*The Two Hundred and Thirty-first Scientific Meeting of the Nutrition Society was held in Guy's Hospital Medical School, St Thomas Street, London, SE*1, *on Friday,* 12 *March* 1971, *at* 15.00 *hours, when the following papers were read:*

Changes in the glucose tolerance test associated with a sucrose-free diet.

By A. J. FRY (introduced by O. G. EDHOLM), *Division of Human Physiology, National Institute for Medical Research, London NW*3

Many workers have studied the effects on glucose tolerance of a change in dietary carbohydrate from sucrose to starch (e.g. Cohen, 1967). However, little work has been undertaken on the long-term effects on glucose tolerance in man of a diet containing sucrose compared with a sucrose-free diet. An opportunity arose to undertake a long-term study when nineteen members of the British Antarctic Survey base at Halley Bay (75°S 26°W) were on a sucrose-free diet for a period of 14 weeks.

Six glucose tolerance tests were carried out at approximately 3-week intervals during the period of the sucrose-free diet and for a similar period of time afterwards on the normal base diet containing sucrose. Blood sugar was determined using a glucose oxidase method (Keston, 1956). The subjects weighed themselves daily before breakfast and no significant changes in body-weight occurred on either diet.

It was found that the blood glucose concentrations were significantly lower 0·5 h after ingestion of the glucose load on the sucrose-free diet towards the end of the diet period compared with values obtained on the normal base diet. This change was reversed at 1 and 1·5 h after ingestion of the glucose load.

The results suggest that, in man, there is a change in glucose tolerance after approximately 9 weeks on a sucrose-free diet compared with results obtained on a normal base diet.

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The influence of dietary fat on the glucose tolerance test.

By J. M. TAYLOR, I. MACDONALD and J. HENDERSON, *Department of Physiology, Guy's Hospital Medical School, London SE*1

In order to study the effect of dietary fat on glucose absorption it was decided to give lipid at the same time as the standard glucose tolerance tests (1 g/kg body-weight) to male students (18–22 years) on a normal diet.

The lipids used were either sunflower-seed oil or coconut oil (1 ml/kg body-weight) and the total amount was made up with water to 4 ml/kg body-weight.

Venous blood was taken at 0, 30, 45 and 60 min and the glucose concentrations were determined using glucose oxidase technique (Faulkner, 1965). Serum insulin was measured by the immuno-assay technique (Hales & Randle, 1963).

The serum glucose concentration for each subject after taking glucose was subtracted from the corresponding value for the meal in which either sunflower-seed oil or coconut oil had been added to the glucose.

The values calculated in this way were combined for all individuals over the 60 min period and showed that when coconut oil was added to the glucose the mean level of serum glucose was significantly greater ($\bar{x}=12$ mg/100 ml, SE $\pm$ 5·2). When sunflower-seed oil was the lipid added to glucose the difference ($\bar{x}=8$ mg /100 ml, SE $\pm$4·0) was not significant. When the values for both the glucose with lipid meals were combined, the increase in serum glucose concentration was significantly greater than when glucose was given alone ($\bar{x}=10$ mg/ml, SE $\pm$ 3·3).

There was a greater increase in serum insulin level after glucose with lipids than after glucose alone.

We are grateful to the volunteers in these experiments.

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Dental caries and fat deposition in rats receiving sucrose or glucose syrup in drinking fluids. By T. H. GRENBY and C. J. LEER, *Department of Oral Medicine and Pathology, Guy's Hospital, London SE1*

Sucrose appears to cause more dental caries than other food constituents. Glucose syrup, a sweet-tasting partial hydrolysate of maize starch, is a possible substitute, especially in drinks.

Glucose syrup was compared with sucrose in two large-scale rat experiments. Weanling rats about 28 d old, drawn from two separate strains, were divided into two balanced groups and fed *ad lib.* on standard Thompson rat cube diet for 8 weeks. Instead of drinking-water, one group was given 20% sucrose solution, and the other 20% spray-dried glucose syrup (D.E. 41). Glucose syrup tastes less sweet than sucrose so, in the second experiment, 0·1% saccharin sodium BP was added to the 20% glucose syrup solution.

Of forty-three rats on each regimen, all survived to 8 weeks in the sucrose group, but four died in the glucose syrup group. In the highly caries-prone Osborne–Mendel strain both the mean caries score and the extent of dental plaque covering the teeth were significantly greater on the sucrose than on the glucose syrup regimen ($P<0{\cdot}002$). On the sucrose regimen there was extensive 'smooth surface' caries

which, unlike the attack resulting from most diets containing sucrose in solid form, did not originate in the fissures of the molar teeth. In a smaller number of rats of the less sensitive Wistar strain, there was no significant difference in the caries and plaque scores between the two regimens.

The average fluid consumption per rat per d over the entire 8-week period was 34 ml of the 20% sucrose fluid, 44 ml of the 20% glucose syrup fluid before it was sweetened with saccharin, and 38 ml after saccharin had been added. The daily weight gain per g carbohydrate consumed in the drinking fluid was consistently greater on sucrose than on glucose syrup (range: sucrose 0·29–0·49 g and glucose syrup 0·24–0·35 g). There was some evidence that the rats on the sucrose regimen had a higher liver fat content than those receiving glucose syrup and the total body fat deposited per g carbohydrate consumed in the drinking fluid was also greater from sucrose than from glucose syrup.

Although substituting spray-dried glucose syrup for sucrose in solid diets did not alter the incidence of caries in some earlier trials, the present results suggest that the reduction in caries when liquid glucose syrup is used in sweetened drinks warrants further investigation.

The effect of disease on nitrogen excretion in the hartebeest. By Pamela ARMAN*, *Makerere University, Kampala, Uganda* and D. HOPCRAFT, *PO Box 14, Athi River, Kenya*

During a series of nitrogen balance trials on East African ruminants, two young male hartebeest (*Alcelaphus buselaphus cokii*) became ill and died. They were fed a pelleted ration containing 13·7% crude protein on which four normal hartebeest were in positive N balance. All had been hand-reared and were introduced to the diet and metabolism cages gradually, but they were excitable and restless.

After 9 d prefeeding, they were caged for faeces and urine collection. Animal A (70 kg) started to lose its appetite after 3 d and its urinary N excretion rose markedly (see Table 1), but its temperature and appearance were normal. On day 7

Table 1. *Nitrogen intake and excretion in normal and diseased hartebeest*

	Animal A							Animal B	
	Expected normal values	Day						Expected normal values	Mean for days 1–8
Daily intake and output		1	2	3	4	5	6		
Dry-matter intake (kg)	1·84	1·99	1·89	1·68	1·04	0·77	0·99	1·46	1·40
N intake (g)	40·4	43·6	41·3	36·7	22·7	16·7	21·6	32·0	30·8
Faecal N (g)	12·7	17·6	17·0	13·5	10·7	7·9	10·1	10·5	12·1
Urine N (g)	14·8	17·6	19·3	21·3	23·0	27·6	36·1	14·4	25·4
N balance (g)	+13	+8	+5	+2	−11	−19	−25	+7	−7
Digestibility of dry matter (%)	60·5		47·4	mean				58·8	50·0
Digestibility of crude protein (%)	68·4		57·9					67·1	60·9

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https://doi.org/10.1079/PNS19710035 Published online by Cambridge University Press

it was removed from the cage and given palatable food. Five days later it looked weak and died on day 12. The cause of death was septicaemia due to *Salmonella typhimurium*. This animal was metabolically abnormal 10 d before death, when compared with the healthy animals.

A month later, a second hartebeest, animal B (59 kg), became ill and died. Its food intake had been normal during the collection period but its urinary N excretion was high and it was in negative N balance (Table 1). It appeared weak on day 9 and was taken out of the cage. Symptoms of septicaemia developed and it died 2 d later. The causal organism was not identified. It was showing abnormal N excretion for at least 10 d before death. Digestibility was low in both diseased animals.

We wish to thank J. G. Debbie and B. Clausen for veterinary assistance and for carrying out the post-mortem examinations.

The effect of meals on serum isoamylases. By BETTY L. COLES, *Department of Physiology, Guy's Hospital Medical School, London SE1*

Serum isoamylases have been separated by electrophoresis on agar gel. The usual pattern shows three bands, corresponding in mobility to one salivary and two pancreatic fractions. The effects of glucose, protein and cooked starch meals on these fractions have been investigated.

This work was supported by the Medical Research Council.

Dietary lipid requirements for hepatic microsomal enzyme induction in the rat. By W. J. MARSHALL, *Department of Chemical Pathology, King's College Hospital Medical School, London SE5*, and A. E. M. MCLEAN, *Department of Experimental Pathology, University College Hospital Medical School, London WC1*

Phenobarbitone induces increased synthesis of hepatic microsomal drug-metabolizing enzymes and the associated cytochrome P-450. Feeding low-protein

Table 1. *The stimulation of cytochrome P-450 induction by dietary oils*

(Phenobarbitone 1 mg/ml in drinking water; diets and phenobarbitone fed for 10 d. Results expressed as means ± SD, with number of animals in parentheses)

Concentration of oil in diet	Cytochrome P-450 (n-mol/g liver)
Fat-free diet	45 ± 8 (28)
10% coconut oil	58 ± 17 (4)
10% olive oil	66 ± 9 (4)
10% maize oil	81 ± 12 (4)
10% linseed oil	92 ± 14 (4)
10% herring oil	111 ± 9 (4)
10% cod-liver oil	111 ± 18 (8)
2·5% cod-liver oil	92 ± 14 (4)
Pellet diet 41B	120 ± 21 (40)

diets depresses the activity of this enzyme system and also greatly reduces the response to phenobarbitone (McLean & McLean, 1966; Marshall & McLean, 1969).

In rats fed high-protein diets, the concentration of cytochrome P-450 achieved with phenobarbitone treatment is determined largely by the nature of the dietary fat (see Table 1). It is apparent that the stimulation of induction increases with the increasing concentration of more highly unsaturated fatty acids in the oils. Because the oils themselves, in the absence of phenobarbitone, are only weak inducers, we describe their effect as permissive, in that they permit induction by phenobarbitone (Marshall & McLean, 1971). The 10% maize-oil diet supplies adequate essential fatty acids for normal growth, yet is insufficient to allow complete expression of the potential for cytochrome P-450 synthesis in rats given phenobarbitone. A diet containing only 2·5% cod-liver oil is equivalent in terms of its permissive effect to one containing 10% maize oil, suggesting some specific role for the more highly unsaturated dietary fatty acids in the determination of microsomal enzyme activity and hence in the response of an animal to its environment.

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The effect of foetal, postnatal and postweaning undernutrition on acetylcholinesterase in regions of the rat brain. By B. P. F. ADLARD and J. DOBBING, *Department of Child Health, University of Manchester*

Acetylcholinesterase (*EC* 3.1.1.7) is localized mainly in the nerve ending particles (synaptosomes) of a brain homogenate. Synaptosomes sediment in the crude mitochondrial fraction (Banik & Davison, 1969) whose other components contain negligible acetylcholinesterase and it is, therefore, possible that the enzyme activity in this fraction may be a good quantitative index of synaptic connexions.

Undernutrition of foetal rats was begun on the 7th day of gestation. During the foetal and suckling periods maternal rats were given a reduced quantity of normal diet each day, representing approximately 50% of the average normal daily intake during each period. Animals further undernourished beyond weaning were given a normal diet, but restricted in quantity, allowing growth at a linear rate towards a body-weight of 70 g at 12 weeks of age compared with about 270 g in controls.

At 3 weeks of age undernourished animals weighed 36% of controls. Enzyme activity (Table 1) was significantly reduced in the forebrain, brainstem and olfactory lobes (but not in the cerebellum) of undernourished animals. These results suggest a deficit in the numbers of synaptosomes per g tissue and, by implication, an impaired or delayed formation of synaptic connexions in young undernourished weanling animals.

Table 1. *Acetylcholinesterase activity in the crude mitochondrial fraction of forebrain (F) and brainstem (B)*

Age (weeks)	No. of animals	Brain region	Weight† (g)		Activity† (n-mol/g per min)		% Change in undernourished	
			Control	Undernourished	Control	Undernourished	Per g wet wt	Per whole region
3	20	F	0·950 ± 0·024	0·778 ± 0·070	2193 ± 118	1843 ± 253	−16%***	−31%***
3	20	B	0·239 ± 0·005	0·196 ± 0·013	2965 ± 179	2742 ± 123	−8%**	−24%***
12	6	F	1·115 ± 0·032	0·854 ± 0·023	4454 ± 291	5349 ± 421	+20%**	−8%NS
12	6	B	0·355 ± 0·012	0·276 ± 0·007	2832 ± 125	3376 ± 112	+19%***	−7%*

NS, not significant; $*P < 0·05$; $**P < 0·01$; $***P < 0·001$
†Mean values and standard deviations.

At 12 weeks of age undernourished animals weighed 26% of controls but had significantly higher enzyme activities per g wet weight in the forebrain and brainstem (Table 1). The total enzyme activity per whole region was not, however, raised, although it had considerably increased in spite of continuing nutritional retardation of growth. It is therefore possible that the ultimate total number of synaptosomes approaches normal although the tissue weight is much reduced.

We gratefully acknowledge support from the Medical Research Council, the National Fund for Research into Crippling Diseases and the Spastics Society.

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The effect of γ-radiation on tryptophan in wheat gluten. By M. T. NADEEM CHAUDRY and R. A. EVANS, *Department of Biochemistry and Soil Science, University College of North Wales, Bangor, Caernarvonshire*

Metta & Johnson (1959) claimed that tryptophan in maize, measured microbiologically, was not affected by up to 9·3 Mrd of γ-radiation, but they did not study this amino acid in wheat gluten. They did not find the digestibility or biological value of wheat gluten for rats was affected by 2·8 Mrd.

Kennedy (1965) showed that treatment of wheat gluten with between 0·2 and 5 Mrd doses reduces the relative nutritive value, measured by the microbiological method of Ford (1960). Amino acid supplementation studies and microbiological assay showed that this reduction could be attributed largely to the loss of availability of methionine. Tryptophan was not studied in this work.

Hepburn, Calhoun & Bradley (1966) showed that the availability of tryptophan in unirradiated wheat gluten is close to 100% when measured by increase in carcass nitrogen or by faecal analysis.

We have analysed samples of wheat gluten, subjected to 0, 0·5, 2·5, 5·0 and 10·0 Mrd γ-radiation, for total and available tryptophan. Total tryptophan was determined

by alkaline hydrolysis and ion-exchange chromatography. Available tryptophan was determined as described by Nadeem Chaudry & Evans (1971). The results, given in Table 1, indicate observable changes occurring below 2·5 Mrd.

Table 1. *Total and available tryptophan in wheat gluten treated with γ-radiation*

Radiation dose (Mrd)	0	0·5	2·5	5·0	10·0
Total tryptophan: g/16 g N	0·80	0·80	0·76	0·73	0·69
% of zero dose	100	100	94	91	86
Available tryptophan: % of zero dose	100	98	73	62	53

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Towards a rapid biological assay for available tryptophan. By M. T. Nadeem Chaudry and R. A. Evans, *Department of Biochemistry and Soil Science, University College of North Wales, Bangor, Caernarvonshire*

A chick biological assay for available tryptophan has been described by Harwood & Shrimpton (1969). The response criterion that was used was the food conversion efficiency from 10 to 18 d of age. There can be little hope of shortening the duration of an assay based upon whole-body response, since this involves relatively long-term changes in components which differ widely in amount and in half-life.

Tyrosine transaminase is a hepatic enzyme which because of its short half-life can be used to measure protein synthesis rapidly. The principal factor controlling the concentration of this enzyme is the dietary protein and, in particular, the tryptophan in that protein. Munro (1970) states that 'the tryptophan content of the amino acid mixture absorbed from the intestine determines the intensity of protein synthesis in the liver during the absorptive period'.

We have measured the increase in tyrosine transaminase produced by feeding a test diet to rats under standard conditions. This increase should reflect the dietary tryptophan which has been absorbed. Wistar rats, approximately 190 g live weight, were fasted for 16 h, then fed 8 g test diet containing 35% protein as a moist paste. This was rapidly and completely consumed. The rats were killed 4 h later, and their livers removed and assayed for tyrosine transaminase activity by a modification of the method of Diamondstone (1966). Wheat gluten was used as a tryptophan standard in the biological assay instead of the free amino acid. A standard curve was obtained by diluting the gluten with zein which has a negligible tryptophan content.

Gluten and four protein meals supplied by the ARC Protein Evaluation Group were analysed for total tryptophan by alkaline hydrolysis followed by ion-exchange

chromatography. The protein meals were then assayed by the tyrosine transaminase procedure to give values for available tryptophan relative to gluten. The results, given in Table 1, show that the meals are ranked in the same order by both criteria.

Table 1. *Relative tryptophan content of protein meals*

	Total tryptophan (g/16 g N)	Available tryptophan (% of gluten)
Gluten	0·80	100
Groundnut meal GN101	0·85	128
Fish meal FM101	0·63	54
Fish meal FM102	0·62	74
Meat meal MM101	0·55	45

This rapid biological procedure may be useful in comparing protein sources for available tryptophan. Further investigation is needed of the most appropriate test animal, of the method of food presentation, and of standard and diluent proteins.

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A comparison of ileal and faecal analysis to determine the availability of dietary amino acids. By SHIRLEY A. VARNISH and K. J. CARPENTER, *Department of Agricultural Science and Applied Biology, University of Cambridge*

Payne, Combs, Kifer & Snyder (1968) have suggested that amino acid analysis of ileal contents rather than of faeces may provide a more reliable estimate of what is unavailable to the animal, since materials may be lost from the large intestine as a result of fermentation rather than direct absorption. A comparison of the methods has now been made with two damaged protein preparations and two of good quality, for which the results of growth assays for available amino acids have already been reported (Varnish & Carpenter, 1970).

For the determination of digestibility by faecal analysis with chicks, the procedure of Nesheim & Carpenter (1967) was followed, using test diets of 20% crude protein and a nitrogen-free diet all containing Cr_2O_3. For ileal analysis, 8-week-old chicks were fed a N-free diet (containing no Cr_2O_3) for 2 d, fasted overnight and then each was given a 15 g meal of one of the diets used for faecal analysis. After 2·75 or 3·5 h the chicks were killed and the ileal contents freeze-dried, acid-hydrolysed and analysed for chromium and amino acids. The 'true' digestibilities by each method, calculated using the results with the N-free diets, are summarized in Table 1.

Table 1. *Percentage digestibility of amino acids from four materials as calculated from ileal (il) or faecal (fc) analysis*

	Chicken muscle				Lactalbumin			
	Control		Heated		Control		Propionylated	
	il	(fc)*	il	(fc)	il	(fc)	il	(fc)
Lysine	98	(98)	52	(60)	93	(94)	73	(81)
Methionine	99	(98)	64	(73)	95	(94)	80	(88)
Means of values for alanine, valine, leucine and isoleucine	99	(98)	52	(60)	95	(94)	81	(86)

*The means in this column are based on only two independent estimates; all the other values are based on three or four estimates.

For the control proteins, digestion was already almost complete at the ileal stage. With damaged materials the digestibility at the ileal stage was 5–9 percentage units less than the corresponding conventional value from faecal analysis. However, the ileal values for the lactalbumin with propionylated lysine groups are not in agreement with the results of earlier growth assays which indicated a very large damage to lysine and no significant damage to methionine and tryptophan. Possibly some of the lysine is being absorbed even before the ileum in an unavailable form.

We thank Dr D. W. T. Crompton for carrying out operations on chicks to allow separate collection of urine and faeces.

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Some effects of a sucrose-free diet on fasting serum lipid levels. By A. M. Roberts (*British Antarctic Survey*), *Department of Physiology, Guy's Hospital Medical School, London SE1*

There is a lack of information on the long-term effects, in healthy young people, of a sucrose-free diet. In this experiment, nineteen male subjects, aged 21–40 years (mean=24·5 years), living on an Antarctic base, were studied for 11 months. Measurements were made of calorie intake, sucrose intake, body-weight and the serum glyceride, cholesterol, and phospholipid concentrations after a 12 h fast. During the period of study, 4 weeks were spent on a normal diet (mean sucrose intake=107 g/d, SE±6·6), followed by 14 weeks on a virtually sucrose-free isocaloric diet (mean sucrose intake=4 g/d, SE±0·3). The study continued for a further 24

weeks on the normal diet (mean sucrose intake=97 g/d, SE±5·7). Serum samples were obtained fortnightly throughout the study.

A fall seen in fasting glyceride concentrations during the sucrose-free period was not significant, but when the normal sucrose-containing diet was resumed a significant rise in the fasting serum glyceride concentration occurred, reaching a peak at 8 weeks, followed by a return to the pre-dietary level over the following 16 weeks. A rise in the fasting serum phospholipid level after resuming a sucrose-containing diet was also found. There were no significant changes in weight or serum cholesterol concentrations, nor was there any significant correlation between weight changes and glyceride changes.

Previous observations in the Antarctic (Easty, 1963) showed that there were no seasonal changes in glyceride concentrations on a normal diet. Other long-term experiments with reduction in dietary sucrose intake (Mann, Truswell, Hendricks & Manning, 1970; Rifkind, Lawson & Gale, 1966) showed greater falls in fasting glyceride concentrations, but their subjects were older and also lost weight during the study. The observations in these latter experiments only continued for 1 month after the resumption of a sucrose-containing diet, whereas the maximum fasting glyceride concentration in the Antarctic subjects did not occur until 8 weeks after the cessation of the sucrose-free diet.

I should like to express my gratitude to the subjects in this study.

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Interrelationship of dietary carbohydrates and fats on serum lipid concentrations. By I. MACDONALD, *Department of Physiology, Guy's Hospital Medical School, London SE1*

It is known that the type of fat in the diet can influence the concentration of serum cholesterol and that the concentration of triglycerides in fasting serum may be affected by the amount, and possibly the type, of carbohydrate in the diet. It was, therefore, decided to study whether the lipid response to the type of dietary fat could be affected by the type of dietary carbohydrate consumed with the fat, and vice versa. Ten healthy male (18–21 years) and eight healthy female (18–20 years) students were given, for 5 d, a diet containing 45·5% carbohydrate calories and 45·5% fat calories. The protein was calcium caseinate. The intake of the diet (made up as a liquid) was approximately 40 kcal/kg body-weight daily and was adjusted to keep the body-weight constant.

The fats used were either sunflower-seed oil or cream and the three carbohydrate mixtures used were (1) two parts fructose : three parts raw maize starch (2) two parts fructose : three parts glucose (3) two parts glucose : three parts raw maize starch. Samples of serum were obtained after a 12 h fast on the morning the diet started and on the penultimate and final mornings before the subject returned to his free-choice diet. Serum cholesterol (Block, Jarrett & Levine, 1965) and triglyceride (Lofland, 1964) estimations were semi-automated.

With sunflower-seed oil in the diet there followed a significant fall in the fasting serum triglyceride concentration, irrespective of the type of carbohydrate eaten and of the sex of the consumer. In the men, the cream caused a significant rise in the fasting serum triglyceride concentration on the fructose–starch and glucose–starch carbohydrate mixtures, but not on the fructose–glucose mixture.

In the experiments on the women, the cream was not associated with any significant change in the fasting serum triglyceride level.

I am very grateful to the volunteers.

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Changes in the concentrations of lipids in human plasma resulting from alterations in the nature of the dietary carbohydrate and in calorie intake. By D. J. NAISMITH, ANNE L. STOCK and JOHN YUDKIN, *Department of Nutrition, Queen Elizabeth College, London W8 7AH*

The replacement of starch with sucrose in the diet of man causes a rise in the fasting plasma concentration of triglycerides (MacDonald & Braithwaite, 1964; Szanto & Yudkin, 1969; Nestel, Carroll & Havenstein, 1970). It has been suggested, however, that the apparent hyperlipidaemic effect of a high-sucrose diet could be explained by a failure to control calorie balance (Antonis, Iles & Pilkington, 1968), and it has recently been confirmed by Nestel *et al.* (1970) that overeating a high-carbohydrate diet can result in a marked increase in the plasma triglyceride concentration.

In the present study, the hypothesis that the hyperlipidaemia associated with the consumption of a high-sucrose diet is due to a concomitant but unrecognized increase in energy intake was examined.

The normal food intakes of twenty-three healthy male students were measured for 7 d. The sucrose intake of each subject was then raised by 200 g at the expense of starch-rich foods and the modified diets were consumed for 14 d. The volunteers then returned to their normal diets for a further 14 d. Blood samples were drawn at the end of each period for lipid analyses.

At the end of the high-sucrose period, the plasma concentrations of triglycerides,

total cholesterol and phospholipids had risen by 18 mg, 14 mg and 19 mg/100 ml respectively, but returned to normal values within 14 d. These changes were statistically highly significant ($P<0.01$).

Although mean values for energy intake were very similar for the normal and high-sucrose diets (3040 and 3094 kcal), individual values showed considerable variations. The subjects were divided into an overeating group (12) who, on average, had increased their energy intake by 360 kcal and an undereating group (11) who had reduced their intake by 279 kcal. No differences were found between the two groups in the plasma lipid response to an increased consumption of sucrose

In the next experiment, the normal food consumption of twelve healthy students was measured for 5 d. Food intakes were then raised by approximately 1800 kcal/d for 21 d, six of the subjects consuming a high-sucrose diet and six a high-starch diet. The volunteers then returned to their normal diets for 12 d. Blood samples were taken at the end of each period and in the middle of the high-energy period for analyses for lipids.

On the high-energy high-sucrose diet, plasma triglycerides, total cholesterol and phospholipids rose by 19 mg, 55 mg and 38 mg/100 ml respectively, but returned to normal values within 21 d. On the high-energy high-starch diet, neither the triglyceride nor cholesterol concentrations showed any significant change, whereas the phospholipid concentration fell by 30 mg/100 ml.

It is concluded that the hyperlipidaemia which results from the consumption of sucrose-rich diets is induced by sucrose itself and not by any change in voluntary calorie intake.

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Chromium and sucrose–induced hyperglyceridaemia. By K. R. BRUCKDORFER, I. H. KHAN and JOHN YUDKIN, *Department of Nutrition, Queen Elizabeth College, London W8 7AH*

There is now considerable evidence that diets high in sucrose play a part in the causation of ischaemic heart disease (Yudkin, 1967). A possible mechanism is that such diets may be low in chromium (Schroeder, 1968), since deficiency of chromium increases the concentration of plasma cholesterol (Schroeder & Balassa, 1965).

Four groups each of eight litter-mate weanling rats of the Sprague Dawley strain were given diets with 68% carbohydrate. The diets contained either maize starch or sucrose, and the drinking-water was with or without the addition of 5 ppm chromium as the acetate. It was calculated that the supplemented animals received about thirty times as much chromium as did the unsupplemented.

The addition of chromium did not affect the cholesterol concentrations, nor did

it significantly reduce the high concentration of triglyceride of the rats given sucrose (Table 1). It produced a significant increase in the concentration of phospholipids in the rats given starch, so that it now became equal to the concentration in rats given sucrose.

Table 1. *Plasma lipid concentrations (median values, mg/100 ml) in rats given diets with 68% starch or sucrose, and supplied with drinking-water with or without 5 ppm chromium as the acetate*

Diet	Phospholipid	Triglyceride	Cholesterol
Starch	120	57	76
Starch + chromium	147	50	81
Sucrose	143	114	83
Sucrose + chromium	146	88	73

The sucrose diets increased the concentration of total lipid in the liver, and this was not affected by chromium supplementation.

Taking all these changes together, we conclude that the effects of chromium in diets containing sugar do not support the view that the atherogenic effect of sucrose depends on the induction of chromium deficiency.

K.R.B. acknowledges the receipt of a Lord Rank Research Fellowship.

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Effects of feeding soya products to pre-ruminant calves. By R. H. SMITH and C. F. WYNN, *National Institute for Research in Dairying, Shinfield, Reading RG2 9AT*

Smith, Hill & Sissons (1970) reported that when calves were, for the first time, given a liquid feed in which all the protein was provided by soya flour, movement of digesta along the alimentary tract was fairly normal. After several such feeds, however, stomach emptying was inhibited, transit times through the small intestine were decreased and abnormally large volumes of effluent left the ileum. These reactions showed similarities to some of the reported effects of dietary wheat gluten believed to be involved in the aetiology of certain malabsorptive states in human subjects. This and the fact that the reactions appeared to occur only after a calf had been sensitized by earlier feeds suggested the possible involvement of a gastro-intestinal allergy.

Circulating antibodies to constituents of enzymic digests of wheat gluten have been demonstrated in patients with coeliac disease (Taylor, Thomson, Truelove & Wright, 1961). A closely similar haemagglutination technique, using tanned red cells coated with a pepsin, trypsin digest of soya flour (55% protein, heat-treated) showed negative reactions with serums from calves given only cow's milk. Serums from calves, aged 4–6 weeks, given diets mainly of cow's milk but with 30% of their

nitrogen intake supplied respectively by added soya flour (55% protein, heat-treated) (three calves) or isolated soya protein (three calves), showed mean titres for circulating antibodies which were low after 2 weeks on the supplemented diets and approached maximum values of 20 000 ± 10 000 and 140 ± 100 respectively after 6–10 weeks. All these calves remained in good health, but in other experiments in which soya products provided the only protein in liquid diets only isolated soya protein proved satisfactory. Such diets containing soya flour (even though heat-treated) usually led to diarrhoea and loss of weight as well as to the disturbances of digesta flow described above; they also led rapidly to high circulating antibody titres.

These results are consistent with an association between antibody production and the digestive disorders often resulting from the ingestion of soya flour.

Formation of circulating antibodies in calves given diets containing soya flours has also been reported by van Adrichem & Frens (1965) and van Leeuwen, Weide & Braas (1969), but the latter workers found that a diet containing isolated soya protein gave even higher antibody titres than a soya-flour diet.

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The effect of mineral deficiencies on energy utilization. By D. S. MILLER and SALLY R. PARSONAGE, *Department of Nutrition, Queen Elizabeth College, London W8 7AH*

Mitchell (1934) proposed that 'the net availability of metabolizable energy of a perfectly balanced ration is maximal'. It follows that any nutrient imbalance such as a dietary mineral deficiency would be expected to increase the thermic effect of such a diet. However, the evidence in the literature is controversial (Kleiber, 1945–6). One of the difficulties arises over the control of food intake. Mitchell undoubtedly intended the food intake of deficient and control animals to be the same, but many subsequent workers did not meet this criterion.

In the present experiments deficient and control rats were pair-fed. The deficiencies listed in the table were investigated for a period of 10 d by the method of Miller & Stock (1969) which is based on the comparative carcass principle.

All the deficient diets caused a decrease in voluntary food intake, the extent depending upon the mineral. Even though the control animals were pair-fed, they gained more weight than the deficient animals. Most of the mineral deficiencies that were studied showed an increased thermic energy % and a reduced net energy of the diet compared with the controls, but the differences are small and in most instances not significant. Potassium deficiency was exceptional in so far as the weight gains were as much as 30% different in favour of the controls, whereas the yields of net energy were very similar. This anomaly arises mainly because of differences in carcass water.

Table 1. *Effect of mineral deficiencies on energy intake, thermic energy (% intake) and net energy*

(Mean values with their standard errors)

Deficient mineral	No. of expts	Mean energy intake (kcal/ d per $W^{0.75}$)	Thermic energy (% intake) Deficient	Thermic energy (% intake) Control	Net energy (kcal/rat per d) Deficient	Net energy (kcal/rat per d) Control	*P** value
Sodium (casein diet)	5	181 ± 6·8	24 ± 2	23 ± 1	22·1 ± 2·0	22·4 ± 2·1	> 0·2
Sodium (wheat diet)	4	211 ± 4·0	37 ± 2	34 ± 1	16·8 ± 0·3	17·8 ± 0·5	0·15
Potassium	8	153 ± 3·2	18 ± 1	20 ± 1	17·7 ± 1·1	17·3 ± 1·1	0·02
Calcium	6	158 ± 4·6	21 ± 2	20 ± 2	17·3 ± 0·3	17·5 ± 0·3	> 0·2
Phosphorus	6	142 ± 2·5	19 ± 1	15 ± 1	14·5 ± 1·0	15·2 ± 1·0	0·04
Trace minerals	5	174 ± 2·5	19 ± 2	16 ± 3	18·0 ± 0·5	18·6 ± 0·5	0·17
All minerals	6	134 ± 6·6	17 ± 2	13 ± 3	14·1 ± 0·9	14·8 ± 0·9	0·05
None	4	210 ± 10·7	—	—	—	—	—

*Student's method for correlated samples.

The variations in the yield of net energy between the different control groups is due to the different levels of food intake. The correlation between thermic energy and energy intake is highly significant (r=0·58, P=0·0002). This factor accounts for most of the discrepancies in the literature, where one can find experiments in which the animals were fed *ad lib.* (Smith & Meyer, 1962), pair-fed (Kahlenberg, Black & Forbes, 1937) and pair-weighed (Kleiber, Boelter & Greenberg, 1940). Whereas we are not inclined to contradict Mitchell so far as minerals are concerned, the magnitude of the effect appears to be not greater than 5%, at least with rats.

This work was supported by a grant from the Science Research Council.

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The antibacterial action of the Ethiopian condiment 'chow'. By J. P. W. Rivers and A. W. Hill, *Queen Elizabeth College, London W8 7AH*

The antibacterial action of many common spices is well known although it is generally considered that at dietary levels this effect is of no practical significance. The mixture of fifteen spices used in Ethiopia called 'chow' is largely chili pepper, *Capsicum frutescens* L. (70%, w/w) and has a pungent flavour which is much prized. It is used for flavouring stew (wat) which is eaten at almost every meal. According to tradition chow is a preservative and Bruce (1820) commends it as a 'most wholesome . . . powerful antiseptic'. We have conducted some preliminary experiments which seem to confirm this view.

The growth of *Staphylococcus aureus*, *Salmonella typhimurium* and *Escherichia*

coli on sterile (121°/15 min) chow-impregnated nutrient agar plates was measured. Inocula of various strengths were used and growth was measured as the number of colonies after 18 h at 37°. Typical results are shown in Table 1.

Table 1

Chow concentration (g/100g)	*Staph. aureus* organisms/plate		*E. coli* organisms/plate		*Salm. typhimurium* organisms/plate
	200	1800	180	450	370
	Growth as % controls				
1	23	48	41	75	79
5	0	11	25	24	39
10	0	4	0	0	0
15	0	0	0	0	0

Even with the heaviest inoculum used, inhibition of growth was almost complete at 10% chow and total at 15%. The mean concentration of chow in 197 wats examined was 11·2% and it seems likely therefore that it may be of considerable use in preventing food poisoning. On those plates on which bacteria did grow, the characteristic pungent smell was lost and a sweet sickly odour was produced, thus providing an indication of the presence of viable organisms.

In a separate experiment, rats were given a stock diet containing chow at different levels and faeces were collected. Nutrient agar containing faeces at the 15% level was autoclaved and plates were prepared and inoculated as above. The faeces had significant antibacterial action which varied with the level of chow in the diet. Since *Klebsiella aerogenes* was found to be relatively insensitive to chow, the presence of antibacterial action in rat faeces is in accord with the work of Meghal & Nath (1962) on similar Indian spices, and the implications of this are being investigated.

It is suggested that chow may play an important part in preventing food poisoning by its action both as a preservative and as an indicator of bacterial growth.

This work is part of the International Biological Programme project in Ethiopia led by D. S. Miller to whom we are grateful for encouragement and advice.

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