QUEENSLAND DEPARTMENT OF PRIMARY INDUSTRIES DIVISION OF PLANT INDUSTRY BULLETIN No. 659

AVAILABLE LYSINE LEVELS IN PREPARED STOCK-FOODS AND STOCKFOOD INGREDIENTS IN QUEENSLAND

By B. W. SIMPSON, B.Sc., and R. M. NOBLE, A.Q.I.T. (Ind. Chem.)

SUMMARY

Using a modification of Carpenter's DNFB method, a survey has been conducted on available lysine levels in some prepared stockfoods and stockfood ingredients available in Oueensland.

There was considerable variation in available lysine level both between and within stockfood types. Vegetable proteins in general had a lower available lysine level than animal protein.

I. INTRODUCTION

Since lysine is one of the most important amino acids, particularly for monogastrics, the level of lysine will give some indication of protein quality. Not all lysine is nutritionally available and for this reason the term "available lysine" has been established. Available lysine is that lysine which has its epsilon-amino group free, i.e. unbound in the peptide chain. It normally refers to the lysine present in the actual protein; however, free lysine, whether indigenous or added, is also nutritionally available (but is not included in most analyses for available lysine).

Several methods have been developed, some chemical (Carpenter 1960; Rao, Carter and Framton 1963; Matheson 1968a, 1968b; Ostrowski, Jones and Cadenhead 1970; Holm 1971), some microbiological (Szmelcmann and Guggenheim 1967) and some biological (Atkinson and Carpenter 1970; Carpenter 1971; Flipot, Belzile and Brisson 1971) to determine the available lysine level in feedstuffs. The most commonly used chemical method is that of Carpenter (1960), which involves the reaction of the available lysine with 1–fluoro 2,4 dinitrobenzene. The final determination is carried out colorimetrically.

Because of the lack of data on locally available feeds, work was carried out by us to obtain results on a range of basic feed ingredients and prepared stockfoods.

II. METHODS AND MATERIALS

The basic method of Carpenter (1960) was followed throughout. The original method recommends prolonged cooling of the hydrolysate before filtering to minimize precipitation of dinitrophenol in the filtrate. This step was modified so that, after hydrolysis, the hydrolysates were allowed to cool to approximately room temperature and then transferred to a volumetric flask. After being adjusted to volume with distilled water and thoroughly mixed, the hydrolysate was then filtered. Precipitation of dinitrophenol did not occur using this procedure.

A similar procedure has been recently reported by Booth (1971), who found the cooling stage unnecessary. Another modification used involving a neutralization step has also been reported by Booth (1971).

Samples of stockfoods were selected at random from a large range of commercial products. A wide range of both pig and poultry foods was selected to cover the two most important monogastric groups. As well as on prepared stockfoods, analyses were carried out on a number of feed ingredients, e.g. grain, meat-and-bone meal and fish meal.

Carpenter's method was originally designed for "high protein" materials, e.g. fish meal and meat meal. On these materials, Carpenter found that the method gave approximately 91% recovery. On this basis he proposed a correction factor of $1\cdot09$ be used. It has since been found (Roach, Sanderson and Williams 1967; Matheson 1968a, 1968b; Booth 1971) that this correction factor is not suitable for many types of sample, e.g. grains and mixed feeds, in which reducing sugars formed cause partial reduction of the DNP-lysine complex. Attempts to establish recovery factors for each class of sample by using added DNP-lysine have proved unreliable, as it has been shown (Booth 1971) that the loss of indigenous available lysine is about half that estimated by loss of added DNP-lysine.

During the course of our work, Booth (1971) proposed the use of a separate factor for those samples containing high levels of carbohydrate. To check the validity of the $1\cdot09$ correction factor for our sample type, a mixture resembling a normal stockfood was formulated by us. This mixture contained sorghum, meatand-bone meal and fish meal (Table 1). Available lysine determinations were carried out on each component and on the mixture itself. DNP-lysine was added to each to estimate the recovery of DNP-lysine.

III. RESULTS AND DISCUSSION

In Table 1 various recoveries are given and compared. The results support the work by Booth in that the results obtained based on either half the loss of DNP-lysine or on those factors proposed by Booth not only agree with one another but also closely match the theoretical value.

These results support the opinion that neither Carpenter's factor of 1.09 nor factors based directly on loss of added DNP-lysine are suitable when dealing with samples containing high carbohydrate levels.

On the basis of this and other work it was decided to use the recovery factors suggested by Booth (Table 1). Although these may not be exact in all cases, they give a far more accurate picture than if Carpenter's factor of $1\cdot09$ was used throughout or if the results were based on recovery of DNP-lysine alone.

_	Meat-and-bone Meal		Sorghum Grain			Fish Meal			Mixture			
Composition in mixture (%) Crude protein (%) Recovery of added DNP-lysine (%)	8·0 48·8 91·1		85·0 8·2 69·6			7·0 63·2 96·3			15·4* 67·2			
	Available Lysine		Available Lysine		Available Lysine		Available Lysine					
Correction Factor	g/16 g N	%	% in Mixture	g/16 g N	%	% in Mixture	g/16 g N	%	% in Mixture	g/16 g N	%	Theoretical %
Nil Carpenter's (1.09) Loss of DNP-lysine added Half loss of DNP-lysine added Booth's factor	4·6 5·0 4·8 4·7 4·9	2·3 2·5 2·4 2·3 2·4	0·18 0·20 0·19 0·19 0·19	1·4 1·6 2·0 1·7 1·7	0·12 0·13 0·17 0·14 0·14	0·10 0·11 0·14 0·12 0·12	6·5 7·1 6·8 6·6 6·8	4·1 4·5 4·3 4·2 4·3	0·29 0·31 0·30 0·29 0·30	3·1 3·4 4·6 3·7 3·7	0·48 0·52 0·71 0·57 0·57	0·57 0·62 0·63 0·60 0·60

^{*} Calculated = 15.3

[†] Meat-and-bone meal and fish meal 1.05 and sorghum and mixture 1.2.

Tables 2 and 3 show results obtained for prepared poultry and pig feeds. The figures show considerable variation in available lysine levels both between and within food types. This illustrates the point that in evaluating a stockfood not only protein quantity but also protein quality must be considered.

TABLE 2

AVAILABLE LYSINE CONTENT OF POULTRY FEEDS

Correction Factor Used	Sample Type	No.	Crude Protein (%)	Mean	g Avail. Lysine/ 16 g N	Mean	Avail. Lysine (%)	Mean
1.2	Chick starter mash	1 2 3 4 5	19·6 21·5 16·6 21·9 17·6	19·4	3·5 3·1 2·7 3·0 2·9	3·1	0·69 0·68 0·45 0·66 0·51	0.60
1.2	Growing all-mash	1 2 3 4 5	15·4 18·4 16·9 19·0 16·8	17·3	3·4 3·0 3·0 2·8 2·4	2.9	0·53 0·55 0·50 0·53 0·40	0.50
1.2	Growing mash	1 2 3 4 5	17·2 17·2 19·1 19·0 17·5	18.0	2·6 3·0 2·5 2·7 2·5	2.7	0·45 0·51 0·48 0·51 0·43	0.48
1.2	Laying all-mash	1 2 3 4 5	22·0 23·6 19·0 17·4 18·4	20·1	3·0 2·3 3·3 2·9 3·3	3.0	0.65 0.55 0.63 0.51 0.60	0.59
1.2	Laying mash	1 2 3 4 5	18·1 21·5 18·5 18·8 19·6	19·3	2·9 2·8 3·3 2·8 3·0	2.9	0·53 0·61 0·60 0·52 0·58	0.57
1.2	Chicken mash	1 2 3 4 5	22·8 20·9 19·5 18·2 21·7	20.6	3·1 3·2 3·3 2·7 2·8	3.0	0·71 0·67 0·65 0·49 0·60	0.62
1.2	Chicken crumbles	1 2 3 4 5	21·0 17·4 21·2 21·4 20·1	20.2	3·5 2·9 3·6 3·5 3·1	3.3	0·74 0·51 0·77 0·76 0·62	0.68

Results are mean of at least duplicate analyses.

Table 4 shows results obtained on high-protein ingredients. It is of interest to note the results for whale solubles. Comparison with fish meals on crude protein basis only indicates a definite advantage in using whale solubles. However, the protein quality of whale solubles based on available lysine levels is significantly lower than that of fish meal. On these data, approximately $1 \cdot 3$ times as much whale solubles as of fish meal would be required to produce the same amount of available lysine.

TABLE 3

AVAILABLE LYSINE CONTENT OF PIG FEEDS

Correction Factor Used	Sample Type	No.	Crude Protein (%)	Mean	g Avail. Lysine/ 16 g N	Mean	Avail. Lysine (%)	Mean
1.2	Sow feed	1 2 3 4 5	16·6 16·1 17·9 16·7 18·1	17·1	2·5 3·1 2·3 3·0 2·9	2.7	0·41 0·49 0·42 0·50 0·52	0.47
1.2	Creep feed	1 2 3 4	21·3 21·4 16·3 22·3	20.3	2·4 2·5 3·2 2·9	2.8	0·51 0·54 0·53 0·64	0.56
1.2	Baconer feed	1 2 3 4 5	16·1 15·6 15·8 17·0 15·5	16.0	3·6 2·6 3·0 2·8 2·9	3.0	0·58 0·41 0·48 0·47 0·44	0.48
1.2	Sow and weaner feed	1 2 3 4 5	16·7 23·0 18·5 17·4 20·3	19·2	3·1 2·9 3·0 2·8 3·0	3.0	0·52 0·68 0·56 0·49 0·62	0.57
1.2	Miscellaneous pig rations	1 2 3 4 5	19·4 15·5 18·3 16·4 17·6	17:4	3·2 2·4 3·6 3·0 3·5	3.2	0·62 0·38 0·67 0·50 0·62	0.56
1.05	Concentrates	1 2 3 4 5 6 7	50·0 49·3 50·1 51·5 52·0 54·6 53·2	51.5	5·3 5·2 5·5 5·3 5·6 6·4 5·4	5.5	2·7 2·6 2·8 2·7 2·9 3·5 2·9	2.9
1.2	-	1 2	36·9 38·9	37.9	4·4 4·7	4.6	1·6 1·8	1.7

Results are mean of at least duplicate analyses.

Vegetable proteins in general have a lower available lysine level than animal proteins. Soybeans provide one of the richest sources of vegetable protein (Table 5). Sorghum grain, which constitutes a major part of both pig and poultry feed, is a good energy source but a relatively poor source of protein (Table 6). It is not only low in available lysine but also low in protein content. For feed purposes, sorghum grain is normally blended with fish meal or meat-and-bone meal to provide a balanced diet.

Obviously, for a more complete evaluation of the nutritional status of basic foods or prepared foodstuffs, many other parameters must be considered, including total amino acid analysis (Payne, Thorn and Packham 1972). Nevertheless, available lysine level remains one of the more important parameters obtainable by single chemical analysis.

TABLE 4
AVAILABLE LYSINE CONTENT OF CONCENTRATES

Correction Factor Used	Sample Type	No.	Crude Protein (%)	Mean	g Avail. Lysine/ 16 g N	Mean	Avail. Lysine (%)	Mea
1.05	Meat-and-bone meal	1 2 3 4 5 6	46·6 47·1 49·6 48·4 47·8 51·4	48·5	4·1 4·2 4·2 4·1 4·4 4·9	4.3	1·9 2·0 2·1 2·0 2·1 2·5	2·1
1.05	Blood meal	1	81.9	-	8.4		6.9	_
1.05	Fish meal (Chilean) Fish meal (S.W. African)	1 2	66·8 66·8	66.8	6·7 7·2	7.0	4·5 4·8	4.7
1.05	Whale solubles	1 2	84·7 82·1	83.4	4·3 4·4	4.4	3·7 3·6	3.7

All results are mean of at least duplicate analyses.

TABLE 5
AVAILABLE LYSINE CONTENT OF SOME GRAINS

Correction Factor Used	Sample Type	No.	Crude Protein (%)	Mean	g Avail. Lysine/ 16 g N	Mean	Avail. Lysine (%)	Mean
1.2	Wheat (off grade) (FAQ)	1 2	16·9 16·0	16.5	2·1 2·0	2.0	0·35 0·32	0.34
1.2	Barley	1 2	14·2 12·3	13.3	2·3 2·3	2.3	0·33 0·28	0.31
1·14	Soybean	1 2 3 4 5	39·0 43·2 34·0 38·4 38·8	38.7	5·3 5·1 5·4 4·9 5·5	5.3	2·1 2·2 1·9 1·9 2·2	2.0
1.2	Millet	1 2 3 4 5	8·4 9·1 10·6 11·3 10·6	10.0	1·9 2·0 1·7 1·7 1·4	1.7	0·16 0·18 0·18 0·19 0·15	0.17
1.14	Linseed meal	1	34.4		2.7		0.94	
1.2	Panicum	1	13·1		1.4		0.19	

Results are mean of at least duplicate analyses.

 $\begin{tabular}{ll} \textbf{TABLE} & 6 \\ \end{tabular} \begin{tabular}{ll} \textbf{AVAILABLE LYSINE CONTENT OF MAIZE AND SORGHUM GRAINS} \\ \end{tabular}$

Correction Factor Used	Sample Type	No. of Samples Analysed	Crude Protein Range (%)	Mean (%)	Avail. Lysine Range (%)	Mean (%)	
1·14	Maize	22	7·2–10·9	9·2	0·17–0·24	0·21	
1·2	Sorghum	15	9·3–13·5	11·2	0·15–0·23	0·21	

REFERENCES

- ATKINSON, J., and CARPENTER, K. J. (1970).—Nutritive values of meat meals. I.—Possible growth depressant factors. J. Sci. Fd Agric. 21:360.
- BOOTH, V. H. (1971).—Problems in the determination of FDNB—available lysine. J. Sci. Fd Agric. 22:658.
- CARPENTER, K. J. (1960).—The estimation of the available lysine in animal protein foods. Biochem. J. 77:604.
- Carpenter, K. J. (1971).—The availability of amino acids in fish meals. Feedstuffs Aug. 7:31.
- FLIPOT, P., BELZILE, R. J., and BRISSON, G. J. (1971).—Availability of the amino acids in casein, fish meal, soya protein and zein as measured in the chicken. Can. J. Anim. Sci. 51:801.
- HOLM, H. (1971).—Micro-method for determination of non-N-terminal lysine in milk and similar products. J. Sci. Fd Agric. 22:378.
- Matheson, N. A. (1968a).—Available lysine I.—Determination of non-N-terminal lysine in protein. J. Sci. Fd Agric. 19:492.
- MATHESON, N. A. (1968b).—Available lysine II.—Determination of available lysine in feedingstuffs by dinitrophenylation. J. Sci. Fd Agric. 19:496.
- OSTROWSKI, H., JONES, A. S., and CADENHEAD, A. (1970).—Availability of lysine in protein concentrates and diets using Carpenter's method and a modified Silcock method. J. Sci. Fd Agric. 21:103.
- Payne, C. G., Thorn, N., and Packham, R. G. (1972).—A survey of essential amino acid composition of livestock dietary ingredients used in eastern Australia. *Proc.* 1972 Australas. Poult. Sci. Conv., Auckland, N. Z.
- RAO, S. R., CARTER, F. L., and FRAMTON, V. L. (1963).—Determination of available lysine in oilseed meal proteins. *Analyt. Chem.* 35:1927.
- ROACH, A. G., SANDERSON, P., and WILLIAMS, D. R. (1967).—Comparison of methods for the determination of available lysine value in animal and vegetable protein sources. J. Sci. Fd Agric. 18:274.
- SZMELCMAN, S., and GUGGENHEIM, K. (1967).—Availability of amino acids in processed plant protein foodstuffs. J. Sci. Fd Agric. 18:347.

(Received for publication March 7, 1973)

The authors are officers of Agricultural Chemical Laboratory Branch, Queensland Department of Primary Industries, stationed at Indooroopilly.