



# Oligosaccharides are a key factor in prediction of amino acid digestibility in soybean meal of different origins when fed to growing pigs

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**Objective:** The objective of this experiment was to determine apparent ileal digestibility (AID) and standardized ileal digestibility (SID) of crude protein (CP) and amino acid (AA) in 15 sources of soybean meal (SBM) produced from soybeans from different countries and subsequently to establish equations for predicting the AID and SID in SBM based on their chemical composition.

**Methods:** Eighteen barrows (57.9±6.1 kg) fitted with a simple T-cannula were allotted into three 6×6 Latin square designs. Each period comprised a 6-d adaption period followed by a 2-d collection of ileal digesta. The 15 test diets included SBM as a sole source of AA in the diet. Another nitrogen-free diet was used to measure basal endogenous losses of CP and AA. Chromic oxide (0.3%) was used as an inert marker in each diet.

**Results:** The AID of lysine in SBM from China and USA tended to be greater than in SBM from Brazil ( $p<0.10$ ). The SID of valine and proline in SBM from China was greater than in SBM from Brazil ( $p<0.05$ ). The SID of lysine, threonine, cysteine and glycine in SBM from China tended to be greater than in SBM from Brazil ( $p<0.10$ ). From a stepwise regression analysis, a series of AID and SID prediction equations were generated. The best fit equations for lysine in SBM were: AID lysine = 1.16 sucrose-1.81 raffinose+82.10 ( $R^2 = 0.69$ ,  $p<0.01$ ) and SID lysine = 1.14 sucrose-1.93 raffinose-0.99 ether extract (EE)+85.26 ( $R^2 = 0.77$ ,  $p<0.01$ ).

**Conclusion:** It was concluded that under the conditions of this experiment, the oligosaccharides (such as sucrose and raffinose) can be used to predict the AID and SID of AA in SBM with reasonable accuracy.

**Keywords:** Amino Acid; Digestibility; Chemical Composition; Oligosaccharides; Pigs; Prediction Equations; Soybean Meal

## INTRODUCTION

Soybean meal (SBM) is the most widely used source of plant protein in swine diets [1]. Considerable quantities of soybeans are imported and processed in China [2]. We have compared the energy value of conventional SBM processed from soybeans grown in different countries in our previous work, in which there were no differences in the digestible energy and metabolizable energy of SBM among the different soybean sources [2]. Frikha et al [3] have determined the correlations between chemical composition and standardized ileal digestibility (SID) of crude protein (CP) and amino acid (AA) of different origin SBM in broilers [3]. However, in pigs limited information is available on comparison and prediction of SID of SBM produced from soybeans grown in different countries, but processed in China. The effect of oligosaccharides on digestibility of AA remains controversial. Some studies have reported that soybean oligosaccharides have negative effects on digestibility, performance, and health of pigs [4-6]. However, one study reported

that the addition of soy oligosaccharides to SBM-based diets minimally affected the apparent or true ileal digestibility of AA [7]. Additionally, digestibility of AA in low oligosaccharide SBM was not different from that in conventional SBM [8,9]. Therefore, the objective of this study was to test the hypothesis that conventional SBM processed from soybeans grown in different countries have different AA digestibility. The second objective was to develop equations to predict AA digestibility in SBM based on chemical composition and oligosaccharides.

## MATERIALS AND METHODS

### Animal care

The experimental protocol used in this study was approved by the Institutional Animal Care and Use Committee at China Agricultural University (Beijing, China).

### SBM sample collection

Fifteen SBM samples used in the current experiment were chosen from 22 SBM samples in our previous work [2]. The SBM sources 1 (China) and sources 8 (USA) and sources 16 and 19 (Brazil) in our previous work were removed due to their similar AA composition. The soybeans used to produce the 15 SBM originated from China (n = 5), USA (n = 5), and Brazil (n = 5). All soybeans were dehulled before crushing, but in some cases, hulls were added back to the meal after crushing. One source of SBM (source 10) had soapstock added after crushing, but the other sources contained no soapstock. Specific processing information, chemical composition and AA content of the 15 SBM are shown in Table

1, 2, and 3, respectively.

### Animals, diets, and experimental design

A total of 18 barrows (Duroc×Landrace×Yorkshire) with an initial body weight (BW) 57.9±6.1 kg were fitted with a simple T-cannula in the distal ileum [10]. After surgery, the barrows were individually placed in stainless-steel metabolism crates (1.4×0.7×0.6 m) in an environmentally controlled room (20°C±2°C). Each crate was installed with a one-hole feeder and a nipple drinker.

The 18 barrows were allotted into three 6×6 Latin square designs according to their initial BW with 6 pigs for each Latin square. Each Latin square contained 5 SBM which were the only source of AA and one nitrogen-free diet which was used to estimate basal ileal endogenous losses of CP and AA. The diets used in this experiment were prepared based on chemical composition of the feed ingredients (Tables 4, 5) in order to above the threshold level of CP and AA [11].

All diets contained 0.3% chromic oxide as an indigestible marker and vitamins and minerals were included to meet or exceed the estimated nutrient requirements for growing pigs recommended by NRC [12]. Each period comprised a 6-d adaption period followed by a 2-d collection of ileal digesta. All pigs were provided daily feed equivalent to 4% of BW, and two equal sized meals were provided every day at 08:00 and 15:00 h. Pigs had free access to water throughout the experiment.

### Sample collection

During each of the 6 periods, the first 6 d were for adaptation to the diet. On d 7 and 8, ileal digesta samples were collected from

**Table 1.** Origin of soybean meal<sup>1)</sup>

No. <sup>2)</sup>	No. <sup>3)</sup>	Source of soybean	Plants <sup>4)</sup>	Location of plants in China	Dehulled/regular <sup>5)</sup>	Special processing
2	1	China	A1	Heilongjiang	Regular	
3	2	China	A2	Heilongjiang	Regular	
4	3	China	B1	Heilongjiang	Regular	
5	4	China	B2	Heilongjiang	Dehulled	
6	5	China	C	Hebei	Dehulled	
7	6	USA	D	Jiangsu	Regular	
9	7	USA	E	Shandong	Regular	
10	8	USA	F	Henan	Regular	
11	9	USA	G	Shandong	Regular	
12	10	USA	H1	Tianjin	Regular	Soapstock added <sup>6)</sup>
13	11	Brazil	I1	Guangdong	Regular	
14	12	Brazil	I2	Guangdong	Regular	
15	13	Brazil	I3	Guangdong	Dehulled	
17	14	Brazil	J	Shandong	Regular	
18	15	Brazil	H2	Tianjin	Dehulled	

SBM, soybean meal.

<sup>1)</sup> Data adjusted from Li et al [2].

<sup>2)</sup> Number of SBM source in Li et al [2].

<sup>3)</sup> Number of SBM source in the current experiment.

<sup>4)</sup> The same capital letter means the soybeans were processed in the same facility.

<sup>5)</sup> Regular means that soybean hulls were added to the crushed meal and dehulled means that no hulls were added after crushing.

<sup>6)</sup> Soapstock was added to the crushed meal from this crushing plant but that was not the case for the other plants.

**Table 2.** Chemical composition of the 15 soybean meals (% of DM)<sup>1)</sup>

Source of soybean	No.	GE (MJ/kg)	DM	CP	EE	AEE	NDF	ADF	CF	Ash	Calcium	Phosphorus	Sucrose	Raffinose	Stachyose
China	1	19.81	89.03	49.35	1.27	1.34	19.45	8.07	6.01	6.61	0.22	0.83	5.35	0.52	1.93
	2	19.38	90.02	47.74	0.94	1.31	17.22	7.91	6.95	6.21	0.36	0.8	5.75	0.57	1.85
	3	19.56	89.24	50.29	0.67	1.34	12.08	5.78	5.56	6.43	0.37	0.84	4.65	0.39	1.46
	4	19.99	89.1	51.69	0.23	0.77	12.69	4.52	3.97	6.47	0.52	0.87	5.04	0.46	1.5
	5	19.23	90.92	51.62	0.76	1.26	14.43	4.14	3.64	6.25	0.36	0.81	5.58	0.66	2.03
	Mean	19.59	89.66	50.14	0.77	1.20	15.17	6.08	5.23	6.39	0.37	0.83	5.27	0.52	1.75
USA	6	19.21	89.99	49.97	0.69	1.66	14.24	6.46	5.82	6.71	0.27	0.74	7.12	0.95	3.27
	7	19.33	89.87	50.09	0.96	1.45	13.76	6.63	5.96	6.85	0.46	0.74	5.71	0.72	2.4
	8	19.66	89.67	48.36	1.41	1.82	19.37	11.33	6.34	6.52	0.47	0.7	6.11	0.9	2.34
	9	19.41	89.41	48.25	1.4	1.71	13.9	6.55	5.59	6.17	0.39	0.68	5.49	0.74	2.6
	10	18.94	90.22	49.44	1.91	2.18	13.14	6.62	5.3	6.84	0.55	0.74	4.78	0.68	2.11
	Mean	19.31	89.83	49.22	1.27	1.76	14.88	7.52	5.80	6.62	0.43	0.72	5.84	0.80	2.54
Brazil	11	19.19	89.56	45.31	1.11	1.64	19.47	11.86	9.9	6.33	0.75	0.6	4.09	0.96	1.58
	12	19.34	89.52	51.23	1.28	1.55	13.1	6.96	5.8	6.82	0.42	0.7	3.73	0.85	1.29
	13	19.61	89.62	53.72	0.29	0.78	12.86	5.94	4.19	6.82	0.45	0.7	3.01	0.99	1.22
	14	19.18	90.06	50.34	1.32	1.76	15.4	7.91	7.33	7.02	0.56	0.72	5.08	1.25	2.14
	15	19.65	89.42	52.32	0.96	1.79	13.86	6.46	5.2	6.87	0.41	0.72	3.36	0.73	1.18
	Mean	19.39	89.64	50.58	0.99	1.50	14.94	7.83	6.48	6.77	0.52	0.69	3.85	0.96	1.48

DM, dry matter; GE, gross energy; CP, crude protein; EE, ether extract; AEE, acid hydrolysed ether extract; NDF, neutral detergent fiber; ADF, acid detergent fiber; CF, crude fiber.

<sup>1)</sup> Data adjusted from Li et al [2].

**Table 3.** Analyzed AA composition (%) of soybean meal (DM basis)<sup>1)</sup>

Source of soybean	No.	Indispensable AA										Dispensable AA							
		Arg	His	Leu	Ile	Lys	Met	Phe	Thr	Trp	Val	Ala	Asp	Cys	Glu	Gly	Pro	Ser	Tyr
China	1	3.74	1.39	3.67	2.12	3.22	0.64	2.36	1.94	0.62	2.30	2.23	5.64	0.68	8.25	2.08	2.46	2.54	1.94
	2	3.80	1.34	3.63	2.07	3.21	0.65	2.36	1.90	0.60	2.28	2.20	5.60	0.71	8.20	2.04	2.30	2.52	1.93
	3	3.79	1.35	3.51	2.04	3.19	0.63	2.29	1.91	0.59	2.27	2.23	5.61	0.69	8.16	2.06	2.22	2.55	1.84
	4	4.06	1.47	3.73	2.16	3.38	0.69	2.53	2.00	0.62	2.41	2.33	5.97	0.76	8.79	2.13	2.32	2.72	1.98
	5	4.06	1.63	4.07	2.41	3.45	0.70	2.71	2.40	0.67	2.61	2.47	6.28	0.76	8.37	2.33	2.60	2.60	1.89
	Mean	3.89	1.44	3.72	2.16	3.29	0.66	2.45	2.03	0.62	2.37	2.29	5.82	0.72	8.35	2.13	2.38	2.59	1.92
USA	6	3.55	1.36	3.79	2.16	3.21	0.65	2.39	1.96	0.61	2.41	2.26	5.59	0.66	8.18	2.11	2.18	2.50	1.80
	7	3.79	1.43	3.91	2.22	3.35	0.68	2.51	2.10	0.65	2.51	2.37	6.04	0.72	8.78	2.23	2.56	2.69	1.96
	8	3.80	1.45	3.74	2.13	3.31	0.66	2.46	1.97	0.62	2.37	2.27	5.73	0.70	8.48	2.07	2.36	2.64	2.03
	9	3.36	1.36	3.48	1.91	3.08	0.63	2.33	1.76	0.57	2.13	2.02	5.18	0.65	7.75	1.84	1.98	2.41	1.80
	10	3.64	1.50	3.86	2.33	3.22	0.69	2.49	2.25	0.64	2.47	2.31	5.92	0.71	7.89	2.28	2.56	2.47	1.70
	Mean	3.63	1.42	3.76	2.15	3.23	0.66	2.44	2.01	0.62	2.38	2.25	5.69	0.69	8.22	2.11	2.33	2.54	1.86
Brazil	11	3.46	1.37	3.65	2.05	3.06	0.61	2.36	1.82	0.57	2.23	2.13	5.39	0.64	7.95	2.01	1.89	2.47	1.78
	12	3.73	1.47	4.00	2.23	3.29	0.69	2.63	1.93	0.65	2.40	2.28	5.82	0.72	8.70	2.05	2.72	2.66	1.96
	13	3.90	1.53	4.16	2.35	3.47	0.69	2.76	2.00	0.65	2.50	2.35	6.07	0.71	9.15	2.12	2.43	2.78	2.03
	14	3.48	1.39	3.81	2.27	3.07	0.62	2.50	2.11	0.58	2.39	2.27	5.67	0.68	7.46	2.21	2.41	2.36	1.85
	15	3.67	1.47	3.91	2.39	3.26	0.63	2.59	2.24	0.64	2.53	2.37	6.03	0.64	7.95	2.29	2.43	2.49	1.78
	Mean	3.65	1.45	3.91	2.26	3.23	0.65	2.57	2.02	0.62	2.41	2.28	5.80	0.68	8.24	2.14	2.38	2.55	1.88

AA, amino acid; DM, dry matter.

<sup>1)</sup> Data adjusted from Li et al [2].

8:00 to 18:00 h. The procedures used to collect digesta followed the description provided by Stein et al [10]. Briefly, digesta was collected in plastic bags attached to simple T-cannula until the bags were full and immediately stored in a -20°C freezer after each collection. Ileal digesta samples were thawed, mixed within animal and diet, and a sub-sample was taken. Digesta samples were lyophilized in a Vacuum-Freeze Dryer (Tofflon Freezing

Drying Systems, Minhang District, Shanghai, China) and ground through a 1-mm screen for further chemical analysis.

### Chemical analysis and calculations

All analyses in the experiment were performed in duplicate and the chemical analyses were repeated if the difference between duplicates were over 5%. The samples of SBM and diets used in

**Table 4.** Ingredient and chemical composition of the experimental diets (as-fed basis)

Items	China					USA					Brazil					N-free diet
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	
Ingredients																
SBM	42.99	43.20	44.08	40.29	39.35	42.43	40.38	42.83	44.03	41.47	43.95	41.64	39.96	43.53	42.08	0.00
Corn starch	42.01	41.80	40.92	44.71	45.65	42.57	44.62	42.17	40.97	43.53	41.05	43.36	45.04	41.47	42.92	73.35
Sucrose	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00	10.00	15.00
Soybean oil	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	3.00
Limestone	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.50
Dicalcium phosphate	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	2.50
Sodium chloride	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.45
Chromic oxide	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Cellulose acetate	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	4.00
Potassium carbonate	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.30
Magnesium oxide	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.10
Mineral and vitamin premix <sup>1)</sup>	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Analyzed composition (%)																
DM	89.82	90.11	89.52	89.82	90.08	90.07	89.61	89.71	89.84	89.89	89.55	89.65	89.61	89.97	89.83	89.47
CP	19.75	18.99	19.05	19.30	18.60	19.35	19.06	18.81	18.59	19.04	19.20	19.71	19.81	19.51	19.57	0.49
EE	2.41	2.15	1.21	1.58	1.93	2.37	1.79	2.58	2.63	3.10	1.72	1.64	1.55	2.52	2.86	1.50
AEE	2.85	2.59	1.70	1.98	2.47	2.82	2.22	2.98	3.04	3.45	2.24	2.12	2.06	2.97	3.38	2.09
NDF	6.18	6.85	6.39	4.74	3.19	5.14	4.72	7.72	7.46	6.60	7.83	5.48	5.63	6.91	6.56	3.62
ADF	2.95	2.97	2.84	1.44	0.91	2.38	2.30	2.42	3.06	2.39	4.18	2.64	2.31	3.38	2.75	2.35

SBM, soybean meal; DM, dry matter; CP, crude protein; EE, ether extract; AEE, acid hydrolysed ether extract; NDF, neutral detergent fiber; ADF, acid detergent fiber.

<sup>1)</sup> Vitamin-mineral premix supplied the following per kg of diet: vitamin A, 5,512 IU; vitamin D<sub>3</sub>, 2,200 IU; vitamin E, 30 IU; vitamin K<sub>3</sub>, 2.2 mg; vitamin B<sub>12</sub>, 27.6 µg; riboflavin, 4 mg; pantothenic acid, 14 mg; niacin, 30 mg; choline chloride, 400 mg; folic acid, 0.7 mg; thiamine, 1.5 mg; pyridoxine, 3 mg; biotin, 44 µg; Mn (MnO), 40 mg; Fe (FeSO<sub>4</sub>·H<sub>2</sub>O), 75 mg; Zn (ZnO), 75 mg; Cu (CuSO<sub>4</sub>·5H<sub>2</sub>O), 100 mg; I (KI), 0.3 mg; Se (Na<sub>2</sub>SeO<sub>3</sub>), 0.3 mg.

**Table 5.** Analyzed AA composition (%) of the experimental diets (DM basis)

AA	China					USA					Brazil				
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Indispensable AA															
Arginine	1.79	1.75	1.75	1.80	1.68	1.69	1.90	1.77	1.61	1.58	1.56	1.63	1.61	1.60	1.66
Histidine	0.57	0.57	0.58	0.56	0.56	0.57	0.60	0.55	0.55	0.54	0.54	0.55	0.54	0.54	0.57
Leucine	1.58	1.59	1.60	1.59	1.55	1.60	1.61	1.51	1.54	1.53	1.53	1.61	1.57	1.56	1.65
Isoleucine	0.92	0.91	0.92	0.92	0.90	0.92	0.94	0.88	0.89	0.89	0.89	0.94	0.91	0.91	0.96
Lysine	1.32	1.34	1.34	1.32	1.28	1.33	1.35	1.26	1.29	1.26	1.25	1.28	1.25	1.26	1.34
Methionine	0.25	0.24	0.22	0.26	0.27	0.23	0.20	0.23	0.25	0.28	0.26	0.25	0.25	0.25	0.25
Phenylalanine	0.91	0.99	1.00	1.00	0.96	0.98	1.12	1.05	0.94	0.95	0.96	1.01	0.97	0.98	1.03
Threonine	0.75	0.75	0.76	0.75	0.75	0.76	0.82	0.77	0.73	0.73	0.72	0.75	0.73	0.73	0.77
Tryptophan	0.26	0.26	0.28	0.25	0.28	0.26	0.26	0.25	0.27	0.25	0.26	0.27	0.28	0.25	0.27
Valine	0.94	0.94	0.94	0.93	0.91	0.94	0.86	0.80	0.90	0.90	0.91	0.94	0.92	0.91	0.96
Dispensable AA															
Alanine	1.02	1.03	1.04	1.02	1.02	1.04	0.92	0.86	1.00	1.00	1.01	1.04	1.01	1.01	1.07
Aspartate	2.27	2.27	2.30	2.28	2.21	2.26	2.31	2.19	2.17	2.15	2.16	2.26	2.20	2.17	2.30
Cystine	0.35	0.35	0.39	0.36	0.38	0.35	0.34	0.35	0.34	0.34	0.33	0.35	0.36	0.34	0.32
Glutamine	3.81	3.84	3.88	3.87	3.72	3.84	3.69	3.50	3.66	3.64	3.65	3.83	3.74	3.66	3.87
Glycine	0.75	0.75	0.76	0.74	0.72	0.75	0.87	0.81	0.73	0.72	0.74	0.74	0.73	0.73	0.76
Proline	0.94	1.00	1.04	1.04	0.96	1.04	1.05	0.97	0.96	0.98	0.99	0.98	0.97	0.94	1.01
Serine	0.95	0.96	0.97	0.95	0.93	0.95	1.00	0.93	0.93	0.91	0.91	0.95	0.92	0.93	0.98
Tyrosine	0.47	0.49	0.47	0.51	0.44	0.51	0.59	0.53	0.45	0.46	0.49	0.52	0.47	0.49	0.50

AA, amino acid; DM, dry matter.

this experiment were analyzed for DM (AOAC 2007, Procedure 930.15), CP (AOAC 2007, Procedure 984.13), ash (AOAC 2007, Procedure 942.05), calcium (Ca; AOAC 2007, Procedure 927.02),

phosphorus (AOAC 2007, Procedure 984.27) [13], and EE (Thiex et al) [14]. Acid hydrolysed ether extract was determined by acid hydrolysis using 3 N HCl followed by crude fat extraction using

petroleum ether (AOAC 2007, Procedure 2003.06) on a Soxtec 2050 Automated Analyser (FOSS North America, Eden Prairie, MN, USA). Crude fiber, neutral detergent fiber, and acid detergent fiber were determined using filter bags and fiber analysis equipment (Fiber Analyser, Ankom Technology, Macedon, NY, USA) following a modification of the procedure of Van Soest et al [15]. The sucrose, raffinose and stachyose in the ingredients were analysed as described by Cervantes-Pahm and Stein [16]. The gross energy in ingredients were analysed using an isoperibol calorimeter (Parr 6400 Calorimeter, Moline, IL, USA) with benzoic acid as a standard.

Fifteen AA were determined after hydrolysis with 6 N HCl at 110°C for 24 h using an AA Analyser (Hitachi L-8900, Tokyo, Japan). Methionine and cysteine were determined as methionine sulfone and cysteic acid after cold performic acid oxidation overnight and samples were then hydrolysed with 7.5 N HCl at 110°C for 24 h using an AA Analyser (Hitachi L-8800, Japan). Tryptophan was determined after LiOH hydrolysis for 22 h at 110°C using high performance liquid chromatography (Agilent 1200 Series, Santa Clara, CA, USA).

Values for apparent ileal digestibility (AID) and SID of CP and each AA were determined according to the method of Stein et al [17] described previously.

$$\text{AID} = [1 - (\text{AA}_d / \text{AA}_f) (\text{Cr}_f / \text{Cr}_d)] \times 100\%$$

In which  $\text{AA}_d$  is the concentration of AA in the ileal digesta (g/kg of DM),  $\text{AA}_f$  is the concentration of AA in the diets (g/kg of DM),  $\text{Cr}_f$  represents chromium concentration in the diet (g/kg of DM), and  $\text{Cr}_d$  represents chromium concentration in the digesta (g/kg of DM). The AID for CP was also calculated using this equation where AA is replaced by the concentration of the CP in the digesta and diets.

The basal endogenous loss of each AA ( $\text{IAA}_{\text{end}}$ , g/kg of dry matter intake) at the distal ileum was determined based on the outflow obtained when pigs were fed the N-free diet using the equation of Stein et al [17]:

$$\text{IAA}_{\text{end}} = \text{AA}_d (\text{Cr}_f / \text{Cr}_d)$$

In which  $\text{AA}_d$  is the concentration of each AA in the ileal digesta collected from pigs fed the N-free diet. The endogenous outflow of CP was determined using the same equation where  $\text{AA}_d$  is replaced by the concentration of the CP in the digesta.

By correcting the AID of each AA that was calculated for each sample for the  $\text{IAA}_{\text{end}}$  of each AA, the SID of each AA was calculated using the equation of Stein et al [17]:

$$\text{SID} = \text{AID} + (\text{IAA}_{\text{end}} / \text{AA}_{\text{diet}}) \times 100\%$$

### Statistical analysis

All data were subjected to analysis of variance using the Proc

MIXED procedure of SAS (SAS Inst. Inc., Carry, NC, USA). The differences among within each country (China, USA, and Brazil) were analysed using source and period as a fixed effect, and pig within source as random effects. To compare the differences among the 3 countries, the country and period were fixed effects, and source within country and pig within country were random effects. The country means and source means were reported as least squares means calculated using the LSMEANS procedure. Means were tested using Protected least significant difference (LSD). In all analyses, the differences were considered significant if  $p < 0.05$  and considered a trend at  $p < 0.10$ .

Prediction equations were developed by the PROC REG procedure of SAS to estimate the AID and SID of CP and AA in the SBM [2,18]. The  $R^2$ ,  $C(p)$ , Akaike information criterion (AIC), root mean square error (RMSE), and p-value of the model were calculated to compare these different equations. The prediction equation with  $C(p)$  criterion closest to the number of predictors in the candidate model +1, the lowest AIC and the lowest RMSE were considered the optimal model.

## RESULTS

### Chemical composition of SBM sources

The chemical composition, AA and coefficient of variation of the SBM sources were discussed in our previous work (Tables 2, 3) [2]. Briefly, the average CP in SBM from China, USA, and Brazil was 50.14%, 49.22%, and 50.58%, respectively. The Chinese SBM contained the least raffinose among the countries ( $p < 0.01$ ). The Brazilian SBM contained the least sucrose ( $p < 0.01$ ) while the USA SBM contained the highest stachyose ( $p < 0.01$ ) (data from Li et al [2]).

### Digestibility of CP and AA

There was a trend ( $p < 0.10$ ) for the AID of lysine in sources of Chinese and USA SBM greater than in sources of Brazilian SBM (Table 6). Differences among the AID of CP and all AA except the methionine and cysteine in the 5 Chinese SBM were not observed ( $p > 0.10$ ). The AID of phenylalanine and tyrosine showed significant differences among the 5 USA SBM ( $p < 0.05$ ). There were no significant differences for AID of CP and all AA in Brazilian SBM ( $p > 0.10$ ).

The SID for valine and proline in SBM from China was greater than in SBM from Brazil ( $p < 0.05$ , Table 7). The SID of lysine, threonine, cysteine and glycine in SBM from China tended to be greater than in SBM from Brazil ( $p < 0.10$ ). The SID of cysteine showed significant differences among the 5 Chinese SBM ( $p = 0.02$ ). Differences among the SID of CP and all AA except the phenylalanine and tyrosine in the 5 sources of USA SBM were not observed ( $p > 0.10$ ). There were no significant differences for SID of CP and all AA in Brazilian SBM ( $p > 0.10$ ).

### Prediction equations for nitrogen and AA digestibility

**Table 6.** Apparent ileal digestibility (%) of CP and AA in soybean meals fed to growing pigs

Items	China (n = 5)				SA (n = 5)				Brazil (n = 5)				SEM	p-value <sup>2)</sup>
	Mean	Minimum	Maximum	p-value <sup>1)</sup>	Mean	Minimum	Maximum	p-value <sup>1)</sup>	Mean	Minimum	Maximum	p-value <sup>1)</sup>		
CP	80.45	77.29	82.30	0.14	80.87	78.77	82.66	0.28	79.45	78.69	80.09	0.93	0.98	0.59
Indispensable AA														
Arginine	93.45	92.82	94.14	0.89	93.36	92.93	93.86	0.97	92.43	92.24	92.75	1.00	0.43	0.22
Histidine	88.35	86.60	89.79	0.41	88.53	87.35	90.23	0.34	86.62	85.96	87.06	0.98	0.84	0.24
Leucine	85.43	83.63	86.98	0.40	85.09	83.75	87.23	0.21	83.56	82.45	84.27	0.75	0.88	0.31
Isoleucine	85.38	83.47	86.77	0.37	85.58	84.30	87.63	0.22	84.18	83.07	84.87	0.61	0.82	0.45
Lysine	86.91 <sup>x</sup>	85.14	88.30	0.22	87.49 <sup>y</sup>	86.17	89.56	0.10	84.85 <sup>z</sup>	84.13	85.52	0.87	0.80	0.09
Methionine	88.17	85.28	90.29	0.07	88.33	85.77	89.89	0.30	87.78	86.99	89.66	0.82	1.27	0.95
Phenylalanine	83.50	81.41	85.27	0.18	84.56	83.03	87.31	0.05	82.76	81.66	83.54	0.69	0.94	0.43
Threonine	78.03	76.68	80.16	0.59	78.26	76.23	81.94	0.14	75.91	74.94	76.67	0.91	0.96	0.20
Tryptophan	82.37	80.87	84.05	0.52	81.79	80.15	84.01	0.46	81.58	79.13	83.20	0.66	1.12	0.87
Valine	83.44	81.18	85.34	0.38	82.74	82.04	84.71	0.63	80.50	78.75	81.69	0.58	0.98	0.13
Dispensable AA														
Alanine	80.54	78.64	82.20	0.52	80.38	79.63	81.89	0.92	78.72	77.94	79.80	0.90	1.07	0.43
Aspartate	83.87	82.36	85.50	0.31	83.77	81.75	86.25	0.18	82.21	81.63	83.60	0.76	0.91	0.38
Cysteine	76.86	72.70	79.65	0.04	73.98	72.89	75.32	0.91	75.28	72.48	77.13	0.48	1.39	0.37
Glutamine	84.76	84.38	85.45	0.99	83.63	82.97	85.39	0.87	84.78	83.33	86.83	0.50	1.06	0.69
Glycine	73.04	70.93	76.13	0.71	73.39	69.68	79.42	0.11	69.48	67.53	71.07	0.87	1.92	0.32
Proline	84.66	82.41	86.37	0.41	83.52	81.43	86.01	0.31	82.77	81.29	83.99	0.62	1.06	0.47
Serine	83.09	80.54	84.95	0.16	82.73	81.01	85.58	0.15	81.32	80.28	82.03	0.82	0.95	0.41
Tyrosine	86.30	83.83	88.25	0.46	86.90	83.52	90.17	0.05	84.37	81.99	86.70	0.45	1.37	0.42

CP, crude protein; AA, amino acid; SEM, standard error of the means; SBM, soybean meal.

<sup>1)</sup> p-value, comparison of the mean of different source SBM.

<sup>2)</sup> p-value, comparison of each country SBM source.

<sup>xy</sup> Means in a row that do not have a common superscript letter tended to be different ( $p < 0.10$ ).

**Table 7.** Standardized ileal digestibility (%) of CP and AA in soybean meals fed to growing pigs

Items	China (n = 5)				USA (n = 5)				Brazil (n = 5)				SEM	p-value <sup>2)</sup>
	Mean	Minimum	Maximum	p-value <sup>1)</sup>	Mean	Minimum	Maximum	p-value <sup>1)</sup>	Mean	Minimum	Maximum	p-value <sup>1)</sup>		
CP	85.36	82.19	87.25	0.13	84.21	82.13	85.95	0.29	83.05	82.29	83.76	0.92	0.98	0.29
Indispensable AA														
Arginine	95.17	94.50	95.82	0.89	94.86	94.45	95.28	0.98	94.20	94.02	94.47	1.00	0.43	0.31
Histidine	90.60	88.80	92.03	0.40	89.95	88.75	91.56	0.37	88.21	87.57	88.69	0.98	0.83	0.16
Leucine	88.38	86.55	89.92	0.40	87.13	85.75	89.20	0.21	85.78	84.70	86.55	0.76	0.88	0.15
Isoleucine	88.41	86.49	89.79	0.37	87.65	86.32	89.63	0.23	86.47	85.38	87.23	0.62	0.81	0.27
Lysine	89.21 <sup>x</sup>	87.40	90.56	0.22	89.25 <sup>x</sup>	87.97	91.25	0.11	86.85 <sup>y</sup>	86.16	87.56	0.88	0.80	0.09
Methionine	90.78	88.24	92.74	0.11	89.57	87.01	90.94	0.31	89.58	88.81	91.50	0.81	1.25	0.74
Phenylalanine	86.81	84.93	88.52	0.22	87.26	85.83	89.74	0.08	85.45	84.38	86.32	0.70	0.91	0.38
Threonine	84.07 <sup>x</sup>	82.16	86.62	0.31	82.79 <sup>xy</sup>	79.12	85.96	0.21	80.46 <sup>y</sup>	79.48	81.86	0.91	1.09	0.09
Tryptophan	86.68	84.93	88.45	0.50	85.14	83.67	87.19	0.52	85.27	83.05	86.72	0.72	1.10	0.56
Valine	87.12 <sup>a</sup>	84.83	88.99	0.38	85.19 <sup>ab</sup>	84.43	87.22	0.57	83.30 <sup>b</sup>	81.62	84.54	0.60	0.99	0.04
Dispensable AA														
Alanine	85.27	83.31	86.92	0.51	83.83	83.05	85.50	0.91	82.54	81.83	83.69	0.90	1.07	0.24
Aspartate	86.50	84.95	88.11	0.31	85.58	83.61	87.98	0.19	84.24	83.59	85.69	0.73	0.91	0.25
Cysteine	83.99 <sup>x</sup>	79.43	86.53	0.02	79.31 <sup>y</sup>	78.06	80.65	0.90	81.14 <sup>y</sup>	78.64	82.71	0.59	1.39	0.09
Glutamine	86.83	86.42	87.51	0.99	85.11	84.45	86.86	0.88	86.48	84.97	88.57	0.48	1.06	0.50
Glycine	82.47 <sup>x</sup>	80.19	85.46	0.71	79.66 <sup>xy</sup>	76.29	84.98	0.19	75.93 <sup>y</sup>	73.79	77.50	0.85	1.86	0.08
Proline	94.50 <sup>a</sup>	92.81	96.12	0.58	86.75 <sup>b</sup>	84.80	89.08	0.35	88.43 <sup>b</sup>	87.17	89.59	0.69	1.02	<0.01
Serine	87.44	84.83	89.28	0.15	85.61	83.89	88.29	0.17	84.38	83.38	85.04	0.83	0.94	0.11
Tyrosine	88.91	86.48	90.78	0.51	88.66	85.50	91.67	0.07	86.33	83.95	88.67	0.45	1.34	0.36

CP, crude protein; AA, amino acid; SEM, standard error of the means; SBM, soybean meal; DM, dry matter.

<sup>1)</sup> p-value, comparison of the mean of different source SBM.

<sup>2)</sup> p-value, comparison of each country SBM source.

<sup>ab</sup> Means in a row that do not have a common superscript letter differ significantly ( $p < 0.05$ ).

<sup>xy</sup> Means in a row that do not have a common superscript letter tended to be different ( $p < 0.10$ ).

The basal ileal endogenous losses were determined (g/kg DM) as CP, 8.45; arginine, 0.28; histidine, 0.10; leucine, 0.38; isoleucine, 0.22; lysine, 0.26; methionine, 0.05; phenylalanine, 0.29; threonine, 0.39; tryptophan, 0.10; valine, 0.27; alanine, 0.40; aspartic acid, 0.48; cystine, 0.21; glutamic acid, 0.66; glycine, 0.53; proline, 0.62; serine, 0.32; tyrosine, 0.10.

Equations were developed to predict the AID and SID of nitrogen (N) and AA for SBM from their chemical characteristics and oligosaccharides (Table 8). The sucrose and raffinose content was the best predictor for AID and SID of N, lysine, threonine, and valine. The sucrose content had a positive correlation with AID and SID of N and AA, however, the raffinose content had a negative correlation with ileal digestibility of N and AA. The best fit equations for lysine in SBM were: AID lysine = 1.16 sucrose-1.81 raffinose+82.10 ( $R^2 = 0.69$ , RMSE = 1.00,  $p < 0.01$ ) and SID lysine = 1.14 sucrose-1.93 raffinose-0.99 EE+85.26 ( $R^2 = 0.77$ , RMSE = 0.86,  $p < 0.01$ ). However, we failed to develop the prediction equations for AID of methionine. The SID of methionine had a rather low  $R^2$  ( $R^2 = 0.40$ ). The other equations for AID and SID of nitrogen and tryptophan also had rather low  $R^2$  and higher AIC.

## DISCUSSION

Previous research indicated that the protein and AA content affected the AID and SID of N and AA [11,19-21]. In order to avoid this effect, the CP and AA content in all test diets in the current experiment were above the threshold level suggested by Fan et al [11].

The AID and SID of N and AA of the 15 sources of SBM were within the range of previously reported values [8,9,12,22,23]. In accordance with previously reported data [24-26], among the AID and SID values of all AA, arginine had the greatest value.

The addition of soapstock to SBM source 10 in USA may increase energy value of SBM [2], however, the ileal digestibility of AA in SBM source 10 was not significantly different from the other samples. Addition of soy hulls may reduce AA digestibility [27]. In the current experiment, the SBM sources 11, 12, and 13 were collected from the same crushing plant, but sources 11 had the greatest amount of hulls added during processing followed by sources 12, whereas sources 13 did not contain hulls. How-

ever, the AA digestibility was not significantly different as the amount of hulls added to the meal increased. These results indicate that, although addition of soapstock or removal of soy hulls may increase the energy value of SBM, it is not a primary factor affecting the ileal digestibility of AA.

Our previous work generated correlation coefficients between chemical composition and ileal digestibility of nitrogen and AA and subsequently established a series of AID or SID prediction equations for peanut meal [25], corn gluten meal [28], sunflower meal [29], and rapeseed meal [30]. Results of these previous studies indicate that correlations between chemical components of these plant protein ingredients and the ileal digestibility are different among plant protein ingredients. Therefore, it is necessary to establish specific ileal digestibility prediction equations for specific ingredients.

The effect of oligosaccharides on digestibility of AA remains controversial. Wiggins [31] noted that oligosaccharides may cause fluid retention and increase the digesta flow rate, leading to an adverse effect on the absorption and utilization of nutrients. Leske et al [32] also suggested that the raffinose family of oligosaccharides can cause shortened transit time, which led to a reduced fiber fermentation. However, the digestibility of AA in low raffinose and stachyose SBM was not different from that in conventional SBM [8,9]. In the current experiment, the sucrose content had a positive correlation with digestibility of AA, however, the raffinose content had a negative correlation with ileal digestibility of AA, which results support the conclusion that raffinose decreases the digestibility of AA. This can explain the results of the greater ileal digestibility of most AA in sources of Chinese SBM in connection with their lower raffinose content. The chemical composition and oligosaccharides of the SBM varied with the origin of the beans, which result in a different ileal digestibility of AA for the different origin of the soybeans, which are in agreement with our hypothesis.

**Table 8.** Stepwise regression equations for prediction of AA digestibility (%) based on the chemical composition (% of DM) of soybean meals fed to growing pigs

No.	Linear regression equations	R <sup>2</sup>	C(p) <sup>1)</sup>	AIC	RMSE	p-value
1	AID nitrogen = 0.74 sucrose+76.61	0.28	-	10.80	1.35	0.04
2	AID lysine = 1.16 sucrose-1.81 raffinose+82.10	0.69	-0.62	2.67	1.00	<0.01
3	AID threonine = 1.57 sucrose-1.16 ether extract+70.81	0.73	-3.50	6.51	1.14	<0.01
4	AID tryptophan = 18.73 tryptophan-3.66 ash+94.51	0.48	1.29	5.53	1.10	0.02
5	AID valine = 1.03 sucrose-3.63 raffinose+79.98	0.65	0.53	7.04	1.16	<0.01
6	SID nitrogen = 0.87 sucrose-1.53 ether extract+81.49	0.43	-	12.24	1.38	0.04
7	SID lysine = 1.14 sucrose-1.93 raffinose-0.99 ether extract+85.26	0.77	1.16	-1.11	0.86	<0.01
8	SID methionine = -0.45 CP-0.70 ADF+117.39	0.40	2.62	11.10	1.33	<0.01
9	SID threonine = 1.59 sucrose-3.67 raffinose+77.31	0.70	-1.98	11.64	1.35	<0.01
10	SID tryptophan = -1.40 ether extract-3.27 ash+108.77	0.59	-0.68	4.29	1.06	<0.01
11	SID valine = 1.04 sucrose-4.48 raffinose-1.42 ether extract+84.89	0.74	2.59	7.91	1.16	<0.01

AA, amino acid; DM, dry matter; AIC, Akaike information criterion, which measures the fit of the model and smaller AIC is a better fit of the model; RMSE, root mean square error is a measure of precision; AID, apparent ileal digestibility; CP, crude protein; ADF, acid detergent fiber; SID, standardised ileal digestibility.

<sup>1)</sup> C(p), Conceptual predictive statistic, the criterion used to determine candidate models that maximize explained variability ( $R^2$ ) with as few variables as possible. Candidate models are those where C(p) is close to the number of predictors in the candidate model+1.

In the current experiment, prediction equations for AID and SID of N and AA were developed from 15 SBM samples. However, considering the lack of additivity of value for AID [17] and the statistical criterion of R<sup>2</sup>, RMSE and AIC, prediction equations 7, 9, and 11 appear useful to evaluate the SID of lysine, threonine and valine in SBM. These equations should, however, be validated using a separate set of SBM samples.

## CONCLUSION

In summary, under the conditions of this experiment, the ileal digestibility of lysine, threonine, valine, cysteine, glycine and proline in SBM from China was greater than in SBM from Brazil. The oligosaccharides (such as sucrose and raffinose) can be used to accurately predict the AID and SID of AA in SBM.

## CONFLICT OF INTEREST

We certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

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