## The nutritive value of groundnut protein

### 1. Some effects of heat upon nutritive value, protein composition and enzyme inhibitory activity

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1. A laboratory-prepared groundnut flour defatted at room temperature (DGF) was subjected to dry heat or to pressure steaming under varying conditions of time and temperature, and its amino acid composition and nutritive value, the latter assessed by a chick growth test the gross protein value (GPV) test, were compared with those of some commercial groundnut meals. Trypsin inhibitor activity, available lysine value (ALV) and 'arachin' and 'conarachin' content and, in some instances, GPV were estimated in the heated samples.

2. The amino acid composition of the DGF and of commercial meals of high, medium and low GPV did not differ markedly, and the GPV of the DGF fell within the range of the three commercial samples.

3. Both dry and moist heat under specified conditions lowered ALV in the DGF and in the 'arachin' fractions, but had little effect on the ALV of the 'conarachin' fraction.

4. Moist, but not dry, heat rapidly removed trypsin inhibitor activity, and dry, but not moist, heat lowered GPV.

5. Neither 'conarachin' content nor trypsin inhibitor activity correlated with GPV in a range of commercial groundnut meals.

6. Dry heat (125° for 5 h) lowered nutritive value and 'conarachin' content but did not reduce the amount of total nitrogen soluble in sodium chloride solution.

7. No trypsin-inhibiting activity was found in the testa (skins) but these did exhibit growthdepressant properties for chicks. This property was removed by mild moist heat treatment.

8. 'Arachin' isolated from a commercial groundnut meal was valueless as a protein supplement for a cereal ration for chicks; 'conarachin' by itself, and mixed with arachin (1:3) was equivalent in GPV to the parent meal.

9. A factor other than those considered here, and possibly unassociated with processing, is primarily responsible for the differences in growth-promoting qualities of the commercial groundnut meals used in this work.

Since 1955 the Rowett Research Institute has participated in the collaborative programme of protein quality tests organized under the aegis of the Agricultural Research Council (Zuckerman, 1959). During the course of this work it became clear that samples of commercially available protein concentrates of a given type differed significantly from one another in nutritive value whether assessed by biological criteria or compared by various chemical and physical parameters (Boyne, Carpenter & Woodham, 1961; Duckworth, Woodham & McDonald, 1961). While the determination chemically of available lysine value (ALV) was shown to be a useful method for discriminating between samples of animal by-products the difficulties involved in applying this test to the plant proteins coupled with their increasing economic importance led us to inquire further into the factors responsible for these differences in nutritive value. It is conceivable that the differences may be attributable entirely to storage and processing but there remains a possibility that factors inherent in the seed may contribute to the final value. These could include the amino acid composition of the whole seed, the ratios between various protein components within the seed, the

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relative activities of enzyme inhibitors, and the levels of toxic components such as goitrogens and haemagglutenins.

The decision to begin these studies with an examination of groundnuts was influenced partly by the economic importance of this seed not only in this country but in a number of the developing countries, and partly by the fact that others had already shown that, unlike other major oilseeds, the bulk of the protein of raw groundnut could be readily extracted and divided into only two main fractions (Johns & Jones, 1916). It is true that each of these fractions can be further divided into numerous components, this having been demonstrated, for example, by electrophoretic methods (Tombs, 1965), but it was felt that any differences in nutritive value due to differences in very minor constituents would probably be small and it was hoped that a study of the two main fractions already known to differ markedly in nutritive value (cf. Sure, 1920; Baernstein, 1938) would be a useful starting-point.

It was decided in the first instance to examine the effect of various heat-treatments upon a meal prepared in the laboratory under conditions which imposed the minimum stress upon the proteins and then, in the light of these results, to examine some commercial groundnut meals.

Some of the results presented here have been briefly reported previously (Woodham & Dawson, 1966; Dawson & Woodham, 1966).

### EXPERIMENTAL

### Materials

Shelled South African groundnuts (variety: Natal Common) were purchased from H. S. Whiteside and Co. Ltd, Parkhouse Works, Camberwell, London, SE 5. Commercial groundnut meals were, with one exception, those collected for the Agricultural Research Council's investigation into protein quality tests, and the code letters and numbers are the same as those used in other publications embodying reports on work carried out with these samples. The country of origin and brief processing details for all of the groundnut meals with the exception of GN 24 and P 923 have been reported previously (Duckworth *et al.* 1961). The two exceptions were additional commercial meals purchased at random.

### Methods

Preparation of defatted groundnut flour (DGF). Groundnuts were skinned by hand and the kernels coarsely ground in a Christy and Norris mill before being stirred with diethyl ether at room temperature for 2 h, drained, re-extracted with fresh ether, air-dried at room temperature and ground. The resulting creamy-white powder contained 8.0% nitrogen, 3.2% oil and 10.4% moisture.

The preparation of heated samples of DGF. The powder was spread on trays to a depth of approximately 1 cm and heated in a forced-draught laboratory oven or in an autoclave under various conditions of time and temperature. Temperature recording was by means of a thermistor probe buried within the powder and coupled to an external recording meter. Dry heating lowered moisture content rapidly, only 1.2% remaining Vol. 22

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after 30 min at 100°. Autoclaving, on the other hand, raised the moisture content to 17% after 15 min.

The extraction of 'arachin' and 'conarachin'. The method was essentially that of Johns & Jones (1916). One part by weight of ground DGF was stirred with 10 parts of aqueous 10% (w/v) NaCl solution for 2 h, the extract filtered off and the filtrate treated with solid ammonium sulphate (30 g/100 ml). After standing for 1 h the 'arachin' was collected by centrifugation and decantation. Trichloroacetic acid was added to the supernatant liquid until no more 'conarachin' precipitated. The latter was collected in the same manner as was the 'arachin' and then both were washed successively with water-ethanol (50/50, v/v), ethanol (99%), ethanol-diethyl ether (50/50, v/v) and finally diethyl ether alone, before air-drying. Alternatively, the 'arachin' may be precipitated by CaCl<sub>2</sub> instead of (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub> (Tombs, 1965) and the 'conarachin' precipitated from the 'arachin'-free filtrate by adjustment of the pH to 2 with hydrochloric acid. The latter is the preferred technique and was used for the latter work.

Amino acid analysis. The ion-exchange chromatographic procedure of Moore, Spackman & Stein (1958) employing a non-automatic apparatus was used, cystine being determined on a separate sample oxidized by performic acid according to Moore (1963).

Trypsin inhibitor activity. The method of Anson (1938–9), as modified by Borchers, Ackerson & Sandstedt (1947), was used. To prepare the extracts for the estimation, a quantity of meal calculated to contain 80 mg nitrogen ground to pass a 50-mesh sieve was stirred vigorously for 15 min with 10 ml of 0.05 N-HCl in a centrifuge tube. The tube was stoppered, kept at 1° overnight and then centrifuged at 3000 rev./min for 10 min. One ml of the extract was mixed with 1 ml of 0.1% trypsin solution, made up to 10 ml and suitable dilutions of this were found by experiment.

ALV. The method of Carpenter (1960) was used.

GPV. The method of Heiman, Carver & Cook (1939) as modified by Duckworth et al. (1961) was used.

### RESULTS

The amino acid composition of the laboratory-prepared DGF was compared with that of several commercial groundnut meals, and the results are shown in Table 1. The nutritive value of the DGF determined by the GPV method—a chick growth method—was intermediate between that of GN 12 and GN 2 which are respectively the best and worst of a range of some twenty commercial meals (Duckworth *et al.* 1961). The DGF, GN 2 and GN 24 were evaluated at the same time (1964), but insufficient of GN 12 remained to allow the original value (obtained in 1957) to be checked. The sample of GN 2 had been kept throughout the intervening period at  $-10^{\circ}$  in an atmosphere of nitrogen. Although there are variations in the contents of individual amino acids between the meals no definite correlation with GPV was noticeable and the contents of the three important essential amino acids—lysine, methionine and cystine—are very similar.

Dry and moist heat over temperature ranges which were not intended to be com-38 Nutr. 22, 4

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parable but which were chosen because they brought about a progressive decline in the quantity of nitrogen soluble in NaCl solution were applied to the DGF, but only with dry heating was this decline accompanied by a decrease in nutritive value as indicated by GPV (Table 2). Dry heat also caused a lowering of the GPV of a commercial groundnut meal from 43 to 24. The 'conarachin' fraction was reduced more rapidly than was the 'arachin' fraction. Whereas the 'arachin' nitrogen was reduced only from 67.0 to 64.8% of the total nitrogen by dry heat ( $125^{\circ}/5$  h), the 'conarachin' nitrogen was reduced from 18.0 to 3.9% of the total.

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Determination	Unheated DGF	GN 12	GN 24	GN 2
gpv (1964)*	48		43	41
GPV (1957)		64		32
Aspartic acid	11.23	11.57	10.39	12.75
Threonine	3.10	2.91	2.76	3.20
Serine	4.90	5.05	4.62	5.48
Glutamic acid	18.40	18.87	16.20	20.10
Proline	4.26	5.26	4.48	5.00
Glycine	5.22	6.44	5.23	6·50
Alanine	3.74	4.11	4.12	5.00
Valine	4.48	4.22	3.28	4.88
Cystine	1.23	1.42	1.32	1.43
Methionine	1.39	I:47	1.31	1.28
Isoleucine	3.67	4.33	3.48	3.72
Leucine	6.53	6.79	5.91	6.42
Tyrosine	3.34	4.29	5.61	4.03
Phenylalanine	5.23	5.67	7.33	5.90
Lysine	3.49	3.97	3.22	4.02
Histidine	2.71	2.44	2.22	2.65
Arginine	15.00	10.97	10.32	16.00
$NH_3$	2.12	2.09	2.18	2.27
	100.16	101.87	94·84	111.04

Table 1. Gross protein value (GPV) and amino acid composition (g|16 g N) of three groundnut meals compared with unheated defatted groundnut flour (DGF)

\* se of means  $= \pm 6$ .

The estimation of both 'arachin' and 'conarachin' fractions in a range of eleven commercial groundnut meals failed to reveal any correlation with GPV (Table 3).

Both dry and moist heat lowered the chemically determined ALV in the DGF and in the isolated 'arachin' fractions, but there was little change in the values obtained for the 'conarachin' fractions (Table 4).

Skins are, of course, normally present in commercial groundnut meals and it is of interest to note the effect of including them. With skins present, the unheated DGF was of poor nutritive value—GPV = 36—but mild moist-heat treatment raised the value to 51—equivalent to unheated skinless DGF. This same mild heating lowered the available lysine value, however, in the DGF as well as in the 'arachin' and 'con-arachin' fractions isolated from it (Table 4).

Moist heat brought about rapid destruction of trypsin inhibitor activity, while even prolonged dry heat did not remove all activity (Table 5). No correlation was found

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between GPV and trypsin inhibitor activity for a range of commercial groundnut meals (Table 6). The units calculated in Table 5 represent absolute values, while 'per cent inhibition' is a simpler measure suited for rapid comparisons of materials of

Table 2. Effect of dry and moist heat on the amounts of protein fractions isolated from defatted groundnut flour (DGF) and from a commercial meal

(Nitrogen as % total nitrogen)

	Total NaCl-soluble	'Arachin'	'Conarachin'	Gross protein
Sample and treatment*	fraction	fraction	fraction	value†
Unheated DGF				
Skinless	95.3	67.0	18· <b>0</b>	48
With skins	93.3	57.1	18.1	36
GN 24	72.5	53.2	7.8	43
	Dry	7 heat		
DGF (skinless)				
75°/1 h	91.3	68.2	21.2	NI
100°/1 h	89.2	70.7	21.1	NI
125°/30 min	93.4	69·1	17.6	47
125°/5 h	90.0	64.8	3.9	32
140°/1 h	62.8	50.4	2.0	NI
140°/2 h	<b>2</b> 7·9	17.8	3.0	NI
150°/1 h	6.7	2.6	2.3	10
GN 24, 140°/1 h	28.3	2.9	1.2	24
	Moi	st heat		-
DGF				
Skinless, 108°/15 min	<b>8</b> 4·4	67.0	16.4	47
With skins,	NI	NI	NI	51
108°/15 min				
Skinless				
108°/30 min	78.4	60.4	9.4	NI
108°/45 min	70.3	56.2	6.6	50
116°/1 h	21.0	Trace	3.0	NI
108°/5 h	5.0	0	ō	44

NI = No information.

\* Further details of the samples and treatments are given on p. 590.

 $\dagger$  se of means  $= \pm 6$ .

# Table 3. Distribution of nitrogen in the globulins extracted from a series of commercial groundnut meals

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Sample*	'Arachin' fraction	'Conarachin' fraction	Total globulins	Gross protein value†
GN 2	9.8	2.8	12.6	32
19	43.2	22.2	65.7	36
15	22.8	9.2	32.0	40
24	39.2	12.6	51.8	43
17	23.2	6.2	29.7	44
I	54·I	19.5	73.6	48
13	56.6	12.1	68.7	49
6	55.3	<b>23</b> .6	78.9	51
9	62.9	12.7	75.6	54
16	53.3	8.6	61.9	60
12	57.9	12.0	69.9	64

### (Nitrogen as % of total nitrogen)

\* Details of the samples are given on p. 590.

 $\dagger$  These values are quoted from Duckworth *et al.* (1961) except for GN 24, which was tested for the first time in 1964. GN 2 was re-checked at this time and was unchanged. SE of means =  $\pm 6$ .

similar type such as the commercial groundnut meals listed in Table 6. The use of both parameters is indicated in Borchers *et al.* (1947).

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In Table 7 it is shown that the 'conarachin' fraction isolated from a commercial groundnut meal, when fed as a supplement to a cereal basal diet for chicks, is equivalent

Table 4. Effect of dry and moist heat on the availability of lysine (ALV) in defatted groundnut meal (DGF) and in commercial groundnut meals and in protein fractions isolated from them

	ALV $(g/16 g N)$			GPV of	
Sample and treatment*	Intact meal	'Arachin' fraction	'Conarachin' fraction	intact meal†	
Unheated DGF					
Skinless	2.91	2.21	4.53	48	
With skins	2.75	2.79	4.01	36	
GN 24	2.95	2.48	3.81	43	
GN P923	3.18	2.37	3.98	31	
	Dry	heat			
DGF (skinless)					
125°/30 min	2.91	2.77	4.32	47	
125°/5 h	2.64	2.14	4.34	32	
150°/ <b>1</b> h	1.23	NI	NI	10	
GN 24, 140°/1 h	1.94	NI	NI	24	
	Mois	t heat			
DGF					
Skinless, 108°/15 min	2.89	2.28	<b>3</b> ·94	47	
With skins, 108°/15 min	2.52	2.49	3.73	51	
Skinless, 108°/45 min	2.28	2.16	3.77	50	
Skinless, 108°/5 h	2.12	NI	NI	44	

\* Further details of the samples and treatments are given on p. 590.  $\dagger$  set of means =  $\pm 6$ .

Table 5. Effect of dry and moist heat on the trypsin inhibitor activity of defatted groundnut flour (DGF)

	Inhibitor units/ml extract
Sample and treatment*	$(\times 10^{-3})$
Unheated soya-bean meal†	30
Unheated DGF	
Skinless	26
With skins	16
Skins only from unheated DGF	0
Dry heat	
DGF (skinless)	
125°/30 min	18, 9
125°/5 h	2-4
140°/2 h	2
150°/1 h	I
Moist heat	
DGF (skinless)	
108°/15 min	0
108°/45 min	o

\* Further details of the samples and treatments are given on p. 590.

† Included as a reference material.

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in nutritive value both to the parent meal and to a 1:3 mixture with 'arachin' obtained from the same meal. The 'arachin', on the other hand, has no growth-promoting value.

Table 6. Trypsin inhibitor activity in a range of commercial groundnut meals

Sample*	( Inhibition	Gross protein value†	Sample†	Inhibition	Gross protein value†
GN 2	8	32	GN 1	12	48
4	12	32	10	9	48
11	17	36	13	ò	49
14	0	36	5	5	50
19	37	36	6	18	51
20	10	36	18	26	51
15	15	40	9	0	54
8	4	42	7	3	56
17	34	44	16	19	60
3	30	47	12	21	64
	* Details	of the samples	are given on p	. 590.	

 $\pm$  sE of means  $= \pm 6$ .

 

 Table 7. Gross protein values (GPV) of a commercial groundnut meal and of protein fractions isolated from it

Sample	GPV	se of differences
GN 24 'Conarachin' fraction 'Arachin' fraction 'Arachin-conarachin' mixture (3:1)	43, 44 39 0 41	±4·1 ±4·1 ±6·4

### DISCUSSION

The amino acid composition of the laboratory preparation (Table 1) and its GPV (Table 3) and ALV (Table 4) compared with that of ordinary commercial samples of groundnut meal suggest that heat applied during processing may not be such an important factor as has been assumed hitherto. The varieties from which the commercial meals have been prepared are not known, and only one known variety was used in this study for the preparation of DGF. It would seem that good nutritive value in a groundnut meal may well be closely associated with some factor inherent in the seed and that comparison of different varieties and growth environments could be profitable. As a corollary we may deduce that total amino acid analysis is of little value by itself in predicting nutritive value in groundnut meals, although this does not exclude the possibility that, taken in conjunction with the other protein constituents of the diet, the analysis may indicate characteristics useful in arriving at an over-all balance of amino acids in the mixed diet. Thus, above-average values for one or more essential amino acids could assume enhanced importance if the groundnut meal were to be fed in mixtures with other protein sources which were deficient in those particular amino acids. Chopra & Sidhu (1967a, b) have found varietal differences to be of little significance in a study of nine Punjab groundnut varieties in which amino acid

composition and nutritive value were examined. The amino acid compositions of these varieties, however, were strikingly similar. The subsequent inclusion of varieties grown in the USA revealed a range in total lysine content from 2.47 to 4.20 g/16 g N (A. K. Chopra, private communication), and a difference of this magnitude nourishes the hope that other significant differences may be found.

The studies on the heated meals demonstrate that, for the range of temperatures under consideration here for the two types of heating, 'dry' but not 'moist' heat depresses nutritive value although both types of processing cause a reduction in the quantity of nitrogen soluble in NaCl solution (Table 2).

The solubility of cottonseed meal nitrogen in aqueous sodium chloride solutions has been shown to be a useful indicator of nutritive value for chicks (Lyman, Chang & Couch, 1953; Barnes & Woodham, 1936). That high nitrogen solubility in groundnut meal is a desirable characteristic has been suggested by Liener (1958). Fontaine, Samuels & Irving (1944) found that moist heat abve 80° caused rapid denaturation of groundnut protein indicated by decreased nitrogen solubility in 1 M-NaCl. A similar trend is shown in our results (Table 2). Fontaine et al. (1944) found that dry heating at 118° for 2.5 h caused no significant change in nitrogen solubility. In the present study, dry heat at 125° for 5 h similarly produced little change in the level of NaCl-soluble nitrogen, but it will be noted that 'conarachin' level and GPV were both considerably affected (Table 2). Barnes & Woodham (1963) demonstrated that nitrogen solubility in NaCl solutions is of only limited value in predicting quality in groundnut meals, and it would appear from the present work that over-all nitrogen solubility in NaCl may give no hint of changes in the protein which affect nutritive value, and which may be detected by fractionation procedures such as those described.

It has already been stated that the particular conditions of heating which were used in this study were chosen solely because they brought about changes in the relative amounts of proteins with similar solubility characteristics which were related to changes in nutritive value. Although an investigation of commercial processing conditions was not intended, it may be of interest to consider to what extent the experimental conditions used are comparable with those used industrially.

Precise details of the commercial processing of groundnuts are difficult to obtain and in any event are variable from mill to mill, but some general information is available. After crushing, the seed is cooked by means of jacket or live steam, the temperature ranging from  $80^{\circ}$  at the top of the cooking kettle to  $95-115^{\circ}$  at the base, and the cooking time may range from 20 to 120 min (Rosen, 1958). If the hydraulic cooking process is used, temperatures may range from 65 to  $105^{\circ}$ , and the time from 15 to 120 min. Drying temperatures vary from about 75° to 115° and the moisture content during cooking from 6% to 15% (Fincher, 1958). If the meal is to be screwpressed, preliminary cooking temperatures may be as high as  $127^{\circ}$  and drying may be carried out at 138° (Fincher, 1958). During screw-pressing, high temperatures are caused by friction. No precise information has been found for groundnut, but for soya beans 150° and for cottonseed 170° have been quoted (Liener, 1958), although the material is only in the barrel of the press for up to 2 min (Fincher, 1958). Moisture

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content during screw-pressing may be 4-6% (Rosen, 1958). Solvent extraction temperatures are themselves low, depending upon the solvent used, but may rise to  $105^{\circ}$  during subsequent stripping of the solvent with live steam.

The moist heat conditions used by us  $(108-116^{\circ} \text{ for } 15-45 \text{ min})$  are substantially within the range quoted by Rosen (1958), and the moisture content during autoclaving (17%) is only marginally higher than the upper limit of 15% quoted by Fincher (1958). Somewhat more drastic conditions than ours were employed by Borchers & Ackerson (1950) (30 min at 121°) and their product did not have a deleterious effect on the performance of rats.

Anantharaman & Carpenter (1965) have reported that dry heating of experimentally prepared groundnut flour at  $121^{\circ}$  for 4 h, but not at  $107^{\circ}$  for 30 min, significantly depressed chick growth and this is in accord with our findings. These workers noted a lowering of chemically determined available lysine in their more severely heated meal, and it is of interest to note that we have shown a substantial reduction in total lysine in the 'conarachin' fraction isolated from DGF dry-heated at  $125^{\circ}$  for 5 h (Dawson & Woodham, 1966) in addition to a reduction in ALV in the heated DGF and in the 'arachin' fraction isolated from it (Table 4). A similar deterioration in nutritive value and ALV was found in a commercial groundnut meal dry-heated at  $140^{\circ}$  for 1 h (Table 4).

The comparative harmlessness of dry heat below  $100^{\circ}$  for up to 20 min has been demonstrated by Cama & Morton (1950). That prolongation of dry heating at a comparatively low temperature may be more harmful than heating for a shorter time at a higher temperature was shown by Mitchell, Hamilton & Beadles (1949). In the present study the effect of prolonging heating time has been examined. Dry heating for 30 min at 125° had little effect on either 'conarachin' level or GPV, but 5 h at the same temperature reduced the 'conarachin' from 17.6% to 3.9% and GPV from 47 to 32. Total globulins and 'arachin' were little affected.

While both dry and moist heat may be met with under commercial processing conditions, the results suggest that the levels of moist heat normally encountered are likely to have little effect upon nutritive value although they may have a considerable effect upon the solubility characteristics of the groundnut proteins. The higher temperatures associated with the dry heating during screw-pressing, on the other hand, could affect the nutritive value of the resulting meal, particularly if the time of heating is prolonged.

That 'arachin' alone does not support normal growth in rats, that 'conarachin' does and that mixtures of the two proteins in the proportions in which they occur in the groundnut produce satisfactory growth have already been demonstrated (Sure, 1920; Baernstein, 1938; Macheboeuf & Tayeau, 1942). Similar results have now been obtained for 'arachin' and 'conarachin' fractions isolated from a commercial groundnut meal in growth experiments in which they have been used as supplements to cereal rations for chickens (Table 7).

The fact that the 'conarachin' fraction was reduced more rapidly than the 'arachin' fraction on heating the parent DGF (Table 2), coupled with the knowledge that whereas the former is of good nutritive value the latter is by itself of little value

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(Table 7), suggested the possibility that the level of 'conarachin' might indicate the amount of heat suffered by the meal and hence its nutritive value. 'Conarachin' is the fraction isolated by the specified precipitation procedure, and fluctuations in the amount of it are presumably due to an alteration in solubility characteristics caused by the heat treatments used. From Table 2 it would seem that the denatured 'conarachin' which is no longer precipitated after the heat treatment is of lower nutritive value than the undenatured material. However, no correlation was found between nutritive value and 'conarachin' content for a range of eleven commercial meals.

It is of interest that the ALV for the isolated 'conarachin' fractions appear to be little affected by the extent of either dry or moist heating applied to the parent meal, whereas the ALV of both the meals and the 'arachin' fractions are depressed. Recoveries of added DNP lysine ranged from  $92 \cdot 5\%$  to 100% for the unheated and heated samples of DGF. Smaller losses occurred with the isolated protein fractions, the lowest recovery being 97%, and in consequence more weight is attached to the absolute values for the fractions than to those for the DGF. In all instances, however, trends should be considered more trustworthy than the absolute values.

The rapid destruction of trypsin inhibitor activity by even small amounts of moist heat is obvious. Borchers et al. (1947) similarly found rapid disappearance of inhibitor activity in soya-beans when they autoclaved a solvent-extracted meal for 45 min at 109°, as well as the relative ineffectiveness of dry heat. After dry heating for 2 or 4 h at 135°, 57% and 28% activity remained respectively. Quantitative results for the destruction of trypsin inhibitor in groundnuts appears to us to be lacking, though some authors (cf. Cama & Morton, 1950) have attributed improvement in feeding value to the inactivation of a trypsin inhibitor. Borchers & Ackerson (1950), investigating eleven legume seeds including groundnut, found no correlation between trypsin inhibitor content and rat growth, after autoclaving at 121° for 30 min. The reason for the depression of nutritive value only by dry heating might, however, be explained in terms of trypsin inhibitor activity. Whereas dry heating brings about a reduction in 'conarachin' content and ALV it is insufficient to significantly affect the trypsin inhibitor activity, the net result being a lowering of nutritive value. Moist heating, however, while causing a reduction in 'conarachin' content and ALV, offsets this by destroying trypsin inhibitor activity completely, the net result in this particular instance being a relatively unchanged GPV. This would also explain the similar results of Borchers & Ackerson (1950), who found that nutritive value judged by rat growth was unaffected by autoclaving groundnut meal though trypsin inhibitory activity was significantly diminished.

The evidence for the presence of a growth inhibitor in the groundnut skins is interesting. No trypsin inhibitor activity was found in them but mild moist-heat treatment removed their growth-depressant properties completely. It can be assumed therefore that ordinary commercial processing will deal with them adequately and there is no case for removing skins as a quality improver in the preparation of feeding meals.

From the results obtained in the work described in this paper it is not possible to decide which factor or factors are chiefly responsible for the variability in nutritive

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value of groundnut meals. If trypsin inhibitor activity and 'conarachin' content be taken as manifestations of such factors, it is of interest that in the experimentally heated samples fluctuations in the levels of these factors parallel both the heat applied and the nutritive value of the samples. It is equally certain, however, that in a range of commercially prepared groundnut meals neither of these parameters, singly or in conjunction, could be used to predict the growth-promoting value of the meals for chicks. In the light of these observations one must deduce that other factors are responsible for nutritive value in commercial groundnut meals. Whereas trypsin inhibitor activity is likely to be affected more by moist heating conditions, 'conarachin' level is affected by both moist and dry heat over the normal range of times and temperatures used in industry. It seems likely then that drastic processing should cause changes in the levels of one or other of the parameters examined, and the fact that neither can be used to predict the nutritive value of commercial meals for the growing chick implies that some other factor, possibly unassociated with processing, may be primarily responsible for quality determination.

The DGF used throughout the work described in this paper has been prepared from a single sample of groundnuts of one variety. The study is now being continued on a number of other varieties and strains obtained from Nigeria, USA and Iraq, as part of a project which forms one of the UK contributions to the International Biological Programme.

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