EQUATIONS FOR PREDICTING PER FROM AMINO ACID ANALYSIS--A REVIEW AND CURRENT SCOPE OF APPLICATION

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The development and application of three equations for predicting protein efficiency ratio (PER) of meat and meat products from their amino acid analysis (Alsmeyer et al., 1974) indicated that equation 3 is a reliable estimator of PER when it is used with products that contain proteins primarily of meat, poultry, grain or yeast origin. New data on meat and on soy products and on combinations of meat, collagen and soy or whey protein concentrate were used to test the three predicting equations. The results indicate that equations 1 and 2 with a few exceptions overestimated PER for soy products. Equation 3 generally underestimates the PER determined by bioassay of all the soy products, except for combinations containing more than 60 percent lean beef.

Results reported suggest that variations in amino acid patterns and digestibility of protein food products may necessitate developing new equations or modifying present ones to fit specific types of food proteins. A discussion of the extended use of the prediction equations is included.

INTRODUCTION

Recently, a new, relatively rapid method was developed for predicting the protein efficiency ratio (PER) of meat and meat products from their amino acid analysis (Alsmeyer et al., 1974). This new method is relatively inexpensive, can be completed in 24-48 hours, and is a reliable estimator of PER when used with products that contain proteins primarily of meat, poultry, grain or yeast origin. It predicts PER within ±0.2 of the value obtained by the AOAC bioassay reference method. This variation is within the experimental error of the reference method.

There is widespread interest in the new PER method for use in implementing quality and regulatory control. The government requires that the protein quality of food products be measured when food protein ingredients in product formulations are replaced or extended with new and/or more economical food protein and when voluntary nutritional labeling is practiced. These requirements, along with increased consumer consciousness of nutritional quality, have generated a large number of samples that require determination of protein nutritive quality. Therefore, there is a need for extending the use of these equations to a variety of other protein products. This paper will present a brief review of the new method, information on its present use, and possibilities for extending it to other protein food products.

REVIEW OF THE NEW METHOD

The most widely used method for determining protein nutritional quality and the only method that has been accepted by the Association of Official Agricultural Chemists (AOAC) is the protein efficiency ratio (PER) test, a biological analysis (AOAC, 1970). This test measures protein nutritional quality by feeding weanling rats a diet containing 10% protein supplied by the test protein for 28 days and measuring their weight gain. The PER is weight gain divided by protein intake. The cost and elapsed time to obtain nutritional data make this method impractical for regulatory and quality control.

Amino acid composition and the PER value for lean beaf, partially defatted (PD) chopped beef, PD beef fatty tissue, and collagen (Happich et al., 1975) determined in research at the Eastern Research Center, USDA, were used by Animal and Plant Health Inspection Service, USDA, to derive three regression equations for predicting PER from amino acid analyses (Alsmeyer et al., 1974).

One multiple regression equation was as follows:

$$PER = -0.684 + 0.456\ (LEU) - 0.047\ (PRO) \qquad (1)$$

A second equation was developed by stepwise regression with backward elimination.

$$PER = -0.468 + 0.454\ (LEU) - 0.105\ (TYR) \qquad (2)$$

A third equation was obtained using a straightforward stepwise regression.

$$PER = -1.816 + 0.435\ (MET) + 0.780\ (LEU) + 0.211\ (HIS) - 0.944\ (TYR) \qquad (3)$$

The regression equation can only be assumed to be linear over the range covered by the values of the variables used in developing the regressions. The ranges in the study were:

Histidine (HIS)	0.8 - 3.8%
Leucine (LEU)	3.8 - 9.3%
Methionine (MET)	0.7 - 2.7%
Tyrosine (TYR)	0.9 - 4.8%
Proline (PRO)	3.6 -13.8%

The amino acid values used in the equations are grams of amino acid residue per 100 grams of total amino acid residues. Therefore, knowledge of the complete amino acid composition of a product, including tryptophan, is a prerequisite for using the equations.

The food industry supplied amino acid composition and PERs for a variety of food products which were used to test the equations. Results from testing the equations indicated that equation 3 predicted the PER (±0.2) of 66 of the 93 food products tested and is a reliable estimator of PER when applied to products that contain proteins primarily of meat, poultry, grain or yeast origin.

Equations 1 and 2 did not predict PER of food containing little or no meat or poultry. All three equations failed to predict PER with accuracy for products containing fish or beans. The reader is referred to the original paper for the variety of food products used for testing the equations.

TESTING EQUATIONS WITH NEW DATA

Widespread interest in the equations has stimulated investigators to test them on a variety of products. Those that have come to our attention include meat food products, horse meat, partially defatted cooked beef fatty tissue residues from rendering at approximately 170°F, and cottonseed and soy protein products. We have tested the equations on a sample of lean beef (#2) and of a soy protein concentrate, and on samples of four textured vegetable protein products, three textured vegetable protein seasoning mix products, and three meat patty products containing textured vegetable protein using amino acid values determined by analysis of the actual products (Happich et al., 1975; Happich, this volume). We have also tested the equations on combinations of lean beef-soy protein, lean beef-collagen protein, and lean beef-collagen-soy or whey protein, using amino acid values calculated for the ten various combinations from the determined amino acid composition of each ingredient used in the combination. Amino acid, digestibility, and PER data on all these samples are found in Table 1.

The data indicate that equation 3 underestimates substantially (0.6-1.2) the PER determined by bioassay of five of the eight soy products and of two others to a lesser extent (0.3 PER) and overestimates the PER for one product by 0.5. It also underestimates the PER of meat patties with textured vegetable protein and of lean beef, collagen and soy combinations by 0.4 to 0.8. Three combinations of soy protein concentrate with 60% or more lean beef fall within +0.2 of the PER determined by bioassay for two of the combinations and +0.3 for the third.

Methionine content is low and methionine is the limiting amino acid in the soy products. Methionine sulfoxide was found in the sample of lean beef (#2), soy protein concentrate, meat patties with textured vegetable protein, textured vegetable protein, and textured vegetable protein seasoning mix products. It was calculated to the equivalent methionine, totaled with the methionine, and this total used in equation 3. In all cases this gave a higher estimated PER and in nearly half of the soy products and soy combinations with meat (7 out of 16) a PER that agreed within ±0.2 and three others within ±0.3 of the PER determined by bioassay.

Equations 1 and 2 tend to overestimate PER for soy products, except for the PER of the soy protein concentrate, individually or in combinations with lean beef and collagen, which agree within ±0.2 of the bioassay. The overestimation for one combination of soy protein concentrate and lean beef and for meat patties containing textured vegetable protein in Table 1 is no greater than 0.3, and in one instance, 0.4 PER. For the seven samples of textured vegetable protein and textured vegetable protein seasoning mix, the overestimation is high (0.4-1.1 PER).

Because "the use of dietary protein by the body for growth and maintenance is dependent on the presence and relative amounts of the essential amino acids and on the digestibility" (Oser, 1959), it may be necessary to use a digestibility factor to extend the use of these equations to products having a nitrogen digestibility much below 90%. The digestibility of the products whose data was used to derive the equations was 90% or above. The nitrogen digestibility of samples of the textured vegetable protein products listed in Table 1 averages about 82%. When this is taken into account and the resulting lower amino acid values are used in equations 1 and 2, the PER estimated by equations 1 and 2 agree within +0.2 PER for 5 and 4, respectively, of the 7 samples. However, more data are needed to adequately test these two equations for use with soy products.

A whey protein concentrate used in our studies has a different amino acid pattern than the products used to derive the equations. It has a higher leucine content (leucine is a key amino acid used in

TABLE 1

Amino Acid and PER Data on Lean Beef, Soy, Whey, and Collagen Proteins and Their Combinations

Food product or protein mixture	Amino acid content (grams of amino acid residue per 100 grams of total amino acid residues)					
	His	Leu	Met	Met & Met sulfoxide[c]	Pro	Tyr
Lean beef #1	3.58	8.30	2.58	2.58	4.18	3.92
Lean beef #2	4.27	8.19	2.70	3.08	4.48	3.80
Soy PC[a]	3.23	7.39	0.66	1.53	5.28	4.18
80% lean beef #2 20% soy PC[a]	4.06	8.03	2.29	2.77	4.64	3.88
70% lean beef #2 30% soy PC[a]	3.96	7.95	2.09	2.62	4.72	3.92
60% lean beef #2 40% soy PC[a]	3.85	7.87	1.88	2.49	4.80	3.95
Meat patties with textured vegetable protein						
1	3.21	7.22	1.39	1.92	6.62	3.55
2	3.29	7.37	1.40	1.99	6.19	3.56
3	2.89	7.32	1.04	1.62	6.35	3.66
Textured vegetable protein						
1	3.42	7.62	1.27	1.81	5.49	4.47
2	3.19	8.19	1.40	1.53	4.72	3.72
3	3.14	8.07	0.97	1.69	4.99	4.36
4	3.25	7.90	0.96	1.80	5.01	4.33
Textured vegetable protein seasoning mix						
1	3.37	7.89	0.82	1.65	4.37	4.26
2	3.16	7.56	0.36	1.30	5.12	3.93
3	2.82	7.13	0.48	1.43	4.75	3.48

Bio-analysis		Estimated PER					
		Equation					
Nitrogen digestibility[d] %	Determined PER[e]	1	2	1 with digestibility[f]	2 with digestibility[f]	3	3 with Met & Met sulfoxide[g]
93	2.8	2.9	2.9	--	--	2.8	--
93	2.8	2.8	2.8	--	--	3.06	3.2
90	2.2	2.4	2.4	--	--	1.0	1.3
92	2.5	2.8	2.8	--	--	2.6	2.8
91	2.6	2.7	2.7	--	--	2.4	2.7
90	2.5	2.7	2.7	--	--	2.2	2.5
87	2.1	2.3	2.4	1.9	2.0	1.7	2.0
87	2.3	2.4	2.5	2.0	2.1	1.9	2.1
87	2.1	2.4	2.5	2.0	2.1	1.5	1.7
82	2.1	2.5	2.5	1.9	2.0	1.2	1.4
81	1.8	2.8	2.9	2.2	2.2	2.3	2.4
84	1.7	2.8	2.7	2.2	2.2	1.4	1.8
82	2.1	2.7	2.7	2.1	2.1	1.4	1.7
83	2.0	2.7	2.7	2.1	2.1	1.4	1.7
80	1.8	2.5	2.5	1.9	1.9	1.2	1.6
82	1.6	2.3	2.4	1.8	1.9	1.3	1.7

TABLE 1 (continued)

Food product or protein mixture	Amino acid content (grams of amino acid residue per 100 grams of total amino acid residues)					
	His	Leu	Met	Met & Met sulfoxide[c]	Pro	Tyr
90% lean beef #1 10% collagen	3.3	7.8	2.4	2.4	5.1	3.6
50% lean beef #1 50% collagen	2.2	5.6	1.6	1.6	8.7	2.4
90% lean beef #1 5% collagen 5% whey PC[b]	3.4	8.1	2.5	2.5	4.7	3.7
50% lean beef #1 25% collagen 25% whey PC[b]	2.5	7.5	1.8	1.8	7.0	3.1
25% lean beef #2 25% collagen 50% whey PC[b]	2.3	8.0	1.6	1.7	7.5	3.0
50% lean beef #2 25% collagen 25% soy PC[a]	3.1	6.7	1.7	2.1	6.9	3.2
25% lean beef #2 25% collagen 50% soy PC[a]	2.9	6.5	1.2	1.7	7.1	3.3
Whey PC[b]	2.1	10.4	1.5	1.5	6.2	3.7
Collagen	0.8	3.0	0.7	0.7	13.3	0.9

[a]Soy protein concentrate, Promosoy-100.

[b]Whey protein concentrate, Enrpro 50.

[c]89.13% of the determined methionine sulfoxide residue which is the molecular equivalent of the methionine residue was totaled with the obtained methionine residue value.

[d]Nitrogen digestibility = N intake - fecal N/N intake X 100. Determined during PER test.

Bio-analysis		Estimated PER					
Nitrogen diges-tibility[d]	Determined PER[e]	Equation					
%		1	2	1 with di-gestibility[f]	2 with di-gestibility[f]	3	3 with Met & Met sulfoxide[g]
92	2.5	2.6	2.7	--	--	2.6	--
89	1.7	1.5	1.8	--	--	1.4	--
93	2.5	2.8	2.8	--	--	2.8	--
90	2.4	2.4	2.6	--	--	2.4	--
89	2.6	2.6	2.8	--	--	2.8	2.8
90	2.2	2.0	2.2	--	--	1.8	1.9
88	2.1	1.9	2.1	--	--	1.3	1.5
90	2.7	3.8	3.9	--	--	3.9	--
88	<0.0	0.06	0.8	--	--	0.15	--

[e]PER = protein efficiency ratio = weight gain (grams)/protein intake (grams). PER values were corrected to that of casein at 2.5.

[f]Amino acid values (grams of amino acid residue per 100 grams of total amino acid residues) were lowered by percent digestibility for use in equations 1 and 2.

[g]Methionine residue + 89.13% of methionine sulfoxide residue totaled and the total used in equation 3 as the methionine value.

all equations) than the range from which the equations were developed although the other key amino acid values fall within the ranges for the equations. All equations overestimate the PER (+1.1 and 1.2) for this whey product, undoubtedly due to its amino acid pattern. All equations estimate the PER of combinations of lean beef, collagen, and whey protein concentrate much closer to the PER of the bioassay, the greatest difference being +0.3 for one combination. Two combinations were predicted within +0.2 PER.

EXTENDING USE OF THE EQUATIONS

It may be necessary to derive a separate equation for each type of food protein (oilseeds, dairy products, noodles, marine products, etc.) based on a group of carefully selected samples from each type which will provide a linear range of amino acid quantities and determined PER values. This would tend to rule out as variables differences in amino acid patterns and digestibility. The latter is inherent in the PER test. With the data already available and new data yet to be developed new mathematical studies could offer a more universal equation.

Our laboratory is continuing studies to extend the development of equations for the greatest possible utilization as indicated above. It will be necessary to establish regression relationships between PER and the amino acid composition and digestibility of bean, soy bean, marine, noodle, dairy, and other products where predicting equations would be useful. This necessitates having available proximate and amino acid composition, digestibility, and PER data on samples from a large number of products. It would be highly advantageous, economically and timewise, to researchers to pool existing data. Such a pool has been started at this laboratory and input from other researchers is invited.

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