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ABSTRACT

The availablility of lysine in sunflower meals was assessed using slope-ration assays for pigs and chicks. Availability was low for pigs (ca. 60%) but high for chicks (ca. 90%). There were no differences in lysine availability for expeller — or 'prepress' solvent-extracted meal. There was also little relationship between the pig results and chemical assays for estimating available lysine.

INTRODUCTION

The availability of amino acids in protein, especially lysine, may be affected by changes induced during processing. The heat and or pressure applied may alter the amino acid structure within the protein molecule. These changes include:

(1) Maillard-type reactions - a reaction between the free E-amino group of basic amino acids and carbonyl groups of reducing sugars (also called browning reactions). It is generally assumed that lysine, being a basic amino acid, is particularly susceptible to this type of reaction.

(2) Cross-linkage reactions - these include the breaking of existing bonds between amino acids within the protein molecule and the formation of new bonds.

(3) Actual destruction of the amino acid

Changes of the nature of (3) are detected by total amino acid analyses. However, changes of the nature of (1) and (2) do not normally affect the estimation of total amino acids as the acid hydrolysis used is capable of hydrolysing these linkages and releasing the amino acids. However, the enzymes used by pigs and chicks are far more specific and are unable to hydrolyse 'unnatural' linkages. Thus any changes to the natural structure of the protein molecule during processing may severely reduce the availability of amino acids. In addition, these changes may not be detected by total amino acid analyses.

Lysine availability is normally determined using the sloperation assay (growth or bio-assay). A diet, deficient in lysine, but adequate in all other nutrients, is formulated. The response to increasing dose levels of standard lysine is assessed. The test protein is incorporated into the basal diet, generally at similar levels to the standard lysine, and the response determined. Availability is expressed as the slope of the response to the lysine in the test protein as a percentage or proportion of the response to standard lysine.

Slope-ration assays are expensive and time-consuming to conduct and a number of chemical tests have been developed for more rapid estimation of lysine 'availability'. These assays are based on an estimation of the number of free \mathcal{E} -amino groups of lysine in a protein sample. The assumption is made that if the \mathcal{E} -amino group is free then that lysine molecule is nutritionally available. The two most widely adopted chemical tests are the Carpenter (1960) and Silcock (Roach *et al.*, 1967) assays. Both techniques are based on the reaction of fluorodinitrobenzene (FDNB) with \mathcal{E} -amino groups of lysine to form E-dinitrophenyl lysine (E-DNP lysine). This complex is resistant to acid hydrolysis and after the protein has been hydrolysed, is measured colorimetrically with the Carpenter (1960) or direct-FDNB assay. The Carpenter (1960) assay was, however, developed with animal proteins and subsequent work indicated that carbohydrates present in vegetable proteins could interfere with the stability of the \mathcal{E} -DNP lysine during acid hydrolysis. The Silcock or indirect-FDNB assay avoids this problem in that it measures the lysine remaining after dinitrophenylation which corresponds to 'unavailable' lysine. This is subtracted from the total lysine in the original sample to give 'available' lysine.

There is little information on the availability of lysine (and amino acids in general) in sunflower meal for pigs and chicks. Eggum (1973) reported true digestibility values of 0.88 and 0.87 for lysine in sunflower meal for rats and pigs respectively. Milner and Westgarth (1973) reported high lysine 'availability' estimates in sunflower meal as assessed by the Silcock (0.95) and Carpenter (1.02) assays. Batterham et al., (1978) reported a high Silcock value (0.92) in sunflower meal but lysine availability, as assessed by the slope-ratio assay, was low for rats (0.64).

The objectives of the work presented in this paper were to (1) determine the availability of lysine in sunflower meals for pigs, chicks and rats and (2) to assess the relative merits of the Carpenter and Silcock assays for estimating lysine 'availability' in these meals.

MATERIALS AND METHODS

The chemical composition of three sunflower meals is presented in Table 1. Two of the meals were 'prepress' - solvent extracted, the third expeller extracted.

Table 1.	Chemical	composition	(g/kg)	of three	sunflower	meals*
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No. Method of processing	1 'Prepress'— solvent	°Prepress' — solvent	3 Expeller
Crude protein	403	318	369
Dry matter	933	920	937
Ether extract	18	4	46
	163	252	160
Crude fibre		64	67
Ash	69		20.4
Gross energy (MJ/kg)	19.8	18.0	20.4
Essential amino acids			
Threonine	14	11	12
Valine	19	16	19
Methionine + cystine	13	11	11
Isoleucine	16	13	12
Leucine	25	18	23
	28	22	18
Phenylalanine + tyrosine		8	8
Histidine	10		11
Lysine	17	12	11
Arginine	35	27	-+
* Pottorham at al (1081)			

⁴ Batterham *et al.*, (1981).

+ Not adequately resolved.

Lysine availability in the meals was assessed using the slope-ratio assays for pigs and rats (Batterham *et al.*, 1981) and for chicks (Major and Batterham, 1981). The Carpenter and Silcock assays were conducted according to Carpenter (1960) and Roach *et al.*, (1967) respectively.

RESULTS

Lysine availabilities in the three meals are presented in Table 2. Lysine availability was lower for pigs (0.54 - 0.66) and rats (0.49 - 0.68) than for chicks (0.93 - 1.01). The chemical estimates of lysine 'availability' were variable with the Carpenter assay (0.46 - 0.88) but consistently high with the Silcock assay (0.87 - 0.94).

Table 2. Availability of lysine (proportion of total) in three sunflower meals as assessed by slope-ratio assays with pigs, rats and chicks and by two chemical tests^{*}.

	Slop	Slope-ratio assays			Chemical tests	
	Pigs	Rats	Chicks	Carpenter	Silcock	
Sunflower meal no. 1	0.59	0.64	-+	0.46	0.92	
2	0.54	0.49	1.01	0.88	0.94	
3	0.66	0.68	0.93	0.71	0.87	

* Batterham et al., (1981); Major and Batterham (1981) and unpublished data. † Not determined.

DISCUSSION

The low availability of lysine in the three meals for pigs indicates substantial damage is being done during processing. This has considerable economic implications in diet formulation for pigs as lysine is normally the first and major limiting amino acid in grower-pig diets (Batterham *et al.*, 1978). It is also particularly important nutritionally in that the total lysine content in sunflower meal protein is characteristically low (approximately 3.6 g/16 g N) compared to higher quality proteins (7 – 9 g lysine/16 g N for fish and milk proteins; 6 g/16 g N for soya-bean meal protein).

The higher lysine availabilities for chicks indicates that considerable species differences exist in their ability to hydrolyse and utilize lysine from different protein sources. This could be due to the chick having a more efficient digestive and absorption system or it may be capable of utilizing forms of lysine that the pig (and rat) cannot. Whatever the reasons, the results indicate that processing conditions do not appear to be having any major detrimental effect on lysine availability in these meals for chicks.

The lower assay results for the Carpenter compared to the Silcock assay is most likely a reflection of instability of the \mathcal{E} -DNP lysine during acid hydrolysis. The higher Silcock values did not reflect the low availability of lysine in these meals for pigs and rats. This indicates that reactions other than those involving the free \mathcal{E} -amino group may be involved in lowering lysine availability. This could include cross-linkage reactions between other amino acids within the proteing molecule. The Silcock values were, howeve, in general agreement with the chick values but the measured range in values was narrow. A wider range would be needed before definite conclusions regarding the suitability of the Silcock assay could be drawn.

The FDNB assays have been used as reference standards in the development of other laboratory techniques for estimating lysine availability. Thus it is likely that these techniques would also be inapplicable for pigs. The rat sloperatio values were, however, in general agreement with the pig results, and this assay could be used for estimating lysine availability in sunflower meals for pigs.

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