

TABLE V

Comparison of Protein Quality as Measured by RPV ^a and EUD		
	RPV ^b	EUD ^b
Poultry meat		
breast	0.762	72.6
back ^c	0.748	74.5
neck ^c	0.646	66.1
wing ^c	0.673	78.6
Lactalbumin (ICN)	0.901	86.7
Casein	0.720	73.9
Blood plasma (conc)	0.695	72.7
Faba beans (raw conc)	0.328	45.4
Sunflower (meal)	0.410	68.7
Sunflower (defatted meal)	0.410	70.1
Correlation coeff. (r)	0.808	
Regression equation $y = 42.7(7.58) + 45.1(11.63)x$		

^aRat response body nitrogen.

^bVersus whole egg protein.

^cMechanically deboned.

shown in Table V together with the correlation coefficients and the correlation equation.

As it can be observed, the values for the animal protein sources fit quite well, while discrepancies emerge for what plant protein sources are concerned. On the whole, the correlation coefficients were satisfactory (0.808) but, owing to differences in the plant protein evaluation, the regression line intercepts the y axis much above the zero point.

Possibly, the discrepancies shown for plant protein sources, which are severely unbalanced, are not to be ascribed to the ultrafiltrate digest methodology per se, but rather to the scheme of calculation, based on the geometric mean of all the essential amino acids, which obscures the effect of the limiting amino acid (11). However, a definitive conclusion about the equivalence of the two methodologies will be drawn only when more data will be collected and carefully analyzed.

With regard to the problem of the best bioassay procedure, the results illustrated indicate that they can all be indifferently used when high quality proteins have to be tested, while the RPV test seems to be the method of choice for low protein quality, as plant proteins in general are.

Two more problems await for a solution. The first is the

problem of the reference protein. All the data reported are relative to the egg protein, whose utilization for rats' growth was better than that of lactalbumin. Moreover, egg protein is the reference protein for the WHO/FAO standard.

However, in a recent paper (12) it was shown that the true safety nitrogen level for human maintenance has a protein quality at least 25% less than egg. If this is so, a more realistic reference protein should be proposed.

The second problem may be formulated as follows: when is it that two proteins must be considered as having different nutritive value? In our laboratory as in others (13), the standard error of RPV was around 8%. Together with the definition of the reference protein, this is of practical importance when recommendations for a minimum value for protein quality are to be made.

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The Nutritive Value of the Same Protein Preparations as Estimated by Human, Rat, and Chemical Assays

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ABSTRACT

Results are summarized from studies in which the protein nutritional values of thirteen protein sources were estimated by human, rat, or chemical assays. Generally, agreement was poor between nutritive value as estimated in adult men and as estimated by various rat assays or by chemical (amino acid) scores. Possible reasons for this lack of agreement are briefly discussed.

INTRODUCTION

Various animal and chemical assays have been developed

for estimating the nutritional value of protein from different sources (1-7). These assays, however, are of little usefulness in human nutrition if they do not accurately predict protein nutritive value for humans. The few published comparisons of nutritive value as estimated by animal or chemical assays and nutritive value as estimated directly in humans with the same protein preparations were reviewed (8,9). In this paper, results from studies in which these comparisons have been made with two different groups of protein sources are summarized.

DESCRIPTION OF STUDIES

In the first group of six protein sources, nutritional value

TABLE I

Relative Net Protein Utilization (Egg = 100) of the Same Protein Sources as Estimated with Men (0.4 g Protein [N x 6.25]/Kg Body Wt/Day) and Rats

Protein source	Humans ^a (A)	Net protein utilization	
		Rats ^b (B)	(A) - (B)
Spray-dried whole egg	(100)	(100)	--
Tuna	93	89	+ 4
Cottage cheese C	97	91	+ 6
Soy isolate B ^c	90	66	+24
Peanut flour	93	62 ^d	+31
Wheat gluten	66	66	0

^aFrom Bodwell (8) and Bodwell et al. (10).

^bFrom Hackler (11).

^cPromine F.

^dHackler, unpublished data.

was estimated by Net Protein Utilization (NPU) as determined with 4-6 men who consumed a single intake level (0.4 g [N x 6.25]/kg body wt/day) of each protein (10), various rat assays (11), the calculated Protein Efficiency Ratio (C-PER) of Satterlee et al. (12), and chemical (amino acid) scores. In the second group of sources, seven protein breads were similarly evaluated except that their nutritional value for humans was estimated by determining the minimal nitrogen intake level required to maintain a "zero" nitrogen balance (13-16). Groups of 17-20 young men consumed each protein bread at N intake levels that varied from 50 mg (all proteins) to 130-200 mg (varied according to protein source) /kg body wt/day (16).

RESULTS

Human Assays vs Rat and C-PER Assays

Relative NPU values (egg values = 100) as estimated in the men and in rats for the first group of protein sources are shown in Table I. Compared to the values from the humans, the NPU values from rats were similar for tuna, cottage cheese and wheat gluten, but markedly lower for the soy isolate and peanut flour. Relative protein value, as estimated by the NPU values determined in the men, was also markedly underestimated by both Relative Protein Values and Relative Nitrogen Utilization values obtained in the rats (Table II).

For the group of seven protein breads, values from three different rat assays are compared in Table III with the estimated nutritive values from the young men. With a value of 100 for egg protein, the Relative Protein Value assay underestimated nutritive value as estimated in the young men (if lactalbumin were assigned a value of 100), then agreement with the values from the men would be good for casein, but the nutritive value of egg white would be overestimated, and of textured soy protein, soy isolate, peanut flour, and wheat gluten, underestimated. The Net Protein Ratio and Relative Nitrogen Utilization values agreed with the estimates from the men for the three animal proteins, but underestimated the nutritive values of textured soy protein, soy isolate, peanut flour, and wheat gluten.

The Protein Efficiency Ratio (PER) values for the first group of protein sources and the NPU values from the men showed little relationship (Table IV). Excluding the values for gluten, the relative NPU values only varied between 93 and 98%. For the same proteins, PER varied from 0.99 to 2.95. The C-PER values were higher for 5 of the 6 proteins

TABLE II

Differences in Relative Protein Value (Egg = 100) as Estimated by Net Protein Utilization (NPU) Determined in Men (0.4 g Protein [N x 6.25]/Kg Body Weight/Day) and by Relative Protein Value (RPV) and Relative Nitrogen Utilization (RNU) in Rats

Protein	NPU estimate (humans) ^a minus RPV estimate (rats) ^b	NPU estimate (humans) ^a minus RNU estimate (rats) ^b
Spray-dried whole egg	---	---
Tuna	+19	+23
Cottage cheese C	+36	+16
Soy isolate B ^c	+40	+35
Peanut flour	+56	---
Wheat gluten	+47	+34

^aBodwell (8) and Bodwell et al. (10).

^bHackler (11) and unpublished data of L.R. Hackler.

^cPromine F.

TABLE III

Relative Protein Nutritive Value (Egg White = 100) of Different Protein Breads as Estimated in Rats and in Young Men

Protein source (Breads)	Assay ^a			Relative value in humans ^b
	RPV	NPR	RNU	
Egg white	(100)	(100)	(100)	(100)
Lactalbumin	76	92	92	91
Casein	78	92	91	91
Textured soy protein ^c	58	78	78	91
Soy isolate B ^d	47	70	69	77
Peanut flour	52	61	61	79
Wheat gluten	25	32	31	75

^aRelative Protein Value (RPV), Net Protein Ratio (NPR) and Relative Nitrogen Utilization (RNU) from Staples et al. (17).

^bBased on minimal nitrogen intake level required for "zero" nitrogen balance (16).

^cSupro 50-4.

^dPromine F.

TABLE IV

Nutritive Value of Six Protein Sources As Estimated by Net Protein Utilization Values (NPU) Determined in Men (0.4 g protein [N x 6.25]/Kg Body Wt/Day), by Protein Efficiency Ratio (PER; Rats) and by Calculated Protein Efficiency Ratio (C-PER)

	NPU ^a (Human)	PER ^b	C-PER ^c
Spray-dried whole egg	(100)	2.95	2.63
Tuna	93	2.28	2.66
Cottage Cheese C	97	2.32	2.43
Soy isolate B ^d	90	1.39	1.81
Peanut flour	93	0.99	1.85
Wheat gluten	66	0.32	0.82

^aRelative values with value for egg = 100 (8,10).

^bFrom Hackler (11) and unpublished data.

^cUnpublished data of L. Satterlee.

^dPromine F.

than the observed PER values. For the three protein breads containing animal protein, the PER values were similar and thus agreed with the estimates of nutritive value from the young men (Table V). However, the PER values for the plant protein breads suggested a much lower relative nutritive value than indicated by their relative values determined in the men. Except for the two soy protein breads, C-PER values were only slightly higher (<0.3 PER units) than the observed PER values.

Human Assays vs Chemical Scores

Calculated chemical scores are compared to the estimates of nutritive value obtained with men in Tables VI and VII. For the first group of proteins (Table VI), when

scores were calculated by use of the 1974 NRC provisional amino acid scoring pattern (4) or the 1973 FAO/WHO provisional pattern (2), agreement with the human NPU values was generally good for the three animal proteins, but not for the plant proteins. The scores calculated by use of the amino acid patterns of egg or human milk were markedly lower than the relative NPU values from the human studies.

For the protein breads (Table VII), the scores calculated by use of the NRC pattern overestimated the relative values from the human studies for lactalbumin, casein, textured soy protein, and soy isolate, and underestimated the values of peanut flour and wheat gluten. With the FAO/WHO pattern as a reference, agreement was good between the scores and the relative nutritive values estimated in the humans for both soy proteins; however, the nutritive values were slightly overestimated for lactalbumin or casein and underestimated for peanut flour and wheat gluten. Except for the value for casein calculated by use of the amino acid pattern of human milk as a reference, the scores calculated by use of either the egg or human milk pattern were much lower than the comparable relative values from humans.

DISCUSSION

On the basis of the limited data presented, the various animal or chemical assays discussed would not appear to accurately predict nutritional value for adult humans. Most of the animal assays use rapidly growing young rats, and values from these assays could be expected to be more closely related to protein nutritional value for infants or children than for adults. However, PER values did not agree with estimates of nutritive value in children (9,13). For the

TABLE V

Protein Efficiency Ratio (PER) Values, Calculated Protein Efficiency Ratio (C-PER) Values and Relative Nutritive Value in Humans of Different Protein Breads

Protein source (Breads)	PER ^a	C-PER ^b	Relative value in humans ^c
Egg white	2.83	2.94	(100)
Lactalbumin	2.59	2.74	91
Casein	2.67	2.76	91
Textured soy protein ^d	2.09	2.64	91
Soy isolate B ^e	1.77	2.48	77
Peanut flour	1.59	1.67	79
Wheat gluten	0.45	0.69	75

^aValues corrected to casein (protein) = 2.50; from Staples et al. (17).

^bCalculated PER; determined by use of 4-enzyme method and Na-caseinate amino acid profile of Satterlee et al. (12); amino acid data used was from a single analysis of each protein source (methionine and cysteine determined as methionine sulfone and cysteine acid).

^cRelative to value for egg white of 100 based on minimal nitrogen intake level required for "zero" nitrogen balance (16).

^dSupro 50-4.

^ePromine F.

TABLE VI

Chemical Scores Calculated According to Different Reference Amino Acid Patterns and Relative Net Protein Utilization Values (Egg = 100) Determined in Men (0.4 g Protein [N x 6.25]/kg Body Wt./Day)

Protein	Reference pattern			Human milk (18)	Relative NPU (humans)
	NRC, 1974 (4)	FAO/WHO, 1973 (2)	Egg ^a		
Spray-dried whole egg	100	100	(100)	79	(100)
Tuna	100	100	68	76	93
Cottage cheese C	100	100	62	82	97
Soy isolate B ^b	85	82	60	66	90
Peanut flour	66	61	45	51	93
Wheat gluten	28	26	21	22	66

^aSpray-dried whole egg (analyses from L.R. Hackler and C.E. Bodwell, unpublished data).

^bPromine F.

TABLE VII

Chemical Scores According to Different Amino Acid Reference Patterns and Relative Nutritive Values of Different Protein Breads as Estimated in Young Men

Protein source (Breads)	Reference pattern				Relative value in humans ^b
	NRC, 1974 (4)	FAO/WHO, 1973 (2)	Egg ^d	Human milk (18)	
Egg white	100	100	89	89	(100)
Lactalbumin	100	100	70	76	91
Casein	100	100	72	96	91
Textured soy protein ^c	100	94	60	76	91
Soy isolate B ^d	90	73	46	62	77
Peanut flour	67	62	49	51	79
Wheat gluten	32	30	24	25	75

^aSpray-dried whole egg (analyses from L.R. Hackler and C.E. Bodwell; unpublished data).

^bRelative to value for egg white of 100 based on minimal nitrogen intake level required for "zero" nitrogen balance (16).

^cSupro 50-4.

^dPromine F.

other animal assays, there are few or no data for making similar comparisons (8,9).

The lack of agreement between nutritive value as predicted by the chemical scores and nutritive value as estimated in our human subjects is not surprising. The NRC and FAO/WHO reference patterns (2,4) were derived as patterns for evaluating protein nutritive value for older infants, children, and adults. Because the estimated requirement levels of total essential amino acids are much higher for the infant or young child than for adults, the scoring patterns are not specifically applicable for prediction of protein nutritive value for adults. Similarly, the essential amino acid levels in egg or milk are much higher than the estimated requirement levels of adults. In addition to these considerations, however, there is also a probable defect in the approach used for the development and use of amino acid reference patterns. By convention, values are summed for the two sulfur amino acids and for the two aromatic amino acids. Particularly with plant proteins, this practice is probably not justified and may contribute a significant amount of error in predicting protein nutritive value for humans.

The problems involved in the use of the PER assay for estimating protein nutritional value for humans have been widely discussed (19-22). It follows that an approach such as the C-PER procedure of Satterlee et al. (12) can be no more useful than the PER assay that it is intended to replace. As indicated by Satterlee et al. (12), however, the procedures developed might be useful in developing an approach for predicting protein nutritive value for humans.

As previously discussed (8), if in vitro digestibility estimates, such as those used in the C-PER procedure (12), were found to correlate with digestibility in humans, the estimates might reflect general differences in amino acid bioavailability. If so, an in vitro estimate of digestibility plus amino acid composition data might be used for estimating the nutritive value of proteins for humans with an accuracy sufficient for practical application.

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