

Some effects on hepatic lipid of varying the dietary carbohydrate intake.

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It has been shown that when adult rabbits of similar weight are put on diets containing the same amount of protein but with varying amounts of sucrose, then the amount of the hepatic lipid present when the animal has lost a third of its weight is greater with the larger sucrose intakes (Macdonald & Gharavi, 1960).

Further experiments have shown that the total amount of lipid present in the liver is related to the mean daily sucrose intakes. When other carbohydrates (glucose and maize starch) replaced the sucrose a similar relationship was found. This accumulation of liver lipid with increased carbohydrate intake is not due to deficient calorie intake. More extensive accumulation of hepatic lipid occurs with sucrose as the dietary carbohydrate than with starch.

Using silicic-acid column chromatography, the liver lipid was divided into four fractions, namely (1) sterol esters, (2) triglycerides and free fatty acids, (3) di- and mono-glycerides and free sterols and (4) phospholipids. Comparison with normal hepatic lipid showed that the composition of the lipid had altered on the excess-sucrose diet. With increase in dietary sucrose the proportion of sterol esters in the total lipid fell while that of the phospholipids rose.

When the amounts of the various hepatic lipid fractions were calculated there seemed to be a relationship between the amount of sterol esters and the mean daily sucrose intake. The other three fractions, however, all showed a significant relationship to the mean daily sucrose intake.

Thus adult rabbits on a low intake of an adequate protein together with varying amounts and types of carbohydrate show that the amount of the hepatic lipid is related to the mean daily carbohydrate intake, and the lipid present under these circumstances does not have the same composition as normal.

REFERENCE

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The One Hundred and Thirty-ninth Meeting of The Nutrition Society (Sixtieth of the Scottish Group) was held in the Strathcona Club, the Rowett Research Institute, Bucksburn, Aberdeen, on Friday, 17 February 1961, at 1.10 p.m., when the following papers were read:

The absorption of food from the gut of the fowl. By W. BOLTON, *Agricultural Research Council Poultry Research Centre, Edinburgh 9*

Adult male fowls were fed P.R.C. Breeders' Pellets (14.6% crude protein, 3.1% ether extract, 47.9% available carbohydrate, 5.6% cellulose) for at least 4 weeks to

obtain equilibration in the gut. Six birds were killed by cervical dislocation. The small intestine was divided at Meckel's diverticulum; each portion was subdivided into three equal lengths. The contents of the six, and of the duodenum, were removed by digital manipulation, avoiding excessive pressure. Each bird was completed within 15 min of death to minimize post-mortem changes.

Absorption and secretion take place continuously in the gut; to relate gut contents to food, cellulose was used as marker and analytical percentages of crude protein and available carbohydrate were calculated as proportions of the cellulose content. The results show that somewhere between the mouth and duodenum, the protein content increased tenfold. Ingesta in the gizzard were fairly dry (33% dry matter); in the duodenum the moisture content had increased enormously (15% dry matter). The duodenum is extremely rich in goblet cells and the increased protein may be due to mucus secretion. Available carbohydrate content decreased from food to duodenum and then increased. Takadiastase would be unlikely to hydrolyse the sugars of mucus, but could do so once mucoprotein had been attacked by proteolytic enzymes. The observation, therefore, supports the suggestion that the increased protein was mainly mucus.

The cellulose content of the duodenal contents was low (0.37%) hence the experiment was repeated with birds fed a diet of 20 parts layers' mash and 80 parts ground oat hulls (crude protein 5.3%, ether extract 3.6%, available carbohydrate 9.0%, cellulose 2.7%). The results confirm the observation for protein.

Crude protein, ether extract and available carbohydrate as proportions of cellulose content (cellulose=1)

		Food	Duodenum	Small intestine					
				1	2	3	4	5	6
Expt 1	Crude protein	2.6	24.7	7.2	2.6	1.9	1.3	1.0	0.8
	Available carbohydrate	8.6	5.5	5.8	2.8	1.5	1.0	0.7	0.7
Expt 2	Crude protein	0.2	10.9	0.6	0.3	0.1	0.2	0.1	0.1

Both experiments indicate tremendous outpouring of protein into the gut, probably by the duodenum, and equally rapid absorption. When a bird scours, re-absorption would be adversely affected and the subsequent loss of protein would explain why such birds lose weight rapidly.

Loss of liver glycogen after administration of protein or amino acids.

By H. N. MUNRO, *Department of Biochemistry, University of Glasgow*, and CATHERINE M. CLARK and G. A. J. GOODLAD, *Department of Biochemistry, St. Salvador's College, University of St. Andrews*

In a previous report (Clark, Goodlad, Chisholm & Munro, 1960) it was shown that the feeding of protein can cause rapid changes in the amount of adenosine triphosphate in the liver. Rats were given a carbohydrate-rich meal in order to produce a high concentration of adenosine triphosphate in the liver over a period of several

hours. When casein was then fed to such animals, the adenosine-triphosphate concentration fell rapidly. Further investigation showed that this fall was accompanied by a considerable reduction in the glycogen content of the liver.

In the present investigation, we have explored the changes in the glycogen content of the liver caused by protein administration. Rats were fed a meal of carbohydrate which caused deposition of glycogen in their livers, and were then given either casein, glycine, alanine, glutamic acid or olive oil. The casein and the amino acids each caused a large loss of glycogen from the liver. This action was already considerable within 1 h of administration. There was no change in glycogen concentration 2 h after feeding olive oil.

Coincident with the loss of liver glycogen after protein or amino-acid administration, there was a fall in blood sugar concentration. There was no change in muscle glycogen concentration.

In view of the well-known glycogenolytic action of adrenaline, the effect of administering casein or glycine to adrenodemedullated rats was examined. Both nutrients caused loss of liver glycogen, indicating that secretion of adrenaline is not an essential part of the mechanism responsible for this loss. A glycogenolytic action has also been observed when insulin is given to rats in the postabsorptive state (Levin & Weinhouse, 1958). In order to determine whether insulin participates in the action of protein and amino acids on liver glycogen concentration, rats were made diabetic with alloxan and glycine was then administered. This caused a loss of glycogen from the liver, indicating that an intact insulin-secreting mechanism is not essential for this action of glycine.

It is suggested that the loss of liver glycogen following administration of protein or amino acids is related to the extra energy required for the specific dynamic action of the absorbed amino acids.

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Further studies of nervous-tissue degeneration resulting from 'conditioned' copper deficiency in lambs. By B. F. FELL, R. B. WILLIAMS and C. F. MILLS, *Rowett Research Institute, Bucksburn, Aberdeen*

The method of producing copper deficiency and ataxia in lambs by feeding Mo and SO_4^{2-} supplements to their ewes has already been published (Mills & Fell, 1960). This report summarizes recent results.

Ataxia was accompanied by demyelination in the spinal-cord motor tracts, by chromatolysis and degeneration of the nuclei of the large motor neurones of the red nucleus in the brain stem and by hypertrophy of oligodendroglia in the caudate nucleus. Mild chromatolysis was found in neurones of the red nucleus in three lambs born of ewes fed the sulphate supplement only. These cases occurred in lambs having less than 18 μg Cu/g dry liver. Liver Cu concentrations of less than 4 μg /g dry

Liver, copper and molybdenum levels, clinical and histological findings in Cu-deficient lambs

Supplement to ewe diet (final dietary concentration indicated)	Lamb no.	Age at killing	Liver ($\mu\text{g/g D.M.}$)		Brain stem, red nucleus: neuronal lesions	Spinal-cord motor-tract: demyelination	Caudate nucleus, satellite cells: hypertrophy	Ataxia	
			Cu	Mo					
Group 1 + sulphate (1%) 1960 (fed last 3 months of pregnancy)	R31	Stillborn	54.1	1.2	-	-	-	?	
	R23A	Stillborn	62.7	1.2	-	-	+	?	
	R23B	24D	29.8	2.8	-	-	-	-	
	R23C	87D	8.1	2.8	*	-	-	-	
	B7A	46D	14.4	1.0	-	-	-	-	
	B7B	110D	6.0	3.6	*	-	-	-	
	R27A	100D	17.2	2.7	*	-	+	-	
	R27B	100D	38.1	3.7	-	-	-	-	
	1959 (fed last 4 months of pregnancy)	329B	100D	9.8	1.8	-	-	-	-
		56B	103D	8.1	3.8	-	-	-	-
38B		87D	9.5	3.1	-	-	-	-	
Group 2 Mo (50 p.p.m.) + sulphate (1%) 1960 (fed last 3 months of pregnancy)	B11A	Stillborn	9.7	17.6	†	-	+	?	
	B27A	27D	4.0	2.5	-	-	+	-	
	B11B	89D	0.2	1.6	*	+	+	Mild	
	B27B	92D	0.5	3.2	†	+	+	Mild	
	Y32A	100D	4.6	3.5	†	+	+	Mild	
	1959 (fed last 4 months of pregnancy)	181B	11D	2.8	3.1	‡	++	-	Severe
		332B	23D	2.9	4.8	‡	++	-	Severe
		46B	77D	4.3	3.8	‡	++	-	Severe
		25B	108D	13.5	4.7	-	-	-	-

-, indicates no lesion; *, few cells involved and these showing only early chromatolysis without nuclear degeneration; †, few cells involved but both chromatolysis and nuclear shrinkage and pyknosis; ‡, numerous cells showing severe damage; + and ++ indicates severity of other lesions.

matter were found in all lambs showing ataxia. In these animals there was no clear correlation between liver Cu level and the time taken to develop ataxia or the extent of degenerative changes in the central nervous system.

REFERENCE

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The natural source of vitamin D for sheep. By J. QUARTERMAN, A. C. DALGARNO and I. McDONALD, *Rowett Research Institute, Bucksburn, Aberdeen*

In theory ruminants at pasture can obtain vitamin D in two ways, from direct solar ultraviolet irradiation of the skin or the sterols in sebaceous secretion or by eating irradiated plants. The importance of direct irradiation might be doubted for sheep whose fleece may shield the skin completely. To provide information on this point assays were made for antirachitic activity throughout the year in the blood of sheep with different amounts of fleece. Seven Blackface ewes were clipped in May 1960 and another seven ewes drawn from the same flock were kept, without clipping, at pasture with the first seven. All the sheep were bled at intervals from May, at the time of clipping, to November. About 150 ml of blood were drawn from each sheep at each bleeding, and the blood samples from subgroups of three or four ewes were pooled before saponification and ether extraction. Assays were made by the standard method with rats (Bourdillon, Bruce, Fischmann & Webster, 1931).

The results (Table 1) show that clipping has a very large effect on the blood level of vitamin D. In June and August the concentration of vitamin D in clipped sheep

was two to three times that in unclipped sheep. By November the difference between the groups had disappeared and the level of vitamin D had sunk almost to the spring, preclipping value.

Table 1. *Estimated blood levels of vitamin D (i.u./100 ml) in clipped and unclipped sheep*

	May 1960	June	August	November
Clipped sheep	15, 28	60, 69	86, 61	18, 28, 16, 23
Unclipped sheep	10, 13	20, 18, 18, 15	30, 30	15, 19, 13, 29

The 95% fiducial limits for the assays are 60–160% of the estimates.

The difference in blood levels of vitamin D between the two groups is greatest during and immediately after the period of most intense sunshine and can only be a result of the greater exposure of skin or fresh sebaceous fat to sunshine following clipping. The summer increase in blood vitamin D in unclipped sheep is probably also largely due to increased insolation. By comparison the contribution due to irradiated provitamin D in herbage is likely to be small. These results also show that the increase in the blood level of vitamin D which occurs in the summer does not last until the end of the year.

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Fermentation of pectin by rumen bacteria. By B. H. HOWARD (introduced by J. DAVIDSON), *Rowett Research Institute, Bucksburn, Aberdeen*

Pectic substances can form a substantial proportion of the carbohydrates in the food of ruminants. It is already known that pectin is decomposed in the alimentary tract of the sheep (Leroy & Michaux, 1949), and in the experiments described here a vigorous fermentation of pectin was brought about *in vitro* by rumen bacteria. A solution of pectin (0.25%) in sheep rumen liquor was incubated at 39°; the substrate was rapidly fermented and had disappeared within 4 h. Fermentation was almost equally as rapid when the rumen liquor had first been lightly centrifuged to remove protozoa, thus demonstrating that the rumen protozoa, which have been shown to decompose pectin (Wright, 1960; Abou Akkada & Howard, 1961) could not be responsible for more than a small proportion of the pectin-fermenting power of whole rumen liquor. Acetic, propionic and butyric acids were the chief products formed from pectin in whole rumen liquor.

Mixed bacteria, separated from the lightly centrifuged rumen liquor by high-speed centrifuging, were suspended in phosphate buffer, pH 6.9, and incubated with pectin solution. Most of the substrate had been decomposed after 5 h incubation. Acetic, propionic and butyric acids, and glucose (as intracellular polysaccharide) accounted for about 80% of the substrate used in the proportions shown in the Table. The bacteria for Expt 1 were taken from a sheep fed on hay and grass; those for

Expts 2 and 3 from sheep fed on hay and concentrates. Most of the pectin was converted into substances already well known as products of fermentation of other carbohydrates in the rumen.

Products of fermentation of pectin by mixed rumen bacteria

Expt no.	Carbon recovery (%)	Products (moles/100 moles pectin-galacturonic acid fermented)				
		Acetic acid	Propionic acid	Butyric acid	Lactic acid	Glucose
1	78	114	38	0	0	21
2	76	96	24	7.5	2	26
3	82	108	48	1.5	0	22

The pectins in the food eaten by ruminants contain methyl-ester groups equivalent, in the case of sheep, to several g methanol/day. Determination of the total methoxyl and free methanol during incubation of pectin with rumen liquor showed that the ester groups were rapidly hydrolysed by the bacteria, and that the resulting methanol was further metabolized at a slower rate. However, rumen liquor from sheep fed on a wide variety of diets was found never to contain more than about 1 µg methanol/ml.

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Skinfold measurements during human pregnancy. By NAN TAGGART, *Obstetric Medicine Research Unit (Medical Research Council), Maternity Hospital, Aberdeen*

Many women say that they 'put on fat' during pregnancy, especially round the hips; metabolic evidence indicates that fat accumulates most rapidly between the 10th and 30th weeks of pregnancy (Thomson & Hytten, 1960, 1961). Skinfolts have been measured with Harpenden skinfold calipers at intervals during pregnancy and postpartum, at seven sites: (1) over triceps, (2) over biceps, (3) below the tip of the scapula, (4) in the mid-axillary line at level of lowest rib, (5) on the abdomen midway between costal edge and iliac crest, (6) mid-thigh anterior, and (7) thigh above knee-cap. Some preliminary results are given.

In ten women, average skinfold thicknesses increased during pregnancy at sites (3), (4), (5), and (6), by 20–50% of the value at 10 weeks of pregnancy. At sites (1) and (2) average skinfold thicknesses decreased by about 5 and 15%, respectively, and at site (7) the average skinfold remained almost unchanged. The distribution of subcutaneous fat therefore appears to change during pregnancy.

The mean value for the sum of all skinfolts increased steadily from 10 to 30 weeks, but there was little further increase between 30 and 38 weeks of pregnancy. The sum of skinfolts at 7 days postpartum was substantially greater than that at the

10th week of pregnancy. Thus the preliminary results conform to the trend of fat deposition suggested by Thomson & Hytten.

The 'net gain' of weight in pregnancy (value at 7 days postpartum minus value at 10th week of pregnancy) showed a high correlation, $r=0.8$, with the net change in the sum of skinfolds. In the periods 10-20, 20-30 and 30-38 weeks the correlations between weight gain and change in the sum of skinfolds were lower, $r=0.4$, 0.5 and 0.2 respectively. The low value from 30 to 38 weeks is consistent with the hypothesis that little of the increase in body-weight during this period is due to the accumulation of maternal body fat, and the high correlation between net gains in weight and skinfolds supports the concept of a net gain in body fat as a result of pregnancy.

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The energy expenditure of sheep in walking on the level and on gradients.

By J. L. CLAPPERTON, *Hannah Dairy Research Institute, Kirkhill, Ayr*

Experiments were made with two sheep to determine the energy cost of horizontal work and the work of ascent. Each sheep was exercised on a treadmill enclosed in a respiration chamber (Clapperton, Armstrong & Blaxter, 1960) at two speeds of 24 and 48 m/min at each of three gradients, 0, 1 in 22 and 1 in 11. Observations were also made when the sheep was not exercised. The maximal work load was a horizontal movement of 12.6 km and an ascent of 1150 m. Two amounts of food were given (each ration being given twice to each sheep) one sufficient to maintain body-weight and the other twice this amount. Metabolism was determined during the 8 h

Energy cost of work

	Maintenance level of nutrition	Twice the maintenance level of nutrition	Mean
Energy cost of horizontal locomotion (cal/horizontal kg m)			
Speed 24 m/min	0.613	0.421	0.517
Speed 48 m/min	0.686	0.640	0.663
Standard error of above means		± 0.104	± 0.074
Mean irrespective of speed and food intake	—	—	0.59 ± 0.05
Energy cost of vertical work (cal/vertical kg m)			
Speed 24 m/min:			
Gradient 1 in 22	5.08	10.90	7.99
Gradient 1 in 11	4.35	5.41	4.68
Speed 48 m/min:			
Gradient 1 in 22	6.38	8.66	7.52
Gradient 1 in 11	5.43	5.38	5.41
Standard error of above means		± 1.32	± 0.92
Mean irrespective of speed, gradient and food intake (cal/vertical kg m)	—	—	6.45 ± 0.47

in which exercise was given and also in the remainder of the 24 h period. Each combination of gradient with speed was imposed for 2 consecutive days.

Exercise during the day had no effect on energy metabolism during the subsequent night interval. The table summarizes the results for the energy cost of work, determined from the 24 h observation.

The energy cost of horizontal locomotion increased with speed and the mean value of 0.59 ± 0.05 cal/horizontal kg m may be compared with the value found for man of 0.54 (Smith, 1922), for the dog of 0.58 (Lusk, 1931) and for the horse of 0.39 cal/horizontal kg m (Brody, 1945). The energy cost of vertical work decreased with speed but was independent of gradient. The mean value of 6.45 cal/vertical kg m may be compared to that found for man of 6.92 cal/vertical kg m (Lusk, 1931). The amount of food given had no effect on the cost of work.

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The food intake and energy expenditure of some elderly men working in heavy and light engineering. By ELAINE C. BLAKE and J. V. G. A. DURNIN, *Institute of Physiology, University of Glasgow*

The nutritive value of the haggis. By D. S. MILLER, D. J. NAISMITH and P. L. PELLETT, *Human Nutrition Research Unit, National Institute for Medical Research, The Ridgeway, Mill Hill, London, N.W.7*

Haggis is an ancient, but still popular Scottish dish, prepared from oatmeal, the heart, liver and lungs of a sheep, onion and seasoning.

The haggis was cooked, freeze-dried, and its nutritive value determined by chemical analysis, microbiological assay (Association of Vitamin Chemists, Inc. 1951), and estimation of the net dietary-protein value (N.D.P. Cals%: Platt & Miller, 1959) of the traditional meal of haggis, swede and potatoes.

Chemical composition of the haggis

Thiamine ($\mu\text{g/g}$)	7.2	Fat (%)	34.2
Riboflavin ($\mu\text{g/g}$)	11.9	Calories (kcal/g)	5.86
Pyridoxine ($\mu\text{g/g}$)	5.2	Protein (%)	23.2
Nicotinic acid ($\mu\text{g/g}$)	60.2	Protein calories (%)	15.8

The beneficial effect of adding liver and heart to oatmeal is clearly shown in the vitamin assay. Oatmeal contributes much of the thiamine to the dish, but contains only $1.0 \mu\text{g/g}$ riboflavin and $10 \mu\text{g/g}$ nicotinic acid.

The content of utilizable protein in the haggis meal was found to be 8.5% N.D-p. Cals, an amount sufficient to satisfy the protein requirements of all groups but the lactating mother.

The protein value of the haggis, like that of most dishes, is limited by its content of the sulphur amino acids. Addition of 0.25% DL-methionine raised the net protein utilization (N.P.U.) of the dish from 56 to 63 and the N.D-p. Cals from 8.5% to 9.6%.

The proteins of oatmeal, like those of other cereals, are known to be limited by lysine; animal proteins, like the proteins of liver, heart and blood, have a relative excess of lysine.

The degree of complementation achieved by combining such proteins was shown by mixing blood with oatmeal.

When 8% of freeze-dried blood (N.P.U. 4) was added to oatmeal (N.P.U. 58; N.D-p. Cals 6.9%), a mixture having a N.P.U. of 66 and N.D-p. Cals content of 9.8% was obtained. With blood added at the 15% level, the values were raised to 68 and 10.4% respectively.

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The utilization of the energy of the same food by cattle and sheep. By K. L. BLAXTER and F. W. WAINMAN, *Hannah Dairy Research Institute, Kirkhill, Ayr*

Sheep are commonly used to measure the digestibility of foods given to cattle, and experiments have shown that this practice leads to little error. The present experiments were undertaken to find whether sheep and cattle utilize the energy of food equally well both for maintenance and for productive purposes. Three adult steers and three adult sheep were used as experimental animals. Each was given the same mixture of a hay containing 9% protein and crushed oats in the proportions 2 : 1 in five (cattle) or six (sheep) different amounts. The lower quantities given led to loss of energy from the body and the highest amounts were equivalent to about twice (cattle) or three times (sheep) the amount necessary for maintenance of energy equilibrium. Each amount was given for periods of 21 days and during the final 4 or 5 days of the period energy losses as heat, in urine, faeces and as CH₄ were determined by indirect calorimetry (Wainman & Blaxter, 1958 *a,b*). The results are summarized in the table.

There were no differences between species in the losses of energy in faeces or in the increase in faecal loss of energy with increasing nutritional level. Methane losses tended to be slightly higher in sheep at low nutritional levels, and there were no differences in urine energy losses/100 kcal food given. No differences of any magnitude occurred in the net availability of food for either maintenance or the production

Utilization of food energy by cattle and sheep

	Nutritional level	Sheep	Cattle
Faecal energy/100 kcal energy in food	Maintenance	30.0	30.6
	Twice maintenance	32.3	33.0
CH ₄ energy/100 kcal energy in food	Maintenance	9.09	8.30
	Twice maintenance	7.55	7.37
Urine energy/100 kcal energy in food	Maintenance	4.51	4.96
	Twice maintenance	4.21	4.51
Net availability of metabolizable energy for maintenance		80.4	80.4
Net availability of metabolizable energy for production of fat with its standard error		53.5 ±1.4	51.5 ±2.6

of fat. The net energy of the ration for production at twice maintenance was 29.9 kcal/100 kcal food for sheep and 28.5 kcal/100 kcal food for cattle, a difference of less than 5% which was not statistically significant. It can be concluded that despite a tenfold difference in their size, the two species utilized the energy of this particular ration equally well.

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Metabolism of radiocalcium and radiostrontium in the ewe. By H. G. JONES, W. S. MACKIE and B. F. SANSOM, *Rowett Research Institute, Bucksburn, Aberdeen*

A number of factors probably contribute to the high retention by hill sheep of ⁹⁰Sr from nuclear fallout. The metabolism of ⁴⁵Ca and ⁸⁹Sr by Blackface ewes fed a diet adequate in calcium and phosphorus during pregnancy and lactation, is described. Data on the effects of low calcium and phosphorus diets are not yet complete.

Weighed quantities of hay and concentrates providing about 6 g calcium and 4 g phosphorus/day were fed to ewes. Single oral doses of ⁴⁵Ca and ⁸⁹Sr were administered; urine, faeces and blood samples were collected for 10 days, after which the sheep were killed and a femur analysed. Experiments were carried out at mid-pregnancy, immediately after parturition and about 12 weeks after parturition. Non-pregnant ewes were used as controls.

Plasma levels indicated a threefold discrimination against ⁸⁹Sr during intestinal absorption of the two nuclides. About three times as much ⁴⁵Ca as ⁸⁹Sr was retained by the femur. This is in agreement with previous results for wether sheep (Jones & Mackie, 1959) and dairy cows (Garner, Jones & Sansom, 1960). About twice as much ⁸⁹Sr as ⁴⁵Ca was excreted in the urine but urinary excretion of both was very low compared with their faecal excretion.

During pregnancy the femur retained more of both nuclides and urinary excretion of both fell. Total absorption of both nuclides increased markedly but absorption of ⁴⁵Ca increased more than that of ⁸⁹Sr. Each sheep produced twin foetuses which retained as much ⁴⁵Ca but about half as much ⁸⁹Sr as the maternal skeleton.

In the postparturition period, skeletal retention of both nuclides was slightly lower than that of non-pregnant sheep. Similar quantities of ^{45}Ca and ^{89}Sr were retained by the skeleton and secreted in milk, but the $^{45}\text{Ca} : ^{89}\text{Sr}$ ratio in milk was rather higher. During late lactation, skeletal retention of both nuclides increased again to levels comparable with those in mid-pregnancy. During early and late lactation, total absorption of ^{45}Ca was proportionately higher than in control sheep.

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Amino-acid supplementation of an experimental diet for laying hens. By
 J. DAVIDSON, GABRIELLE M. ELLINGER and A. W. BOYNE, *Rowett Research
 Institute, Bucksburn, Aberdeen*

According to the (U.S.A.) National Research Council: Committee on Animal Nutrition (1954) the proportions of methionine + cystine, and of lysine required in laying diets containing 15% protein are 3.5% and 3.3% respectively of the total protein. Experimental rations having a critical percentage of protein around 10%, with 2½% provided by protein supplements, contained by calculation, 3.5% and 4.5% respectively of sulphur amino acids and lysine when the supplement was white fish meal and 3.2% and 3.4% when it was groundnut meal.

However, both sulphur amino-acid and lysine values calculated for the groundnut rations are sufficiently near requirement to suggest that individual variation may decide whether egg production will be improved when these amino acids are added. Preliminary work showed that a groundnut meal (GN1) with the high gross protein value (GPV) (Duckworth, Woodham & McDonald, 1961) of 54, indicating good processing, could not be improved by methionine addition. However a Burmese groundnut meal (GN2) ('jungle cake') with the low GPV of 36 was improved by addition of methionine and was used in a series of three laying experiments involving supplementation with methionine and lysine.

Egg production on diets supplemented with white fish meal, or groundnut meal of low gross protein value with and without amino-acid addition

Experiment/year Experimental weeks	1/1957-8			2/1958-9			3/1959-60		
	(a) Wt eggs per bird starting (kg)	(b) Food eaten (kg)	(c) Wt eggs adjusted to food intake (kg)	(a) (kg)	(b) (kg)	(c) (kg)	(a) (kg)	(b) (kg)	(c) (kg)
White fish meal	4.77	22.2	4.62	—	—	—	5.94	25.6	5.67
GN ₂	3.81	21.3	3.86	4.2	22.8	4.5	4.68	23.6	4.95
GN ₂ +0.1% MET	4.12	21.1	4.22	5.1	24.4	5.0	5.15	25.0	5.04
GN ₂ +0.1% LYS	—	—	—	4.0	22.6	4.3	—	—	—
GN ₂ +0.1% MET +0.1% LYS	—	—	—	5.9	26.9	5.2	5.63	24.1	5.74
S.E. of difference	±0.36	±1.54	±0.16	±0.41	±1.55	±0.24	±0.42	±1.44	±0.20

Results in the table show that a supplement of white fish meal supported about 20% higher egg production than a supplement of GN2. Addition of 0.1% DL-methionine (L. Light & Co.) can raise egg production on the GN2 diet by about 10% while 0.1% L-lysine (as monohydrochloride, E. I. du Pont) in addition to the methionine can bring production up to that with white fish meal as supplement. 0.1% L-lysine alone did not improve egg production on the GN2 diet.

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- Duckworth, J., Woodham, A. A. & McDonald, I. (1961). *J. Sci. Fd Agric.* **12**, 407.
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The One Hundred and Forty-second Meeting of The Nutrition Society was held at the Royal Society of Medicine, 1 Wimpole Street, London, W.1, on Friday, 26 May 1961, at 1.30 p.m., when the following papers were read:

The copper content of the liver and hair in kwashiorkor. By I. MACDONALD and P. J. WARREN, *Departments of Physiology and Chemical Pathology, Guy's Hospital Medical School, London, S.E.1*

It has been suggested that changes in the blood and pigmentation in kwashiorkor may be related to a copper deficiency and a marked hypocupraemia has been found (Lahey, Behar, Viteri & Scrimshaw, 1958; Edozien & Udeozo, 1960). In view of this the copper content of liver and hair in kwashiorkor was studied to determine whether the copper concentration in these tissues is altered.

The results showed a significant reduction in the copper concentration in both liver and hair (Table). The results also show that no copper was present in the liver lipid in the cases studied.

Mean values and standard deviations for the copper concentration ($\mu\text{g Cu/g tissue}$) in liver and hair

	Kwashiorkor	Control	Comparison between kwashiorkor and control P
Dry liver	6.7 \pm 1.2	20.4 \pm 8.2	0.005
Dry fat-free liver	17.3 \pm 3.9	24.4 \pm 7.3	0.05-0.025
Hair	13.9 \pm 2.7	18.5 \pm 4.2	0.05-0.025

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- Edozien, J. C. & Udeozo, I. O. K. (1960). *J. trop. Pediat.* **6**, 60.
 Lahey, M. E., Behar, M., Viteri, F. & Scrimshaw, N. S. (1958). *Pediatrics, Springfield*, **22**, 72.