

I A F M M

international association of fish meal manufacturers

Hoval House, Orchard Parade, Mutton Lane, Potters Bar, Hertfordshire, EN6 3AR
Tel: (Potters Bar) 0707 42343

No.1 OCTOBER 1970

AVAILABLE AMINO ACID CONTENT OF FISH MEALS

Summary. The Food and Agriculture Organization of the United Nations has recently issued a paper entitled "Available Amino Acid Content of Fish Meals" in which the most recent data on this subject have been evaluated. The data have been compared with figures for soyabean meal and meat meal. From animal experiments the mean percentage availability for lysine, methionine and tryptophan is estimated as 93, 97, 103 for fish meals; 86, 75, 77 for meat meals; 90, 101, 103 for soyabean meals respectively. These figures correspond quite closely to the true digestibility values for the amino acids which are 91% for fish meal, 91% for soyabean meal and 71% for meat meals. This suggests that indigestibility is a major cause of incomplete availability. Thus the evidence indicates that fish meals and soyabean meals have comparable high amino acid availability, whilst meat meals have a lower amino acid availability.

Some time ago the Food and Agriculture Organisation of the United Nations initiated the collection of analytical data for various feedstuffs, including fish meal, to be used in the formulation of feeds. At the beginning of this year Dr. E.L. Miller of the University of Cambridge, U.K. was commissioned to prepare a review paper, entitled "Available Amino Acid Content of Fish Meals", so that the most important amino acid data could be collected together and evaluated. The 66-page document has now been published and is available from F.A.O. Headquarters in Rome¹.

The paper presents the mean values for the amino acid composition of fish meals determined in the last 10 years by ion exchange chromatography (Table 1). These figures are based on results of analyses of 159 samples by a number of different laboratories. The standard deviations appropriate to the mean values of the fish meals listed in Table 1 are presented in Table 2. The standard deviations were determined either from results obtained in one particular laboratory or from results obtained from a number of laboratories; thus the standard deviations are expressed as either "within laboratory" or "between laboratory" respectively. The question arises as to which is the appropriate standard deviation. If it is desired to know the confidence limits within which an analysis from any laboratory should fall, then the "between laboratory" standard deviation is appropriate. On the other hand if it is desired to know the variability between fish meals of a similar type, then the "within laboratory" standard deviation is more appropriate, since variability due to the laboratory

TABLE 1

MEAN VALUE OF TOTAL AMINO ACID COMPOSITION (g/16gN) OF FISH MEALS DETERMINED MAINLY BY ION-EXCHANGE CHROMATOGRAPHY

	Herring meals	Anchovy meals	Pilchard & Maasbanker meals	Tuna (mixed species) offal meals	Menhaden meals processed by wet reduction	White fish meals
Lysine	7,73	7,75	7,94	7,30	7,56	6,90
Methionine	2,86	2,95	2,71	2,75	2,82	2,60
Cystine	0,97	0,94	0,95	0,79	0,90	0,93
Tryptophan	1,15	1,20	1,02	1,05	1,07	0,94
Histidine	2,41	2,43	3,02	3,41	2,32	2,01
Arginine	5,84	5,82	5,95	6,43	6,04	6,37
Threonine	4,26	4,31	4,38	4,34	3,97	3,85
Valine	5,41	5,29	5,41	5,31	5,10	4,47
Isoleucine	4,49	4,68	4,48	4,46	4,40	3,70
Leucine	7,50	7,62	7,30	7,20	7,14	6,48
Phenylalanine	3,91	4,21	3,91	4,10	3,95	3,29
Tyrosine	3,13	3,40	3,23	3,28	3,22	2,60
Aspartic acid	9,10	9,49	9,37	9,30	9,07	8,54
Serine	3,82	3,84	4,27	4,18	3,61	4,75
Glutamic acid	12,77	12,96	12,92	11,93	12,70	12,79
Proline	4,15	4,17	4,52	5,43	4,58	5,34
Glycine	5,97	5,62	6,92	8,15	6,78	9,92
Alanine	6,25	6,31	6,17	6,76	5,94	6,31
Crude protein %	73,6	65,4	65,4	53,24	62,01	65,01
Moisture %	6,93	8,01	9,0	6,20	8,25	8,49
Ash %						20,92

is more or less excluded. "Within laboratory" standard deviations are given for all meals except white fish meal, and whilst the analyses of pilchard and maasbanker meals came from one laboratory, they were the work of different authors at different times.

However, a feed, even with such a good balance of amino acids as is found in fish meal, will be of little value if the nutrients are not available to the animal ingesting it. The paper considers four possible reasons for the failure of an animal to make full use of a dietary amino acid, even though it is the one limiting protein quality.

1. In the course of digestion, proteins are hydrolised by digestive enzymes to their constituent amino acids. The free amino acids are absorbed from the digestive tract and passed to the liver via the bloodstream. Some proteins, due to their chemical structure, are resistant to the digestive process. Heat damaged proteins are also more slowly digested. Clearly, an amino acid which is not digested is also unavailable.
2. All the free amino acids must be present simultaneously at the site of protein synthesis. If amino acids are digested and absorbed from the intestinal tract at different rates, so that the optimum ratio of amino acids at the site of protein synthesis is not maintained, the absorbed amino acid will be utilised with impaired efficiency. In practice, however, with animals fed *ad libitum*, proteins in various stages of digestion will always be present in the digestive tract, and it seems unlikely that any differential rate of digestion of the proteins from a single meal will result in differential availability of amino acids at the site of protein synthesis.
3. It is not sufficient for amino acids to be present simultaneously at the site of protein synthesis. They must be present in proportions not too far different from those finally required in the synthesized protein. Excess of certain amino acids, by creating an imbalance, prevents the optimum utilisation of other amino acids. For example, high levels of lysine in the diets of chicks are known to increase the requirement for arginine. Again, amino acid imbalance is unlikely to be important in practical diets which contain a mixture of proteins from various feedstuffs, but imbalance effects may be important in laboratory procedures used to determine protein quality and available amino acids.
4. Amino acids may be present in foods in some combined form so that they are unable to partake in metabolic reactions, although digested and absorbed at a normal rate. Such amino acids may be released by acid hydrolysis and be included in the total amino acid value by most present-day analytical techniques. For example, lysine is known to react with carbonyl groups of reducing sugars and of oxidised lipids, thus decreasing its availability, but nevertheless it is analysed by ion exchange chromatography.

In recent years a great deal of experimental work has been carried out in order to determine the availability of amino acids in feedstuffs. Combs² in 1968 reviewed much of the data and produced feedstuffs analysis tables in which the amino acid data had been modified by what he described as an availability factor. Soyabean meal and corn were given an arbitrary value of 100. Other feed ingredients were expressed relative to this figure. Fish meal was given an availability figure of 90, together with meat meal and meat and bone meal. Dr. Miller has reviewed the various methods that have been used to estimate this availability factor and reassessed the situation in the light of more recent data.

There are three basic methods for estimating the availability of amino acids – biological procedures using live animals, microbiological procedures and chemical methods. Assessing the practical validity of the results of these methods is not an easy task, for in some cases meals of extreme quality – good and bad – are selected for experimental purposes and these, therefore, do not reflect the quality of meals available commercially. Furthermore, some of the older references reflect the quality of the meal at the time of the experiments, but improvements in feed processing techniques since that time relegate these results to mere historical significance. Also, “between laboratory” variation in the data is high and can be misleading. Finally, the methods themselves are by no means established, and currently there is a great deal of work being undertaken to examine the validity of the various techniques, attempting to correlate results of biological tests with those of microbiological and chemical tests. Accepting these difficulties, Dr. Miller has reviewed the literature of the past 10 years, and, wherever possible, compared the mean availability figures of fish meal with those of soyabean meal and meat meal.

Available lysine, methionine and tryptophan values determined by biological procedures are summarised in table 3 for fish meal, meat meals and soyabean meals. Where authors have given the total amino acid content for individual meals, the percentage availability has been calculated. Where no total values were reported, mean values given in table 1, or by F.A.O.³ were used to calculate availability. Unweighted mean values for percentage availability for lysine, methionine and tryptophan are respectively 93, 97, 103 for fish meals; 86, 75, 77 for meat meals; 90, 101, 103 for soyabean meals. Owing to the great variability between results from different bio-assay procedures, little weight can be given to the individual values. However, the indication from the mean values is that fish meals and soyabean meals have a comparable high amino acid availability, whilst meat meals have a lower amino acid availability.

The value of a protein when fed as the sole source of nitrogen depends upon the available level of the limiting amino acid. Supplementation studies have shown the sulphur amino acids to be limiting in both fish meal and soyabean meal when fed as the sole protein to rats, and, therefore, the net protein utilisation (N.P.U.) of fish meals and soyabean meals should be a measure of available methionine-plus-cystine. The results of 28 N.P.U. carcass experiments were averaged and compared with calculated N.P.U. values based on the total content of methionine-plus-cystine in both fish meal and soyabean meal. The percentage by which the experimentally determined N.P.U. values were smaller than the calculated values gave a measure of the availability of the sulphur containing amino acids. The results of this exercise confirmed the equal availability of the sulphur amino acids in fish meal and soyabean meal.

The same conclusion can be drawn from the results of the large number of published “biological value” experiments with these two protein sources. The results are summarised in Table 4 where it is seen that the N.P.U. value is approximately 25% greater for the protein of fish meal than of soyabean meal – this is, of course, to be explained by the higher content in fish protein of the two sulphur-containing amino acids.

TABLE 4

Mean Values (with their standard errors) for % Nitrogen Digestibility, Biological Value, and Net Protein Utilisation (N.P.U.) of Fish Meal and Soyabean Meal

	Fish Meal	Soyabean Meal
N Digestibility (TD)	91,1 ± 0,51 (51) *	86,0 ± 1,56 (15)
Biological Value (BV)	80,4 ± 1,06 (37)	67,5 ± 2,15 (14)
N.P.U. (BV x TD)	73,9 ± 1,18 (35)	59,3 ± 2,16 (14)

* Number of samples for which values are reported.

AVAILABLE AMINO ACIDS DETERMINED BY BIOASSAY IN FISHMEALS, MEAT MEALS AND SOYABEAN MEALS

Animal	Dose - Response Metameters	Assay Conditions	Lysine			Methionine			Tryptophan		
			Total (g/16gN)	Available (g/16gN)	Availability (%)	Total (g/16gN)	Available (g/16gN)	Availability (%)	Total (g/16gN)	Available (g/16gN)	Availability (%)
1. HERRING MEALS											
Chick	1	C	7,5	7,3	97	2,6	2,7	104	1,1	1,3	118
Chick	2	B	7,7	8,7	113	-	-	-	-	-	-
Chick	2	A	-	-	-	3,1	3,1	100	-	-	-
Chick	3	D	-	6,8	[88]	-	-	-	-	-	-
Chick	2	A	-	-	-	-	2,6(4)	[91]	1,15(4)	0,76(4)	66
Chick	4	B	7,6(12)	7,2(12)	95(12)	2,8(12)	3,3(12)	118(12)	-	-	-
Chick	4	B	7,9(9)	7,8(9)	100(9)	3,0(8)	3,4(8)	113(8)	-	-	-
2. ANCHOVY MEALS											
Chick	1	C	-	6,8	[88]	-	-	-	-	1,4	[117]
Chick	2	B	7,6	8,7	114	-	-	-	-	-	-
Chick	2	B	6,7	4,3	64	-	-	-	-	-	-
Chick	4	B	7,8(31)	7,0(31)	89(31)	3,0(14)	3,0(14)	101(14)	-	-	-
Chick	4	B	7,8(8)	7,9(8)	101(8)	3,0(5)	3,2(5)	104(5)	-	-	-
3. MENHADEN MEALS											
Chick	1	C	9,6	6,1	64	3,0	2,2	73	0,8	1,0	125
Chick	1	C	7,6	7,8	103	2,3	2,4	104	-	-	-
Chick	1	C	7,2	6,6	92	2,6	2,3	88	0,9	1,1	122
Chick	4	B	7,6(19)	7,3(19)	97(19)	2,8(14)	3,4(14)	120(14)	-	-	-
4. U.K. WHITE FISH MEAL											
Chick	2	A	-	-	-	2,7	2,5	93	-	-	-
Chick	2	D	-	-	-	2,7	2,9	107	-	-	-
Chick	3	D	-	6,4	[93]	-	-	-	-	-	-
Rat (Lysine)	2	A	-	6,8	[99]	-	2,4	[92]	-	0,64	[68]
Chick (Methionine)											
Chick (Tryptophan)											
5. S. AFRICAN PILCHARD MEAL											
Chick	3	D	-	6,4	[80]	-	-	-	-	-	-
6. TUNA MEAL											

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Chick	1	C	7	5,2	91	1,9	2,4	26	0,5	0,6	120
Chick	4	B	7,4(7)	7,3(7)	99(7)	2,8(5)	3,3(5)	118(5)	-	-	-

7. ROSEFISH MEAL

Chick	1	C	7,6	7,2	95	2,9	2,6	90	1,0	0,9	90
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8. MEAT MEALS

Chick	1	C	5,3	5,5	104	1,4	1,9	136	0,8	0,5	62
Chick	2	A	-	-	-	1,2	0,8	67	-	-	-
Chick	2	A	-	-	-	1,2	0,9	75	-	-	-
Chick	5	A	-	-	-	1,8	1,24	68	-	-	-
Chick	5	A	-	-	-	1,0	0,56	56	-	-	-
Chick	5	A	-	-	-	2,0	0,93	47	-	-	-
Chick	3	D	-	3,4(3)	[65]	-	-	-	-	-	-
Chick	2	A	-	-	-	-	1,08(6)	[77]	0,63(2)	0,61(2)	97(2)
Rat (Lysine)	2	A	-	4,2	[80]	-	0,94	[67]	0,66	0,48	73
Chick (Methionine)											
Chick (Tryptophan)											
Chick	4	B	-	5,0(4)	[95]	-	1,2(4)	[86]	-	-	-

9. SOYABEAN MEALS

Rat	6	A	-	-	-	-	-	-	1,26	1,53	121
Rat	6	A	-	-	-	1,4	1,1	80	-	-	-
Chick	1	C	6,3	6,7	106	1,35	1,6	118	1,5	1,6	107
Chick	3	D	-	3,3(3)	[54]	-	-	-	-	-	-
Chick	5	A	-	-	-	1,8(3)	1,9(3)	105	-	-	-
Chick	4	B	-	6,3(13)	[103]	-	1,4(10)	[100]	-	-	-
Chick	4	E	6,6	6,5	98	-	-	-	-	1,25	[81]

() Number of samples.

[] Calculated assuming total lysine, methionine and tryptophan of meat meal 5,25, 1,4, 0,88 and of soyabean meal 6,1, 1,4, 1,54 (FAO, 1969). Total value for fish meals taken from Table 1.

Dose-response metameters

1. Assumed amino acid requirement; weight gain.
2. % amino acid supplement; food conversion efficiency.
3. Logarithm % amino supplement; gN retained/gN ingested.
4. g. available amino acid eaten; weight gain.
5. Logarithm % amino acid supplement; food conversion efficiency.
6. % amino acid supplement; weight gain.

Assay Conditions

- A. Test protein added at expense of carbohydrate; crude protein and amino acid balance not constant.
- B. Test protein added at expense of protein; crude protein constant, amino acid balance not constant.
- C. Crude protein constant, amino acid balance of test and positive control similar.
- D. Test protein added at expense of identical amino acids; crude protein and amino acid balance constant.
- E. Test protein added to crystalline amino acid basal; crude protein and amino acid balance not constant.

Recent collaborative studies in the U.K. on microbiological methods for estimating available amino acids have shown considerable variability in results, making the confidence limits on the tests too wide for the values to be of use in discriminating between commercial fish meals. Consequently, the values obtained for various protein concentrates by these methods have not been reviewed.

Probably, the most widely reported chemical test for measuring the availability of amino acids is the chemical estimation of available lysine with the reagent, fluorodinitrobenzene (FDNB). The hypothesis that only lysine molecules with reactive $\epsilon\text{-NH}_2$ groups are nutritionally available is the basis of this chemical method. A number of published papers have shown a good correlation between FDNB-available lysine values for fish meal and the biological availability determined by the gross protein value (G.P.V.) procedure. However, there are other reports, particularly with commercial fish meals, which do not demonstrate a significant correlation between G.P.V. or other biological assays and FDNB-available lysine, but in these reports the range of values obtained by feeding experiments is quite small. It is possible that such results can be explained, not only by the inaccuracy of the chemical procedure, but also by the variability and non-specificity of the biological tests resulting in a failure to detect slight changes in the biological availability of lysine. In spite of the uncertainty of the significance of the FDNB-available lysine test, some of the most recently reported values for fish meals are given in Table 5. Part of the variation in these values can be attributed to differences in analytical techniques in various laboratories, but sufficient data are available from "within laboratories" to suggest a coefficient of variation within each type of meal of about 9%.

Incomplete digestion is only one of four possible causes of unavailability. However, it is likely to be the most important cause. Certainly, availability cannot be any better than the digestibility. Values for amino acid digestibility, therefore, set an upper limit to possible availability figures. Amino acid digestibility is determined by analysing the food and the faeces for amino acids. Amino acids measured in the faeces have to be corrected for metabolic contributions (e.g. digestive enzymes) in order to calculate "true digestibility". The determination of the metabolic contribution may be done by feeding a nitrogen-free diet, or by feeding a low level of egg protein, which is assumed to be completely digested, or by regression analysis of amino acid excretion against amino acid intake. A more important source of error is brought about by the activities of bacteria in the intestinal tract, which may ferment undigested feed proteins with the liberation of ammonia and the synthesis of bacterial proteins and nucleic acids. Thus, there are less amino acid residues in the faeces of normal animals than there would be with germ-free animals, and consequently the amino acid digestibility values are greater with the former. Therefore, digestibility values obtained with animals supporting a normal bacterial flora must be regarded as maximal values. Estimates of "true digestibility" of amino acids obtained with rats or with birds, colostomised to facilitate separate collection of faeces, are given in table 6 in which the data suggest that the amino acids of fish meal are 91% digestible. This is the same as the estimate of digestibility for the amino acids of soyabean meal, and better than the estimate of 71% for the amino acids of meat and bone meal. The bio-assays indicate that values for the percent availability of amino acids are similar to those for digestibility; this suggests that indigestibility is the major, if not the only, cause of incomplete availability.

The evidence does not support the suggestion of Combs that the amino acids of normal commercial fish meals are 10% less available than those of soyabean meal. Rather, the evidence indicates that there is no difference in availability of amino acids between fish meal and soyabean meal; furthermore, fish meal protein contains a higher content of lysine and sulphur-amino acids than soyabean protein. The evidence also indicates that the amino acids in fish meal are more available than those in meat meal.

TABLE 5

THE FDNB-AVAILABLE LYSINE CONTENT (g/16gN) OF FISH MEALS

Meal	Number of Samples	Mean	Standard Deviation	Range	References
Herring Denmark	32	7,4	—	5, 6-8, 1	Pritchard <i>et al.</i> (1964). J.Sci.Fd Agric. 15, 690
Herring Norway	15	6,2	—	5, 5-7, 0	Pritchard <i>et al.</i> (1964). J.Sci.Fd Agric. 15, 690
Herring Iceland	23	6,8	—	6, 0-7, 7	Pritchard <i>et al.</i> (1964). J.Sci.Fd Agric. 15, 690
Herring Germany	6	6,5	—	6, 1-7, 2	Pritchard <i>et al.</i> (1964). J.Sci.Fd Agric. 15, 690
Herring U.K.	11	7,1	—	6, 6-7, 7	Pritchard <i>et al.</i> (1964). J.Sci.Fd Agric. 15, 690
Herring Canada	21	6,4	± 0,37	5, 9-7, 5	March <i>et al.</i> (1966). J.Fish.Res.Bd Can. 23, 395
Herring Norway	12	6,8	± 0,49	5, 8-7, 4	Combs & Kifer (1970). Personal communication
Herring Canada	9	6,4	± 1,06	4, 4-7, 3	Combs & Kifer (1970). Personal communication
Herring Canada	7	7,8	± 0,50	7, 3-8, 6	Power <i>et al.</i> (1969). Fish.Res.Bd Can.Technical Rept.No.114
6 Anchovy	25	6,4	—	4, 7-7, 3	Pritchard <i>et al.</i> (1964). J.Sci.Fd Agric. 15, 690
Anchovy	5	6,3	± 0,55	5, 8-6, 9	March <i>et al.</i> (1966). J.Fish.Res.Bd Can. 23, 395
Anchovy,Peru, Chile		6,2	± 0,26	5, 8-6, 7	Brüggemann <i>et al.</i> (1969).Tierphysiol. Tierernähr. Futtermittelk. 25, 128.
Anchovy Peru	31	6,85	± 0,71	5, 3-7, 8	Combs & Kifer (1970). Personal communication
Anchovy Chile	8	6,2	± 0,70	5, 3-7, 1	Combs & Kifer (1970). Personal communication
White Fish U.K.	31	6,2	—	5, 0-8, 3	Pritchard <i>et al.</i> (1964). J.Sci.Fd Agric. 15, 690
White Fish S. Africa	7	7,3	—	6, 95-7, 6	Amoraal (1964). Rep.Fishg.Ind.Res.Inst.Cape Tn. 18, 73
Fish Meal S.Africa	17	6,9	—	6, 7-7, 2	Amoraal (1964). Rep.Fishg.Ind.Res.Inst.Cape Tn. 18, 73
Fish Meal S.W. Africa	17	7,0	—	6, 8-7, 4	Amoraal (1964). Rep.Fishg.Ind.Res.Inst.Cape Tn. 18, 73
Fish Meal S. Africa	52	7,1	—	5, 8-7, 7	Pritchard <i>et al.</i> (1964). J.Sci.Fd.Agric.15. 690
Fish Meal S. Africa	25	7,1	—	5, 1-7, 7	Brookes & Atkinson.(1966).Rep.Fishg.Ind.Res.Inst. Cape Tn., 20, 62.
Fish Meal S. Africa	8	6,9	—	6, 2-7, 6	Reid.(1968).Rep.Fishg.Ind.Res.Inst., Cape Tn., 22, 75.
Fish Meal S. Africa	8	7,1	—	6, 5-7, 6	Reid & Marshall (1969). Rep.Fishg.Ind.Res.Inst.,Cape Tn., 23,72
Menhaden U.S.A.	19	6,4	± 0,49	5, 0-7, 0	Combs & Kifer.(1970).Personal communication
Tuna U.S.A.	7	5,8	± 0,84	4, 8-7, 4	Combs & Kifer.(1970). Personal communication

In the long term, total amino acid data should be reduced by an availability factor. However, this should only be done when the appropriate factors are known for all feed ingredients. It would be wrong to propose adjusted values for fish meals if similar adjustments were not made for all other feedstuffs, since it is the relative values of one feedstuff to another that really matter. Also, where nutrient requirements for livestock have been calculated from the total amino acid levels in diets based on practical feeds, the requirement values will also need adjustment.

References

1. F.A.O. (1970). Available Amino Acid Content of Fish Meals. F.A.O. Fish. Rep., (92).
2. Combs G.F. (1968). Proc. Maryland Nutrition Conference for Feed Manufacturers. P.86.
3. F.A.O. (1970). Amino Acid Content of Food and Biological Data on Proteins. F.A.O. Nutritional Studies, No.24.

TABLE 6

TRUE DIGESTIBILITY (%) OF AMINO ACIDS

Animal	U.K. White fish meal	Meat and Bone meal	Soyabean meal	Soyabean meal	Soyabean meal
	Colostomised Hen	Colostomised Hen	Colostomised Hen	Rat	Rat
Lysine	93,5	73,4	90,9	87,2	
Methionine	91,7	73,6	87,6		91,1
Cystine	83,0	58,7	94,9		
Tryptophan	—	—	—		
Histidine	95,5	73,2	94,2		
Arginine	93,3	77,7	93,5		
Threonine	94,0	68,7	91,6		
Valine	92,9	74,9	91,9		
Isoleucine	93,2	75,2	91,3		
Leucine	93,5	75,4	90,8		
Phenylalanine	92,9	77,7	92,7		
Tyrosine	95,4	76,8	92,8		
Aspartic acid	86,1	52,4	91,0		
Serine	91,8	67,5	92,7		
Glutamic acid	90,9	70,7	92,9		
Proline	—	—	—		
Glycine	87,3	70,9	91,4		
Alanine	87,8	70,7	87,4		



GRAPHIC DESIGN PRINTING FINISHING

**91 The Maltings, Stanstead Abbots,
Nr. Ware, Herts. SG12 8HG
Telephone: (0920) 870355**
