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ABSTRACTS OF COMMUNICATIONS

The Two Hundred and Eighty-second Scientific Meeting of the Nutrition Society (One Hundred and Twelfth of the Scottish Group) was held at Trinity College, College Green, Dublin, on Friday, 5 September 1975, at 14.00 hours, when the following papers were read:

Food and nutrient consumption trends in Ireland 1961–71: a decade of change. By FRANK M. CREMIN and PATRICK A. MORRISSEY, *Department of Dairy and Food Chemistry, University College, Cork, Republic of Ireland*

Using values obtained from the Irish Statistical Bulletin (1972) and standard food composition tables, we have calculated changes in the mean per capita daily consumption of the thirteen major foodstuffs and nutrients from 1961 to 1971. A comparison of the intakes of the major nutrients with the recommended daily intakes of these nutrients indicates that the population as a whole is: (1) changing its preferences with respect to the major foodstuffs, the consumption of bread, flour and potatoes being considerably reduced while pig-meat, beef, poultry and margarine consumption is increased; (2) continuing to increase its consumption of protein (the mixture obtained from vegetable and animal sources yields an essential amino acid pattern which is 'ideal'); (3) receiving the recommended dietary allowances of the principal nutrients with the exception of vitamin D and riboflavin and the minerals sodium and magnesium, from the thirteen principal foodstuffs in the diet; (4) receiving an ever-increasing proportion of its daily energy intake from animal fat; and (5) now receiving 35% of its total energy from fat, the maximum level recommended by the American Heart Association ((US) National Research Council, 1974).

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A study of energy balance on ten male Ethiopian labourers on low energy intakes. By J. V. G. A. DURNIN, J. WOMERSLEY and R. CAMPBELL, *Institute of Physiology, The University, Glasgow G12 8QQ*

Considerable interest has been aroused recently by the possibility that many individuals, and perhaps even large populations, may be able to exist in reasonable physical equilibrium on comparatively low food energy intakes. These people often have muscular physiques, and their way of life requires at least moderate physical activity, yet they maintain energy balance on apparent intakes much below the

accepted standards (e.g. Miller & Rivers, 1969; Norgan, Ferro-Luzzi & Durnin, 1974).

However, there is often a lingering doubt as to the validity of the apparent food intakes, even when these are done by an accepted and reputedly accurate technique. Thus Ashworth (1968), in a study in a metabolic ward on some Jamaicans from a rural community, found that half of her sample of subjects were unable to maintain energy balance when they were fed the amounts of energy previously measured, in their homes, as their normal intake.

In the present experiment ten male Ethiopian labourers, representing those having the lowest intakes of a group of thirty-three men, were kept for 4 weeks in a metabolic unit of the Ethiopian Nutrition Institute in Addis Ababa. During this time, each individual was given a diet approximating to his normal in content and energy value, as had previously been measured in his home environment. Physical activity was maintained near the normal average level throughout the period. Total daily energy expenditure was measured during the entire 4 weeks. The body-weight of each man, unclothed, was taken first thing each morning. Skinfold thicknesses were also measured on several occasions throughout the 4 weeks.

On energy intakes ranging from 6·28 to 9·71 MJ (1500–2320 kcal)/d, in only three of the ten men was energy balance maintained: that is, in seven out of ten, on energy intakes which were apparently 'normal' for the individual, body-weight was being consistently lost. However, two men on 6·28 MJ (1500 kcal)/d stayed at a constant weight and one man on 8·37 MJ (2000 kcal)/d gained some weight, while carrying out a routine of moderate physical activity.

These findings, together with their relationship to some discrepancies in the calculated energy balance, will be discussed.

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Oral glucose tolerance test results in various eating disorders. By A. DARRAGH, *Psycho-Endocrine Centre, St James's Hospital, Dublin* 8, *Republic of Ireland* (introduced by C. W. M. WILSON)

There are various methods of evaluating the results obtained by the oral glucose tolerance test (OGTT). The glucose tolerance sum and the insulin efficiency index have been selected as simple and relatively precise methods for analysing OGTT results.

These parameters of evaluation have been applied to the results of the OGTT performed on patients in various clinical states: obesity (before and after weight loss); anorexia nervosa (untreated and treated); and subjects with a history of minimal carbohydrate intake before and after a period of carbohydrate loading.

An improvement in OGTT results following weight loss in obese subjects has been demonstrated. We have shown a contrast between the increased glucose tolerance in anorexia nervosa and decreased glucose tolerance following prolonged

dietary restriction of carbohydrate intake, and the normalization of the OGTT following successful treatment of anorexia nervosa or compulsive carbohydrate intake restriction.

Obesity and anorexia nervosa in relation to pituitary function. By J. G. DEVLIN, *St Laurence's Hospital, North Brunswick Street, Dublin 7, Republic of Ireland*

We have studied the hypothalamic–pituitary response to the hypoglycaemic stimulus in comparable groups of obese and anorectic patients, comparing both groups with a group of matched control patients. An abnormal growth hormone pattern was observed in the anorectic group; blunting of the growth hormone response was noted in the obese group, and no obvious evidence of insulin insensitivity noted. Comparisons will be made between this study and studies of patients with defined organic syndromes such as pituitary tumours and chromosomal abnormalities and the results discussed in the light of the possible role of hypothalamic–pituitary function in the genesis of obesity and anorexia.

Fasting metabolic rate of overweight adult rats. By K. J. MCCRACKEN, *Agricultural and Food Chemistry Research Division, Department of Agriculture, Northern Ireland, and The Queen's University of Belfast, Belfast BT9 5PX*

Dietary control of hyperlipidaemia. By VIVIEN REID, I. GRAHAM, N. HICKEY and R. MULCAHY, *Cardiac Department, St Vincent's Hospital, Dublin 4, Republic of Ireland*

A total of 110 adult out-patients entered a prospective study of the effect of dietary control of hyperlipidaemia between 1 January 1971 and 31 December 1973, and were followed until 30 June 1974. The mean age was 52 years (range 36–76). Seventy-nine were male and thirty-one female. Eighty-four patients had primary type II and twenty-six had primary type IV hyperlipidaemia. Secondary hyperlipidaemia was excluded in all cases.

Each patient was interviewed by the same dietitian before treatment. All patients were seen at least every 6 months. Diets were based on the recommendations of Fredrickson, Levy & Jones (1970). Patients with type II hyperlipidaemia were treated with a diet low in saturated fat and cholesterol, with substitution of polyunsaturated fats. Additional carbohydrate restriction was applied to patients with type IV hyperlipidaemia. Further energy restriction was applied to overweight patients.

In type II patients, mean serum cholesterol fell from 8·34 to 7·41 mmol/l, and body-weight from 72·3 to 69·1 kg over the first 6 months of treatment. In type IV patients, cholesterol fell from 8·50 to 7·28 mmol/l, triglyceride from 3·89 to 2·41 g/l and body-weight from 75·5 to 70·9 kg, also over the first 6 months of treatment. All these reductions were statistically significant ($P<0·05$). No statistical correlation was observed between reductions in body-weight and lipid

levels when examined by means of 'scattergrams' and calculation of correlation coefficients.

Long-term results were not suitable for statistical analysis since those patients who entered the study at a later date had not completed the follow-up period. However, over 3 years of follow-up study, lipid levels and body-weight continued to decrease at a more gradual rate, suggesting that the dietary treatment continued to be effective. No tendency for lipid levels or weight to 'rebound' was noted. It is concluded that diet can produce a significant reduction of lipid levels over 6 months, and that this effect may continue at a lesser rate in the long term.

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The effects of pregnancy on uptake and distribution of copper in the rat.

By N. T. DAVIES and R. B. WILLIAMS, *Rowett Research Institute, Bucksburn, Aberdeen AB2 9SB*

During pregnancy in rats, an increase in copper storage occurs in the maternal body in addition to Cu accumulation in the foetuses (Spray, 1950). However, it is not known whether this increased demand must be met by an increased dietary Cu content, or implies a more efficient utilization of dietary Cu. A study has therefore been made of uptake and distribution of ^{64}Cu, following intragastric dosing in pregnant and non-pregnant rats.

Female rats (18 d pregnant), previously maintained for 4 d on a diet low but not deficient in Cu, were dosed intragastrically with 10 μg Cu labelled with ^{64}Cu while under brief diethyl ether anaesthesia. After 6, 12 or 18 h they were killed and the whole bodies, whole guts plus contents and the uteruses with foetuses were separately assayed for ^{64}Cu. Non-pregnant animals were similarly injected with ^{64}Cu and killed 6 h after dosing.

Uptake of ^{64}Cu into the bodies of pregnant rats 6 h after dosing was double that in non-pregnant animals (Table 1). At all times studied, 92% or more of the administered dose could be accounted for, and the amount retained in the whole bodies with guts removed appeared to be independent of the numbers of foetuses. Thus in the 18 h group, where the foetal numbers ranged from two to eleven, the amount of Cu deposited in the maternal bodies, including the products of conception, varied between 6·85 and 7·55 μg and was not related to foetal numbers. However, the amount of this Cu transferred to the conception products 18 h after dosing appeared to be governed by the foetal number, as indicated by a significant correlation between foetal number (x) and μg Cu transferred to the conception products (y), which could be described by the equation $y=0{\cdot}084x+0{\cdot}037$ (r 0·99, $P<0{\cdot}01$). The amount of ^{64}Cu retained by the whole bodies (with gut removed) including the conception products appeared maximal at

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Table 1. *Partition of an intragastric dose of 10 μg ^{64}Cu in pregnant and non-pregnant rats, and its transfer to the products of conception*

(Mean values with their standard errors; no. of animals in parentheses)

Group		Time after dosing (h)	^{64}Cu recovered in whole bodies less guts and contents (μg)		Accumulation of ^{64}Cu in uteruses and foetuses (μg ^{64}Cu/g tissue)	
			Mean	SE	Mean	SE
Non-pregnant	(5)	6	2·64	0·61	—	—
Pregnant	(5)	6	5·39	0·44***	0·0115	0·0009
Pregnant	(4)	12	6·63	0·37	0·0155	0·0021
Pregnant	(4)	18	7·22	0·15	0·0222	0·0007

Results were calculated as μg Cu from the initial specific activity of ^{64}Cu.
Difference from non-pregnant group significant: ***$P<0·001$.

about 12 h after dosing, while transfer of ^{64}Cu to the conception products continued to increase at least up to 18 h after dosing.

The results of this study suggest that at the stage of pregnancy examined, that is, 18 d after conception, there is a more efficient utilization of dietary Cu in order to satisfy the increased demand for this nutrient.

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Zinc absorption in pregnancy and lactation. By N. T. DAVIES and R. B. WILLIAMS, *Rowett Research Institute, Bucksburn, Aberdeen AB2 9SB*

The rate of deposition of zinc in the products of conception during the later stages of pregnancy in the rat, and in the litter before weaning, proceeds at a greater rate than it does at any other stage of postnatal growth (Williams & Davies, unpublished results).

This suggests an increased demand for Zn by the pregnant and lactating animal to meet her own requirements and those of her young, which might be met by an increase in the absorption of Zn by the maternal gut.

The absorption of labelled Zn (^{65}Zn) was measured, in situ, in isolated loops of the duodenum (150 mm segments distal to the pyloric sphincter) of rats at different stages of pregnancy and lactation (Table 1) by the method of Davies & Nightingale (1975). A comparison of the rate of absorption with the accretion of Zn by the maternal–concepta complex at different times in pregnancy revealed that a significant correlation existed between demand for and rate of absorption of this metal, which could be described by the equation $y=0·00263x+0·286$ (r 0·96, $P<0·01$) where y is μg Zn absorbed/loop per 15 min and x is mean daily retention of Zn (μg) by the maternal–concepta complex.

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Table 1. *Rate of zinc absorption (μg absorbed/loop per 15 min) by isolated rat duodenal loops in situ at various stages of pregnancy and lactation*

(Mean values with their standard errors; no. of animals in parentheses)

Day of pregnancy	Pregnant animals		
	Mean	SE	
12	0·37	0·02	(5)
15	0·47	0·06	(4)
18	0·55	0·05	(6)••
21	0·72	0·05	(5)•••
Time post partum (d)	**Lactating animals**		
	Mean	SE	
2	0·67	0·07	(5)•••
6	0·81	0·14	(5)•••
14	0·91	0·11	(5)•••
28	0·36	0·04	(6)†††
All controls‡	0·38	0·03	(13)

Significance of differences between non-pregnant controls and pregnant or lactating animals: ••$P<0·01$, •••$P<0·001$; difference between animals at 14 d post partum and 28 d post partum: †††$P<0·001$.

‡Zn absorption in control animals of similar age was measured at intervals during the course of the experiment. No differences were found and all control values were pooled.

During lactation, however, further increases in the rate of absorption of Zn appeared to be a reflection of an increase in weight of the loop tissue which is consistent with hypertrophy of the gut, as is known to occur during lactation (Boyne, Fell & Robb, 1966). By the 28th day post partum, when litters were fully weaned and lactation had ceased, the absorption of Zn had declined to control values.

No evidence was obtained that any increase in Zn absorption was a consequence of the state of pregnancy itself.

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Leaf-protein concentrate as a source of vitamins. By WALTER BRAY (introduced by F. AYLWARD), *Department of Food Science, University of Reading, London Road, Reading RG1 5AQ*

Leaf protein has been recognized for its nutritional value and its use to provide supplemental protein in diets has long been suggested. However, in spite of the fact that leaf-protein concentrates (LPC) contain about 400 g non-proteinaceous components/kg, this aspect of the product has received little attention.

As part of a wide-ranging study of LPC in human nutrition we have examined a number of these compounds, particularly with reference to the ability of LPC to provide lipids, vitamins and minerals in the diet.

Green plants, especially the leafy portions, are well recognized as an important source of various vitamins. That LPC would also be rich in vitamins is to be expected, but the amount will depend on: (a) the plant materials used as a source; (b) the conditions of cultivation and harvesting; (c) the method used to obtain juice from the leaves and the amount extracted; and (d) the methods and conditions used for recovery of the LPC.

We have determined the concentration of vitamins and some related components in samples of lucerne LPC and have calculated the daily contribution of 25 g LPC (the amount supplying 15 g protein in the diet) as a percentage of the recommended daily allowance for 11–14-year-old males ((US) National Research Council, 1974). In the materials examined 25 g LPC would supply over 400% of the vitamin A, as β-carotene; over 50% of the vitamin E, over 35% of the riboflavin and nicotinic acid (+ tryptophan), and significant amounts (over 20%) of pteroylmonoglutamic acid, pyridoxine and pantothenic acid.

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The effect of vitamin C-deficient diet and leptazol on the regional distribution of brain ascorbic acid in guinea-pigs. By A. ODUMOSU and C. W. M. WILSON, *Department of Pharmacology, Trinity College, Dublin 2, Republic of Ireland*

Ascorbic acid (AA) is generally present at higher levels in the brain than in the plasma of guinea-pigs (Odumosu & Wilson, 1973). No other naturally occurring active compounds are present in comparable concentrations in guinea-pig brain. A scorbutogenic (vitamin C-deficient) diet induces weight-loss and reduced tissue AA concentrations in guinea-pigs after 27 d (Odumosu & Wilson, 1970*a*,*b*). Brain AA falls to 48% of normal in guinea-pigs given a scorbutogenic diet for 30 d.

Three groups of guinea pigs were given for 27 d a scorbutogenic diet either unsupplemented (group S), supplemented with AA (30 mg/kg) (group N) or supplemented with AA, followed by a single convulsant dose of leptazol (60 mg/kg body-weight) (group NL). The animals were then slaughtered and AA distribution in the brain was compared with that of an initial slaughter group (group C).

The results for male and female guinea-pigs are given in Table 1. Before administration of the scorbutogenic diet (group C), AA concentration was maximal in the mid-brain. A normal AA intake (group N) increased brain AA concentrations in both sexes and the elevation was most pronounced in the mid-brain ($P<0.02$).

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Table 1. *Regional distribution of ascorbic acid (AA) in brain of guinea-pigs given for 27 d diets deficient in vitamin C (group S), supplemented with 30 mg AA/kg (group N) or supplemented with AA and given Leptazol (60 mg/kg body-weight) (group NL) compared with that in an initial slaughter group (group C)*

(Mean values and standard deviations for six animals/group)

Group		Brain AA concentrations (mg/kg)					
		Male			Female		
		Forebrain	Mid-brain	Hindbrain	Forebrain	Mid-brain	Hindbrain
C		48·6±8·0	69·2±4·3	30·1±3·8	50·2±5·4	78·9±7·6	28·4±5·4
	P*	<0·05	<0·02	<0·05	<0·05	<0·02	<0·05
N		82·3±6·4	148·9±10·5	49·2±6·2	88·2±9·0	139·2±9·7	27·9±6·3
	P	<0·02	<0·02	<0·05	<0·05	<0·02	<0·05
S		21·9±3·9	62·5±4·9	8·1±2·9	34·5±4·3	65·4±2·8	1·2±0·5
	P	<0·02	<0·05	<0·05	<0·05	<0·05	<0·05
NL		82·7±2·0	87·2±7·4	32·9±1·1	84·8±4·1	65·6±5·9	32·2±0·4

*Significance of difference between adjacent groups.

The scorbutogenic diet (group S) reduced mid-brain AA to levels of 42 and 49% of the supplemented values in males and females respectively. The administration of leptazol with AA (group NL) reduced brain AA to 59% of the level in supplemented guinea-pigs ($P<0.01$). Brain AA concentrations were higher in the male guinea-pigs receiving leptazol (group NL) than in those of group S ($P<0.05$). This indirect evidence indicates that mid-brain AA plays a metabolic role in the homoeostatic function of the brain during the stress induced by convulsions in guinea-pigs.

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The metabolism of ascorbic acid in rheumatoid arthritis. By A. MULLEN and C. W. M. WILSON, *Department of Pharmacology, Trinity College, Dublin 2, Republic of Ireland*

There is increased need for, or increased destruction of, ascorbic acid (AA) in rheumatoid arthritis (Rhinehart, Freenberg, Baker & Christie, 1936). The purpose of this study was to determine whether AA metabolism differs between rheumatoid-arthritic patients and normal healthy controls. The AA metabolism of patients with rheumatoid arthritis, defined by the criteria of the American Rheumatism Association (1959), was compared with that of a healthy control group. The patients were referred from a rheumatology clinic in Dublin: 80% were living at home and 20% were hospitalized during the investigation. Twelve were

male and thirty were female, with an age range of 26–77 years and a mean age of 53 years. The duration of illness varied from 2 months to 11 years, and the disease was classified as being in acute, sub-acute or chronic stages. The healthy control group consisted of an age-related population of twenty-one volunteers, none of whom were taking supplementary AA.

The rheumatoid patients had a mean plasma AA value of 3·0 mg/l; 85% had plasma concentrations below the normally accepted range of 5–25 mg/l; their levels were compatible with a diagnosis of subclinical scurvy. The mean plasma value in the normal volunteers was 10·4 mg/l, SD 4·1. The plasma level was significantly lower in the rheumatoid subjects ($P<0.001$). Leucocyte AA levels were also significantly lower in the rheumatoid group ($P<0.001$). Of rheumatoid patients, 42% had leucocyte levels compatible with subclinical scurvy, corresponding to levels of less than 20 μg/10^8 cells. Urinary excretion of ascorbic acid was measured during a 24 h period in both groups. Their excretion did not differ significantly. Dietary intake of AA was also estimated. It did not differ significantly between the groups. AA concentrations were measured in plasma and leucocytes at 0, 0·5, 1 and 2 h after administration of 500 mg AA. The consequent rise in plasma values did not differ between the two groups. It therefore appears that rheumatoid patients can absorb the vitamin at the same rate as normal subjects. However, since intake and excretion of the vitamin did not differ between the groups it is probable that rheumatoid subjects utilize AA at a faster rate.

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Ascorbic acid values in malignant disease. By S. C. KAKAR and C. W. M. WILSON, *Department of Pharmacology, Trinity College, Dublin 2, Republic of Ireland*

Plasma and platelet ascorbic acid (AA) concentrations are diminished in leukaemic patients (Lloyd, Davis, Emery & Lander, 1972) and plasma and leucocyte AA values are significantly reduced in leukaemic children during remission, in comparison with the values in normal children (Kakar & Wilson, 1974). Leucocyte and plasma AA levels are also low in patients with neoplastic growths (Krasner & Dymock, 1974). Tumours have a higher AA content than the normal organs from which they originate (Goth & Littman, 1948). Leucocyte and plasma AA concentrations have been compared between normal adults and patients with a variety of neoplastic lesions (Table 1). Leucocyte and plasma values were significantly lower in the cancer patients. Tissue was removed from the centre of the neoplastic growths after operation. AA concentrations were compared in the neoplastic tissue with those in the adjoining normal tissue. In the skin tumours AA level was found to be significantly elevated and AA was concentrated 3·3 times in

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Table 1. *Leucocyte and plasma ascorbic acid (AA) levels in normal subjects and cancer patients, and AA levels in tumour tissue and adjoining normal tissue of cancer patients*

(Mean values and standard deviations where given)

	No. of subjects	Leucocyte AA level ($\mu g/10^8$ cells)	Plasma AA level (mg/l)
Normal subjects	10	45·2±14·8	7·0±4·0
Skin cancer	8	25·4± 5·9*	2·0±1·0*
Pulmonary cancer	7	25·9±10·7*	2·0±1·0*
Buccal region cancer	11	23·1± 8·9**	1·0±1·0**
Bladder neoplasm	2	20·5	1·3
Breast cancer	1	10·4	1·6
Rectal cancer	2	32·6	4·0

	No. of subjects	Normal tissue (μg/g)	Tumour tissue (μg/g)
Skin cancer	7	44±20	145±53***
Cancer of cervix	1		

Significance of differences between normal and cancer subjects, or between normal and tumour tissue: $*P<0.01$, $**P<0.002$, $***P<0.001$.

the neoplastic tissue. The value for AA in normal skin from the tumour patients was one-twentieth of that reported in skin samples taken from autopsies, or biopsies from non-cancerous patients during operation (Barton, Laing & Barisoni, 1972). It can be concluded that AA accumulates preferentially in neoplastic tissue. A corresponding absolute reduction in AA concentrations takes place in adjoining normal tissue and in plasma. The labile stores of AA in the leucocytes also become depleted.

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Effect of pharmacological doses of ascorbic acid on copper acetate-induced ovulation in the rabbit. By S. C. SHARMA, P. ROBINSON and C. W. M. WILSON, *Department of Pharmacology, Trinity College, Dublin 2, Republic of Ireland*

Prostaglandins may act as intraovarian mediators of physiological responses to luteinizing hormone (LH) (Armstrong, 1970). Indomethacin, an inhibitor of prostaglandin synthesis, can effectively inhibit ovulation in the rabbit (Grinwich, Kennedy & Armstrong, 1972; O'Grady, Caldwell, Auletta & Speroff, 1972). This effect can be reversed by the administration of prostaglandins (Orczyk & Behrman, 1972; Sato, Iesaka, Tyujo, Taya, Ishikawa & Igarashi, 1972). Recently it has been shown that ascorbic acid (AA) reduces synthesis of prostaglandin F (Pugh, Sharma

& Wilson, 1975). In this pilot study we have investigated whether administration of pharmacological doses of AA to the rabbit inhibits ovulation. Eighteen individually housed female New Zealand white rabbits were divided into four groups. Ovulation was induced by intravenous administration of copper acetate (0·3 mg/kg body-weight) and laparotomies were performed 24 h later. Intravenous AA (100 mg/kg body-weight) was administered at the same time as the copper acetate, or at varying intervals thereafter. Group A did not receive AA; group B received AA simultaneously; group C received AA 5 h after, and group D received AA in two doses at 3 h intervals, after administration of copper acetate.

Table 1. *Effect of administration of ascorbic acid (AA) (100 mg/kg body-weight) at varying intervals relative to copper acetate administration (0·3 mg/kg body-weight) on incidence of ovulation in rabbits*

Group	No. of animals	Total no. of corpora lutea recorded	No. of corpora lutea/ovary	
			Mean	SE
A (Control, no AA)	7	64	4·57	1·07
B (AA simultaneously)	2	14	3·5	—
C (AA 5 h later)	7	14	1·0	0·39
D (AA in two doses at 3 h intervals)	2	7	1·7	—

Results for the corpora lutea recorded are given in Table 1. Group C had significantly fewer corpora lutea than group A ($P<0·05$) AA evidently had an inhibitory effect on ovulation which was most marked when given 5 h after the copper acetate. It is not clear whether AA is acting at the ovarian, pituitary or hypothalamic level. Sufficient LH is released from the pituitary gland 1 h after coitus in the rabbit to permit ovulation (Fee & Parkes, 1929). If the stimulus due to copper acetate is acting in same way as natural mating, then the inhibition of ovulation 5 h after AA administration cannot be ascribed to its effect on LH release. The most reasonable interpretation is that AA acts by inhibiting synthesis of prostaglandins involved in the process of ovulation. However further work involving more animals and measurements of prostaglandins in the ovary is required.

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Effect of restricted postnatal nutrition on the growth rate and carcass composition of fattening lambs. By M. J. LAWLOR, M. A. CARROLL and D. B. R. POOLE, *An Foras Taluntais (Agricultural Institute), Dunsinea Research Centre, Castleknock, Co. Dublin, Republic of Ireland*

Twenty-four Suffolk cross lambs of uniform birth weight were weaned at 48 h and reared on a ewe milk-replacer. The lambs were individually penned and were randomized onto two treatments. The first consisted of a low daily milk allowance which amounted to an average of 125 g milk-replacer dry matter (DM) (0·625 l milk)/d. In the second treatment each lamb was given an average of 240 g milk-replacer DM (1·20 l milk)/d. The lambs were weaned off the milk-replacer at 8 weeks of age and six lambs from each treatment were slaughtered. A complete dissection and carcass analysis was carried out on each carcass. The remaining six lambs on each treatment were subsequently given a pelleted creep feed *ad lib.* until they reached a slaughter live weight of 37 kg. A dissection and carcass analysis was carried out on one half of each carcass from the fattened lambs.

The mean daily live-weight gains of the lambs given the low milk allowance was 87 g during the first 8 weeks compared with 187 g for those on the high milk allowance. Food conversion ratio (kg milk-replacer DM/kg live-weight increase) was poorer for the lambs on the low milk intake.

During the postweaning phase of the trial the mean daily live-weight gains of the lambs were 317 and 347 g for those previously given the low and high milk allowances respectively. The food conversion ratio was identical for both groups. The mean number of d required to fatten the lambs reared on the low milk allowance was significantly higher ($P<0·001$), being 88 d compared with 65 d for those on the high milk allowance.

The mean hot carcass weights of the pre-slaughter lambs were 4·5 and 7·9 kg for those reared on the low and high milk allowances respectively. The corresponding values for carcass bone, lean and fat (g/kg) were 272, 681 and 48 compared with 212, 683 and 106. The differences in bone and fat content were highly significant ($P<0·001$) No differences in carcass composition were observed for the lambs reared from either treatment which were fattened to a live weight of 37 kg.

Factors affecting the melting point and fatty acid composition in the carcass fat of lambs given cereal-rich diets. By J. L. L'ESTRANGE and C. SPILLANE, *Department of Agricultural Chemistry, University College Dublin, Glasnevin, Dublin 9, Republic of Ireland*

In earlier studies (L'Estrange & Mulvihill, 1975), it was observed that the melting point (MP) of both subcutaneous and perinephric fat of young lambs fattened indoors on diets rich in cereals was lower than that of older grass-fed lambs and the difference was related mainly to lower proportions of stearic acid (18:0) and higher proportions of oleic acid (18:1) in the fat of the concentrate-fed lambs. Garton, Hovell & Duncan (1972) also reported unusually soft carcass fat in

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young lambs fattened on high-barley rations but, in contrast, they associated the softness with low levels of stearic acid and high levels of odd-numbered and branched-chain fatty acids in the fat.

To examine this problem further two experiments were carried out in which a number of factors were examined. In Expt 1, carried out from January to April, the effect of age was studied, by using thirty-two lambs (Finn–Galway × Dorset Horn) which were weaned at 5–7 weeks and then fattened to slaughter at 35–40 kg, and sixteen 1-year-old hoggets (Cheviot × Suffolk) fattened for 11 weeks before slaughter. In addition, the effects of ground maize and ground barley as the cereal source and of vitamin E and cobalt sulphate supplementation were compared. In Expt 2, carried out from June to September, the effects of whole and ground barley and of roughage supplementations with chopped hay (100 or 200 g/kg) and chopped straw (75 or 150 g/kg) were compared using young lambs (Galway × Suffolk), weaned at 5–7 weeks and fattened to slaughter at 35–40 kg bodyweight.

In Expt 1, the MP of subcutaneous and perinephric fat of the lambs averaged 30·8° and 38·7° respectively and were about 4° lower than corresponding values for the hoggets. In subcutaneous fat this was associated with lower proportions of palmitic (16:0) and stearic acids and higher proportions of odd-numbered and branched-chain fatty acids, the amounts of these unusual acids being similar to the values reported by Garton *et al.* (1972). However in Expt 2 the MP and fatty acid composition of both fats, even on the rations without roughage supplementation, were similar to those of the hoggets in Expt 1, and high proportions of odd-numbered and branched-chain fatty acids were not observed.

In both experiments the effects of dietary treatments on fat characteristics were small.

The results indicate that age, breed of lamb, and the time of year of feeding are important factors in relation to MP and fatty acid composition of the fat of concentrate-fed lambs.

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Food intake and utilization by growing lambs with parasitic damage to the abomasum or small intestine. By A. R. Sykes and R. L. Coop, *Moredun Research Institute, Edinburgh EH17 7JH*

An attempt was made to induce chronic parasitism by continuously dosing 4-month-old lambs, reared parasite-free from birth, with either a small-intestinal (*Trichostrongylus colubriformis;* 2500 larvae/d) or abomasal (*Ostertagia circumcincta;* 4000 larvae/d) parasite. In each trial, conducted in consecutive years, an initial control group (C) was killed, two groups were fed *ad lib.*, one of which was infected (I) and a fourth group (PF) were pair-fed to the intake of the infected group. The complete diet 'Ruminant A', described by Wainman, Blaxter & Pullar

(1970), was used; balance trials were conducted in infected and pair-fed sheep and the sheep were killed after 13 weeks on trial. Body energy content was calculated using values of 38·9 and 22·2 MJ/kg respectively for the energy values of fat and protein (Blaxter & Rook, 1953).

Parasitism of the abomasum caused a 22% ($P<0.001$ from week 2) reduction in food intake, a slight reduction in energy digestibility (Table 1) and a marked reduction in nitrogen digestibility, particularly in the early stages of infection. N and fat deposition were reduced and the efficiency of energy utilization was reduced by 30%. No overt clinical signs of disease were seen.

Table 1. *Energy and nitrogen digestibilities, carcase composition and energy utilization in growing lambs infected (I) with* Trichostrongylus colubriformis (2500 *larvae/d*) *or* Ostertagia circumcincta (4000 *larvae/d*) *compared with pair-fed (PF) controls and an initial slaughter group (C).*

		O. circumcincta				*T. colubriformis*			
Digestibility	Weeks	No. of animals	I	PF	LSD	No. of animals	I	PF	LSD
Energy	2–3	8	0·550	0·575	0·016				
	6–8	8	0·531	0·549	0·024	8	0·524	0·526	0·031
	12–13	8	0·504	0·522	0·030	6	0·534	0·557	0·026
N	2–3	8	0·440	0·593	0·086				
	6–8	8	0·547	0·641	0·075	8	0·559	0·570	0·029
	12–13	8	0·614	0·668	0·036	6	0·624	0·597	0·036
		Group				Group			
Carcase analysis (kg)		C	I	PF	LSD	C	I	PF	LSD
Fat		1·87	5·41	7·01	1·31	2·51	6·31	7·89	1·570
Protein		3·25	4·12	4·36	0·848	3·63	4·25	5·22	0·460
Retention of digestible energy (%)		nd	11·9	17·0	—	nd	11·9	18·2	—

LSD, least significant difference; nd, not determined.

Parasitism of the small intestine produced clinical signs in one lamb, after 10 weeks. In general, food intake was reduced by 9%, particularly in the later stages of the trial. Apparent digestibility of energy and N were unaffected, but deposition of N and fat were severely impaired (Table 1). Efficiency of energy utilization was reduced by 35%.

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The eating behaviour of steers offered grass silage *ad lib.* in troughs with and without a barley supplement. By R. K. WILSON and A. V. FLYNN, *An Foras Taluntais (Agricultural Institute), Dunsinea Research Centre, Castleknock, Co. Dublin, Republic of Ireland*

Trough-feeding is a method used to feed grass silage to groups of cattle in this country. The time animals spend eating, ruminating and resting under these conditions has received little attention.

Animals from a twelve-treatment production trial (three silages, early, late and regrowth, × two methods of conservation, unwilted and wilted, × two levels of supplementation, 0 and 3 kg barley/d) were available. The animals (ten Friesian steers/treatment) were offered silage *ad lib.* in troughs. The times the animals spent eating, ruminating and resting were recorded by radio-telemetry during the 83 d feeding trial (October–February). Complete 24 h recordings were obtained for thirty-four animals (three/treatment except for two treatments).

There were no significant differences in animal eating behaviour due to type of silage. The over-all means (±SD) were (h/d): eating 5·1±1·3, ruminating 7·8±1·8 and resting 11·1±2·3. No differences in eating behaviour were noted for wilted and unwilted silages or for early- and late-harvested silages. Barley supplementation tended to reduce eating time, 4·6±0·8 *v.* 5·50±0·3 h/d, and ruminating time, 7·30±0·3 *v.* 8·1±1·1 h/d.

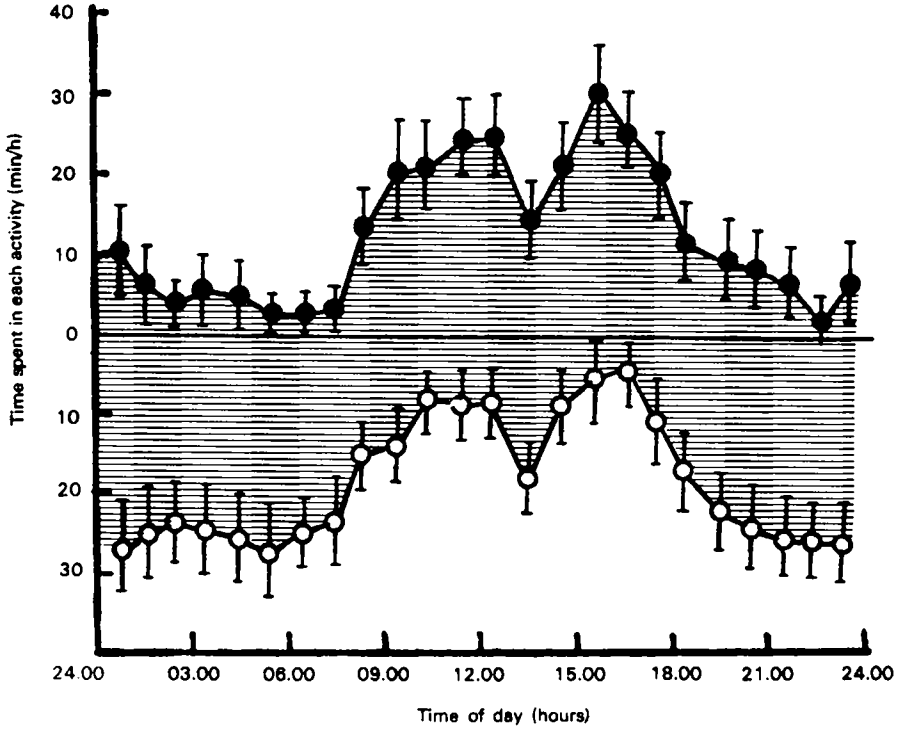

Fig. 1. Average times (min/h), with 95% confidence limits (vertical bars), spent eating (●——●) and ruminating (○——○) by beef cattle offered grass silage *ad lib.* in troughs during the period October–February. ▣, Time spent eating and ruminating.

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Animals showed definite eating and ruminating patterns: 71% of eating and 23% of ruminating were between 08.00 and 18.00 hours (Fig. 1). The most intense period of eating was just before nightfall, as observed in feed-lot cattle (Ray & Roubicek, 1971). Time given to resting (absence of jaw movement) was remarkably constant (27·5±3·5 (SD) min/h).

The results obtained are similar to those for stall-fed cattle (Wilson & Flynn, 1974). The low live-weight gains of cattle eating silage, relative to summer grazing, may be partly due to animals restricting their period of active eating.

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Some non-protein nitrogen compounds in silage and their metabolism in the bovine rumen. By T. W. GRIFFITHS and R. K. WILSON, *An Foras Taluntais (Agricultural Institute), Dunsinea Research Centre, Castleknock, Co. Dublin, Republic of Ireland*

Hughes (1970) has shown that in well-fermented grass silage the protein fraction is extensively degraded, with the production of large amounts of free amino acids, and particularly alanine and glycine. Chalmers & Hughes (1969) found that whilst alanine and lysine were slowly deaminated in the rumen, glycine was not deaminated and disappeared very slowly from the rumen of sheep.

As part of an investigation into possible reasons for the low voluntary intake of silage by cattle, two silages, with and without barley, were offered to two heifers of approximately 400 kg live weight, fitted with rumen cannulas and trained to wear a device which measured jaw movements and recorded time spent eating, ruminating and resting (Wilson & Flynn, 1974). The silages, which were offered at rates just below *ad lib.* intake, contained 51 and 45% of the total nitrogen as non-protein-N but similar amounts of glutamic acid, glycine, alanine, valine and leucine. Most of the amino acids found in the silages were identified in rumen liquor 1–3 h after the diets were offered: maximum concentrations were found after 2 h and, with the exception of alanine (which was present in amounts >2·0 mmol/l), all had disappeared from the rumen after 6 h. The correlation coefficients between time spent eating (/h) and total N, alanine and glycine in rumen liquor were 0·91, 0·97 and 0·98 respectively. Polyethylene glycol administered directly into the rumen 1 h after the diets were offered decreased linearly with time. The results suggest that all amino acids were rapidly metabolized in the rumen of animals given silage and were not likely to affect voluntary intake.

Rumen microbial protein was isolated from rumen liquor using the technique of Pearson & Smith (1943). Selected amino acids determined in this material (mg/g total amino acid N) for the silage and silage–barley diets respectively were: glutamic acid 145, 147; glycine 71, 71; alanine 111, 106; valine 59, 61; leucine 75, 78 and lysine 64, 62. The material isolated contained a higher proportion of alanine and glycine (present in considerable amounts in the silages) and a lower

level of lysine (almost absent from silage extracts) than that obtained from non-silage diets (Ibrahim & Ingalls, 1972; Liebholz, 1972).

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Observations on the ensilage and feeding to sheep of partially dewatered rumen contents. By R. K. WILSON, W. SHEEHAN* and M. F. MAGUIRE, *An Foras Taluntais (Agricultural Institute), Dunsinea Research Centre, Castleknock, Co. Dublin, Republic of Ireland*

The disposal of rumen contents from slaughtered animals poses a problem at most meat-processing factories. The preservation of rumen contents by ensilage has been demonstrated (Nilsson, 1969) but a filler (milled straw) is required. In the work reported here rumen contents were dewatered and ensiled and the acceptability of the silages measured with sheep.

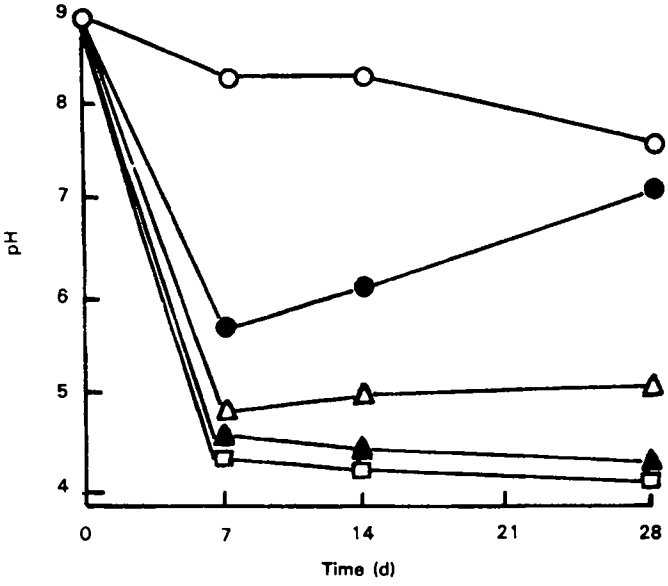

Fig. 1. Change in pH of rumen-fibre silage on storage. Rumen fibre was ensiled with 0 (○), 30 (●), 60 (△), 90 (▲) or 120 (□) g barley–malt meal/kg.

Rumen fibre was obtained from rumen contents by passing the latter through an endless-belt press. This process raised the dry matter (DM) from 130 to 250 g/kg. Rumen fibre was mixed with 0, 30, 60, 90 and 120 g barley–malt meal

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(milled barley–malt meal, 5:1)/kg and ensiled in 500 g lots in laboratory silos. The silages were assessed after 7, 14 and 28 d. At the 90 and 120 g/kg levels of supplementation, stable lactic acid-type silages were produced. The pH changes are shown in Fig. 1; other characteristics followed the trends expected from the pH results.

A rumen-contents silage (Nilsson, 1969) and a rumen-fibre silage were offered to two groups of ten sheep for 15 d. DM intakes over the last 9 d were (mean±SD) 61·9±11·9 and 57·9±8·4 g DM/kg body-weight$^{0·75}$.

In a second trial rumen-fibre silage, grass silage and grass silage plus barley were offered to three groups of six sheep for 45 d. DM intakes over the last 40 d were (mean±SD) 63·6±2·9, 48·2±2·6 and 62·1±2·8 g DM respectively. Changes in live weight over the latter trial were (mean±SD) −0·5±1·3, −0·5±1·0 and 2·7±0·6 kg respectively. Rumen-fibre silage had the following composition (g/kg): DM 269, crude protein (nitrogen × 6·25) 145, ash 86, acid-detergent fibre 283, IVD 441.

Ensilage and subsequent feeding to ruminants offers a method of disposal of rumen contents.

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Effects of dietary hydrochloric acid on voluntary food intake of rats.

By J. L. L'ESTRANGE and P. K. UPTON, *Department of Agricultural Chemistry, University College Dublin, Glasnevin, Dublin* 9, *Republic of Ireland*

Mineral acids, potentially useful as silage additives, have been shown to have an adverse effect on voluntary food intake of ruminants (L'Estrange & Murphy, 1972; L'Estrange & McNamara, 1975). This effect has been partly ascribed to palatability associated with dietary pH and partly to metabolic disturbance associated with metabolic acidosis (L'Estrange & McNamara, 1975). The response of the rat to dietary hydrochloric acid was examined in this study with a view to elucidating the problem more fully.

In Expt 1, weanling rats were given commercial rat pellets alone or supplemented with HCl up to 560 mmol/kg dry matter (DM). In Expt 2, adult rats were given a similar diet alone, or supplemented with HCl up to 1250 mmol/kg DM. The results are summarized in Table 1, and show that the levels of dietary HCl used did not affect food intake of the weanling rats, despite lowering dietary pH to 3·50 at the high level. With the adult rats, there was no effect on food intake with levels up to 625 mmol/kg DM, which lowered dietary pH to 2·84. At higher levels dietary HCl decreased food intake and caused a high mortality rate.

The results are in contrast with the more gradual and marked effects of dietary HCl on food intake of sheep (L'Estrange & McNamara, 1975). They indicate a difference between the simple-stomached animal and the ruminant in the mode of action by which mineral acids affect food intake.

Table 1. *The effect of dietary hydrochloric acid on food intake, water intake and live-weight gain in weanling rats given HCl for 7 weeks (Expt 1) and in adult rats given HCl for 9 weeks (Expt 2)*

(Mean values for eight rats/treatment)

	Supplement to basal diet (mmol HCl/kg DM)	pH of diet	DM intake (g/d)	Water intake (ml/d)	Live-weight gain (g/d)
Expt 1	None	5·90	14·6	28·9^{b}	3·93
	280	4·60	14·2	33·6^{a}	4·20
	420	4·10	14·6	36·5^{a}	4·07
	560	3·50	14·4	36·3^{a}	3·96
SE		—	0·50	0·87	0·23
Expt 2	None	5·80	14·8^{a}	33·9^{b}	−0·31^{c}
	312	4·17	15·8^{a}	35·7^{b}	−0·22^{c}
	625	2·84	15·3^{a}	44·8^{a}	−0·47^{c}
	938	2·23	10·7^{b*}	44·2^{a*}	−2·40^{a*}
	1250	1·82	9·56^{b*}	25·9^{c*}	−1·72^{b*}
SE		—	0·38	1·47	0·15

DM, dry matter.
Mean values without a common superscript letter are significantly different ($P<0·05$).
*Where some or all of the animals died during the experiment, the values refer to means obtained during the period while the animals survived.

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A comparison between the effects of dietary hydrochloric and lactic acids in the rat. By P. K. UPTON and J. L. L'ESTRANGE, *Department of Agricultural Chemistry, University College Dublin, Glasnevin, Dublin 9, Republic of Ireland*

An increase in consumption by cattle and sheep of silage made without mineral acid additives, brought about by neutralization of the silage with sodium bicarbonate (McLeod, Wilkins & Raymond, 1970), indicates that organic acids in silage have an adverse effect on food intake of ruminants. Previous work in this laboratory showed, however, that the effect of lactic acid on food intake of sheep and cattle was much less than that of hydrochloric acid (Morgan & L'Estrange, 1973). In this study, the effects of dietary lactic acid and HCl on food intake and metabolism of animals were further studied using the rat as an experimental animal.

Weanling rats, weighing approximately 60 g, were offered commercial rat pellets alone or supplemented with HCl or lactic acid at levels up to 900 mmol/kg dry matter (DM). The results (Table 1) show that lactic acid at the level used had no effect on food intake while HCl at a level of 900 mmol/kg DM reduced food intake and live-weight gain and increased blood acidity. This experiment further supports the suggestion that increasing HCl concentrations in the diet are well tolerated up to threshold levels, beyond which increased supplementation results in a serious decline in food intake.

Table 1. *Effects of lactic and hydrochloric acids on food intake, water intake, live-weight gain and blood acid-base status in weanling rats*

(Mean values for three rats/treatment)

Dietary supplement (mmol/kg DM)	pH of diet	Food intake (g DM/d) (day 0–84)	Water intake (ml/d) (day 0–84)	Live-weight gain (g/d) (day 0–84)	Blood pH	Plasma CO_2 (mmol/l)	Plasma base excess (mmol/l)
None	5·81	15·4 ^{a}	39·2	2·57 ^{a}	7·36	22·0 ^{ab}	−3·20 ^{a}
300 lactic acid	4·60	14·9 ^{a}	40·9	2·80 ^{a}	7·36	22·2 ^{ab}	−3·70 ^{a}
600 lactic acid	4·04	16·1 ^{a}	39·2	2·80 ^{a}	7·39	21·6 ^{ab}	−2·87 ^{a}
900 lactic acid	3·71	14·8 ^{a}	37·5	2·58 ^{a}	7·35	20·2 ^{ab}	−5·62 ^{a}
300 HCl	4·27	15·0 ^{a}	41·3	2·52 ^{a}	7·35	22·7 ^{a}	−3·89 ^{a}
600 HCl	3·09	14·0 ^{a}	44·9	2·21 ^{a}	7·37	19·4 ^{b}	−5·69 ^{a}
900 HCl	2·54	7·2 ^{b}	33·2	0·84 ^{b}	7·24	15·3 ^{c}	−12·97 ^{b}
SE	—	0·85	1·98	0·19	0·09	0·95	1·16
F-test †	—	•••	NS	•••	NS	•••	••

DM, dry matter.

†Where the *F*-test shows a significant treatment effect, mean values without a common superscript letter are significantly different ($P<0·05$).

NS, not significant; ••$P<0·01$, •••$P<0·001$).

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Effects of high intakes of dietary sodium sulphate and sodium chloride on voluntary food intake of rats. By P. K. UPTON and J. L. L'ESTRANGE, *Department of Agricultural Chemistry, University College Dublin, Glasnevin, Dublin* 9, *Republic of Ireland*

In an earlier series of experiments on the mechanism by which mineral acids and sulphate salts affect food intake of ruminants (L'Estrange, Clarke & McAleese, 1969; L'Estrange, Upton & McAleese, 1972) it was shown that supplementary sulphur in the diet of sheep, at levels above 5 g/kg dry matter (DM), progressively decreased food intake. Studies by Bird (1972) suggested that this effect was associated with the microbial reduction of sulphate to sulphide in the rumen and the subsequent absorption of this more toxic form of S.

In two experiments carried out with rats, the direct effect of dietary sulphate on food intake of the animals was examined on the assumption that the rat is unlikely to metabolize sulphate to sulphide prior to absorption. Expt 1 was carried out with weanling rats fed over a 6-week period on a commercial rat diet alone, or supplemented with sodium sulphate up to 10 g S/kg DM. In Expt 2, adult rats were given a similar diet for 6 weeks alone, or supplemented with sodium sulphate up to 20 g S/kg DM. In each experiment one level of sodium chloride equivalent to the high level of sodium sulphate was also included. The results are summarized in Table 1. Over-all, dietary supplementation with sodium sulphate did not significantly affect food intake and live-weight gain of rats. The results support the

Table 1. *Effects of dietary sodium sulphate and sodium chloride on dry matter (DM) intake, water intake and live-weight gain in weanling rats (Expt 1) and adult rats (Expt 2) treated for 6 weeks*

(Mean values for eight rats/treatment)

	Supplement to basal diet (g/kg)	DM intake (g/d)	Water intake (ml/d)	Live-weight gain (g/d)
Expt 1	None	13·4	$28·0^{c}$	4·54
	5 sulphur as Na_2SO_4	13·4	$35·6^{ab}$	4·50
	7·5 S as Na_2SO_4	13·6	$34·9^{b}$	4·61
	10 S as Na_2SO_4	13·5	$36·2^{b}$	4·49
	36·6 NaCl	13·4	$38·7^{a}$	4·15
	SE	0·43	0·98	0·13
Expt 2	None	13·8	$36·8^{d}$	−1·42
	10 S as Na_2SO_4	15·7	$51·2^{c}$	−1·25
	15 S as Na_2SO_4	13·7	$50·9^{c}$	−1·73
	20 S as Na_2SO_4	15·2	$57·4^{b}$	−1·95
	73·1 NaCl	13·4	$66·2^{a}$	−2·07
	SE	0·59	1·62	0·30

Mean values without a common superscript letter are significantly different ($P<0·05$).

suggestion that the adverse effect of dietary sulphate in the ruminant is caused by its reduction to the more toxic sulphide form in the rumen.

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The effect of dietary molybdenum and sulphate on plasma copper distribution in sheep. By I. BREMNER, *Rowett Research Institute, Bucksburn, Aberdeen AB2 9SB*

Increase in the dietary concentration of molybdenum and sulphate in sheep causes an increase in plasma copper concentration which is partly associated with the occurrence of a novel Cu-binding fraction (Suttle & Field, 1968), characterized *inter alia* by its insolubility in dilute trichloroacetic acid (TCA) (Smith & Wright, 1975). As the nature of this potentially important fraction has not been established, some preliminary studies on its isolation and identification have been undertaken.

Heparinized plasma was collected at intervals from 1 to 30 weeks from three groups each of four sheep maintained on a semi-purified diet (Suttle & Field, 1968) containing 10 mg Cu and 0·8 g sulphur/kg. One group was given a supplement of 25 mg Mo/kg (as ammonium molybdate) and another group 25 mg Mo/kg+5 g sulphate/kg (as Na_2SO_4). The plasma samples were analysed for various Cu-containing fractions and for Mo (Table 1). Caeruloplasmin (*EC* 1.16.3.1) activity was not significantly affected by the dietary treatments. Only in the animals

receiving Mo plus sulphate were there any changes in the concentration or distribution of Cu. About half the Cu in these samples was in a TCA-insoluble form, the absolute concentration tending to vary with plasma Mo concentration. No such relationship existed in the sheep receiving Mo alone, as all the Cu was in a TCA-soluble form despite the greatly increased Mo concentrations.

Table 1. *Concentrations of copper and molybdenum in sheep plasma*
(Mean values with their standard errors)

Treatment	Concentration in plasma (μg/ml)							
	Total Cu		TCA-soluble Cu		Direct-reacting Cu		Total Mo	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Control	0·90	0·03	0·93	0·05	0·17	0·02	ND	
+Mo	1·05	0·04	1·03	0·04	0·16	0·01	16·3	0·9
+Mo+sulphate	1·45	0·07	0·70	0·03	0·68	0·11	0·90	0·08

TCA, trichloroacetic acid; ND, not detected.

Two Cu-containing fractions were isolated from the plasma of the Mo plus sulphate-supplemented sheep by gel filtration on Sephadex G-75. One was eluted at the void volume of the column, and contained all the caeruloplasmin activity and mainly TCA-soluble Cu. The second fraction, of lower molecular weight, contained most of the TCA-insoluble Cu and usually at least 75% of the plasma Mo. A similar association of the TCA-insoluble Cu with Mo was obtained on fractionation of the plasma on DEAE-Sephadex A-50, which also yielded two principal Cu fractions, one consisting mainly of caeruloplasmin.

The yields of the TCA-insoluble Cu during fractionation tended to be low, except when procedures were carried out under nitrogen, suggesting that this fraction was labile and subject to oxidative change. This was confirmed by its almost complete conversion into a TCA-soluble form by aeration of a plasma sample.

These results suggest that Cu and Mo are closely associated in this novel protein fraction, which binds Cu firmly only when oxidation is prevented and whose formation is dependent on increased dietary S intake.

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Predicting the effects of dietary molybdenum and sulphur on the availability of copper to ruminants. By N. F. SUTTLE and M. MCLAUCHLAN, *Moredun Research Institute, Edinburgh EH17 7JH*

Although molybdenum is known to act as a biological antagonist of copper, attempts to correlate the incidence of bovine hypocuprosis with the Mo content of pastures have yielded conflicting results. Since dietary sulphur potentiates the Cu–Mo antagonism, a closer correlation has been sought by relating the availability of dietary Cu for sheep to the S and Mo concentrations in a large number of experimental diets. The results came mostly from nine published

experiments (e.g. Suttle, 1973, 1974*b*) in which the true availability (TA) of Cu was predicted from responses in plasma Cu, using a repletion technique (Suttle, 1974*a*). Concentrations of S and Mo in the basal semi-purified diet varied within the normal ranges for herbage (i.e. 1·0–4·0 g S and 0·5–4·5 mg Mo/kg dry matter (DM)). A further unpublished experiment involved lambs on a bruised-oat–blood-meal diet, Mo concentrations of 0·5–16·5 mg/kg and accumulation of Cu in liver as the measure of the TA of Cu.

Table 1. *Multiple regression equations relating the true availability of dietary copper* (y) *to molybdenum and sulphur concentrations (in mg and g/kg dry matter, respectively) in the diet of sheep*

Equation no.	Dependent variable	Independent variables	Constant	Coefficient			Variance accounted for (%)
				Mo	S	Mo×S	
1	y	Mo, S, Mo×S	0·070	−0·0015	−0·0094***	−0·0002	68·0
2	$\log y$	Mo, S, Mo×S	−1·153	−0·0019	−0·0755***	−0·0131*	85·7
3	y	log Mo, log S, log Mo×log S	0·059	−0·0056	−0·0485***	−0·0235	75·5
4	$\log y$	log Mo, log S, log Mo×log S	−1·234	−0·313	−0·494***	−0·576***	83·3

Significance of coefficients: *$P<0.05$, ***$P<0.001$; others $P>0.05$.

Of the relationships examined (Table 1), the best fit was obtained from a regression of log TA Cu *v.* S and S×Mo in the diet (Equation 2; r 0·93, $P<0.001$, 28 df): this equation implies that S exerts a predominant and independent effect on Cu availability, whereas Mo has a lesser and S-dependent effect. Responses to dietary S and S×Mo were exponential rather than linear, indicating that increments at the lower end of the normal ranges of concentrations have relatively large depressing effects on Cu availability. The accuracy with which such equations predict the availability of Cu in natural diets will depend on the extent to which other factors, such as soil ingestion, exert independent effects on availability or affect the course of the Cu–Mo–S interaction. Equation 2 predicts a high Cu availability (0·059) for cereal-rich diets which are associated with susceptibility to Cu poisoning (Todd, 1972) and a low availability (0·019) for herbage of marginally high Mo content (6 mg/kg DM) from areas associated with subclinical bovine hypocuprosis (Thornton, Kershaw & Davies, 1972).

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An approach to the rapid measurement of 'reactive lysine' in foods by dye binding. By R. F. HURRELL and K. J. CARPENTER, *Department of Applied Biology, University of Cambridge, Cambridge CB2 3DX*

Methods to determine reactive or 'available' lysine in food proteins, using fluorodinitrobenzene (FDNB) or trinitrobenzene sulphonic acid (TNBS) followed

by hydrolysis of the proteins, are relatively complicated (cf. Carpenter & Booth, 1973). Measurements of dye-binding capacity (DBC) are simple and rapid (cf. Lakin, 1973). Hurrell & Carpenter (1975) found that for many food materials the DBC with Acid Orange 12 was equivalent to the sum of the total histidine, arginine and reactive lysine. We have now used the difference in DBC of materials, before and after the masking of their reactive lysine groups by propionylation, as a measure of lysine specifically. Since starting this work we have seen descriptions of two similar procedures in which lysine is blocked by either ethyl chloroformate (Sandler & Warren, 1974) or TNBS (Jones, 1974) but propionylation may have advantages.

Propionylation was carried out by vigorously shaking a sample containing *c.* 5 mg lysine for 15 min with 0·2 ml propionic anhydride and 2 ml half-saturated sodium acetate solution (or double quantities for low-lysine samples) in a glass tube stoppered at each end. The suspension was then subjected to the 'Foss' DBC procedure as before (Hurrell & Carpenter, 1975). The ordinary DBC measurement was made with 2 ml acetate solution added to the 40 ml dye solution. Typical results for a series of vegetable materials and pure proteins, comparing 'dye-binding difference' (DBD) values (Jones, 1974) with direct values for FDNB-reactive lysine (Booth, 1971) using a standard '×1·09' correction factor, are given in Table 1.

Table 1. *Reactive lysine concentrations (mmol lysine/kg crude protein (nitrogen×6·25)) in vegetables and pure proteins determined by 'dye-binding difference' (DBD) and by reaction with fluorodinitrobenzene (FDNB)*

	DBD	FDNB		DBD	FDNB
Wheat gluten	110	100	Broad bean (*Vicia faba*)	330	340
Whole wheat	190	180	Soya bean	370	390
Rice	200	200	Chick-pea (*Cicer arietinum*)	440	410
Sweet corn (*Zea mays rugosa*)	220	220	Wing bean (*Psophocarpus tetragonolobus*)	500	470
Groundnut flour	230	220	Bovine plasma albumin	810	830

These results show good agreement but fish meals and meat meals have usually given DBD values only 0·7–0·9 of the corresponding FDNB values. This may be due to incomplete propionylation under the conditions used so far, and to the practical difficulty of transfer from a small vessel to one suitable for the dye-binding reaction.

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Dietary habits of lactating women and infant feeding practices in the Mid-Western State of Nigeria. By OSAFU OGBEIDE, 20*B Willenhall Road, London SE*18

Malnutrition and death amongst children are major problems in Nigeria. The nutrient intake of the Nigerian population has been shown to be unsatisfactory (Omololu, 1972). There is a general low intake of protein, riboflavin and thiamin (Olusanya & Omololu, 1972). The energy intake is marginal with the lowest intakes found in the Mid-Western State of Nigeria. Thus lactating women must be consuming foods which are inadequate to meet their increased physiological needs: this could result in malnutrition in the breast-fed babies. The type of local foods and imported weaning foods used for feeding babies and the way these foods are given could add to the nutritional problems of infants. Improvements in food production and nutrition are seriously hindered by the traditional food practices, preferences and habits of particular sectors of the population. This paper will therefore focus on the relationship between dietary habits of mothers, feeding practices of infants and malnutrition among children in Benin with an overview to the Mid-Western State of Nigeria.

We interviewed 250 mothers of apparently healthy children from the Urban Health Centre (1–5 years clinic), Ministry of Health, Benin and 230 mothers of malnourished children from the Nutrition Rehabilitation Unit, Ministry of Health, Benin, using questionnaires.

It was observed that in the past, the tradition in the area ensured an adequate, nutritious diet and sufficient rest for about 3 months after parturition for lactating women. The survey showed a shift from this practice, resulting in a general poor standard of nutrition of these mothers. The factors which were observed to be responsible for this were over-work due to breakdown of extended family units, customs, food taboos, irregular feeding habits and poor food intake as early as 1–2 months post partum. All these factors hinder lactating mothers from obtaining an adequate diet to meet their requirements and that of the breast-fed infants, thus initiating malnutrition in the babies at an early age.

Although the traditional patterns of weaning are far from perfect, new ideas which have been introduced have worsened the situation. The most important of these were found to be early supplementation and sudden weaning of infants. Malnutrition among children in the Mid-Western State of Nigeria is, therefore, associated with poor nutrition of lactating mothers, bad weaning practices and low weaning ages of infants in the community.

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