The nutritional value of poor proteins fed at high levels

2.* Species differences

BY K. ANANTHARAMAN AND K. J. CARPENTER

School of Agriculture, University of Cambridge

AND M. C. NESHEIM

Department of Poultry Science, Cornell University, USA

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1. The same six high-protein diets that were fed to rats (Carpenter & Anantharaman, 1968) have now been fed to chicks.

2. The net nitrogen retention by chicks, per 100 kcal consumed, was for each diet greater by 20–50% than the retention by rats. For a diet based on a mixture of commercial protein concentrates the NDpCal % was 19·1 (±0·18); this value greatly exceeded the theoretical maximum of 14·6 obtained from the equations of Miller & Payne (1963).

3. Although groundnut protein plus lysine has a calculated chemical score of only 56, chicks receiving this at a high level retained N at the same rate (NDpCal % of 17·5–17·9) as those receiving a diet which included egg protein at the level (26% of the dietary ME) predicted to be optimal for them.

4. The 'endogenous+metabolic' losses of N were in almost the same proportion to metabolic size for chicks as for rats.

The preceding paper (Carpenter & Anantharaman, 1968) has reported the results of a rat-feeding experiment in which two protein sources of low chemical score, groundnut (+lysine) and wheat gluten, each supported better growth at high levels than was predicted from the original equations of Miller & Payne (1961, 1963). A further prediction of Miller & Payne (1963), based on the results of Summers & Fisher (1961) and others, was that chicks would show the same NDpCal % value as rats for any given diet balanced in its content of nutrients other than protein. The present paper reports the results of feeding to chicks the same six diets as were used in the previous experiment with rats.

EXPERIMENTAL

Diets

The diets A–E, G and H used for the main feeding experiment were further portions of those already fed to rats (Carpenter & Anantharaman, 1968). Diet J used in the digestibility trial was made up of groundnut meal (X. 512) 38, partially hydrogenated fat 5, cellulose 5, mineral mix 6, vitamin mix 1, lysine hydrochloride 0·2, choline chloride 0·3, chromium bread (Kane, Jacobson & Moore, 1950) 1, and maize starch to 100. In diet L the level of groundnut meal was 55 and of lysine hydrochloride 0·3. These two diets differed from diets D and E respectively in their levels of groundnut meal and in the ratio of lysine to groundnut protein.

* Paper no. 1: *Br. J. Nutr.* (1968), **22**, 183. Some of these results have been communicated in a preliminary form (Anantharaman & Carpenter, 1967).

Digestibility determination

Chicks were operated on to allow separate collection of faeces and urine, and the experiment was conducted to determine the apparent digestibility of nitrogen in the manner already described (Nesheim & Carpenter, 1967; Expt 4). The procedure was carried out satisfactorily with three birds for diet J, and with four for diet L.

The metabolizable energy (ME) values of diets A–E, G and H used in the main feeding experiment, were determined from analyses of mixed excreta from birds in that experiment (described below). Excreta were collected separately from four of the six cages allocated to each dietary treatment over the last 4 days of the experiment. The analyses for chromium and N and the determinations of gross energy followed the procedures already described (Carpenter & Anantharaman, 1968). The ME values of the diets (kcal/g) were corrected to N equilibrium by making a deduction of 8·22 cal for each mg N retained per g diet fed (cf. Hill & Anderson, 1958). The retention over the collection period was calculated as the difference between the N content of 1 g food and the N content of that quantity of excreta which contained the same weight of chromium as was present in 1 g food.

Main feeding experiment and carcass analysis

One hundred and fifty cockerels of a fast-growing broiler strain (Light Sussex × Rhode Island Red) were purchased at 1 day old and reared for 10 days on a commercial diet. They were then weighed and ninety-six of medial weight were selected, the remainder being discarded. Of the selected birds, the sixteen lightest formed the first stratum, the sixteen next in weight formed the second stratum and so on for six strata. The birds of each stratum were randomized into eight pairs. The first pair (treatment K) were killed immediately and the remainder were allocated at random to seven cages each of which received a different experimental treatment (A–E, G or H). The cages have been illustrated and the use of stratified designs of this kind has already been described (Carpenter, March, Milner & Campbell, 1963).

The birds (six cages per treatment) received their diets *ad lib.* for 10 days, and the experimental procedure then followed that used for the rats including the carcass analysis at the end of the feeding period (Carpenter & Anatharaman, 1968). As before, two adjacent strata were pooled to give a unit of four birds for each mincing. In this way three values were obtained for each dietary treatment.

Calculations of net N retention

In general in this experiment the mean initial live weights of the chicks assigned to each dietary treatment differed by less than 1 g from the mean weight (124 g) of those killed at the beginning of the experiment, and it has been assumed that their initial N contents were the same. The chicks allocated to diet D were exceptional in having a mean initial weight 1·8 g greater than those of treatment K. If allowance had been made for this the net N retention per chick would have been reduced by 0·05 g or approximately 2 %.

The chicks receiving diets E, G and H more than doubled their weight during the period of the trial. It was therefore decided to estimate the 'endogenous + metabolic N loss' of the experimental birds as being proportional to their metabolic size, rather than to assume that the loss was the same as that determined with the protein-free group in each stratum. Mean metabolic size was, in turn, estimated as the mean of the initial and final weights raised to the power 0·73. Calculated in this way, the mean metabolic size of the fastest-growing birds (diet H) was 1·7 times that of the birds receiving the protein-free diet.

The values in Table 2 are set out so that the estimates of net protein utilization can, if it is wished, be re-calculated without this adjustment, as was done for the rats (Carpenter & Anantharaman, 1968). Even with diet H, the effect would be to reduce the value for NPU by only 2·8 units or less than 5 %.

RESULTS

Digestibility determinations

The results for the apparent digestibility of the N in diets J and L are set out in Table 1. The overall mean estimate of the digestibility of the N in the form of 'groundnut plus lysine' was 82 %. This was similar to the corresponding value of 83 % obtained with rats (Carpenter & Anatharaman, 1968).

Table 1. *Apparent digestibility for individual chicks of the nitrogen in diets containing high levels of groundnut plus lysine*

	Diet J	Diet L
	81·9	81·8
	80·0	72·5
	87·4	82·6
		83·0
Mean	83·1	80·0

The results of the metabolizable energy determinations are set out in Table 2, in comparison with those previously found with rats. The diets containing groundnut again gave much the lowest values in the series, but overall the values tended to be slightly lower, by an average of 0·09 kcal/g (i.e. by approximately 3 %), than those found with the rats. Metabolism of digested N to uric acid rather than to urea (and the consequent difference in the correction of the value to N equilibrium) accounts for up to 0·03 kcal/g of the difference.

N retention experiment

The experiment was carried out as planned and none of the birds had to be removed for any reason. No problem was encountered in reducing the carcasses, including feathers, to a sufficiently homogenous state for sampling. Agreement between replicated N analyses was good and the final standard error of the treatment means (of 2·7–3·0 g N/100 g carcass) was 0·022, representing a coefficient of variation of less than 1 %.

The essential results are summarized in Table 2. The standard errors shown were

Table 2. *Live weight and nitrogen content (treatment K) and performance of the chicks receiving diets A–E, G and H in the main feeding experiment*

	Treatment K (killed at start)	Diet A (protein-free)	Diet B (wheat gluten)	Diet C (wheat gluten)	Diet D (groundnut)	Diet E (groundnut)	Diet G (egg)	Diet H (mixed protein)	SE of treatment means
N × 6·25 on diets (%)	—	0·66	18·7	29·6	17·4	28·1	25·0	26·1	—
Metabolizable energy of diets (kcal/g)	- -	—	3·33	3·32	2·84	2·53	3·31	3·19	±0·020
(Corresponding result with the rat)*			(3·38)	(3·39)	(2·83)	(2·63)	(3·49)	(3·35)	—
Final live weight/chick (g)	(123·9)	95·3	137·5	150·4	180·5	271·6	280·3	329·1	±6·4
Food eaten/chick (g)	—	84·1	114·7	112·1	185·3	273·1	253·7	280·1	±6·5
Metabolizable energy eaten/chick (kcal)	—	—	381	369	526	691	840	893	±22
Gain (g)/g food eaten	—	—	0·129	0·248	0·296	0·511	0·679	0·786	—
N/carcass (g)	(3·22)	2·75	3·92	4·34	4·93	7·44	8·33	9·26	±0·17
Estimated 'endogenous and metabolic' N loss/chick (g)	—	0·47	0·54	0·56	0·60	0·72	0·76	0·80	—
Net N gained/chick (g)	—	0	1·24	1·68	2·31	4·94	5·88	6·84	±0·18
NPU_{op} of diets	—	—	36·5	32·0	44·5	40·4	57·9	54·2	±0·69
(Corresponding result with the rat)*			(24·3)	(23·9)	(37·1)	(34·0)	(46·5)	(39·7)	
NDpCal % of diets†	—	—	8·21	11·4	11·0	17·9	17·5	19·1	±0·18

* Values obtained for the same diets fed to rats (Carpenter & Anantharaman, 1968).

† Calculated here as '25 × net N retained (g) per 100 kcal ME eaten', using the determined ME values shown in the table.

obtained from an analysis of variance in which strata effects were removed; they had 12 degrees of freedom when measures for diet A were included, and 10 degrees when they were not. It is seen that the results for net protein utilization (NPU_{op} with the chicks were, for each diet, higher than those obtained with the rats.

The chicks receiving the protein-free diet (A) apparently lost 0·47 g N during the 10-day experimental period. The loss found with the rats was 0·22 g N (Carpenter & Anantharaman, 1968). In terms of relative metabolic size expressed as $W^{0.73}$ the equivalent value for the 'protein-free' chicks with an average weight of 110 g (as compared with 44 g for the rats) would be 0·43 g. There was not therefore any significant difference between our values for the two species when they were compared in this way.

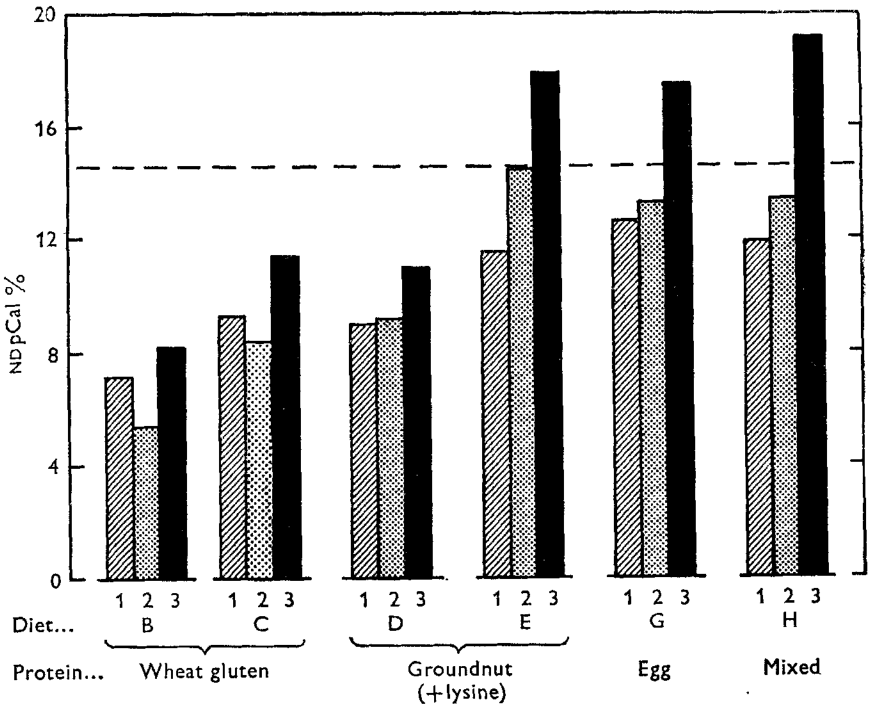

Fig. 1. NDpCal % (based on determined ME values) for the six diets under investigation; 1, as predicted for rats by the equation NDpCal % = $PS \times 1{\cdot}25/(100+0{\cdot}064PS)$ (P. R. Payne, private communication); 2, as determined with rats (Carpenter & Anantharaman, 1968); 3, as determined with chicks; ——, level of theoretical maximum from equation of Miller & Payne (1961). S has been taken as 40 for wheat gluten, 56 for groundnut plus lysine, 100 for egg and 73 for 'mixed' protein.

DISCUSSION

The high NDpCal % obtained with diet E containing a high level of groundnut (plus lysine) as the protein source is a confirmation of the indirect estimates (from live-weight gain alone) obtained previously by Carpenter & De Muelenaere (1965). As is seen from Fig. 1, the value obtained was higher than that obtained with rats, which was itself higher than the value predicted to apply to either species. It is seen that the other diets also gave more efficient N retention with the chicks than with the rats. This is contrary to the hypothesis of Miller & Payne (1963).

The N retentions by chicks receiving diets E, G and H were also all above an NDpCal value of 14·6 %, the theoretical maximum obtainable according to the original general prediction equation (Miller & Payne, 1961), though there is not the same upper limit in the modified equation (P. R. Payne, private communication) shown in the footnote to Fig. 1. From the values given by Carew, Hopkins & Nesheim (1964) we have calculated that their broiler chicks also attained an NDpCal value of approximately 18 % on diets similar to diet H of the present series.

Differences between the rat and the chick for a particular material could, of course, be explained by differences in the make-up of the ideal protein for the two species, and thus by differences in the scoring of the material for the two species. However, this could not explain our results with diet G based on egg protein, which has been considered optimal for the rat (Miller & Payne, 1961), but has given an even higher NDpCal % value with the chick.

Another possible explanation for the higher values obtained with chicks is that their response curve to increasing levels of protein is not identical with that of the rat, and that it falls off less rapidly. This might well be expected in a species that grows, in proportion to its size, at a faster rate than the rat. The discrepancy between the two species examined here naturally throws a doubt on the utility of equations worked out with rats, for predicting the relative efficiency of utilization of proteins at different dietary levels in human diets (Platt, Miller & Payne, 1961). Certainly there seems to be no theoretical basis for assuming that with two species growing at such different rates the results obtained with one can readily be applied to the other.

The low ME value of groundnut flour

The diets (D and E) containing high levels of groundnut meal have given ME values with both rats and chicks that show them to contain a considerable portion of indigestible material. Thus the ME of diet E is 65·7 % of the gross energy with rats (Carpenter & Anantharaman, 1968) and 63·2 % with chicks, as compared with the expected 86·5 % from the formula of Miller & Payne (1959).

The formula of Miller & Payne (1959) assumes 95 % digestibility of the gross energy of all ingredients, including proteins, and subtracts 7·5 kcal/g dietary N, the generally assumed gross energy of the urine per g N excreted under condition of N balance in mammals. Our 'determined' value for rats represents a direct measure of the digestibility of the gross energy, with the same correction applied for digested N as in the formula used for calculation. If we had corrected the digestible energy just by the gross energy of the urine produced, the retained protein would be credited with its full gross energy, rather than its physiological fuel value.

Since it is only in diets D and E that large discrepancies were found between determined and predicted ME values, the discrepancies must arise from the presence of the groundnut flour. Our sample (X. 512) had a gross energy of 4·52 kcal/g DM and in a preliminary experiment with chicks (not described in detail here) we determined its ME as 2·56 kcal/g DM, or 56·7 % of the gross energy. With this value, the calculated ME for diet E was very close to that determined. Zablan, Griffith, Nesheim, Young & Scott (1963) have reported a ME value of 2·93 kcal/g DM and Rajaguru, Vohra & Kratzer (1966) a value of 2·68 kcal/g DM. Our own value is therefore the lowest of the three values, but all three are well below the value calculated from the standard formula (Miller & Payne, 1959).

Since the apparent digestibility of the crude protein of our groundnut flour was 82 %, and the true digestibility presumably about 87 %, some other substantial components must have been less well digested. Probably these were the structural carbohydrates.

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