Adequacy of linear equations to predict apparent available amino acid contents in compound diets and feed ingredients for fish

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SUMMARY

Predicted apparent available amino acid contents in compound diets and feed ingredients used in fish nutrition were evaluated for accuracy and precision against observed values from independent studies with the use of linear regression and mean prediction error techniques. Linear relationships between observed and predicted values, which were obtained with 21 to 43 compound diets in 13 studies with 6 fish species, showed that available contents of most amino acids can be predicted with high precision (R^2 above 0.80). Among essential amino acids in compound diets, mean prediction errors varied from 0.1 (arginine) to 0.19 (tryptophan), with most (>0.72) of the mean square prediction error attributed to a failure to predict the pattern of fluctuations across observed values (random bias). Prediction equations underestimated apparent available contents of individual essential amino acids in feed ingredients (n = 31 to 122, feed ingredients = 18, studies = 20, fish species = 16) with mean prediction errors mostly less than 0.14. However, R^2 between observed and predicted contents of essential amino acids were all above 0.94. This study concluded that previous determined linear regression equations can be used to predict apparent available contents of individual amino acids from dietary contents with high accuracy and precision, which can be utilise in effective feed formulation for fish species.

Keywords**:** amino acids, linear regression, mean prediction error analysis

INTRODUCTION

For feed formulations to be successful, the animal's dietary need for nutrients requires quantification according to digestible contents (Moughan, 2003). Faecal collection to determine digestibility coefficients is lengthy,

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laborious, tedious and prone to error. These have resulted in several attempts to predict digestible nutrient contents of diets and feed ingredients from its chemical composition in terrestrial animals (see Sales, 2008; 2009a).

In fish, where the aquatic environment further complicated faecal collection (Cho et al., 1982; Glencross et al., 2007), few studies have concentrated on prediction of digestible nutrient contents from chemical composition (Kirchgessner et al., 1986; Anderson et al., 1991; Sklan et al., 2004; Sales, 2008; 2009a; 2009b). Sales (2008) presented evidence that apparent digestible crude protein content and available contents of individual amino acids in fish diets and feed ingredients can be predicted from its dietary contents across a wide range of fish species, feed ingredients, feed types, nutrient levels, life stages and rearing conditions with the use of linear regression equations. This facilitates different digestibilities at variable nutrient contents, and eliminates the use of a constant value that does not account for endogenous losses. Dietary protein, and its amino acid components, have received priority in fish nutrition studies due to its impact on animal growth and high cost (Sales, 2008).

It is of utmost importance that models should be evaluated for adequacy before widely applied (Oldick et al., 1999). However, evaluation of the accuracy and precision of linear prediction equations presented by Sales (2008) with independent observed values was limited to crude protein. However, since new information and accessibility of literature have increased. The aim of the current study was to quantify the error associated with the use of the linear prediction equations established by Sales (2008) for prediction of apparent available contents of individual amino acids in compound diets and feed ingredients for fish.

Material and Methods

Description of data sets

Information were compiled on dietary contents and apparent availability of individual amino acids in compound diets obtained in 13 studies and in feed ingredients in 20 studies (Table 1). These studies were not included in Sales (2008) during the calculation of prediction equations. All fish species used to evaluate amino acid availability in compound diets (Table 1) were carnivorous, and rainbow trout occurred in 8 studies. Studies on feed ingredients included several omnivorous species (bluegill, channel catfish, Chinese sucker, Nile tilapia, pacu, rohu).

Table 1. Studies that presented dietary contents and apparent availability of individual amino acids in compound diets and feed ingredients for fish and have not been included in Sales (2008).

Reference	Fish species		n^a
	Common name	Scientific name	
Compound diets			
Aas et al. (2006)	Rainbow trout	*Oncorhynchus mykiss*	4
Bharadwaj et al. (2002)	Sunshine bass	*Morone chrysops* x *M. saxatilis*	8
Dabrowski and Dabrowska (1981)	Rainbow trout	*Oncorhynchus mykiss*	3
Dabrowski et al. (1980)	Rainbow trout	*Oncorhynchus mykiss*	3
Dabrowski et al. (1989)	Rainbow trout	*Oncorhynchus mykiss*	4
Mambrini et al. (1999)	Rainbow trout	*Oncorhynchus mykiss*	6
Nordrum et al. (2000)	Atlantic salmon	*Salmo salar*	1
Perera et al. (1995)	Rainbow trout	*Oncorhynchus mykiss*	2
Quartararo et al. (1998)	Red seabream	*Pagrus auratus*	2
Rawles et al. (2006)	Hybrid striped bass	*Morone saxatilis* x *M. chrysops*	4
Riche and Williams (2010)	Florida pompano	*Trachinotus carolinus*	2
Romarheim et al. (2006)	Rainbow trout	*Oncorhynchus mykiss*	3
Stone et al. (2008)	Rainbow trout	*Oncorhynchus mykiss*	1
Feed ingredients			
Abimorad et al. (2008)	Pacu	*Piaractus mesopotamicus*	6
Barrows et al. (2008)	Rainbow trout	*Oncorhynchus mykiss*	2
Borghesi et al. (2008)	Nile tilapia	*Oreochromis niloticus*	3
Borghesi et al. (2009)	Dourado	*Salminus brasiliensis*	4
Gaylord et al. (2010)	Rainbow trout	*Oncorhynchus mykiss*	24
Guimarães et al. (2008)	Nile tilapia	*Oreochromis niloticus*	8
Kitagima and Fracalossi (2011)	Channel catfish	*Ictalurus punctatus*	6
Lin et al. (2004)	Orange-spotted grouper	*Epinephelus coioides*	5
Liu et al. (2009)	Siberian sturgeon	*Acipenser baerii*	7
Luo et al. (2009)	Gobius	*Synechogobius hasta*	4
Masagounder et al. (2009)	Bluegill	*Lepomis macrochirus*	7
	Largemouth bass	*Micropterus salmoides*	4
Metts et al. (2011)	Sunshine bass	*Morone chrysops x M. saxatilis*	6
Noreen and Salim (2008)	Rohu	*Labeo rohita*	8
Rawles et al. (2010)	Sunshine bass	*Morone chrysops x M. saxatilis*	3

Riche and Williams (2010)	Florida pompano	*Trachinotus carolinus*	6
Skrede et al. (1998)	Atlantic salmon	*Salmo salar*	1
Stone et al. (2008)	Rainbow trout	*Oncorhynchus mykiss*	3
Yamamoto et al. (1997)	Rainbow trout	*Oncorhynchus mykiss*	4
Yamamoto et al. (1998)	Japanese flounder	*Paralichthys olivaceus*	4
Yuan et al. (2010)	Chinese sucker	*Myxocyprinus asiaticus*	7

[a] Number of diets or feed ingredients used from study.

Of the 43 compound diets used, 86% included fish meal as protein source, and 51% included soybean meal. All compound diets contained more than 1 protein source, and inclusion of diets from studies that concentrated on feed ingredient digestibility (Stone et al., 2008; Riche and Williams, 2010) in the compound diet data set was limited to reference diets, if containing practical ingredients. Faeces collection with compound diets was done by dissecting, settling, siphoning and stripping. Yttrium oxide was used as indigestible marker in 6 studies, chromic oxide in 6 studies, and 1 study (Bharadwaj et al., 2002) utilised barium carbonate. Amino acid contents were quantified in 8 studies with an amino acid analyzer, whereas 3 studies used high performance liquid chromatography, and 2 studies did not mentioned the method of analysis.

A wide range of feed ingredients were evaluated, some such as bacterial meal, barley, coconut meal, fish offal, flaxseed, rice concentrate, sunflower meal and wheat bran which were not included to establish prediction equations by Sales (2008). However, justification of inclusion of these feed ingredients in the current data set was based on the use of dietary and available amino acid contents as variables when prediction equations were computed (Sales, 2008), without accounting for individual feed ingredients. Studies on feed ingredient evaluation dominantly used stripping (9 studies) or settling (7 studies) to collect faeces, and chromic oxide (15 studies) as indigestible dietary marker. High performance liquid chromatography was used in 10 studies to analyze amino acids, an amino acid analyzer in 8 studies, and the method of analysis was not reported in 2 studies. Three studies (Yamamoto et al., 1997; 1998; Masagounder et al., 2009) used single protein source diets. Others used a 30:70 feed ingredient to test diet combination to calculate availability of amino acids in feed ingredients, mostly (11 studies) with the equation proposed by Forster (1999).

Calculations and statistical analysis

Apparent available contents of individual amino acids (*y*; g/kg dry weight) in diets and feed ingredients in the current studies were predicted from dietary contents (*x*; g/kg dry weight) with the use of linear prediction equations (Table 2) presented by Sales (2008). The accuracy and precision of predicted values

were evaluated by linear regression analysis with the PROC REG model procedure of SAS version 9.2 (SAS Institute Inc., Cary, NC). Predicted values are deterministic with no random variation (Tedeschi, 2006) and were plotted in the *x*-axis, with observed values in the *y*-axis. The coefficient of variation (R^2) was used to illustrate the portion of the total squared error that was explained by the model (precision), with the root mean square error (*RMSE*) included as a measure of the magnitude of variation. In addition, mean prediction error (*MPE*) analysis was conducted, with the mean square prediction error (*MSPE*) differentiated into the error in central tendency (mean bias), error due to regression (line bias) and error due to disturbance (random bias), as detailed in Sales (2010). All values were converted to dry weight if reported on a wet weight basis.

Table 2. Linear regression equations to predict apparent available amino acid content (y; g/kg dry weight) from dietary content (x; g/kg dry weight), as presented by Sales (2008).

Amino acid	Compound diets	Feed ingredients
Essential		
Arginine	-1.0659 + 0.9402*x*	-1.7172 + 0.9510*x*
Histidine	0.0517 + 0.8697*x*	1.2716 + 0.7759*x*
Isoleucine	0.6189 + 0.8403*x*	-1.6818 + 0.9411*x*
Leucine	3.8661 + 0.7654*x*	-1.5768 + 0.9185*x*
Lysine	-0.6491 + 0.9242*x*	-1.1048 + 0.9233*x*
Methionine	0.0032 + 0.8892*x*	-0.1499 + 0.8955*x*
Phenylalanine	1.2693 + 0.8151*x*	-1.0869 + 0.9183*x*
Threonine	0.1496 + 0.8524*x*	-1.0865 + 0.9107*x*
Valine	-1.1566 + 0.9218*x*	-0.9081 + 0.8959*x*
Tryptophan	0.0063 + 0.8232*x*	-0.2196 + 0.9309*x*
Non-essential		
Alanine	2.5724 + 0.7628*x*	-1.3896 + 0.9179*x*
Aspartic acid	1.7803 + 0.7713*x*	-1.9240 + 0.8954*x*
Cystine	-0.8569 + 0.9507*x*	-0.5092 + 0.8595*x*
Glutamic acid	8.8697 + 0.7682*x*	-6.0194 + 0.9670*x*
Glycine	-0.3851 + 0.8561*x*	-0.2266 + 0.8537*x*
Proline	-0.1532 + 0.8803*x*	0.9985 + 0.8449*x*
Serine	-0.2145 + 1.2017*x*	-0.7323 + 0.8957*x*
Tyrosine	-0.2805 + 0.8904*x*	-0.8161 + 0.9234*x*

RESULTS AND DISCUSSION

Compound diets

In the current plot format with predicted values indicated on the *x*-axis, points below and above the *y* = *x* line indicate over-and underestimation by the prediction equations, respectively (Tedeschi, 2006). Available contents of the

essential amino acids lysine and methonine, and the non-essential amino acids alanine, aspartic acid, cystine, glutamic acid and tyrosine presented intercepts and slopes for the linear relationship between observed and predicted values that differed (P<0.05) from 0 and 1, respectively (Table 3). Non-significance of intercepts from 0 and slopes from 1 might indicate that equations lacked accuracy through an inability to predict the correct values. However, the aforementioned amino acids presented a high degree of precision of the prediction equations. This was illustrated by R^2 of above 0.9200, which indicated an ability of the equations to predict similar values constantly (Tedeschi, 2006).

Table 3. Intercepts and slopes (± standard error) from linear regression analysis between observed (y) and predicted (x) apparent available amino acid contents (g/kg dry weight) in compound fish diets.

Amino acid	n[a]	Intercept	Slope	R^2	$RMSE$[b]
Essential					
Arginine	43	1.9867 ± 1.3816	0.9241 ± 0.0545	0.8754	2.3588
Histidine	43	-0.5079 ± 0.5697	1.0709 ± 0.0659	0.8657	0.8385
Isoleucine	43	-2.1964 ± 1.2741	1.1388 ± 0.0847	0.8150	1.7525
Leucine	43	-4.9764 ± 2.5020	1.1608 ± 0.0902	0.8014	3.1919
Lysine	43	-3.0755 ± 1.2738[c]	1.1285 ± 0.0513[c]	0.9219	2.2246
Methionine	42	-1.2428 ± 0.3492[c]	1.1407 ± 0.0358[c]	0.9622	0.8939
Phenylalanine	43	0.0531 ± 1.4412	0.9494 ± 0.0909	0.7271	1.9682
Threonine	43	1.9930 ± 1.1873	0.8078 ± 0.0837[c]	0.6946	2.2886
Valine	43	1.3088 ± 1.1872	0.9221 ± 0.0667	0.8236	2.2445
Tryptophan	21	0.0212 ± 0.4741	0.9709 ± 0.1201	0.7748	0.7364
Non-essential					
Alanine	23	-3.1882 ± 1.1433[c]	1.1717 ± 0.0530[c]	0.9588	1.4219
Aspartic acid	21	-9.7252 ± 1.6316[c]	1.3974 ± 0.0578[c]	0.9686	1.8560
Cystine	23	0.7317 ± 0.1798[c]	0.8824 ± 0.0315[c]	0.9739	0.5010
Glutamic acid	21	-18.4048 ± 1.9033[c]	1.3340 ± 0.0350[c]	0.9871	2.3875
Glycine	23	-0.2202 ± 1.4528	1.0180 ± 0.0728	0.9031	1.7752
Proline	23	-1.1016 ± 0.9752	1.0510 ± 0.0501	0.9545	1.5649
Serine	23	-0.2497 ± 0.6340	0.7310 ± 0.0284[c]	0.9693	1.3157
Tyrosine	23	-2.1715 ± 0.7315[c]	1.1928 ± 0.0638[c]	0.9434	1.0708

[a] Number of values.
[b] *RMSE*, root mean square error.
[c] Different (P<0.05) from 0 for intercept and 1 for slope.

Ambiguity of null hypothesis tests was stated by Mitchell (1997) as an inability of linear regression analysis to evaluate the adequacy of models. Limited dispersion of points will result in small standard errors and high computed values for the test statistics for intercept and slope, which in turn will cause values that are likely to be significant from 0 and 1, respectively. In

contrast, with points that are scattered, falsification of the null hypothesis might fail, either because the intercept or slope is really not different from 0 or 1, respectively, or there is too much dispersion of points around the line.

Table 4. Mean prediction errors (MPE) and components of the mean square prediction error (MSPE) between observed and predicted apparent available amino acid contents (g/kg dry weight) in compound fish diets.

Amino acid	n[a]	$\sqrt{MSPE}$	*MPE*	Bias[b]	Proportion of *MSPE*		
					Mean bias	Line bias	Random bias
Essential							
Arginine	43	2.3607	0.0959	-0.1264	0.0029	0.0452	0.9520
Histidine	43	0.8350	0.0980	-0.0892	0.0079	0.0271	0.9615
Isoleucine	43	1.7732	0.1219	0.1560	0.0077	0.0609	0.9313
Leucine	43	3.2909	0.1238	0.6027	0.0335	0.0695	0.8970
Lysine	43	2.3326	0.0974	-0.0008	0.0000	0.1328	0.8672
Methionine	42	1.0275	0.1143	-0.0194	0.0004	0.2789	0.7207
Phenylalanine	43	2.0635	0.1396	0.7324	0.1260	0.0066	0.8675
Threonine	43	2.4523	0.1893	0.6140	0.0627	0.1069	0.8304
Valine	43	2.2279	0.1308	0.0194	0.0001	0.0322	0.9677
Tryptophan	21	0.7069	0.1949	0.0868	0.0151	0.0030	0.9819
Non-essential							
Alanine	23	1.7088	0.0805	-0.3890	0.0518	0.3160	0.6322
Aspartic acid	21	3.4930	0.1225	-1.1488	0.1082	0.6364	0.2554
Cystine	23	0.6447	0.1335	-0.1859	0.0831	0.3655	0.5514
Glutamic acid	21	5.5447	0.1080	0.9431	0.0289	0.8033	0.1678
Glycine	23	1.7035	0.0876	-0.1272	0.0056	0.0029	0.9915
Proline	23	1.5407	0.0847	0.1663	0.0117	0.0465	0.9419
Serine	23	6.3563	0.4396	5.6632	0.7938	0.1671	0.0391
Tyrosine	23	1.2275	0.1131	0.0659	0.0043	0.3023	0.6948

[a] Number of values used.
[b] Predicted - observed.

Mean prediction error analysis is frequently used in animal nutrition to evaluate the origin (mean bias, linear bias, random bias) of deviations of model predicted values from observed values (e.g. Benchaar et al., 1998; Oldick et al., 1999; Halas et al., 2004; Hirooka et al., 2007; Peripolli et al., 2011; Wang et al., 2011). A drawback of *MPE* analysis is that it does not provide any information on precision of the prediction equations (Mitchell and Sheeby, 1997). In the current evaluation prediction equations underestimated available contents of 8 amino acids, with overestimations obtained with other amino acids (Table 4). According to the root of the mean square prediction error (*RMSPE*), which can be expressed in the same units as the output (Theil, 1966), under-or overpredictions varied from 0.71 (tryptophan) to 3.30 (leucine) g/kg dry weight

for the individual essensial amino acids. When the *RMSPE* was expressed as a fraction of the observed mean to illustrate the *MPE* (Theil, 1966), a value of 0.11 indicated that the prediction equation for glutamic acid overestimated its available contents with more than 5.50 g/kg dry weight. However, an overestimation of 6.36 g/kg dry weight for available serine contents represented an *MPE* as high as 0.44

Random bias was responsible for the major proportion (>0.72) of the *MSPE* with essential amino acids (Table 4). Random bias presented the proportion of the *MSPE* unrelated to the errors of the prediction (Halas et al., 2004) and cannot be eliminated by linear corrections of the predictions (Theil, 1966). With serine, a high proportion (0.79) of mean bias indicated a consistent overprediction of values. This also occurred to a lesser extent (mean bias of above 0.10) with phenylalanine and aspartic acid. With these amino acids a portion of the error could thus be eliminated by a correction factor. Linear bias that presented more than 0.27 of the *MSPE* with the essential amino acid methionine and non-essential amino acids alanine, aspartic acid, cystine, glutamic acid and tyrosine showed that high proportions of the *MSPE* resulted from proportional bias due to inadequate presentation of the relationships involved (Benchaar et al., 1998). This implies that slopes of the relationships between observed and predicted values differed from 1 with these amino acids (Hirooka et al., 2007).

As accentuated by Sales (2008; 2009a), extrapolation outside ranges used for development is not recommended with empirical linear prediction models. From 3 (tryptophan, proline, tyrosine) to 15 (threonine) values in the current studies were outside the ranges of dietary contents and availability used by Sales (2008) to calculate the respective prediction equations. Elimination of these values increased the accuracy of prediction equations by changing slopes and intercepts from significant to non-significant from 0 and 1, respectively, with the non-essential amino acids alanine, cystine, glutamic acid and tyrosine (data not shown). However, the precision (R^2) with cystine and serine was decreased with 0.19 and 0.17 units, respectively, which could probably be related to a more narrow range of values created by the elimination process. With threonine, the slope changed to non-significant from 1, the R^2 increased with 0.10 units and the *RMSE* decreased with 0.77 units. Omission of values outside ranges lowered the *MPE* to less than 0.10, with a corresponding decrease in the *RMSPE* of more than 0.50 g/kg dry weight, with leucine, phenylalanine, valine, aspartic acid and glutamic acid (data not shown). With cystine, glutamic acid and tyrosine the major proportion (>0.72) of the *MSPE* after elimination of values was found in random bias, whereas more than 0.20 of the *MSPE* could be attributed to mean bias with histidine, alanine and cystine. However, based on the lack of an increase in accuracy of prediction

equations for especially essential amino acids as evaluated by both linear regression and prediction error analysis, linearity outside ranges used to establish prediction equations for available contents of individual amino acids was assumed.

Feed ingredients

Prediction equations for the available contents of the essential amino acids arginine, isoleucine, phenylalanine, threonine and valine, and all non-essential amino acids with the exception of cystine, glutamic acid and tyrosine, presented a high degree of accuracy, as indicated by non-significant intercepts and slopes from 0 and 1, respectively (Table 5). Furthermore, R^2 of above 0.94 showed high precision of predictions with essential amino acids.

Table 5. Intercepts and slopes (± standard error) from linear regression analysis between observed (y) and predicted (x) apparent available amino acid contents (g/kg dry weight) in feed ingredients.

Amino acid	n[a]	Intercept	Slope	R^2	*RMSE*[b]
Essential					
Arginine	122	0.3119 ± 0.4336	1.0129 ± 0.0121	0.9831	2.0989
Histidine	119	-1.7976 ± 0.2109[c]	1.1754 ± 0.0169[c]	0.9764	1.1471
Isoleucine	122	0.3245 ± 0.3887	1.0095 ± 0.0193	0.9581	1.9278
Leucine	122	2.4864 ± 0.0973[c]	0.9520 ± 0.0213[c]	0.9436	5.4471
Lysine	119	-0.4247 ± 0.3042	1.0425 ± 0.0102[c]	0.9889	1.8102
Methionine	113	-0.3234 ± 0.1761	1.0652 ± 0.0165[c]	0.9742	1.0193
Phenylalanine	122	0.9007 ± 0.5174	0.9799 ± 0.0210	0.9476	2.5415
Threonine	114	0.0564 ± 0.3783	1.0268 ± 0.0191	0.9626	1.7414
Valine	122	0.0646 ± 0.5714	1.0010 ± 0.0217	0.9466	2.9891
Tryptophan	31	0.6326 ± 0.0975[c]	1.1351 ± 0.0302[c]	0.9799	0.2725
Non-essential					
Alanine	88	-0.0287 ± 0.8703	0.9946 ± 0.0254	0.9469	3.6364
Aspartic acid	82	-0.6796 ± 1.3677	1.0358 ± 0.0325	0.9269	4.5432
Cystine	80	0.4710 ± 0.1934[c]	0.9156 ± 0.0208[c]	0.9614	1.2593
Glutamic acid	82	8.6061 ± 3.1138[c]	0.8736 ± 0.0400[c]	0.8563	9.4386
Glycine	88	0.4322 ± 2.1386	0.9759 ± 0.0605	0.7514	10.4471
Proline	88	-0.2677 ± 0.8626	0.9908 ± 0.0275	0.9380	3.7590
Serine	88	0.0919 ± 0.5414	1.0152 ± 0.0194	0.9697	2.7117
Tyrosine	114	0.9894 ± 0.3986[c]	0.9561 ± 0.0233	0.9378	2.0321

[a] Number of values.
[b] *RMSE* = root mean square error.
[c] Different (P<0.05) from 0 for intercept and 1 for slope.

Equations underpredicted observed available contents of all essential amino acids (Table 6). Available contents of arginine, lysine and threonine were underpredicted with less than 10% of the observed mean (*MPE*). The

prediction equation for tryptophan presented a *MPE* of 0.28, which could be translated to an underestimation of 1.07 g/kg dry weight. More than 0.78 of the *MPSE* in all amino acids but histidine and tryptophan could be attributed to random bias. Line bias was evident (0.48) in the prediction of available histidine content, and mean bias was dominant (0.90) with tryptophan.

Table 6. Mean prediction errors (MPE) and components of the mean square prediction error (MSPE) between observed and predicted apparent available amino acid contents (g/kg dry weight) in feed ingredients.

Amino acid	n[a]	$\sqrt{MSPE}$	*MPE*	Bias[b]	Proportion of *MSPE*		
					Mean bias	Line bias	Random bias
Essential							
Arginine	122	2.2143	0.0673	-0.7275	0.1079	0.0084	0.8837
Histidine	119	1.5809	0.1446	-0.1026	0.0042	0.4781	0.5177
Isoleucine	122	1.9771	0.1068	-0.4961	0.0630	0.0019	0.9351
Leucine	122	5.5492	0.1396	-0.6061	0.0119	0.0403	0.9477
Lysine	119	2.0266	0.0790	-0.6379	0.0991	0.1165	0.7844
Methionine	113	1.1108	0.1202	-0.2622	0.0557	0.1171	0.8272
Phenylalanine	122	2.5713	0.1143	-0.4581	0.0317	0.0073	0.9610
Threonine	114	1.8213	0.0991	-0.5344	0.0861	0.0157	0.8982
Valine	122	2.9659	0.1275	-0.0878	0.0009	0.0000	0.9991
Tryptophan	31	1.0671	0.2803	-1.0105	0.8968	0.0422	0.0610
Non-essential							
Alanine	88	3.6010	0.1181	0.1940	0.0029	0.0005	0.9966
Aspartic acid	82	4.5785	0.1149	-0.7213	0.0248	0.0146	0.9606
Cystine	80	1.3704	0.2170	0.0678	0.0024	0.1742	0.8233
Glutamic acid	82	9.9098	0.1364	0.6653	0.0045	0.1105	0.8850
Glycine	88	10.3415	0.3462	0.2958	0.0008	0.0018	0.9973
Proline	88	3.7553	0.1377	0.5245	0.0195	0.0013	0.9792
Serine	88	2.7278	0.1132	-0.4509	0.0273	0.0069	0.9658
Tyrosine	114	2.0722	0.1347	-0.3285	0.0251	0.0300	0.9448

[a] Number of values used.
[b] Predicted - observed.

In contrast to the data set on compound diets, few values with feed ingredients were outside the ranges used by Sales (2008) to compute prediction equations. Elimination of the contents of available leucine (observed dietary content of 114.71 g/kg dry weight and availability of 44.5%) and glutamic acid (observed dietary content of 147.87 g/kg dry weight and availability of 43.6%) reported for wheat gluten by Yamamoto et al. (1998), changed the intercepts and slopes of the linear relationship between observed and predicted values for these 2 amino acids to non-significant from 0 and 1, respectively. Furthermore, it decreased the *MPE* to 0.07 and 0.08 for leucine

and glutamic acid, respectively. Omission of available glycine content from poultry byproduct meal with an availability of -50% (Rawles et al., 2010) decreased the *MPE* from 0.35 to 0.11, and the *RMSPE* from 10.34 to 3.48 g/kg dry weight. Although it increased the R^2 to 0.97, it changed the slope of the linear relationship between observed and predicted values to significant from 1.

Conclusions

It should be accentuated that the present study did not investigate the effect of factors such as fish species, water type, water temperature, feed habit, fish size, or feed ingredients on apparent amino acid availability, but evaluated the accuracy and precision of previous determined linear equations to predict contents of available amino acids. Linear regression and mean prediction error analysis presented evidence that established linear prediction equations can be used to predict the apparent available contents of individual amino acids from its dietary contents in compound diets and feed ingredients with a high degree of accuracy and precision. This would be possible for a wide range of fish species, which are reared under different dietary, environmental and physiological conditions, and holds promise for feed formulators to create effective fish diets without the need to conduct lengthy, expensive, and tedious digestibility experiments.

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