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Digestible indispensable amino acid score (DIAAS) and protein digestibility corrected amino acid score (PDCAAS) in oat protein concentrate measured in 20- to 30-kilogram pigs

Jerubella J Abelilla, Yanhong Liu and Hans H Stein*

Abstract

BACKGROUND: Oat protein concentrate is often used in human food, but the quality of this protein has not been characterized. Therefore, the objectives of this experiment were to determine the standardized ileal digestibility (SID) of crude protein (CP) and amino acids (AA) in oat protein concentrate and to determine differences in protein quality estimates between the protein digestibility-corrected AA score (PDCAAS) and the digestible indispensable AA score (DIAAS) when using growing pigs for both measurements.

RESULTS: For infants, the most limiting AA in oat protein concentrate was the aromatic AA (Phe + Tyr), for which the DIAAS value was 41 and the PDCAAS was 43. For children (6 months to 3 years) and children older than 3 years, the most limiting AA in oat protein concentrate was Lys, for which the DIAAS was 56 and 67 and the PDCAAS was 58 and 69, respectively.

CONCLUSION: The DIAAS value for oat protein concentrate was close to the calculated value for PDCAAS, but below the recommended intake for protein. Therefore, to satisfy the daily human AA requirement, oat protein needs to be complemented by other proteins of higher quality and specifically with greater lysine concentrations.

Keywords: amino acid digestibility; DIAAS; oat protein concentrate; pigs; PDCAAS

INTRODUCTION

Oats ranks sixth in cereal grain production in the world, with an annual global production of approximately 22.5 million metric tons.[1] More than 50% of the global production of oats is used as animal feed, but the remaining is used for human consumption.[2] For human consumption, oats are processed to generate rolled oat groats or oat flour, or used in functional or medicinal products.[2,3]

After harvest, the hulls are removed from the oat kernel to produce dehulled oats, which is also known as oat groats. Dehulled oats (uncooked) have a greater apparent ileal digestibility (AID) and standardized ileal digestibility (SID) of crude protein (CP) and most amino acids (AA) than most other cereal grains, and the highest digestible indispensable AA score (DIAAS) among all cereal grains.[4] However, instead of being consumed as oat groats, oats may also be processed and separated into oat bran, oat starch, oat fiber and oat protein concentrate.[3] Both wet milling and dry grinding procedures may be used in the processing of oats,[5–7] and the resulting oat protein concentrate typically contains 500–700 g kg^{-1} CP.[8,9]

For the last 25 years, the quality of proteins in human foods has been evaluated using the protein digestibility-corrected AA score (PDCAAS).[10] The PDCAAS system uses a digestibility value based on total tract digestibility of CP determined in rats and multiplies this value by the concentration of the first limiting AA, which is identified by comparing the AA profile of the test protein with the profile of the presumed requirement for AA for 2- to 5-year-old children.[10] Values for PDCAAS obtained in rats have been used extensively to evaluate human proteins and to determine if a specific protein meets the requirement for limiting indispensable AA for humans. However, a number of problems with PDCAAS values have been identified and inaccurate values are sometimes calculated using this procedure.[11–14] To overcome the limitations of the PDCAAS system, the Food and Agriculture Organization of the United Nations (FAO) now recommends that protein quality of human foods be evaluated using values for DIAAS. Unlike PDCAAS values, DIAAS values are calculated by multiplying the digestibility of each indispensable AA by the concentration of that AA in the protein and then comparing these values to a scoring pattern.[12] The FAO has also stated that in the absence of human data the pig is a better model than the rat to determine

* *Correspondence to: HH Stein, Department of Animal Sciences, University of Illinois, 1207 West Gregory Drive, Urbana, IL 61801, USA. E-mail: hstein@illinois.edu*

Department of Animal Sciences, University of Illinois, Urbana, IL 61801, USA

the ileal digestibility of AA in food proteins.[11] However, to our knowledge, there are no published data for DIAAS values of oat protein concentrate. The objective of this experiment, therefore, was to determine the difference in protein quality of oat protein concentrate estimated from PDCAAS or DIAAS if both values are obtained from digestibility data in 20- to 30-kg pigs rather than in rats.

EXPERIMENTAL

The protocol for the experiment was reviewed and approved by the Institutional Animal Care and Use Committee at the University of Illinois. Pigs used in the experiment were the offspring of L 359 boars that were mated to Camborough females (Pig Improvement Company, Hendersonville, TN, USA).

Ten growing barrows (initial body weight: 19.7 ± 2.3 kg) were equipped with a T-cannula in the distal ileum[15] and randomly allotted to a two-period crossover design with two diets and five replicate pigs per diet in each period. No pig received the same diet more than once during the experiment. Pigs were housed in individual pens (1.2 × 1.5 m) in an environmentally controlled room. Each pen was equipped with a feeder and a nipple drinker and had fully slatted tribar floors.

One source of oat protein concentrate was procured (Tate and Lyle Ingredients Americas LLC, Hoffman Estates, IL, USA) and was included in one diet as the only AA-containing ingredient (Table 1). A nitrogen-free diet that was used to measure basal endogenous losses of AA and protein was also formulated. Vitamins and minerals were included in both diets to meet or exceed current requirement estimates.[16] All diets also contained 4 g kg^{-1} titanium dioxide as an indigestible index and all diets were provided in a meal form. Pigs were fed their assigned diets in a daily amount that was calculated to be equal to three times the estimated energy requirement for maintenance (i.e. 824 kJ ME kg$^{-0.60}$).[16] Two equal meals were provided every day at 0800 and 1600 h. Water was available at all times.

Table 1. Ingredient composition (g kg^{-1}, as-fed basis) of experimental diets.

Ingredient	Oat protein diet	N-free diet
Oat protein concentrate	330.0	–
Sucrose	100.0	200.0
Corn starch	491.0	677.0
Ground limestone	4.0	7.0
Dicalcium phosphate	24.0	20.0
Choice white grease[a]	40.0	40.0
Solka floc[b]	–	40.0
Sodium chloride	4.0	4.0
Magnesium oxide	–	1.0
Potassium carbonate	–	4.0
Vitamin–mineral premix[c]	3.0	3.0
Titanium dioxide	4.0	4.0
Total	1000.0	1000.0

[a] Choice white grease is a source of fat.
[b] Fiber Sales and Development Corp., Urbana, OH, USA.
[c] Provided the following quantities of vitamins and micro minerals per kilogram of complete diet: vitamin A as retinyl acetate, 11 136 IU; vitamin D_3 as cholecalciferol, 2208 IU; vitamin E as DL-α-tocopheryl acetate, 66 IU; vitamin K as menadione dimethylprimidinol bisulfite, 1.42 mg; thiamine as thiamine mononitrate, 0.24 mg; riboflavin, 6.59 mg; pyridoxine as pyridoxine hydrochloride, 0.24 mg; vitamin B_{12}, 0.03 mg; D-pantothenic acid as D-calcium pantothenate, 23.5 mg; niacin, 44.1 mg; folic acid, 1.59 mg; biotin, 0.44 mg; Cu, 20 mg as copper sulfate and copper chloride; Fe, 126 mg as ferrous sulfate; I, 1.26 mg as ethylenediamine dihydriodide; Mn, 60.2 mg as manganese sulfate; Se, 0.3 mg as sodium selenite and selenium yeast; and Zn, 125.1 mg as zinc sulfate.

Data and sample collection

Pig weights were recorded at the beginning of each period and at the conclusion of the experiment. The amount of feed supplied each day was recorded. Each period lasted 9 days, with the initial 6 days as an adaptation period to the diet. Fecal samples were collected in the mornings of days 7 and 8 by anal stimulation and immediately frozen at −20 °C. Ileal digesta were collected for 8 h (from 0800 to 1600 h) on days 8 and 9 using standard operating procedures.[15] These procedures generally follow the procedures recommended by FAO,[17] with the exception that the test diet was formulated to contain approximately 200 g kg^{-1} CP, the daily feed provision was calculated as three times the energy required for maintenance and no antimicrobial agent was included in collection bags. On completion of the first experimental period, pigs were deprived of feed overnight and new experimental diets were offered the following morning.

At the conclusion of the experiment, ileal samples were thawed and all digesta collected over the 2 days were pooled within animal, and a subsample from each pig was lyophilized and ground. Fecal samples were dried at 65 °C in a forced-air oven and ground through a 1 mm screen in a Wiley mill (model 4; Thomas Scientific, Swedesboro, NJ, USA). The oat protein concentrate and all samples of diets, ileal digesta and feces were analyzed for DM (AOAC method 930.15)[18] and CP using the combustion procedure (AOAC method 90.03)[18] on an Elementar Rapid N-cube protein/nitrogen apparatus.[18] Diets and ileal digesta samples were analyzed for AA (AOAC method 982.30 E [a, b, c]).[18] Samples were also analyzed for titanium after wet-ash preparation based on the procedure by Myers *et al.*[19]

Values for AID and SID of AA and CP,[20] apparent total tract digestibility (ATTD) and standardized total tract digestibility (STTD) of CP,[14] and PDCAAS and DIAAS[10,11] were calculated. Data were tested for outliers and normal distribution using the UNIVARIATE procedure of SAS (SAS Institute Inc., Cary, NC, USA). Means were calculated using the LS Means procedure in SAS, with the pig being the experimental unit.

RESULTS AND DISCUSSION

The analyzed nutrient composition of the diet containing oat protein concentrate was close to the expected values (Table 2). The oat protein concentrate used in the present experiment contained 597.0 g kg^{-1} CP, 21.3 g kg^{-1} Lys, 10.6 g kg^{-1} Met, 18.7 g kg^{-1} Thr and 6.9 g kg^{-1} Trp.

Values for AID and SID of most AA in the oat protein concentrate (Table 3) are in agreement with values obtained for dehulled oats[4] and indicate that the majority of the non-hull proteins in oats were included in the oat protein concentrate. The basal ileal endogenous losses of CP and AA that were determined in this experiment are within the range of values previously determined in growing pigs.[21] The relatively high values for SID of the indispensable AA indicate that oat protein is well digested, as has been demonstrated in the past.[4]

Table 2. Analyzed nutrient composition (g kg^{-1}) of oat protein concentrate and experimental diets, as-fed basis.

Items	Oat protein concentrate	Oat protein diet	N-free diet
DM	958.2	934.5	929.8
CP	597	202.0	2.2
Indispensable amino acids			
Arg	41.1	13.3	0.1
His	13.5	4.6	0.0
Ile	24.1	8.6	0.2
Leu	48.9	16.7	0.3
Lys	21.3	7.5	0.2
Met	10.6	3.9	0.1
Phe	35.2	12.0	0.1
Thr	18.7	6.3	0.1
Trp	6.9	2.6	< 0.2
Val	34.0	11.3	0.1
Total	254.3	86.8	1.4
Dispensable amino acids			
Ala	25.5	8.9	0.2
Asp	43.2	15.1	0.2
Cys	12.4	4.3	0.1
Glu	126.6	45.3	0.5
Gly	23.7	8.4	0.1
Pro	31.7	10.6	0.3
Ser	22.9	8.0	0.1
Tyr	20.2	6.2	0.1
Total	306.2	106.8	1.6

Table 3. Apparent ileal digestibility (AID) and standardized ileal digestibility (SID) of CP and AA, and apparent total tract digestibility (ATTD) and standardized total tract digestibility (STTD) of CP in oat protein concentrate, as-fed basis[a].

Item	Oat protein concentrate			
	AID	SID[†]	ATTD	STTD[b]
CP, %	74.3 ± 3.3	83.7 ± 3.3	84.4 ± 1.9	89.5 ± 1.9
Indispensable, AA %				
Arg	85.9 ± 1.8	91.9 ± 1.8	–	–
His	75.9 ± 3.2	80.8 ± 3.2	–	–
Ile	78.1 ± 2.9	83.0 ± 2.9	–	–
Leu	81.0 ± 2.8	84.5 ± 2.8	–	–
Lys	79.2 ± 3.9	86.0 ± 3.9	–	–
Met	80.8 ± 2.2	83.2 ± 2.2	–	–
Phe	82.7 ± 2.2	85.8 ± 2.2	–	–
Thr	72.4 ± 5.2	82.4 ± 5.2	–	–
Trp	89.2 ± 3.8	95.0 ± 3.8	–	–
Val	77.6 ± 3.1	81.6 ± 3.1	–	–
Mean	80.4 ± 2.8	85.3 ± 2.8	–	–
Dispensable, AA %				
Ala	75.6 ± 3.4	82.9 ± 3.4	–	–
Asp	66.2 ± 3.6	72.0 ± 3.6	–	–
Cys	80.9 ± 4.0	86.0 ± 4.0	–	–
Glu	84.4 ± 1.7	86.7 ± 1.7	–	–
Gly	63.6 ± 8.1	87.8 ± 8.1	–	–
Ser	76.6 ± 3.5	83.7 ± 3.5	–	–
Tyr	80.9 ± 2.9	85.6 ± 2.9	–	–
Mean	72.4 ± 2.5	83.4 ± 2.5	–	–
Total AA	76.0 ± 2.6	84.2 ± 2.6	–	–

[a] Values are means ± standard deviation; $n = 10$.
[b] Values for SID were calculated by correcting the values for AID for basal ileal endogenous losses. Basal ileal endogenous losses were determined (g kg^{-1} DMI) as CP, 20.44; Arg, 0.85; His, 0.24; Ile, 0.45; Leu, 0.63; Lys, 0.55; Met, 0.10; Phe, 0.40; Thr, 0.68; Trp, 0.16; Val, 0.48; Ala, 0.69; Asp, 0.93; Cys, 0.24; Glu, 1.14; Gly, 2.18; Ser, 0.61; Tyr, 0.31; total indispensable AA, 4.53; total dispensable AA, 6.09; total AA, 10.62. Values for STTD were calculated by correcting the values for ATTD for basal total tract endogenous losses. Basal total tract endogenous loss of CP was 11.02 g kg^{-1} DMI.

The ATTD and STTD of CP in oat protein concentrate were 84.36% and 89.46%, respectively, whereas the AID and SID values were 74.29% and 83.74%, respectively. The reason for the greater values for ATTD and STTD of CP compared with AID and SID of CP is most likely that ammonia was absorbed from the hindgut of the pigs, which reduced the excretion of N in the feces. However, ammonia does not contribute to the protein status of pigs or humans[11] and absorbed ammonia will, therefore, be used in urea biosynthesis in the liver and excreted in the urine. The reason the difference between AID and SID of N is greater than the difference between ATTD and STTD of CP is that the total tract endogenous loss of N was much less than the ileal endogenous loss of N, which is also a consequence of hindgut absorption of N. In the PDCAAS calculation, the value for STTD of CP is used to calculate the digestibility of the first limiting AA, but because of the greater value for STTD of CP compared with the SID of most AA, the PDCAAS value likely overestimates the absorption of AA.

The PDCAAS value for oat protein concentrate based on the PDCAAS scoring pattern for a 2- to 5-year-old child was 57, with Lys being the first limiting AA (Table 4). However, if calculated based on the scoring patterns for DIAAS,[11] the PDCAAS values of oat protein concentrate for infants (birth to 6 months), children (6 months to 3 years), and older children, adolescents and adults was 43, 58, and 69, respectively (Table 5). The corresponding values for DIAAS were 41, 56, and 67, respectively. Values for DIAAS and PDCAAS for infants (birth to 6 months) were determined by the concentration of Phe + Tyr, whereas the DIAAS and PDCAAS values for children (6 months to 3 years) and older children, adolescents and adults were determined by the concentration of Lys. These observations indicate that the first limiting AA in oat protein concentrate consumed by individuals older than 6 months is Lys, which is in agreement with results of previous studies showing that Lys is the first limiting AA for children from 6 months to 3 years old in cooked rolled oats, uncooked dehulled oats and most other cereal grains.[4,13] The PDCAAS value of the oat protein concentrate used in this experiment was less than the values (61) that have been reported for cooked rolled oats if determined in rats.[12]

The DIAAS value for oat protein concentrate was greater than the reported DIAAS for cooked rolled oats (54),[12] but both of these values are less than in uncooked dehulled oats (77)[4] if the DIAAS value is calculated based on the scoring pattern for older children, adolescents and adults. This observation indicates that cooking and processing of oats may reduce digestibility of AA, resulting in a reduced DIAAS value. However, the value for cooked rolled oats was determined in rats, whereas the values for dehulled oats and oat protein concentrate were determined in pigs, which may also have contributed to the different values observed.

Regardless of the scoring pattern used, the DIAAS values for the oat protein concentrate used in this experiment did not satisfy the cut-off value (75) for DIAAS that has been suggested as

Table 4. Protein digestibility corrected amino acid score (PDCAAS) based on the PDCAAS scoring pattern (2- to 5-year-old children)[a].

Item	Protein digestibility corrected amino acid reference ratio									PDCAAS
	His	Ile	Leu	Lys	Sulfur amino acid	Phe + Tyr	Thr	Trp	Val	
2–5 years										
Reference scoring pattern, mg g^{-1} protein	19	28	66	58	25	63	34	11	35	–
PDCAAS, %	1.07	1.36	1.12	0.57	1.11	0.64	0.82	1.05	1.43	57 (Lys)

[a] The value for PDCAAS was calculated according to FAO.[10]

Table 5. Digestible indispensable amino acid score (DIAAS) and protein digestibility corrected amino acid score (PDCAAS) for oat protein concentrate[a].

Item	Digestible indispensable amino acid reference ratio									Amino acid score
	His	Ile	Leu	Lys	Sulfur amino acid	Phe + Tyr	Thr	Trp	Val	
Infants (birth to 6 months)[b]										
DIAAS, %	0.88	0.64	0.73	0.46	0.80	0.41	0.58	0.72	0.83	41 (Phe + Tyr)
PDCAAS, %	0.97	0.69	0.77	0.48	0.84	0.43	0.63	0.68	0.91	43 (Phe + Tyr)
Children (6 months to 3 years)[b]										
DIAAS, %	0.92	1.10	1.06	0.56	0.98	0.74	0.83	1.44	1.06	56 (Lys)
PDCAAS, %	1.02	1.19	1.12	0.58	1.03	0.78	0.90	1.35	1.16	58 (Lys)
Older children, adolescents, adults[b]										
DIAAS, %	1.15	1.18	1.15	0.67	1.15	0.94	1.03	1.85	1.14	67 (Lys)
PDCAAS, %	1.27	1.27	1.21	0.69	1.2	0.98	1.12	1.74	1.25	69 (Lys)

[a] Values for both DIAAS and PDCAAS were calculated using the scoring patterns recommended by FAO.[11]
[b] The reference scoring pattern (mg g^{-1} protein) for infants are: His, 21; Ile, 55; Leu, 96; Lys, 69; sulfur AA, 33; aromatic AA, 94; Thr, 44; Trp, 17; Val, 55.[10] The reference scoring pattern (mg g^{-1} protein) for children are: His, 20; Ile, 32; Leu, 66; Lys, 57; sulfur AA, 27; aromatic AA, 52; Thr, 31; Trp, 8.5; Val, 43.[10] The reference scoring pattern (mg g^{-1} protein) for older children, adolescents, and adults are: His, 16; Ile, 30; Leu, 61; Lys, 48; sulfur AA, 23; aromatic AA, 41; Thr, 25; Trp, 6.6; Val, 40.[11]

the minimum value for making claims for the protein quality of foods.[11] It is suggested that foods with a DIAAS value less than 75 should not make any claims related to protein quality.[11] This indicates that the oat protein concentrate used in this experiment will not satisfy the AA requirement of humans if consumed as the sole source of AA.[11] However, if oat protein concentrate is consumed in combination with other protein sources containing greater concentrations of Lys and other indispensable AA, a balanced meal that satisfies the AA requirement may be formulated.

The pig was used to calculate PDCAAS values in this experiment, although the rat is the recommended model for these calculations,[10] and it is possible that a different result would have been obtained if PDCAAS values had been determined in rats. But because it is recommended that DIAAS values be determined in pigs, and to avoid confounding results by using two different animal models, we decided to determine PDCAAS as well as DIAAS values in pigs. Nevertheless, the differences between PDCAAS and DIAAS that were calculated in this experiment, where both values were calculated in pigs, were relatively small if the same scoring patterns were used, which is in contrast to the results of an experiment in which PDCAAS and DIAAS were compared for high-quality proteins.[14] However, for all measurements and regardless of the scoring pattern used, values for PDCAAS were greater than values for DIAAS. This is a reflection of the elevated value for the STTD of CP compared with the SID of AA that is a result of hindgut absorption of ammonia, as discussed above. This observation is also in agreement with previous data indicating that for low-quality proteins PDCAAS will overvalue the quality,[12] whereas the opposite is the case for high-quality proteins.[14]

CONCLUSION

For infants (birth to 6 months), the most limiting AA in oat protein concentrate was Phe + Tyr, and the DIAAS was 41, whereas the PDCAAS was 43. For children (6 months to 3 years) and children older than 3 years, the most limiting AA in oat protein concentrate was Lys; the DIAAS was 56 and 67 and the PDCAAS was 58 and 69 for oat protein concentrate, respectively. Regardless of the scoring pattern used, the DIAAS values of the oat protein concentrate used in this experiment did not satisfy the minimum value that is recommended to make claims for protein quality of foods,[11] indicating that oat protein concentrate needs to be supplemented with proteins with greater concentrations of Lys such as soy protein or animal proteins, to fulfill the AA requirements of children.

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