA, the lowest was Ala (84.88%), the highest was Pro (96.83%), and the average was 91.73%. The average SID of TAA on d 28 was 90.45%.

The effect of age on the SID of AA is shown in Table 6. The results show that broiler age affected the SID of all AA ($P < 0.05$) except Tyr and Cys. The SID of most AA was higher on day 13 than on d 28.

Correlation Analysis

Tables 7 and 8 show the correlation between the chemical properties and SIDAA of wheat in this experiment. On d 13, the SID of all AA was positively correlated ($P < 0.05$) with CP, and there was a positive correlation ($P < 0.05$) between NDF and Ala, Val, Met, Ile, Leu, His, Lys. All amino acids had a negative relationship ($P < 0.05$) with ST except Ala, Ile, Leu, and Phe. On d 28, there was a negative correlation ($P < 0.05$) between EE and the SID of Phe, Glu. There was a positive correlation ($P <0.05$) between Ash and Cys, Tyr, Ile. ADF had a positive correlation ($P < 0.05$) with the SID of Asp, Ser, Cys, Tyr, Pro, Gly, DAA, Thr, Val, Ile, Leu, Phe, Arg, IAA, and TAA. A positive correlation ($P < 0.05$) was observed between NDF and the SID of Tyr, Val, Ile, Leu, Lys.

Prediction Equations for AA

Stepwise regression equations for predicting the SIDAA for broilers on d 13 and 28 based on the chemical properties of wheat are presented in Tables 9 and 10. On d 13, the R^2 value of stepwise regression equations for predicting the SIDAA was the best for Glu, $SID_{Glu} = 82.340 + 0.545CP + 0.408NDF + 1.568CF - 0.715EE$ ($R^2 = 0.969$, $P < 0.01$) and least

Table 7. Correlation analysis between chemical properties and coefficients of standardized ileal amino acid digestibility of wheat at d 13.

Item	CP	EE	Ash	CF	NDF	ADF	ST
Arg	0.502**	−0.107	−0.097	0.038	0.292	−0.162	−0.424**
His	0.528**	−0.107	−0.036	0.043	0.321*	−0.145	−0.396*
Ile	0.387*	−0.150	−0.030	0.098	0.441**	−0.019	−0.248
Leu	0.373*	−0.120	0.023	0.100	0.479**	0.004	−0.193
Lys	0.454**	−0.154	−0.079	0.055	0.368*	−0.070	−0.350*
Met	0.498**	−0.197	−0.097	−0.032	0.375*	−0.091	−0.412*
Phe	0.520**	0.055	−0.128	0.170	0.304	−0.044	−0.295
Thr	0.454**	−0.077	−0.063	0.127	0.278	−0.152	−0.350*
Trp	0.498**	−0.109	0.067	0.009	0.245	−0.263	−0.400*
Val	0.480**	−0.101	−0.051	0.093	0.333*	−0.121	−0.349*
IAA	0.489**	−0.112	−0.053	0.076	0.354*	−0.114	−0.359*
Ala	0.421**	−0.113	−0.072	0.090	0.390*	−0.055	−0.303
Asp	0.448**	−0.132	−0.134	0.079	0.265	−0.179	−0.430**
Cys	0.578**	−0.076	−0.039	0.009	0.250	−0.224	−0.459**
Glu	0.524**	−0.079	−0.046	0.055	0.280	−0.159	−0.397*
Gly	0.518**	−0.103	−0.062	0.050	0.288	−0.179	−0.422**
Pro	0.477**	−0.092	−0.070	−0.012	0.307	−0.120	−0.393*
Ser	0.554**	−0.048	−0.072	0.093	0.182	−0.271	−0.469**
Tyr	0.480**	0.033	−0.250	0.226	0.149	−0.162	−0.392*
DAA	0.514**	−0.081	−0.105	0.088	0.275	−0.175	−0.423**
TAA	0.501**	−0.100	−0.074	0.081	0.323*	−0.139	−0.386*

*0.01< $P \leq 0.05$.
**0.001< $P \leq 0.01$.Abbreviations: Ala, Alanine; Asp, Aspartic acid; Arg, Arginine; Cys, Cystine; DAA, dispensable amino acid; Glu, Glutamic acid; Gly, Glycine; His, Histidine; IAA, Dispensable amino acid; Ile, Isoleucine; Leu, Leucine; Lys, Lysine; Met, Methionine; Phe, Phenylalanine; Pro, Proline; Ser, Serine; TAA, Total amino acid; Thr, Threonine; Trp, Tryptophan; Tyr, Tyrosine; Val, Valine.

Table 8. Correlation analysis between chemical properties and coefficients of standardized ileal amino acid digestibility of wheat at d 28.

Item	CP	EE	Ash	CF	NDF	ADF	ST
Arg	0.022	0.268	−0.213	−0.075	0.254	0.335*	0.106
His	0.166	0.254	−0.163	−0.158	0.238	0.219	−0.041
Ile	−0.045	0.138	−0.364*	−0.048	0.340*	0.439**	0.094
Leu	0.037	0.218	−0.222	−0.123	0.315*	0.376*	0.077
Lys	0.104	0.113	−0.220	−0.149	0.350*	0.266	−0.033
Met	0.221	0.155	−0.153	−0.111	0.287	0.214	−0.037
Phe	0.051	0.314*	−0.212	−0.069	0.243	0.338*	0.088
Thr	−0.051	0.187	−0.292	0.084	0.286	0.442**	0.211
Trp	−0.192	0.042	−0.190	0.109	0.106	0.287	0.235
Val	0.061	0.167	−0.289	−0.082	0.344*	0.396**	0.078
IAA	0.026	0.191	−0.260	−0.053	0.304*	0.370*	0.098
Ala	0.084	0.181	−0.178	−0.147	0.291	0.268	0.028
Asp	−0.018	0.191	−0.21	−0.032	0.236	0.328*	0.138
Cys	−0.010	0.149	−0.300*	0.141	0.282	0.428**	0.204
Glu	0.152	0.342*	−0.135	−0.124	0.184	0.251	0.032
Gly	0.061	0.247	−0.263	−0.094	0.236	0.311*	0.040
Pro	0.094	0.220	−0.240	−0.023	0.288	0.366*	0.105
Ser	−0.049	0.227	−0.250	0.077	0.233	0.428**	0.230
Tyr	−0.077	0.094	−0.325*	−0.028	0.415**	0.499**	0.161
DAA	0.019	0.208	−0.256	−0.032	0.292	0.383**	0.128
TAA	0.023	0.199	−0.259	−0.044	0.300*	0.377*	0.111

*0.01< $P \leq 0.05$.
**0.001< $P \leq 0.01$.Abbreviations: Ala, Alanine; Asp, Aspartic acid; Arg, Arginine; Cys, Cystine; DAA, dispensable amino acid; Glu, Glutamic acid; Gly, Glycine; His, Histidine; IAA, Dispensable amino acid; Ile, Isoleucine; Leu, Leucine; Lys, Lysine; Met, Methionine; Phe, Phenylalanine; Pro, Proline; Ser, Serine; TAA, Total amino acid; Thr, Threonine; Trp, Tryptophan; Tyr, Tyrosine; Val, Valine.

Table 9. Prediction equations of standardized ileal digestibility of some amino acids based on the chemical properties of wheat at d 13.

Prediction equations	R^2	*P*-value
$SID_{Asp} = 80.920 + 0.793CP + 6.187CF + 1.752NDF - 0.557ST$	0.785	<0.01
$SID_{Ser} = 54.592 + 1.509CP + 4.584CF + 0.849NDF - 1.988EE$	0.836	<0.01
$SID_{Lys} = 11.471 + 2.465CP + 2.293NDF + 7.500CF - 3.791EE$	0.841	<0.01
$SID_{Ala} = 32.876 + 1.152CP + 3.223NDF + 6.061CF - 4.00ADF$	0.845	<0.01
$SID_{Gly} = 38.696 + 1.869CP + 1.416NDF + 5.440CF - 2.633EE$	0.845	<0.01
$SID_{Glu} = 82.340 + 0.545CP + 0.408NDF + 1.568CF - 0.715EE$	0.852	<0.01
$SID_{DAA} = 53.183 + 1.389CP + 4.345CF + 1.054NDF - 1.746EE$	0.854	<0.01
$SID_{His} = 48.626 + 1.526CP + 1.219NDF + 4.315CF - 2.143EE$	0.855	<0.01
$SID_{Va}l = 43.773 + 1.59CP + 1.436NDF + 5.046CF - 2.076EE$	0.862	<0.01
$SID_{TAA} = 47.101 + 1.515CP + 1.300NDF + 4.702CF - 1.970EE$	0.862	<0.01
$SID_{IAA} = 42.233 + 1.615CP + 1.495NDF + 4.988CF - 2.134EE$	0.863	<0.01
$SID_{Trp} = 58.282 + 0.594CP + 2.934NDF + 4.307CF - 6.408ADF$	0.866	<0.01
$SID_{Arg} = 54.692 + 1.342CP + 1.041NDF + 3.852CF - 6.408ADF$	0.870	<0.01
$SID_{Ile} = 43.133 + 2.646NDF + 0.808CP + 4.845CF - 2.783ADF + 2.687Ash$	0.877	<0.01
$SID_{Phe} = 38.396 + 1.562CP + 5.289CF + 1.598NDF$	0.907	<0.01
$SID_{Cys} = 53.198 + 1.532CP + 0.963NDF + 3.866CF - 2.163EE$	0.912	<0.01
$SID_{Met} = 53.901 + 1.431CP + 1.158NDF + 3.465CF - 2.661EE$	0.932	<0.01
$SID_{Tyr} = 78.315 + 0.956CP + 6.841CF + 1.278NDF - 0.445ST$	0.940	<0.01
$SID_{Leu} = 81.065 + 2.069NDF + 7.754CF + 14.188Ash - 1.024ST + 3.411ADF$	0.972	<0.01

Abbreviations: Ala, Alanine; Asp, Aspartic acid; Arg, Arginine; Cys, Cystine; DAA, dispensable amino acid; Glu, Glutamic acid; Gly, Glycine; His, Histidine; IAA, Dispensable amino acid; Ile, Isoleucine; Leu, Leucine; Lys, Lysine; Met, Methionine; Phe, Phenylalanine; Pro, Proline; Ser, Serine; TAA, Total amino acid; Thr, Threonine; Trp, Tryptophan; Tyr, Tyrosine; Val, Valine.

Table 10. Prediction equations of standardized ileal digestibility of some amino acids based on the chemical properties of wheat at d 28.

Prediction equations	R^2	*P*-value
$SID_{His} = 73.046 + 0.931NDF + 2.728EE$	0.678	<0.01
$SID_{Met} = 75.492 + 0.905NDF + 1.913EE$	0.703	<0.01
$SID_{Ile} = 76.645 + 3.716ADF + 1.955EE$	0.707	<0.01
$SID_{IAA} = 73.90 + 3.221ADF + 2.218EE$	0.723	<0.01
$SID_{TAA} = 75.414 + 3.110ADF + 2.169EE$	0.739	<0.01
$SID_{Cys} = 76.262 + 3.358ADF + 1.832EE$	0.758	<0.01
$SID_{DAA} = 77.306 + 2.970ADF + 2.107EE$	0.759	<0.01
$SID_{Tyr} = 79.232 + 3.802ADF + 1.543EE$	0.786	<0.01
$SID_{Lys} = 50.266 + 2.014NDF + 3.571EE$	0.790	<0.01
$SID_{Pro} = 81.925 + 2.453ADF + 0.319CP + 0.993EE$	0.824	<0.01
$SID_{Thr} = 56.301 + 6.958ADF + 4.265EE$	0.877	<0.01
$SID_{Leu} = 80.032 + 3.487ADF + 2.630EE - 1.926CF$	0.899	<0.01
$SID_{Phe} = 78.947 + 3.202ADF + 3.133EE - 1.578CF$	0.910	<0.01
$SID_{Arg} = 77.412 + 3.304ADF + 2.960EE - 1.715CF$	0.941	<0.01
$SID_{Asp} = 74.641 + 4.357ADF + 3.579EE - 2.198CF - 2.056Ash$	0.943	<0.01
$SID_{Ser} = 71.811 + 5.679ADF + 2.117EE + 0.388CP - 0.501NDF - 0.631CF$	0.977	<0.01
$SID_{Ala} = 53.364 + 0.794NDF + 2.976EE - 3.935CF - 6.903Ash + 0.494ST + 0.402CP$	0.981	<0.01
$SID_{Val} = 41.157 + 1.173CP + 0.612EE - 4.254CF - 11.365Ash + 0.920ST$	0.992	<0.01
$SID_{Gly} = 69.932 + 1.960ADF + 2.524EE + 0.292ST - 2.885CF - 5.796Ash + 0.414CP$	0.995	<0.01
$SID_{Glu} = 86.749 + 1.453EE + 1.742ADF - 0.693CF + 0.169CP$	0.998	<0.01

Abbreviations: Ala, Alanine; Asp, Aspartic acid; Arg, Arginine; Cys, Cystine; DAA, dispensable amino acid; Glu, Glutamic acid; Gly, Glycine; His, Histidine; IAA, Dispensable amino acid; Ile, Isoleucine; Leu, Leucine; Lys, Lysine; Met, Methionine; Phe, Phenylalanine; Pro, Proline; Ser, Serine; TAA, Total amino acid; Thr, Threonine; Trp, Tryptophan; Tyr, Tyrosine; Val, Valine.

for Asp, $SID_{Asp} = 80.920 + 0.793CP + 6.187CF + 1.752NDF - 0.557ST$ ($R^2 = 0.785$, $P < 0.01$). On d 28, the R^2 value of stepwise regression equations for predicting the SIDAA was the best for Gly, $SID_{Gly} = 69.932 + 1.960ADF + 2.524EE + 0.292ST - 2.885CF - 5.796Ash + 0.414CP$ ($R^2 = 0.995$, $P < 0.01$), and least for His, $SID_{His} = 73.046 + 2.728EE + 0.931NDF$ ($R^2 = 0.678$, $P < 0.01$).

DISCUSSION

In the present study, there were great differences in the chemical properties of 8 sources of wheat. This indicated that the wheat samples were representative. Different properties of wheat can be attributed to the growing environments (Lasek et al., 2020), variety (Azhar et al., 2019), and so on. Based on chemical analyses, the W-6 sample appeared to be of better quality than others, with high TAA content. Generally, the chemical properties of the samples were within the range previously reported for wheat (Lasek et al., 2020; Lu et al., 2020).

At d 13, the SID of Met, Leu, Lys, and Trp measured in this experiment were close to those reported by Szczurek et al. (2020) in 14-day-old broilers; the SID of Glu, Thr, Leu, and Arg were close to those reported by Barua et al., (2021b) in 14-day-old broilers. At d 28, the SID of Ala, Glu, Met, Leu, Phe, and Lys were close to the values measured by Lu et al. (2020) in 28-day-old broilers. The SID of Glu, Ala, Cys, Pro, Met, and Leu were close to those reported by Bandegan et al. (2011) in 21-day-old broilers, and the value of Trp was similar to that reported by Osho et al. (2019) in 21-day-old broilers. Except for the AA already mentioned, the SIDAA values measured in our experiment differed from those reported in each literature.

Several factors may contribute to these differences. First, the different growing environments and varieties of wheat may be the primary factor. On d 13 and 28, we also found that the SID of some amino acids varied among wheat samples from different sources. Due to the different growing environments and varieties of wheat, different sources of wheat have different chemical properties (Azhar et al., 2019). It had been shown that SIDAA was influenced by the protein content in wheat (Lu et al., 2020). The sources of wheat may also impact the anti-nutritional factors (Choct, 2002) that have many adverse effects on SIDAA. Anti-nutritional factors can increase intestinal viscosity and encapsulate nutrients (Jha and Mishra, 2021), reducing the digestibility of raw materials in broilers (Gutierrez et al., 2008) and thus affecting SIDAA. Secondly, it could be related to the breed of experimental broilers, as different strains have different physiological structures and digest AA differently. Finally, the variations in IEAA loss resulting from different ratios of corn starch to glucose or sucrose in nitrogen-free diet formulations (Zhou et al., 2022) may lead to differences in SIDAA.

Based on the results of this study, it was concluded that 14-day-old broilers had a higher SIDAA than 28-day-old broilers, and the SIDAA decreased with advancing age. According to the literature, there are different results of the effect of broiler age on the ileal digestibility of AA in raw materials. The results are the ileal digestibility of raw amino acids increased with broiler age (Batal and Parsons, 2002; Garcia et al., 2007; An and Kong, 2022), decreased with broiler age (Kim and Corzo, 2012; Zuprizal and Chagneau, 1992; Adedokun et al., 2008), and remained unchanged with broiler age (Barua et al., 2021b). These contradictory experimental results may be due to the content of anti-nutritional factors in the feed raw materials and the development of the intestinal tract in poultry. The wheat samples in our experiment contain low anti-nutritional factors with almost no negative impact on the intestine, while the intestine of poultry reaches its maximum digestive and absorptive capacity at about 2 wk (Nitsan et al., 1991) and then tends to decrease (Bryan and Jared D., 1992). So, the SIDAA is higher in younger broilers than in older broilers. However, the intestinal tract of young broilers is not fully developed, such as the small intestinal villi are not fully developed and the intestinal flora is not fully established (Nitsan et al., 1991; Iji et al., 2001; Kers et al., 2020). Older broilers have more mature intestinal tracts and are more tolerant to anti-nutritional factors. The SIDAA for feed ingredients containing higher anti-nutritional factors was higher in older broilers than in younger ones (Barua et al., 2021a).

The effect of the chemical properties of wheat on the SID of some AA was variable. For example, on d 28, the SID of Ile was positively correlated with CP, while the SID of Leu was negatively correlated with CP. The results are in agreement with the results reported by Wang et al. (2020) in corn distillers dried grains with solubles (DDGS) and Ebadi et al. (2011) in sorghum. These may be due to the different digestion processes of the various amino acids. In the process of establishing the prediction equation, we found that using only one predictor cannot establish the prediction equation for the SIDAA in wheat. In addition, with the introduction of predictors in the equation, the predictive ability of the equation has been improved. These suggested that the SIDAA in wheat may be influenced by multiple chemical properties.

CONCLUSIONS

The results of this study indicate that chemical properties and the SID of most AA in wheat showed a huge difference. Wheat source and broiler age can affect the SIDAA of wheat. There was a correlation between SIDAA and the chemical properties of wheat. We could develop the prediction equation for SIDAA based on the chemical properties of wheat samples.

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DISCLOSURES

We declare that no financial and personal relationships with other people or organizations can inappropriately influence our work, and there is no professional or other personal interest of any nature or kind in any product, service, and/or company that could be construed as influencing the content of this paper.

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