

The One Hundred and Twenty-fifth Meeting of The Nutrition Society was held at Queen Elizabeth College, University of London, on Friday, 29 May 1959, at 4.15 p.m., when the following papers were read:

Pancreatic fibrosis and calcification in Uganda Africans. By A. G. SHAPER, (introduced by Z. A. LEITNER), *Makerere College Medical School, Kampala, Uganda*

Protein deficiency has been considered to be responsible for the pancreatic calcification seen in young people in Indonesia (Zuidema, 1955). Diffuse pancreatic lithiasis has been demonstrated in eleven adult Africans in Kampala, Uganda, and these cases are described and the possible relationship to protein malnutrition is discussed. Two patients, aged 21 and 24 years, presented with severe malabsorption states; steatorrhoea, hypoalbuminaemia, skin and hair changes, oedema and fatty liver were present in both subjects. Their nutritional background was fairly good although low in protein. A second group of five males aged 23–45 years presented with diabetes mellitus and were shown to have pancreatic calcification. Steatorrhoea was present in two of these patients but with no clinical evidence of malabsorption. The dietary history was poor in four of this group, the remaining subject having a good protein intake. The third group comprised four subjects in whom the pancreatic calcification was a finding at routine autopsy examination.

The histological appearances in five cases suggest a low-grade inflammatory reaction which has given rise to replacement fibrosis of the exocrine elements. All cases show varying degrees of fibrosis, the earliest phase of which appeared to be periductal and perilobular, followed by interacinar fibrosis until only isolated remnants of the exocrine tissue remained. The islets showed mainly hypertrophy, hyperplasia, fibrosis and hydropic degeneration. The presence of dilated ducts, ductular secretion and calcification and acinar dilatation suggests that the cause may be obstructive in nature.

While there is evidence that severe protein deficiency in children and adults can depress pancreatic function and produce fibrosis in the pancreas, the demonstration of pancreatic fibrosis and calcification in an area where protein malnutrition is common is only circumstantial evidence of a relationship between the two conditions. It may be that a variety of agents may produce lesions in a pancreas rendered more susceptible to injury by low protein intakes, but the possibility must be considered of altered protein metabolism leading to abnormally viscid secretions and ultimately to pancreatic lithiasis.

I am grateful to Professor Kenneth Hill, Royal Free Hospital Medical School, for his report on the pancreatic histology.

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The relationship between excitability of the central nervous system, food intake and body growth. By J. LÁT (introduced by E. M. WIDDOWSON), *Laboratory of Physiology, and Pathophysiology of Metabolism, Prague*

The relationship between excitability of the central nervous system and growth rate was studied under different conditions in rats. Excitability was experimentally defined in terms of the frequency of spontaneous reactions in unit time under standard stimulation. It was shown that the individual excitability level is a constant and a constitutional factor, and it determines to a certain degree the speed of learning, the discrimination capacity, as well as the speed of adaptation to traumatizing agents. It is also a factor in longevity. Further findings were as follows. (1) Under standard conditions the higher the individual excitability level the faster the exponential growth rate and the shorter the time of growth. (2) The excitability level can be predicted to a certain degree from the growth rate in the first 10 days of life. (3) There are close reciprocal relationships between central nervous system excitability and metabolic (nutritional and endocrinological) processes. Excitable animals show a higher caloric intake, and they choose spontaneously large amounts of carbohydrates and NaCl. Non-excitable animals prefer protein, fat and KCl. (4) The excitability level can be experimentally elevated by a high-carbohydrate diet, while a high-protein diet decreases it. (5) A close relation exists between central nervous system excitability and adrenal activity. (6) The isolation of rats in separate cages slows down their growth rate. The greater the excitability level of the animal the greater the loss of appetite and weight under these conditions. (7) When external stimulation (optic, acoustic, olfactory or tactile) was increased the animals ate more food, particularly more carbohydrate, and grew faster than when the stimulation was less. These findings may have some bearing on the more rapid growth rate of children now than during the early part of this century.

The effect of dietary sorbitol and related substances on the vitamin B requirements of the rat. By VALMAI M. HEDLEY and JOHN YUDKIN, *Department of Nutrition, Queen Elizabeth College, University of London*

Rats, 28 days old, were fed a carbohydrate-free diet, deficient in thiamine, to which were added various levels of glucose, fructose, sorbose, sorbic acid, ascorbic acid, glycerol, mannitol or sorbitol. It was found that only sorbitol or sorbose supported growth similar to that of animals fed the fully supplemented diets. Those animals fed the thiamine-deficient diets with either ascorbic acid, fructose, glucose, mannitol or sorbic acid died within 3–6 weeks from the beginning of the experiment and usually in the order given. The animals fed glycerol, on the other hand, showed a little growth in the first few weeks and then maintained that weight for at least 12 weeks.

In order to determine whether sorbitol produced the same effect with B vitamins other than thiamine, 28-day-old rats were fed synthetic diets containing either 10% sorbitol or 10% glucose, but with single deficiencies in each case of thiamine, riboflavin, pantothenic acid or pyridoxine. No signs of deficiency developed with

any of the animals fed sorbitol, except those on a pyridoxine-deficient diet. Signs of deficiency occurred, however, with animals on the glucose diets, deficient in thiamine, riboflavin, pantothenic acid or pyridoxine. In a further experiment, animals 9 weeks old were placed on synthetic diets containing either glucose or sorbitol at the 20% level, but deficient in pyridoxine. It was found that there was a gradual loss of weight and finally death with the animals on the glucose diet, and a gradual increase in weight with the animals on the sorbitol diet.

Thus sorbitol induces in the rat the production of thiamine, riboflavin, pantothenic acid, and in the older animal pyridoxine, when these vitamins are absent from the diet. A likely site of formation would seem to be the gut.

Avoidance of sucrose by rats deprived of thiamine. By JOHN YUDKIN,
Department of Nutrition, Queen Elizabeth College, University of London

This work was inspired by the classical paper of Harris, Clay, Hargreaves & Ward (1933), who showed that thiamine-deprived rats developed an appetite for foods containing the vitamin. Most of the published work on nutritional choice has been designed to answer the question, 'Will an animal choose a food which will prevent or cure nutritional deficiency?'. We have attempted to answer the question, 'Will an animal avoid a food which will produce nutritional deficiency?'

We have shown previously that rats will survive, though poorly, in the absence of thiamine, provided they are given a diet free from carbohydrate (e.g. Yudkin, 1951). We have thus more specifically asked the question, 'If such animals are allowed access to carbohydrate, separately from their thiamine-free, carbohydrate-free diets, will they eat it and die, or will they avoid it and live?'. The problem was studied with albino rats placed in one of three different dietary situations. All the animals were allowed unrestricted access to carbohydrate-free diet. In the first series of experiments, the diet was given without thiamine for varying periods, and then the animals offered sucrose. In the second, the diet was given with thiamine; after a time, the thiamine was stopped and sucrose was offered. In the third series, the animals were given thiamine and allowed access to sucrose from the beginning of the experiment; later, the thiamine was removed.

In each series of experiments, the animals exhibited a range of behaviour when they were consuming sucrose in the absence of thiamine. Some animals died within 3 or 4 weeks, whilst others survived for 3 or 4 months or longer. In general, those animals survived longest which ate least sucrose. There was also a good relationship between consumption of sucrose, consumption of carbohydrate-free diet, and the weight of the animal. Whenever the consumption of sucrose rose, there was a fall in the consumption of diet and a fall in weight of the animal. The results are best explained by a slight extension of the 'learning theory' of Harris, so as to take account of the palatability of foods.

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Avoidance of sucrose, and choice of sorbitol, by rats deprived of thiamine.

By T. B. MORGAN and JOHN YUDKIN, *Department of Nutrition, Queen Elizabeth College, University of London*

We have shown that rats receiving sorbitol become independent of dietary thiamine (Morgan & Yudkin, 1957). We have made use of this fact in extending our work on dietary choice by rats fed a carbohydrate-free diet without thiamine.

On such a diet, rats survive but do not grow. When thiamine is given, they grow well. If such rats receiving thiamine were allowed access to sorbitol, they ate about 1 g daily for 2 or 3 weeks, but then ate only occasional amounts to an average of 0.2 g daily. On the other hand, rats given the diet without thiamine ate, after the initial 2 or 3 weeks, an average of 0.7 g sorbitol daily. These rats grew much better than those receiving the thiamine-free diet without access to sorbitol, though not so well as those receiving thiamine.

In a further series of experiments, rats on the thiamine-free, carbohydrate-free diet were allowed access to sorbitol and to sucrose in separate vessels. They chose to eat both, and grew well. They were thus eating sucrose, which would have produced polyneuritis, but also sorbitol, which provided them (indirectly) with thiamine. In the conditions of these experiments, rats offered sucrose but no sorbitol ate the sucrose and developed signs of deficiency, with loss of weight, polyneuritis and death within 40 days.

The results cannot be explained on the basis of a specific appetite for a dietary essential, since sorbitol contains no thiamine. The simplest explanation is that the rat learns to associate an improvement or deterioration of its deficiency symptoms with the food it has consumed. The effect on its condition however does not necessarily follow immediately on the consumption of the food. With choice of thiamine, or avoidance of sucrose, the beneficial or harmful effects occur rapidly. But there must be at least several hours delay before sorbitol can exert its beneficial effect, since this effect is an indirect one. Sorbitol leads to increased excretion of thiamine in the faeces, which have to be consumed by the rat before the thiamine is available to it. We have shown that, if coprophagy is prevented, sorbitol no longer protects against thiamine deficiency.

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A comparison of results obtained for protein value of U.K. diets using chemical (F.A.O.) and biological (N.D-p.V.) methods. By ELIZABETH ANNE DRURY, *Scientific Adviser's Division (Food), Ministry of Agriculture, Fisheries and Food, Great Westminster House, London, S.W. 1* and D. S. MILLER, *Human Nutrition Research Unit, Nutrition Building, National Institute for Medical Research, Mill Hill, London, N.W. 7*

It will be seen that, although the diets are ranked in a similar order, the chemical method gives higher values than the biological method, and it is considered that this is chiefly due to the inverse relationship between biological value and level of protein

Diets*	1957 All	1957 A	1957 All	1957 C & D ₁	1957 C & D ₁	1957 All	1952 All
Year of survey	1957	1957	1957	1957	1957	1957	1952
Social class	All	A	All	C & D ₁	C & D ₁	All	All
Household composition	All	Younger couples	Old age pensioners	1m + 1f	1m + 1f	1m + 1f	1m + 1f
kcal/head/day	2587	2998	2528	a + c	> 3c	> 3c	> 3c
Protein (g/head/day)	75	93	72	68	57	59	65
Animal protein as percentage of total protein	58	64	57	53	50	53	45
Protein calories as percentage of total calories	11.6	12.4	11.3	11.3	11.1	11.1	12.0
Chemical method†							
Protein score (%)	87	87	86	86	88	87	86
R.P. (g/head/day)	65	81	62	58	50	51	56
R.P. calories as percentage of total calories	10.0	10.8	9.8	9.6	9.6	9.5	10.3
Biological method‡							
N.P.U. (operative) (%)	65	61	59	63	63	71	62
N.D-p.v. (g/head/day)	49	57	42	43	36	42	40
N.D-p.v. calories as percentage of total calories	7.6	7.6	6.6	7.1	6.9	7.9	7.4
Adequacy (intake as percentage of requirement)							
Chemical method	175	230	178	128	152	159	177
Biological method	148	186	136	105	124	150	143
B.M.A. scale§	100	127	112	79	80	85	95

*Ministry of Food: National Food Survey Committee (1954); Ministry of Agriculture, Fisheries and Food: National Food Survey Committee (1959). Classification: capital letters refer to social class, and lower-case letters refer to composition of family (m = adult male, f = adult female, c = children, and a = adolescents).

†F.A.O. (1957) using the amino-acid figures of Widdowson (1959). R.P. = Reference Protein.

‡Platt & Miller (1959). N.P.U. (operative) = net protein utilization (operative); N.D-p.v. = net dietary-protein value.

§British Medical Association: Committee on Nutrition (1950).

in the diet (Platt & Miller, 1958); even Reference Protein would not be completely utilized for anabolism in diets containing more than 4% protein. The proportion of protein from animal sources has no influence on the values obtained by either method. The adequacy of the diets by both methods in relation to the 'safe practical allowances' of F.A.O. is also given, together with the assessment, used in the National Food Survey, based on the B.M.A. (British Medical Association: Committee on Nutrition, 1950) scale. Further, the balance of protein to the energy content of the diets has been calculated and this may be compared with the allowances suggested (Platt & Miller, 1959) for an adult (4.6% R.P. or N.D-p.v. calories as percentage of total calories), children, aged 8 years (5.9%) aged 2 years (7.8%), adolescents (8.4%) and lactating women (9.5%).

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Effect of diet on intestinal phytase in the rat. By A. HEULWEN ROBERTS and JOHN YUDKIN, *Department of Nutrition, Queen Elizabeth College, University of London*

An increase in phytate in human dietaries leads to diminished absorption of calcium. There is evidence that this effect gradually diminishes with time, and that the organism becomes in some way adapted to the high dietary phytate. One way in

which this could occur is by an increased hydrolysis of the phytate from an increased amount of the enzyme phytase in the small intestine, produced under the stimulus of the increased amounts of phytate.

This study was begun in order to test the hypothesis that intestinal phytase is an adaptive enzyme in rats. The addition of bran or of sodium phytate to cereal diets produced changes in phytase which could not be interpreted as a simple adaptation to dietary phytate. It appeared that there was some relationship between the enzyme and the degree of calcification of the bone. We therefore studied the effect, both on phytase and on calcification, of a variety of dietary factors. The diets were based either on cereal, or on purified ingredients. They were fed alone or with the addition of vitamin D, calcium carbonate, sodium phytate or sodium citrate.

The addition of vitamin D invariably increased the amount of phytase. The addition of phytate increased the amount of phytase in only one set of conditions; in most of the other conditions, it decreased the amount of phytase. From the range of experiments in which inorganic phosphate was not a limiting factor, the combined results show a relationship between phytase and calcification, and appear to be governed by the amount of available calcium. The remaining experiments, in which there was little inorganic phosphate, showed lower calcification for a given enzyme value. The results throw light on the way in which the phytates of cereals may produce their rachitogenic effect, and on the way this in turn may be influenced by vitamin D, soluble phytate and citrate.

The chemistry of growth in the newly born. 1. Retentions of dietary constituents. By JEAN E. SLATER, *Medical Research Council Department of Experimental Medicine, University of Cambridge*

Nitrogen, potassium, calcium and phosphorus balances have been carried out on normal, full-term, male infants during the 6th, 7th and 8th days of life while receiving either breast milk or a proprietary brand of modified, dried cow's milk. More nitrogen, calcium and phosphorus was retained by the infants fed on the cow's-milk preparation than those on breast milk. The same amount of potassium was retained on both types of feeding.

Absolute intakes, absorptions and retentions (mg/kg body-weight/24 h) of N, K, Ca and P of infants fed on cow's-milk preparation or on breast milk, and the relation between the two types of feeding

	Nitrogen			Potassium			B.M.	Calcium		Phosphorus		
	B.M.	C.M.	$\frac{C.M.}{B.M.}$	B.M.	C.M.	$\frac{C.M.}{B.M.}$		C.M.	$\frac{C.M.}{B.M.}$	B.M.	C.M.	$\frac{C.M.}{B.M.}$
Intake	400	580	1.45	89.2	154.4	1.7	41	127	3.0	18.7	100.8	5.4
Absorption	325	510	1.57	71.5	131.4	1.8	23	78	3.4	16.0	80.0	5.0
Retention	210	321	1.53	49.3	51.1	1.0	16	72	4.5	15.5	41.2	2.6

In all cases the relation between the intake and absorption was the same. The fact that there was no increase in the retention of potassium, but large increases in the amounts of calcium and phosphorus retained on the cow's-milk preparation suggest that the extra nitrogen retained is associated with the calcium and phosphorus in the further development of bone during this period.

The chemistry of growth in the newly born. 2. The background of chemical structure. By J. W. T. DICKERSON (introduced by E. M. WIDDOWSON),
Medical Research Council Department of Experimental Medicine, University of Cambridge

Thigh muscle, skin and a femur have been analysed from 20–22 week human foetuses, newborn and 4–7 month old babies and thigh muscle and skin from young male adults.

The amount of water in skeletal muscle fell from 890 to 800 g/kg fresh tissue before birth and a further fall to 760 g/kg took place between birth and some time after 7 months. The amount of chloride in the muscle fell from 66 to 43 m-equiv./kg fresh tissue before birth and then continued to fall to 22 m-equiv./kg in the mature muscle.

The amounts of nitrogen, potassium and phosphorus per kg fresh muscle increased during growth. The largest increase in these constituents took place during the first 4–7 months of postnatal life when there were increases of 8 g nitrogen, 30 m-equiv. potassium and 20 m-moles phosphorus per kg fresh muscle. The deposition of these amounts per kg were therefore required to change the composition of the muscle and larger amounts were required for growth.

Between 20–22 weeks' gestation and a postnatal age of 4–7 months, the total nitrogen in the skin increased from 11.9 to 58 g/kg fat-free skin. Of this increase 38 g were deposited in each kg of fat-free skin during the 4–7 months after birth. The proportion of the total nitrogen accounted for by collagen increased from 20.6% in the foetuses to 71.5% at 4–7 months.

The average weight of a femur at the three ages was 2.5 g in the 20–22 week foetus, 16.4 g at birth and 46.3 g at 5–7 months. The percentage of water fell from 73.5% in the foetal bones to 61.5% in those at 5–7 months. The percentage of total nitrogen in the fat-free bones increased from 2.23 to 3.20 and that of calcium from 4.26 to 5.83 over the same period. Thus, 340 mg nitrogen and 812 mg calcium were deposited in a femur during the period before birth and 1100 mg nitrogen and 1800 mg calcium were deposited during the first 5–7 months after birth.

The results on skeletal muscle form part of a more detailed study on this tissue which is being published elsewhere (Dickerson & Widdowson, 1959).

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Metabolic studies on large and small eaters. By GEOFFREY A. ROSE and R. T. WILLIAMS, *St. Mary's Hospital, London, W. 2*

In any group of people living apparently similar lives it is a common observation that the calorie intake varies enormously. We have carried out a series of laboratory experiments to see if there are any constitutional differences between big eaters and small eaters, which might explain the different energy requirements.

Forty medical students provided detailed records of their diets for 1 week and from them six pairs of subjects were chosen so that the members of each pair were

approximately matched in weight but differed as widely as possible in calorie intake. The average body-weight of the large eaters was 5.3 kg less than that of the small eaters.

Oxygen consumptions were measured by means of a Benedict-Roth spirometer in the basal state and whilst sitting, standing and performing a standardized exercise. Using a Douglas Bag further oxygen consumptions were measured during a standardized steps exercise and during 'free style' walking and stair climbing. Each subject was then given a meal providing 840 kcal and oxygen consumption measured at $\frac{1}{2}$ h intervals for 4 h. Records were also made of pulse, oral temperature and blood pressure.

A rough estimate was made of the normal daily activities of each subject by questioning and by use of a pedometer for 1 week. The large eaters tended to be slightly more active, but this could not account directly for more than a small proportion of their excess calorie intake.

It was shown that there was wide variation in oxygen consumption between different individuals under the same conditions. But there was no systematic trend in these differences and individuals, who were 'costly' in some exercises, were 'economic' under other conditions. There was no evidence that, as a group, the big eaters tended to use more energy than the small eaters in these laboratory tests. Similarly, the specific dynamic action of the standard meal did not differ between the two groups.

The big eaters tended to show higher resting pulse rates and blood pressures a greater rise of pulse rate after exertion and a greater postprandial temperature and pulse rise. Some of these differences were statistically very significant. The big eaters, too, showed a significantly faster rate in the 'free style' walking exercise. The big eaters, then, seemed to be more sprightly and more physiologically reactive individuals and it is possible that these characteristics in the course of the numerous small activities of daily life could account, in part, for their increased energy requirements.

Urinary excretion of vitamin A in pregnancy. By D. S. McLAREN, *East African Institute for Medical Research, Mwanza, Tanganyika*

It is known that vitamin A may be excreted in considerable quantities in the urine in certain diseases, and it has been reported to occur in normal pregnancy (Gaetgens, 1937). It was thought that if this occurred in areas where vitamin A deficiency is rife, it might be an important contributory factor. No such investigation seems to have been made previously.

The results were as follows: (1) Urine samples from fifty healthy pregnant African women of varying parity, without exception, contained no vitamin A. (2) In a further sixty-four women, the effect of a large dose of 100 000 i.u. vitamin A by mouth was studied. In all cases, urine samples taken before dosing, and also 48 h after dosing, were negative for vitamin A. This group included 29% showing abnormalities in the urine, such as albumin (17%), red cells (12%), pus cells (9%) and schistosome

ova (9%). (3) Plasma carotenoids and vitamin A levels were determined in the case of twenty-four of the above-mentioned sixty-four women. This was done before the vitamin A was given, and the antimony-trichloride method was used. Compared with the ranges found in healthy United States adult males ((U.S.A.) National Research Council, 1954), two of the carotenoid and ten of the vitamin A values were low. After allowing for the generally lower values in women, and the fall in vitamin A level during the third trimester, nine of the vitamin A values were still abnormally low.

It is concluded that in a part of Africa where vitamin A deficiency occurs, there was no evidence that the vitamin was being lost in the urine.

I am grateful to Roche Products Ltd and Glaxo Laboratories Ltd for supplies of vitamin A.

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Essential fatty-acid (E.F.A.) requirement of the pig. By W. M. F. LEAT,
School of Agriculture, Cambridge

The pig is considered a species relatively refractory to E.F.A. deficiency (Deuel, 1957). Witz & Beeson (1951) described a skin scaliness with rations low in fat (0.12%) but Hill, Warmanen, Hayes & Holman (1957) noted no skin abnormality.

My observations arose on investigating the tocopherol needs of pigs receiving low-fat diets. Eight 17-day-old litter-mate piglets were weaned and divided into four pairs. The first pair (A) received a low-fat diet with composition adjusted to body-weight. It contained white fish meal (up to 40 lb weight only), dried skim milk, extracted palm-kernel cake, dried brewer's yeast, cassava, minerals, vitamins A and D, and analysed as follows.

	Body-weight		
	Up to 40 lb	40-120 lb	120-200 lb
Ether extract (%)	1.25	0.87	0.77
(Iodine value)	(37)	(28)	(29)
Linoleic acid (%)	0.027	0.030	0.030

The remaining pairs (B, C and D) received the same diets supplemented as shown below. They made the weight gains shown over a period from 4 weeks old to slaughter weight (21-25 weeks). Back-fat samples taken at slaughter had the analyses shown in the table.

	Group				
	A	B	C	D	
Olive-oil supplement (ml/100 g diet)	—	—	2	2	
α -tocopheryl succinate (mg/100 g diet)	—	7	—	7	
Linoleic-acid intake (g/100 g diet)	0.03	0.03	0.22	0.22	
Scaliness of skin	++	++	—;	—;	
Growth rate/pig (lb/day)	1.34; 1.58	1.43; 1.44	1.40	1.46	
Food consumption/pig (lb/day)	3.64; 3.80	3.76; 3.66	3.58	3.32	
Back-fat	analysis	{	Iodine value	... ; 52.5	... ; 54.1
analysis			Linoleic acid (g/100 g)	... ; 0.27	... ; 1.07

After 13 weeks, pairs A and B showed a dry flaking skin on the back and particularly over the shoulders, while the remaining pigs had a sleek appearance. One 'A' pig that received 2% olive oil from the 18th to 23rd week showed a markedly improved appearance while an unsupplemented control did not.

These preliminary observations suggest that under these conditions (i.e. with early weaning, etc.) a dietary level of 0.03% linoleic acid may satisfy the E.F.A. requirement for normal growth but is marginal for normal skin development.

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Defective blood coagulation in scorbutic guinea-pigs. By A. N. HOWARD, *Dunn Nutritional Laboratory, University of Cambridge and Medical Research Council*, and P. T. FLUTE, *Department of Medicine, University of Cambridge*

Haemorrhage, which is a typical feature of scurvy, has been attributed to increased capillary permeability due to defective vascular membranes (Wolbach & Howe, 1926). Barkhan & Howard (1959) showed however that the blood coagulation is impaired by a deficiency of plasma thromboplastin generation.

Exposure of human plasma to contact with glass produces an acceleration of blood coagulation (Margolis, 1957); and congenital syndromes in which there is a failure to react to contact are associated with defective thromboplastin generation (Biggs, Sharp, Margolis, Hardisty, Stewart & Davidson, 1958). The activation of coagulation by kaolin and glass surfaces was therefore investigated in twenty normal and ten scorbutic guinea-pigs.

In scurvy, a defective reaction to glass contact was always found as judged by the two-stage recalcification technique (Margolis, 1957); activated scorbutic plasma failed to shorten the recalcification time of control intact plasma to the same degree as normal activated plasma. Likewise, increased thromboplastin generation (as measured by the method of Hicks & Pitney, 1957) which occurred in normal plasma following activation by glass was not observed in scorbutic plasma.

The kaolin clotting time (Margolis, 1958) was also prolonged. Addition of reagents which contain factors necessary for contact activation such as serum adsorbed with $Al(OH)_3$ corrected the defect. Whole blood clotting time was normal, plasma prothrombin time increased, and serum prothrombin consumption reduced.

It is generally accepted that the initial stimulus for coagulation *in vitro* is contact with a foreign surface. A similar physico-chemical change may initiate intravascular coagulation. Defective ability to respond to the initial stimulus may be important in the production of haemorrhage in scurvy, but the significance of the *in vitro* measurements to the *in vivo* bleeding cannot as yet be assessed. The defect may be secondary to the haemorrhage rather than its primary cause.

Defective ability to react to glass contact in man is associated with a congenital deficiency of Hageman factor or plasma thromboplastin antecedent (Biggs *et al.*

1958). Only in the latter does abnormal haemorrhage occur (Ramot, Singer, Heller & Zimmerman, 1956; Rosenthal, Dreskin & Rosenthal, 1955). The defect in scurvy resembles these deficiencies but differs in that it is acquired.

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Nutritional ketosis and stress. By R. PASSMORE, *Department of Physiology, University of Edinburgh*

Ketosis develops in most subjects during recovery after a 10 mile walk in the post-absorptive state and a positive Rothera reaction is found in the urine from 2 to 6 h after the end of the walk (Courtice & Douglas, 1936; Passmore & Johnson, 1958). This might be related simply to the calorie deficiency; alternatively stress associated with the exercise might be a factor. To test this, post-exercise ketosis was studied for periods of 6 h on two occasions in four young men in the postabsorptive state (*a*) after a 10 mile walk at 4 miles/h on the level with no load and (*b*) after a 5 mile walk at 4 miles/h up a gradient of 6% with a load of 10 kg. The two walks involved about the same total calorie expenditure; on the level this was spread over 150 min and was carried out with no difficulty or obvious stress: uphill with the load the exercise lasted only 75 min, but the rate of energy expenditure was over 600 kcal/h and the subjects were taxed almost to the limits of their endurance. The experimental details were as described by Passmore & Johnson (1958). All the subjects were fed on a fixed artificial diet for 2 days before each walk.

Table 1. *Ketonuria during two periods of exercise of approximately the same calorie cost, but of different duration, and a recovery period of 6 h*

Subject	Weight (kg)	Duration of exercise (min)	Calorie expenditure during walk (kcal)	Ketonuria during walk and recovery (m-moles)
H	68	150	810	0.6
		75	770	0.8
P	59	150	770	1.5
		75	690	1.2
M	63	150	810	2.3
		75	770	1.8
G	63	150	1050	9.5
		75	900	2.6

Table 1 shows the results. Although the degree of ketonuria was different in the four subjects, in none was it significantly greater after the hard exercise than after the moderate exercise. It can be concluded that the ketosis was related primarily to

the calorie deficiency and that other stresses associated with exhausting work contributed little.

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The effects of intraruminal infusions of acetic and propionic acids on the yield and composition of the milk of the cow. By J. A. F. ROOK and C. C. BALCH, *National Institute for Research in Dairying, Shinfield, Reading*

The experiment reported is the first of a series designed to study the effects of intraruminal infusions of volatile fatty acids, individually and in combination, on the secretion of milk and its major constituents. Three cows were placed on a 3 × 3 Latin square design, in their 2nd month of lactation. Periods were of 28 days' duration. Throughout the first experimental period, they were given rations of hay and dairy cubes calculated, from their previous performance, on the basis of Woodman's (1954) feeding standards, and then at the end of each period the rations were decreased by 0.7 lb S.E. and 0.16 lb D.C.P. The three treatments were: daily intraruminal infusions of (1) 100 lb water, (2) 100 lb water + 900 ml acetic acid and (3) 100 lb water + 900 ml propionic acid.

Mean values for milk yield and the concentration and yield of major milk constituents over the last 8 days of each period, with the standard error of the difference between two means, are given in the table.

Treatment	Yield (lb/day)	Fat		Solids-not-fat		Lactose		N · 6.38		Casein N (mg/100g)
		%	lb/day	%*	lb/day	%*	lb/day	%*	lb/day	
Control	22.7	3.41	0.76	8.73	1.91	4.77	1.05	3.02	0.65	360
Acetic-acid supplement	25.7	3.55	0.92	8.63	2.15	4.78	1.10	2.90	0.72	353
Propionic-acid supplement	24.8	3.09	0.76	8.95	2.15	4.87	1.17	3.16	0.76	376
S.E. of difference between means	±0.39	±0.17	±0.05	±0.09	±0.04	±0.04	±0.02	±0.08	±0.02	±10

* Calculated on a fat-free milk basis.

In this experiment the propionic-acid followed the acetic-acid treatment and the observed effect of propionic acid on milk yield may, therefore, be in part a carry-over effect and requires further evaluation with the order of treatments reversed.

The trends observed are consistent with the following theoretical possibilities: that acetic acid has a general effect on the synthesis of all milk constituents, and the output of water is increased in accordance with osmotic requirements (see Rook & Wood, 1959); that acetic and propionic acids have specific effects on milk-fat synthesis, and that propionic acid has a specific effect on the synthesis of solids-not-fat (see Rook, 1959).

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The physiological significance of the fluid consistency of faeces from cattle grazing spring pasture. By J. A. F. ROOK and C. C. BALCH, *National Institute for Research in Dairying, Shinfield, Reading*

The fluid consistency of the faeces from cattle grazing spring pasture is widely believed to indicate a high loss of water by the faecal route and consequently an abnormal loss of minerals, particularly sodium and potassium, and other food constituents.

Recently (Rook, Balch & Line, 1958; Rook & Balch, 1958) four cows were first given 56 lb silage, 6 lb hay and 16–18 lb concentrates daily, and later young, freshly cut Cocksfoot herbage *ad lib*. With grass the average total daily intake of water, in food and by drinking, increased from 153 lb to 168 lb; faecal water fell from 74 lb to 69 lb, but urine output increased from 24 lb to 52 lb. At the same time, the intake of dry matter decreased from 32 lb to 27 lb and the output of faecal dry matter fell from 11 lb to 7 lb. The change to grass increased the intake of sodium from 14 g to 32 g and of potassium from 226 g to 368 g. The faecal excretion of sodium increased only from 5.8 g to 9.7 g representing 41% and 30% of the intake, and that of potassium changed from 71.2 g to 64.0 g representing 32% and 17% of the intake. These values were typical for some twenty cows. With a few others the changes were larger. In one cow the water intake rose with grass from 180 lb to 296 lb and urine production from 48 lb to 186 lb, but faecal water fell from 64 to 40 lb; later she drank 251 lb water and produced 285 lb urine in 24 h. The ruminal digesta and faeces were not abnormal and her appetite was good.

The rate of passage of stained hay through the hind gut of the four cows was not markedly or consistently different with the two diets.

Thus, although the faeces from cattle grazing spring pasture have a high percentage water content, this does not indicate the daily excretion of an increased amount of water, or of sodium and potassium in the faeces. The fluid consistency of the faeces is due to a decreased output of faecal dry matter with a virtual absence of mature plant structural components, and results from physical rather than from physiological causes.

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The behaviour of the proteins of maternal milk in the normal infant rat's stomach. By T. GUMINSKI and D. J. NAISMITH, *Human Nutrition Research Unit, Nutrition Building, National Institute for Medical Research, Mill Hill, London, N.W.7*

In an earlier study, attention was drawn to the separation of some of the constituents of milk in the stomach of the suckling rat (Platt, 1954). Milk from successive feeds was shown to be clotted and disposed, roughly, in concentric layers, the outer layer being formed by the latest feed, and it was inferred, from the rate of disappearance of lactose and water, that the greater part of the milk serum leaves the stomach within 30–40 min of the feed.

In the present experiments, the partition in the stomach of casein and 'whey proteins' was studied. Six litters of 10-day-old albino rats were allowed to suckle for 20 min, after having been separated from their mothers for 18 h, then killed immediately, or at various intervals up to 3 h after ingestion. The period of fasting was necessary, since milk curd may remain in the infant rat's stomach for as long as 20 h after ingestion (Platt, 1954). In addition, to obtain a figure for a normally fed rat, one of each litter was left with the mother throughout the experiment. Stomach contents were weighed, and the proteins fractionated; casein was separated by rennin coagulation at pH 4.6, and 'whey proteins' by precipitation with trichloroacetic acid.

The mean values for weights of stomach contents decreased rapidly, from 1.15 g immediately after suckling to 0.31 g 3 h after ingestion. During this period, there was a noticeable loss of water from the milk curds. At the same time the ratio casein-N/'whey protein'-N rose from 2.5 to 5.9 after a 3 h period of 'digestion'. The value for the normally fed animals was 11.4.

A pooled sample of rat's milk was found to contain 6.8% casein and 2.4% 'whey proteins' (ratio 2.8).

It would appear that, after a meal, the 'whey proteins', like lactose, rapidly leave the stomach in the serum fraction of the milk, while casein, the major protein fraction (74%) is retained, to be delivered at a constant rate to the duodenum. The significance of this finding may lie in the limited functional capacity of the proteolytic enzyme systems in the digestive tract of the infant animal (Werner, 1948).

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The nutritive value of kushuk—an Iraqi fermented milk-wheat product.

By W. FRANKUL and P. L. PELLETT, *Human Nutrition Research Unit, Nutrition Building, National Institute for Medical Research, Mill Hill, London, N.W.7*

Kushuk or similar products are made locally throughout the Middle East. Approximately 1 part by weight of dried parboiled wheat grits is mixed with 2 parts by weight of yogurt and fermented for about a week. Curd from an equal volume of milk is then added and the whole fermented for a further period of 4-5 days. The product is sun-dried and ground to a powder, which can be stored for long periods before it is eaten.

The chemical composition of an Iraqi sample is shown in the table. Also shown are the compositions of (a) a laboratory-made sample, prepared in the same manner, (b) milk and (c) parboiled wheat. The main point of interest is in the very low content of riboflavin, particularly in the Iraqi product; this is almost certainly due to destruction of the vitamin during drying in sunlight.

Chemical composition of kushuk

	Content/100 g dry weight					
	Moisture (%)	Protein (g)	Fat (g)	Ribo- flavin (mg)	Thia- mine (mg)	Nicotinic acid (mg)
Kushuk (Iraqi)	7.4	17.4	8.6	0.05	0.21	3.5
Kushuk (laboratory)	5.2	19.7	11.1	0.16	0.22	3.7
Milk (whole)	88.0	25.0	25.0	1.16	0.48	0.53
Wheat (parboiled)	9.0	12.2	1.8	0.14	0.24	4.0
Milk-wheat* mixture	—	16.6	9.7	0.35	0.26	3.3

*Calculated: 1 g wheat + 4 ml milk for fat and protein;
1 g wheat + 2 ml milk for vitamins (whey rejected).

Investigations into changes in the vitamin content during fermentation showed that there were increases of 78 and 50% respectively in the contents of riboflavin and nicotinic acid, but that the concentration of thiamine remained fairly constant.

Rat-growth experiments showed that the supplementation of poor Iraqi diets with 15% kushuk increased the protein-efficiency ratios. The ratio for a rice-okra diet was increased from 1.30 to 1.98, and that for a wheat-chickpea diet from 1.47 to 1.85 by supplementation.

The nutritive value of a similar Egyptian product has been presented recently (El-Sadek, Zawahry, Mahmoud & Abdel-Motteleb, 1958), and van Veen (1957) has discussed the general importance of fermented foods in the Middle East. The food technological importance of this and similar materials as a means of preserving milk in hot climates has been recognized by Adolf (1954).

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'Riboflavin metabolite.' By J. MONTGOMERY, E. C. OWEN and R. PROUDFOOT,
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Crossland, Owen & Proudfoot (1958*a,b*) described a number of lyochromes in goat urine, one of which was 'riboflavin metabolite', some properties of which have already been reported. Chromatograms were painted with the aqueous concentrates of goat urine and the strips which contained the 'riboflavin metabolite' were extracted with ether to remove the developing solvents. The metabolite was eluted from the strips with water. The solution appeared on being re-tested in the Crammer (1948) system to be pure, but the shape of its spectral absorption in aqueous solution varied and chromatography in the system of Kilgour, Felton & Huennekens (1957) showed a blue-fluorescent impurity. 'Riboflavin metabolite' was soluble in methyl benzoate and in chloroform but not so soluble as in water. Chloroform, in view of its low boiling point and great density, was used for continuous extraction of the metabolite from aqueous eluate of Crammer chromatogram strips. Ultraviolet illumination showed that the yellow-fluorescent 'riboflavin metabolite' was being extracted into

the chloroform and that the water eluate contained a blue-fluorescing impurity which remained behind in the aqueous phase. Testing of the watery eluate in the system of Kilgour *et al.* (1957) confirmed the presence of both blue- and yellow-fluorescing components in the strip eluates from the Crammer system and also showed that the blue-fluorescing material was not extracted by the chloroform.

In the Unicam spectrophotometer the chloroform solution of the concentrate of 'riboflavin metabolite' showed peaks of maximal absorption at 272, 338 and 447 m μ (two different samples) with a distinct trough on each side of the 338 m μ peak. After concentration by evaporation, solutions of the metabolite in both water and CHCl₃ crystallized when stored at -18° but as yet we have not sufficient material for elementary analysis because only a small proportion of a 2 g dose of riboflavin is converted by the goat into the 'riboflavin metabolite'.

Its partition between water and chloroform by photometry at 447 or 338 m μ is about 3:1 in favour of the water. It is microbiologically inactive (Crossland *et al.* 1958*b*) but it cannot be lumichrome. On irradiation in neutral solution it gives a spot in the Crammer system in the same place as lumichrome produced by irradiation of riboflavin.

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