EFFECTS OF PASTURE SILAGE ON YIELD AND COMPOSITION OF MILK FROM DAIRY COWS

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ABSTRACT

Factors affecting milk yield and composition were reviewed and it was concluded that feeding level and diet type (notably unwilted pasture silage) may have independent effects on yield and composition of milk.

The objective of this research was to investigate the separate effects of forage type and feeding level on milk yield and composition and to study some of the mechanisms that may effect the efficiency of utilization of DE in these diets for milk synthesis.

Initially effects on milk yield and composition and nutrient utilization were studied in a series of experiments in which dairy cattle were individually fed in stalls on varying levels of forage diets. Unwilted pasture silage was compared with pasture, wilted silage, formalin treated silage, and unwilted silage supplemented with pasture, maize silage or protein concentrates.

These experiments showed that both level of feeding and the type of forage diet offered to dairy cows can affect milk yield and composition.

Increasing the intake of pasture, increased the yields of milk, fat, protein and lactose, and increased milk protein percentage and decreased milk fat percentage. Similar results were obtained with unwilted silage except that no relationship was found between silage intake and milk protein concentration.

Cows offered unwilted pasture silage produced less milk containing a lower concentration of fat and protein than cows offered pasture at the same intake of DE. Neither ration affected milk lactose percentage.

The efficiency of utilization of DE for milk synthesis by cows fed silage was improved by either reducing protein degradation during ensiling, or by providing pasture or protein concentrates as supplements. Maize silage as a supplement had no effect. These studies indicated that the amount of protein entering the duodenum of cows offered unwilted silage was limiting milk protein synthesis.

Subsequently cows were surgically prepared with abomasal cannulae to test this hypothesis. Consistent and significant increases in milk yield, milk protein concentration and milk protein yield by cows fed unwilted silage were obtained when abomasal infusions of sodium caseinate were given in a series of studies. Further studies showed no responses were obtained with abomasal infusions of glucose which indicated that the response in milk protein synthesis was due to amino acids *per se*.

The magnitude of the responses in milk protein synthesis to abomasal infusions of casein were higher for cows offered silage in comparison to those fed pasture at similar intakes of DE. The difference in the responses of the cows on the two rations was due apparently to an inadequate supply of essential amino acids for cows fed silage in comparison with those fed pasture.

Abomasal infusions of L-methionine increased milk yield, milk protein percentage and milk protein yield to the same extent as casein suggesting that methionine might be the major essential amino acid that was limiting milk protein synthesis of cows fed unwilted pasture silage.

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LIST OF ABBREVIATIONS

b, B	partial regression coefficients
CH_4	methane
DE	apparent digestible energy
DEI	apparent digestible energy intake
DM	dry matter
DMI	dry matter intake
DN	apparent digestible nitrogen
DNI	apparent digestible nitrogen intake
df	degrees of freedom
d	day
FCM	fat corrected milk
G.L.C.	gas liquid chromatograph
g	grams
GE	gross energy
GEI	gross energy intake
h	hours
HAc	acetic acid
HPr	propionic acid
HBu	butyric acid
nHBu	normal butyric acid
IHBu	iso-butyric acid
nHVa	normal valeric acid
IHVa	iso-valeric acid
I.D.	internal diameter
kg	kilograms
LW	live weight
$\triangle$ LW	live weight change

milk N + NR	productive nitrogen
ml	millilitres
mg	milligrams
MJ	megajoules
meq	milliequivalents
MAD fibre	fibre extracted by the modified acid detergent method
milk protein	milk crude protein (N x 6.38)
NPN	non protein nitrogen
N	nitrogen
NR	nitrogen retention
NH_3-N	ammonia nitrogen
OM	organic matter
PCV	packed cell volume
r	simple correlation coefficient
R	Multiple correlation coefficient for overall regression model
RSD	residual standard deviation of regression model.
r.p.m.	revolutions per minute
SS	sums of squares
SEM	standard error of mean
SD	standard deviation
SED	standard error of difference between means
SNF	solids not fat
TCA	tricarboxylic acid
VFA	volatile fatty acids.

1. INTRODUCTION

Over the past 20 years pastoral products have provided about 80 % of New Zealand's (N.Z.) export earnings. One-third of this was obtained from dairy products. By world standards the N.Z. dairy industry is small but dairy products exported by N.Z. represent about one-third of the total international dairy trade.

N.Z. Dairy farming is based on grazing perennial pastures and is seasonally orientated. Surplus pasture is conserved either as silage or hay and is fed to cows as supplements in times of herbage scarcity. This farming system which has a low cost structure is highly competitive with systems in many overseas countries where expensive concentrates are extensively used to feed dairy cows.

Up to the present time, payments for milk have been on the basis of milk fat production. Therefore farmers have sought to maximise milk fat production by selection and breeding, and by increasing stocking rates and manipulating pasture supplies with strategic use of pasture supplements to equate them with cow requirements.

During the last 20 years the N.Z. dairy industry has undergone profound changes especially in diversification of products. Butter and cheese were the major salable products, but there has been a steady increase in the value of non-fat constituents, particularly milk protein which alone or in combination with other milk products form the basis of all dairy products other than butter. Thus in 1956, milk fat in the form of butter provided over 80 % of N.Z. dairy export earnings whereas in 1975 it had decreased to 43 %. Conversely milk protein products have changed from being of minor

importance to being the major earner of export income, reflecting the rising world demand for high quality protein.

The increasing importance of milk protein has led to schemes being developed for payment to be made to farmers for protein as well as fat production. A scheme has already been implemented by the Rangitaik Plains Dairy Company to encourage farmers to improve the protein content of milk. Improvements have been made, to a certain extent, by a partial change in the breed structure of the industry from the Jersey to the Friesian breed, the latter having a higher protein to fat ratio. However rising energy costs involved in the cartage of milk and manufacture of milk products is accentuated by the lower fat and solids not fat (SNF) in Friesian milk, and increases the need for farmers to consider ways in which they might now improve the concentration of these milk constituents, especially protein, to offset additional costs.

In the long term, breeding based on selection for high milk yields and high concentrations of fat and protein in milk is probably the most important way a farmer can exert his control. In the short term a farmer may be able to influence milk composition by manipulation of the diet. However the effect of diet on milk composition is not well understood and current feeding standards based on net energy and digestible crude protein do not specify product quality.

Research has shown that milk composition can be altered by varying the amount and quality of forage fed (Sections 2.1, 2.2). Stocking rate experiments have shown an increase in stocking rate reduces per cow milk yields and milk protein concentration but increases milk fat concentration (Section 2.2.1). Presumably both

the level of feeding and differences in pasture quality were implicated. Lower milk yields and concentrations of protein in milk have also been observed when the proportion of pasture silage has been increased in a forage ration but again it was not possible to differentiate between diet and level of feeding effects, since silage resulted in reduced food comsumption (Flux and Patchell, 1950).

It is therefore probable that on N.Z. dairy farms milk composition is altered by the feeding management imposed by the farmers. Selecting and breeding cows with potentially high milk yields coupled with increasing stocking rates have made adequate feeding more difficult especially in early lactation. This places greater reliance on the use of pasture supplements. Hence the traditional use of pasture and its conserved products, especially pasture silage, to provide low cost production has possibly led to lower intakes and poor utilization of digested nutrients for milk synthesis.

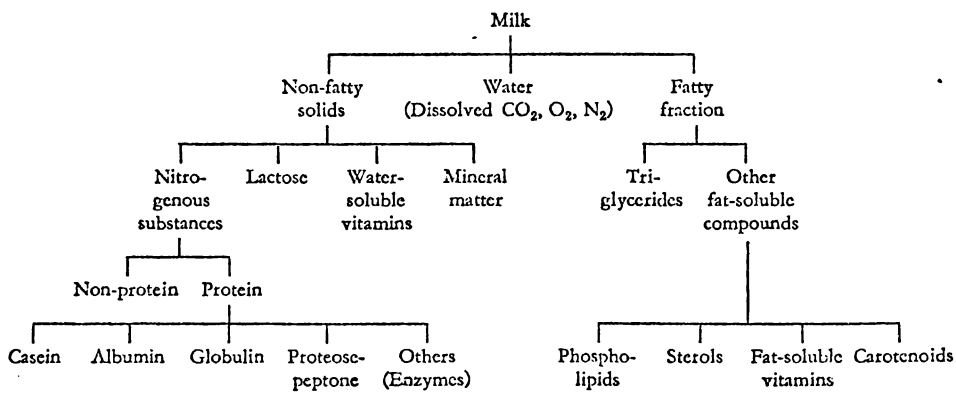
The present investigation was initiated to provide more information on the effects of pasture and high moisture silage and the level of feeding of these rations on milk yield and composition. Since milk synthesis is closely associated with digestion and metabolism of absorbed nutrients, the investigations included studies of the effects of modifying dietary nutrients and altering the site of digestion on the utilization of absorbed nutrients.

2. REVIEW OF LITERATURE

FACTORS AFFECTING MILK YIELD AND COMPOSITION

This review examines current knowledge of the factors effecting variations in the yield and chemical composition of cows' milk. Particular emphasis is given to the forage diets commonly used in N.Z. and to the major milk constituents, fat, protein, and lactose. The main constituents of milk as summarized by Ling, Kon, and Porter (1961) are shown in Table 1.

TABLE 1. Components of Milk



2.1 NON DIETARY FACTORS EFFECTING MILK YIELD AND COMPOSITION

A number of non nutritional factors affect milk yield and composition. These have been reviewed by Smith (1959), Jenness and Patton (1959), Rook (1961), Ling, et al. (1961), Laben (1963), Corbin and Whittier (1965), and only a brief account will be given here.

2.1.1 Breed and genetic structure of the individual animal

Studies by Overman, Garret, Wright and Sanmann (1939), have defined the composition of milk of the major breeds of dairy cow.

Jerseys produce milk usually with a higher fat content than Friesians and Ayrshires (see Table 2) but exhibit greater variability among individuals. Jersey milk often has a higher protein concentration than Friesian milk but the lactose contents are similar. Milk from Jerseys also contains a much higher proportion of carotene but less vitamin A than other breeds (Ling et al. 1961) and considerable differences have been shown between the breeds in protein composition (Rolleri, Larson and Touchberry, 1956).

Individual animals within a breed exhibit wide differences in composition of milk and within breed standard deviations are higher for fat, than for protein and lactose (Legates, 1960). Foot (1964) has shown that between herd differences in milk production and composition are due more to environmental and management factors than to the genetic potential of the animals.

Estimates of the correlations of fat and protein contents for different lactations of the same cow have been shown to be much larger than is the case of lactation yields of fat and protein (Touchberry, 1974). Heritability estimates of the percentages of milk constituents have also been shown to be approximately twice those for the lactation yield traits. This provided evidence that variations in environment has much less effect on the percentages of the various constituents of milk than on the lactation yields of milk and its constituents. Investigations by Wilcox, Gaunt and Farthing (1971) have shown that fat and protein contents in milk are not inherited individually and have an average genetic correlation of 0.45. Lactation milk yield and percentages of milk constituents are negatively correlated indicating that breeding for milk yield decreases the concentration of milk solids.

TABLE 2. Mean production of the main N.Z. dairy breeds.

Season	No. cows	Av. milk (l)	Av. milkfat %	Av. milkfat (kg)	Av. days in milk	Av. milkfat all tested cows (kg)
Ayrshires						
1951-52	7,454	2,970	4.18	124	253	129
1955-56	8,410	3,059	4.18	128	258	132
1959-60	8,848	3,297	4.22	139	262	143
1965-66	9,267	3,487	4.33	151	267	152
1969-70	10,340	3,222	4.28	138	252	129
1970-71	10,400	3,256	4.27	139	264	134
1971-72	10,252	3,446	4.29	148	267	146
1972-73	10,960	3,312	4.31	143	254	139
1973-74	10,797	3,230	4.27	138	252	135
1974-75	10,589	3,239	4.23	137	261	138
Friesians						
1951-52	6,683	3,888	3.70	144	262	129
1955-56	7,735	4,061	3.74	152	271	132
1959-60	9,782	4,317	3.80	164	273	143
1965-66	13,818	4,499	3.98	179	277	152
1969-70	19,728	4,283	3.97	170	268	129
1970-71	20,692	4,306	3.97	171	277	134
1971-72	21,942	4,468	4.03	180	280	146
1972-73	24,251	4,312	4.02	174	270	139
1973-74	25,669	4,253	4.04	172	271	135
1974-75	26,599	4,245	4.02	171	275	138
Jerseys						
1951-52	43,776	2,551	5.49	140	260	129
1955-56	49,781	2,617	5.46	143	264	132
1959-60	49,576	2,782	5.57	155	271	143
1965-66	51,898	2,866	5.76	165	272	152
1969-70	54,002	2,553	5.56	142	246	129
1970-71	52,884	2,593	5.59	145	259	134
1971-72	50,158	2,772	5.63	156	269	146
1972-73	51,455	2,677	5.58	149	254	139
1973-74	50,874	2,607	5.56	145	253	135
1974-75	49,506	2,640	5.50	145	259	138
M. Shorthorns						
1951-52	3,416	3,001	4.10	123	249	129
1955-56	3,160	3,076	4.13	127	257	132
1959-60	3,061	3,211	4.11	132	258	143
1965-66	2,481	3,408	4.17	142	262	152
1969-70	2,100	3,143	4.07	128	249	129
1970-71	1,777	3,145	4.10	129	258	134
1971-72	2,063	3,192	4.14	132	263	146
1972-73	2,047	3,153	4.11	130	248	139
1973-74	2,172	2,957	4.06	120	242	135
1974-75	2,138	3,170	4.03	128	257	138

Source: Anon. (1975)

2.1.2 Stage of lactation

Changes in milk yield and composition during lactation have been described by Rook (1961) and Lampo, Willems and Vanschoubroek (1966). The initial secretion of colostrum for about 4-5 days after parturition contains more minerals, total protein, casein and whey proteins with variable amounts of fat and less lactose than milk. Peak milk yield is usually attained 2-8 weeks after calving. Fat and protein decline in concentration during the first 6 weeks of lactation, then gradually increase until close to the end of lactation when they increase rapidly. Inverse changes occur in lactose content. N.Z. data (J.B. Hutton, unpublished) depicting these trends are shown in Figs. 1 and 2, include climatic and nutritional influences. The increase in protein after the sixth month of lactation is associated with pregnancy since the change only occurs if the cow is pregnant (Wilcox, Pfau, Mather and Bartlett, 1959). Hutton (1958) suggested these changes in milk yield and composition with advancing pregnancy were associated with increasing levels of blood oestrogen.

2.1.3 Age of cow

Several studies (Waite, White and Robertson, 1956; Legates, 1960; Rook, 1961) have indicated that fat content decreases by about 0.2 percentage units and SNF by about 0.4 percentage units over the first 5 lactations. The fall in the SNF content has been shown to be due to a drop in lactose percentage (Waite et al. 1956) and in casein (Robertson, Waite and White, 1956). N.Z. data (Anon., 1975) clearly shows that milk fat concentration declines and milk fat yield increases with age (Table 3).

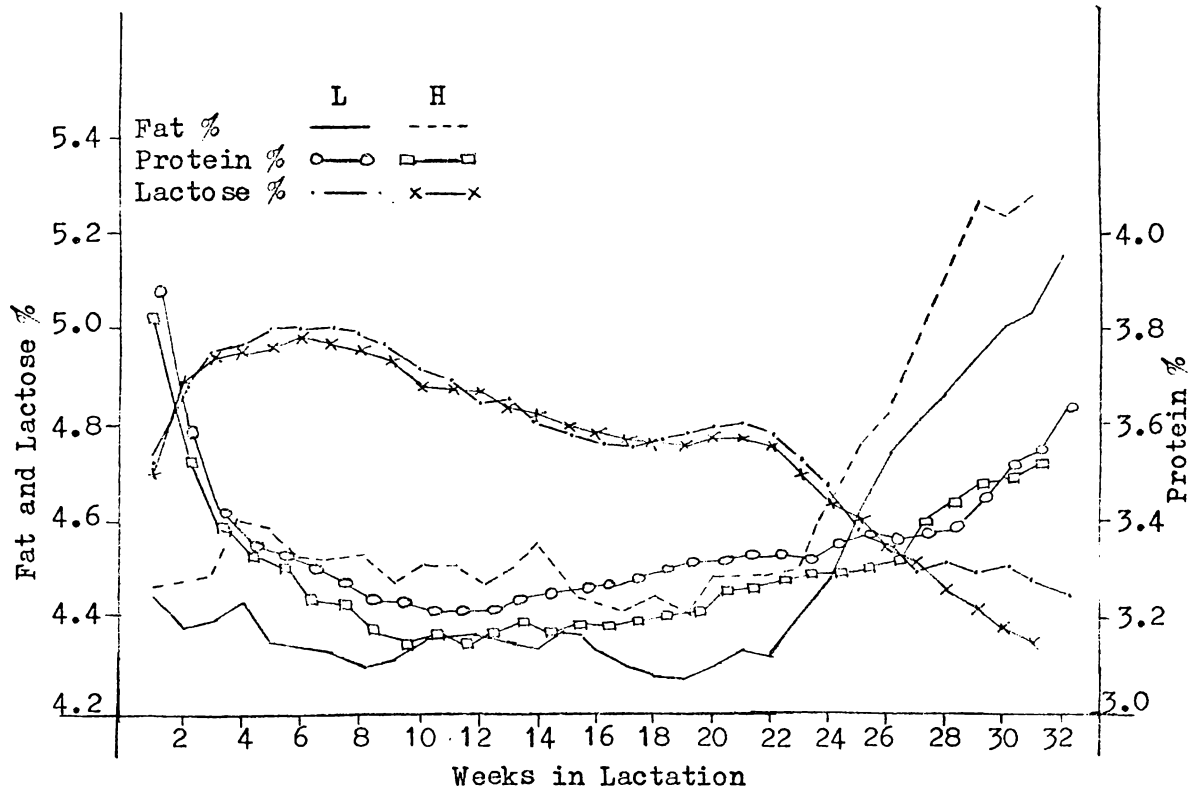


Fig.1 Lactational trends in milk composition of 18 sets of monozygotic twin cows at two stocking rates (4.1 (L) and 5.0 (H) cows/ha).

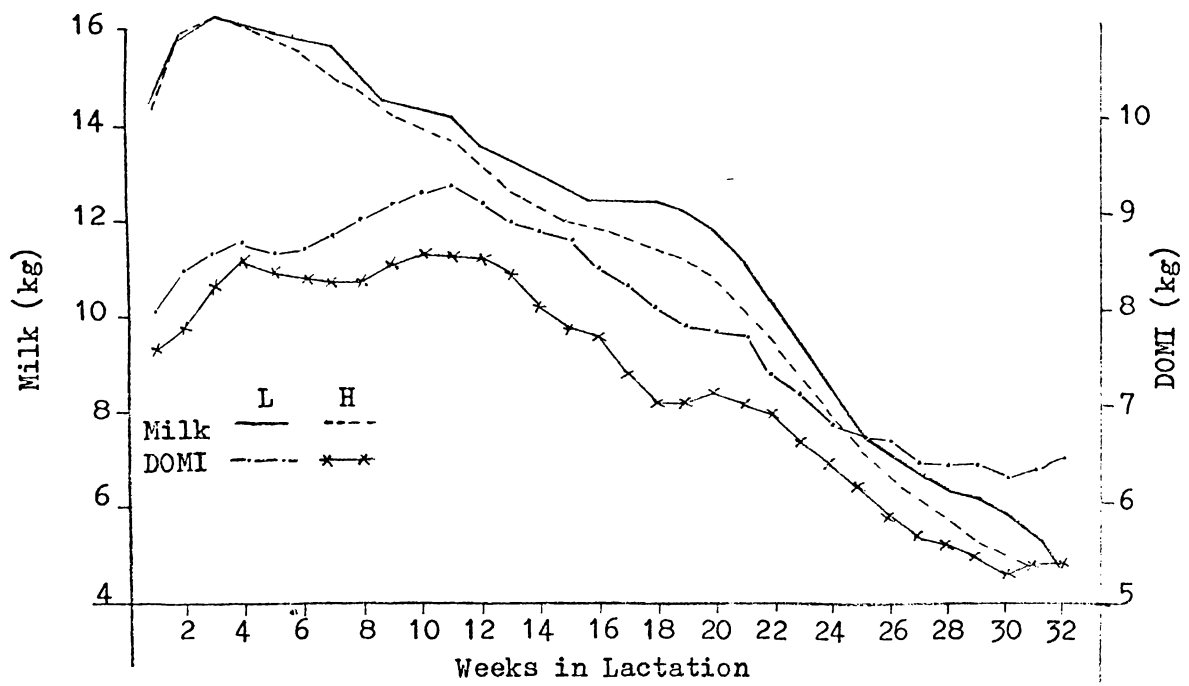


Fig.2 Lactational trends in milk yield and digestible organic matter intake (DOMI) for the same cows.
Source: J.B. Hutton (unpublished).

TABLE 3. Effect of age on milk fat yield and concentration.

Age of Cow	AVERAGE PRODUCTION											
	Ayrshire			Friesian			Jersey			M. Shorthorn		
	M'fat %	Milkfat kg	Days in milk	M'fat %	Milkfat kg	Days in milk	M'fat %	Milkfat kg	Days in milk	M'fat %	Milkfat kg	Days in milk
2 years	4.30	113	261	4.09	138	273	5.51	122	259	4.16	100	249
3 „	4.26	128	257	4.07	159	274	5.57	137	256	4.10	117	254
4 „	4.24	139	260	4.06	177	275	5.55	149	257	4.07	129	254
5 „	4.22	148	263	4.02	188	277	5.54	159	261	4.03	142	263
6 „	4.22	157	265	4.01	193	276	5.51	162	262	4.01	143	262
7 „	4.20	155	264	3.95	192	277	5.47	163	261	3.97	146	260
8 „	4.15	153	262	3.94	194	278	5.46	165	264	3.97	145	263
9 „	4.20	155	262	3.94	191	276	5.41	161	261	3.89	143	266
10 years and over	4.14	145	258	3.89	180	272	5.31	153	259	3.87	134	261
Average—all ages	4.23	137	261	4.02	171	275	5.50	145	259	4.03	128	257

Source: Anon. (1975)

Legates (1960) suggested these changes in composition may reflect udder deterioration either as the result of increasing incidence of mastitis, or of slight physical damage with age. O'Donovan, Dodd and Neave (1960) found little depression in milk fat content, but an average decrease of SNF content by 0.1 percentage units between consecutive infection-free lactations indicated this may be a specific effect related to age.

2.1.4 Climatic effects

Pronounced variations in milk yield and composition occur over seasons in N.Z. as shown in Table 4 and Figs. 1 and 2, but these effects are complicated by nutritional factors as well as stage of lactation.

Campbell, Flux and Patchell (1955) have shown that SNF values are relatively low in winter and summer for autumn and spring calving cows, respectively, but are high during spring when pasture of high quality is abundant.

Environmental temperature between 18 to 30 ^{o}C is without effect on milk composition (Wayman, Johnson, Merilan and Berry, 1962) although effects have been reported at temperatures outside this range (Cobble and Herman, 1951).

2.1.5 Disease

Mastitis can greatly influence the composition of milk by altering the permeability of udder tissue and its ability to synthesize milk constituents. Infection decreases fat, casein and lactose content and results in blood proteins appearing in the milk (Barry and Rowland, 1953). Recovery is rapid after therapy

TABLE 4. Seasonal variations in the daily yield and composition of milk.

Trait	Breed	TEST DAY AVERAGES								Est. 240-day lact. average
		Sept.	Oct.	Nov.	Dec.	Jan.	Feb.	March	April	
YIELD (lb. day)										(lb.)
Milk	J	24.5	24.6	23.3	20.3	20.4	16.0	9.4	6.2	4,341
	F	33.0	28.7	28.8	24.8	27.1	17.1	14.0	7.2	5,420
Total solids	J	3.55	3.62	3.43	3.03	3.02	2.45	1.50	1.05	649
	F	4.19	3.64	3.75	3.17	3.50	2.21	1.89	1.09	703
Fat	J	1.24	1.29	1.22	1.10	1.11	0.92	0.61	0.47	239
	F	1.27	1.12	1.17	0.98	1.10	0.73	0.63	0.41	223
Solids not fat	J	2.31	2.33	2.21	1.93	1.91	1.50	0.89	0.58	410
	F	2.92	2.52	2.58	2.19	2.40	1.48	1.26	0.68	480
Protein	J	0.93	0.94	0.90	0.79	0.80	0.63	0.38	0.28	170
	F	1.09	0.95	1.00	0.83	0.94	0.56	0.51	0.22	182
Lactose	J	1.18	1.21	1.13	0.98	0.98	0.75	0.43	0.25	207
	F	1.54	1.36	1.36	1.16	1.25	0.79	0.67	0.24	252
% COMPOSITION										%
Total solids	J	14.5	14.7	14.7	14.9	14.8	15.1	15.9	16.9	14.9
	F	12.9	12.7	13.0	12.8	12.9	12.9	13.5	15.1	13.0
Fat	J	5.1	5.2	5.2	5.4	5.4	5.8	6.5	7.6	5.5
	F	3.8	3.9	4.1	4.0	4.1	4.3	4.5	5.7	4.1
Solids not fat	J	9.4	9.5	9.5	9.5	9.4	9.4	9.4	9.7	9.4
	F	8.9	8.8	8.9	8.8	8.8	8.6	8.9	9.1	8.9
Protein	J	3.8	3.8	3.9	3.9	3.9	3.9	4.1	4.5	3.9
	F	3.3	3.3	3.4	3.3	3.4	3.3	3.6	4.1	3.4
Lactose	J	4.8	4.9	4.8	4.8	4.8	4.7	4.5	4.2	4.8
	F	4.7	4.7	4.7	4.7	4.6	4.5	4.7	4.0	4.7

NOTE: (1) Figures are for two-year-olds only—808 Jerseys (34 herds) and 149 Friesians (6 herds) tested in alternate months.
(2) F denotes Friesian and Friesian-cross cows.
J „ Jersey cows.

Source: Anon. (1969)

(Rowland, Neave, Dodd and Oliver, 1959) and as a consequence the overall effect of mastitis on milk composition is small. Although the influence of undetected subclinical mastitis on milk yield can be significant (Philpot, 1976), its effect on milk composition remains in question. Other metabolic diseases such as ketosis may also be associated with changes in milk yield and milk composition (Campbell et al.1955; McCarthy, Porter and Griel, 1968; Kronfeld, 1976).

2.1.6 Effects of milking

Cows milked three or four times daily produce more milk than when milked twice daily (Elliot, 1959) with no apparent change in milk composition. The results are apparently due to the stimulatory effect on milk synthesis of milk removal (Elliot, 1961).

During milking, fat content increases with little change in the contents of protein or lactose (Johansson, 1952). Milk yield and composition at a single milking are affected by the duration of the milking interval. When cows are milked twice daily, a longer interval at night has been shown to result in higher yields of milk in the morning with lower concentrations of milk solids, especially fat (Johansson and Claesson, 1957; Ormiston Spahr, Touchberry and Albridge, 1967). The differences in milk yield and milk fat content have been attributed to "residual milk"* effects rather than rate of synthesis which has been shown to be linear up to approximately 16 hours (Schmidt and Trimberger, 1963). "Residual milk" has an exceptionally high milk fat content which increases with a longer milking interval (Wheelock, Rook, and Dodd, 1965).

* Residual milk is that not removed by normal milking processes and amounts to 10-20 % of the original volume.

It follows that at any one milking, incomplete removal of milk will lower the fat content of milk removed. In practice however there will normally be compensation at the next milking. Ormiston et al. (1967) compared equal and unequal milking intervals with twice daily milkings and showed there were no consistent overall differences in milk composition. Only when there is failure to remove milk over extended periods such as two successive incomplete milkings, or by missing a milking, will there be a decrease in net milk yield (Dodd and Clough, 1962) and in milk composition (Wheelock, Rook, Dodd and Griffin, 1966).

2.1.7 Excitement

Periods of excitement or stress caused by environmental or physiological factors such as noise or oestrous have variable effects on the yield of milk and milk composition. The effects, mediated by hormones, are only temporary (Flux, Folley and Rowland, 1954; Cross, 1966; Dorotyuk, 1965).

2.2 NUTRITIONAL FACTORS AFFECTING MILK YIELD AND COMPOSITION

It is widely recognised that alterations in feeding levels and composition of the diet induce changes in the yield of milk and its constituents (Campbell and Dolby, 1959; Rook, 1961; Huber and Bowman, 1966a; Kirchgessner, Friescke and Koch, 1967; Broster, 1969, 1972; Balch, 1972; Thomas, 1975). The majority of evidence presented in these reviews has been based on work carried out overseas on forage-concentrate diets. As a consequence, two features require emphasis; first is the lack of information on the effects of forages on milk yield and composition when fed as sole

rations or in conjunction with other forages. Secondly, in many cases, the different treatments were obtained by alterations to the level of supplement or type of supplement. Also supplements were often fed according to milk yield, therefore the effects of level of feeding and type of diet were generally confounded.

2.2.1 Variations in milk yield and composition

Nutrition is known to be an important factor influencing variations in the yield of milk and milk composition. Stocking rate experiments conducted by Hancock (1958); McMeekan and Walshe (1963), and Hutton (1974) have shown that increasing stocking rates have been associated with lower yields of milk and fat produced per cow. Figs. 1 and 2 illustrate the average yield of milk and milk composition of 18 sets of monozygous twins at two different stocking rates. Milk yield and milk protein concentrations are consistently less and milk fat concentrations are higher for cows at the higher stocking rate. These differences are associated with a consistently lower intake of digestible nutrients. However, quantitative differences in nutrient intake were possibly not the only cause of the variations in milk yield and composition, since variations in pasture quality may also have affected responses in milk production.

2.2.2 The effect of level of feeding on milk yield and composition

2.2.2.1 Effect of precalving feeding level

It is widely recognised that the level of food intake in late pregnancy affects the subsequent level of milk synthesis (Broster, 1971). This generalization has been supported by N.Z. work with diets of pasture or mixtures of pasture with hay or silage

(Campbell and Flux, 1948; Lees, McMeekan and Wallace, 1948; Flux, 1950; Patchell, 1957; Wallace, 1958; Hutton and Parker, 1972).

In all cases, increased intakes of pasture or conserved pasture in late pregnancy consistently increased milk yield and percentage of fat and SNF and increased live weight loss in early lactation.

2.2.2.2 Effect of feeding level after calving

Riddet, Campbell, McDowall and Cox (1941), reduced the amount of concentrate offered with a basal ration of hay to cows and observed a significant fall in milk yield, and the concentrations of SNF and protein, but no effect on milk fat percentage. Similar results were found for cows in both early and in late lactation.

In subsequent experiments Flux and Patchell (1954, 1957) and Patchell (1957) varied post calving feeding by varying the quantities of pasture and hay or silage offered to groups of cattle. The results agreed with those of Riddet et al. (1941) with the exception of milk fat percentage which increased when the level of feeding decreased.

Further evidence of an effect of pasture intake on milk yield and composition was provided by Hutton and Parker (1966) who found that increasing grazing time from four to twenty hours per day resulted in cows producing more milk with a lower fat, and a higher SNF percentage.

In another experiment Hutton and Parker (1972) varied the amounts of pasture offered to groups of cattle in early lactation so that the feeding levels approximated that expected to be available under moderate and high stocking rates over the first eight weeks of

lactation. Differences in milk yield and milk composition were small. In the second year of the experiment cows at the lower feeding level had a lower milk fat percentage in the second month of the treatment period. It is of interest that the lower level of pasture feeding in this trial produced effects on milk composition which contrast with those of Patchell (1957) who also imposed two levels of pasture feeding. Further evidence of the effects of grazing cows in early lactation at different grazing intensities was reported by Hutton (1973, 1975). Increasing grazing intensities to reduce intake by 20 % resulted in a drop of 13 % in milk yield and a decline in milk protein content.

In a recent experiment (T.E.Trigg, unpublished) identical twin cows were grazed each day on areas of pasture calculated to provide either sufficient food to satisfy appetite or one half of that amount. Over the first 5 weeks of lactation the cows offered more food produced 46 % more milk with a significantly higher milk protein percentage and milk lactose percentage and a lower milk fat percentage.

In contrast Fisher, Donnelly, Hutton and Duganzich, (1975) fed dairy cows a mixture of pasture and silage (1:1 on a dry matter (DM) basis) at 100 %, 75 %, or 50 % of their individual intakes established on that food in a preliminary period, and found that although milk yield decreased with feeding level, the changes in fat and protein contents were small and not significant. Lactose percentage was however significantly less at the restricted feeding levels in comparison to unrestricted feeding.

The results of these experiments suggest that feeding different levels of forages are associated with variations in the

yield and composition of milk. This is not surprising as variations in intake are likely to be reflected as differences in the quantity and quality of precursors for milk synthesis reaching the mammary gland. Differences in forage quality and extent of body tissue mobilization may account in part for variations in the magnitude and nature of the responses between the experiments.

2.2.3 Effects of diet on milk yield and composition

2.2.3.1 Effects of concentrates fed with roughages

Variations in milk yield and milk composition have been shown to be associated with supplementing roughages with concentrates. Powell (1938, 1941) showed that a reduction in the proportion of roughage in the diet of dairy cows resulted in a fall in milk fat concentration without any change in milk yield. Subsequently McClymont and Paxton (1947) observed that cows grazing young leafy oats and receiving concentrates had an abnormal low milk fat percentage unless a coarse roughage was also fed. Although intakes were not measured, these experiments indicated that diet may have a specific effect on milk fat in contrast to the previous belief that milk fat percentage was inversely related to yield (McDowell, 1936). Nordfeldt (1966) fed cows at different levels on different ratios of hay or silage with concentrates, where the roughages constituted 70, 55, 40, and 25 % of the total diet. Maximum milk yields were achieved when the roughages constituted 40 % of the diet indicating a specific diet effect. This experiment also indicated that as the level of silage in the ration increased, the concentrations of fat and protein in milk decreased.

Hay had no such effect. This suggests that there may be a difference between forages in their influence on milk yield and composition.

Further evidence of the influence of concentrates in the ration on milk production was provided by Hotchkiss, Jacobsen and Cox, (1960). Different ratios of hay and concentrates were offered at several feeding levels. Cows fed rations with a high proportion of hay (75 %) produced less milk with a lower protein content but had no effect on milk fat percentage. A similar experiment was carried out by Flatt, Moe, Moore, Hooven, Lehmann, Ørskov and Hemken (1969) who fed cows on mixtures of alfalfa hay and concentrates (60:40; 40:60; 20:80). They found that increasing the proportion of concentrates in the ration did not increase the intake of digestible energy by the cows, but it did lower the percentage of fat in the milk, and reduce tissue mobilization. The yield of milk and SNF percentage was not altered by diet. Where the diet is rich in fermentable carbohydrates and the amount of roughage is low, depression in milk fat percentage may be pronounced. Emery, Brown and Thomas (1964) found when concentrates formed 80 % of the diet, milk fat content fell, irrespective of the roughage type. The effect on fat content may also be caused by reducing the particle size of the fibre and can occur independantly of a change in milk yield. This "low milk fat syndrome" has been discussed elsewhere (Davis and Brown, 1970; Storry, 1970; Annison, 1976) and the response apparently varies widely between animals and is associated with a marked increase in the proportion of propionic acid in the rumen (Armstrong and Prescott, 1971).

Evidence that concentrate supplements have a specific effect on milk yield and composition of cows on pasture is limited. Hutton and Parker (1967) fed different quantities of concentrates to cows at a high and low grazing intensities. The results showed slight increases in milk yield and decreases in fat percentage were obtained with increasing amounts of concentrates but these effects were only significant when the pasture supply was low.

Taparia and Davey (1970) found that when cows were fed ad libitum on pasture, the addition of concentrates had no significant effects on milk yield, milk fat percentage or SNF percentage.

2.2.3.2 Effect of forage diets on milk yield and composition

Although there is evidence that qualitative factors in mixed forage and concentrate diets can influence milk yield and composition much less is known about the effects of forages when fed alone or in combination.

(a) The effects of pasture

Wilson and McDowell (1966) observed that cows grazing Ruanui ryegrass (Lolium perenne, "Grasslands Ruanui") in the spring produced less milk with a higher milk fat percentage than two other ryegrass varieties grazed at a similar stage of growth. Wilson and Dolby (1967, 1969) observed differences in the composition of milk from cows grazing different varieties of ryegrass, pasture differing in maturity, and pasture receiving different rates of N fertilizer. Hutton and Parker (1966) observed that increasing the proportion of hay in a hay and pasture ration resulted in less

milk with a higher content of fat and lower content of SNF. However in all these experiments the effects of type of ration are inseparable from those of level of intake.

(b) The effects of silage

The feeding of pasture silage ad libitum to grazing cows during a summer drought was observed to arrest the decline in milk and fat yields but made little difference to the decline in the concentration of SNF in milk (Campbell, Flux and Patchell, 1955). Wallace and Parker (1966) reported a study in which two groups of cows were fed either pasture or pasture silage to appetite in the first 6 weeks of lactation. In that period the cows offered silage produced only half the level of milk fat of the other group and had very low intakes of silage, averaging only 6.3 - 7.3 kg DM per day.

Further comparisons of silage offered to appetite as a sole diet and with either meal or choumoellier (Brassica oleracea), showed milk yields and the SNF and protein contents in milk were significantly less from feeding silage alone than from more adequate diets of silage plus meal, (Flux and Patchell, 1950). Since the low responses with silage alone were associated with a marked drop in dry matter intake it was not possible to determine if differences in ration quality contributed to decreased productions.

The consequences of providing approximately 25 % of a ration as maize silage or pasture silage for cows grazing pasture restricted to about 75 % of appetite was compared with cows fully fed on pasture (Hutton, 1975). Both supplements prevented the fall in

milk yield and protein percentage observed when no supplements were fed. However it was calculated that more of the supplements were required than pasture to produce each additional increment of milk above that obtained from 75 % of full feeding of pasture. In a further experiment (Hutton, 1975) groups of cows were rationed pasture so that it provided either $\frac{1}{3}$ or $\frac{2}{3}$ or their requirements and the balance was provided either as maize silage or pasture silage. Milk fat production was the same for both silages, but grass silage significantly depressed milk protein percentage at both levels of supplementation. Since the digestibility of the dry matter was found to be similar for each of the rations, this suggests qualitative differences between the rations may have caused the responses in milk production.

Bryant and Donnelly (1974) compared cows fully fed on pasture with cows fed 75 % pasture and 25 % silage. Supplementing with silage resulted in a significant fall in milk yield but an associated fall in milk protein content, and a rise in milk fat content, were not significant. Because silage caused a significant fall in digestible energy intake, any qualitative effects of the ration could not be discerned. In the same experiment, increasing the proportion of maize silage to pasture fed cows was also found to be associated with decreasing intakes, milk yields and milk protein contents in the first year of the trial. In the second year however, cows fed on the 1:3 maize silage and pasture mixture produced similar yields of milk with the same protein content as cows on pasture despite significantly lower intakes of digestible energy, indicating in this case the response was possibly mediated by diet quality.

Fisher, et al. (1974) fed lactating cows a constant ration of pasture and silage (1:1, on a DM basis) at levels of 100 %, 75 %, 50 % of appetite. Milk yields and milk lactose percentage decreased but milk fat percentage and milk protein percentage was unaltered despite an average decrease in digestible energy intake (DEI) from 31.3 to 16.5 MJ per day. These effects of feeding level on milk fat percentage and milk protein percentage are in marked contrast to those observed for pasture fed cows (Hutton, 1975; T.E. Trigg, unpublished) which increased milk fat percentage and decreased milk protein percentage as food intake decreased. This suggests silage may have a specific effect on milk fat and protein synthesis.

Further evidence of silage having a quality affect on milk composition was indicated in two experiments which compared silages of different chemical composition arising from treatment variations in consolidation and air exclusion during manufacture (Hutton, Jury, Hughes, Parker and Lancaster, 1971; Lancaster, Hutton, Hughes and Marshall, 1974). Small but significant increases in intake were observed for those silages regarded as the least stable, having the highest pH and highest levels of ammonia. The increases in intake were consistently related to increased milk fat percentage but not to SNF percentage or milk protein percentage. The SNF percentage and milk protein percentage of cows on all silages were consistently low during the experiments in comparison to the levels obtained in the pre and post experimental periods when the cows were grazing pasture.

(c) Recapitulation

Evidence from grazing trials suggests that level of pasture intake may effect milk yield and composition. There is much less evidence to suggest that qualitative factors in pasture may affect milk yield and composition independantly of intake. However several observations suggest that silage may have specific dietary effects on milk composition. Milk protein and SNF content of cows' milk appear to be consistently low when cows are fed on silage diets, and these constituents do not appear to be related to food intake in contrast with other diets. Moreover the limited evidence indicates milk fat content may be also affected by qualitative factors in silage since variations with intake appear to be inconsistent.

Evidence on which to base possible mechanisms to account for these effects will now be examined. The effects of diet and level of feeding on milk synthesis, digestibility, metabolizability, and on the extent, rate, and site of energy, protein and fat digestion are considered. Calorimetric studies have shown that diet can markedly affect the efficiency of utilization of metabolizable energy for milk synthesis (Coppock, Flatt and Moore, 1964a,b; Tyrrell, Moe and Flatt, 1970; Moe and Tyrrell, 1971; Tyrrell and Moe, 1972). However these studies in themselves do not necessarily indicate the mechanisms accounting for the effect of diet on milk composition.

2.3 EVIDENCE INDICATING POSSIBLE MECHANISMS ACCOUNTING FOR EFFECTS OF LEVEL OF FEEDING AND TYPE OF DIET ON MILK COMPOSITION

2.3.1 Effect of diet and level of feeding on milk synthesis

Milk is synthesized in the mammary gland from precursors absorbed from the blood. The main precursors are acetic acid, B-hydroxy butyrate, glucose, essential and non essential amino acids and long chain fatty acids. They are either the end-products of digestion, in some cases modified by metabolism in the rumen epithelium or liver, or the result of catabolism of body tissue. Since diet and feeding level effect the absolute and relative quantities of nutrients available for absorption it follows these factors will have a direct influence on the quantities and the proportions of nutrients in plasma available for mammary uptake.

Although indentification of the main precursors has helped to clarify the importance of the products of digestion in milk synthesis, the mechanisms for the control of synthesis are still not clearly established. Many of the investigations of diet on milk composition have been undertaken with diets specially selected to exaggerate the changes in milk composition. In general these have contained high levels of grain and may give a misleading impression of the quantitative importance of dietary effects where forages are used exclusively. Furthermore much of the present knowledge of diet on products of digestion, absorption, and utilization has been obtained in experiments involving sheep and goats, rather than cows; with limited numbers of animals, and extensive surgical modification. Proposed mechanisms to account for the effect of diet on milk composition must be regarded as speculative when applied to lactating cows under grazing conditions.

2.3.1.1 Mechanisms involved in milk protein synthesis

Increases in milk protein synthesis have been observed in response to abomasal infusions of casein in cows receiving diets containing adequate protein according to nutritional standards (Broderick, Kowalczyk and Satter, 1970; Hale, Jacobsen and Hemken, 1972; Spechter, 1972; Clark, Spires and Derrig, 1973; Spires, Clark and Derrig, 1973; Derrig, Clark and Davis, 1974; Vik-mo, Emery and Huber, 1974; Spires, Clark, Derrig and Davis, 1975). Since casein infusions were not serving primarily as an energy source in the studies of Clark et al. (1973), Spires et al. (1973), Vik-mo et al. (1974), these results indicate milk protein synthesis was limited by the supply of one or more amino acids.

Although abomasal infusions of casein have consistently increased milk protein yield, the degree to which they have affected milk yield or milk protein content has been variable. Thomas (1975) suggested the variation possibly reflects differences in the degree to which casein affects lactose synthesis, and hence milk yield, by changes in the supply of glucogenic amino acids. Administration of essential amino acids either singly or in combination by post-ruminal or intravenous infusion has not given consistent responses in milk protein synthesis (Teichman, Caruolo and Mochrie, 1969; Williams, Martz and Hilderbrand, 1970; Fisher, 1969, 1972; Schwab and Satter, 1973, 1974; Schwab, Satter and Clay, 1976) and the factors in the casein causing the increase have not been identified.

Variation between cows and diets may account for the variable responses obtained. The amino acids available for milk production and/or milk protein synthesis will be affected by feed proteins,

which differ in their amino acid composition, and the extent to which they are degraded in the rumen (Schwab et al. 1976). Diets that increase rumen proportions of propionate and depress milk fat content may also increase milk protein content (Huber and Bowman, 1966b). Rook and Balch (1961), and Rook, Balch and Johnson (1965) and Wilson, Davey and Dolby (1967) have shown that rumen VFA's may have specific effects on milk protein synthesis. Intra-ruminal infusions of propionic acid given to cows increased milk protein content with little change in milk yield whereas infusions of acetic acid increased milk yield with no change in protein content. Thomas (1975) suggested acetic acid improved milk protein yield by providing additional ATP since the yield of lactose and milk was also increased. The effect of propionate is obscure since it would have also provided additional glucose to the udder but the yield of milk or lactose was not affected. This may be due to the limited use of glucose in the udder for ATP production (Smith, 1971). Halfpenny, Rook and Smith, (1969) and Clark, (1975) have suggested in those instances where increased milk protein yields have been associated with increased rumen propionate levels, the increase in milk protein yield may have been due to more propionate being available for glucose synthesis, sparing gluconeogenic amino acids for protein synthesis.

There is no evidence in the literature as to why milk protein content increases with increasing intake of rations of constant composition.

2.3.1.2 Mechanisms involved in milk fat synthesis

Milk fatty acids may be derived by de novo synthesis from acetate and B-hydroxy butyrate (C_4-C_{16} acids) or directly from plasma triglycerides (C_{12}-C_{18} acids) which are derived from body reserves, de novo synthesis, or diet (Storry, 1970; Bickerstaffe, 1971; Storry, 1972).

Present evidence indicates that the fat content in milk is relatively insensitive to dietary change when there is no pronounced effect on the proportions of ruminal VFA's (Storry, 1970; Thomas, 1975; Thomas and Kelly, 1976). Intraruminal infusion of acetic or butyric acids have produced increases in fat yield and content (Rook and Balch, 1961; Wilson et al. 1967). It is possible that variations in de novo synthesis of milk fat occur primarily through variations in the supply of acetate and B-hydroxy butyrate which have specific roles in the synthesis of C_4-C_{16} fatty acids in milk fat (Smith, McCarthy and Rook, 1974).

On milk-fat depressing diets there is only partial recovery to intraruminal infusions of acetic or butyric acid (Storry and Rook, 1966). This has been explained as an indirect effect of propionate, since on these diets increases in ruminal propionate proportions accompany falls in acetate and/or butyrate (Storry and Rook, 1966; Storry, 1970).

Annison, Bickerstaffe and Linzell (1974) observed when diets which increase ruminal propionate levels were offered to cows, glucose entry rates and blood glucose levels increased, and levels of acetate and B-hydroxy butyrate in blood decreased. The decreased fat content of the cows milk was associated with a reduced mammary uptake of acetate, B-hydroxy butyrate and plasma

triglycerides. Annison et al. (1974) suggested that increased glucose production stimulates lipogenesis and decreases lipolysis in adipose tissue which in turn would reduce lipoprotein synthesis in the liver and the availability of other fat precursors to the udder. This is supported by the observations that intraruminal infusion of propionic acid (Rook and Balch, 1961; Wilson et al. 1967), intravenous infusion of glucose (Vallance and McClymont, 1959; Storry and Rook, 1965), and intra-abomasal infusion of glucose (Ørskov and Fraser, 1974) depress milk fat secretion. Moreover Baldwin, Lim, Cheng, Cabrera and Ronning (1969) have shown increased activities of enzymes involved in lipogenesis in adipose tissue of animals on fat depressing diets.

2.3.1.3 Mechanisms involved in milk lactose synthesis

Glucose is a specific precursor for lactose synthesis in the udder (Dimant, Smith and Lardy, 1953; Reiss and Barry, 1953; Struss, Peterson and Mix, 1964; Bickerstaffe, Annison and Linzell, 1974). Since only small amounts of glucose are absorbed from the gut of animals on forage diets (Hungate, 1966) and glucose is indispensible for milk production, VFA's are utilised as a major source of glucose or to spare glucose use. Gluconeogenesis is therefore a major metabolic activity in ruminants, and propionate and amino acids (Bergman, Rowe and Kon, 1966; Leng, Steel and Luick, 1967; Annison, 1970; Leng, 1970a; Wolff and Bergman, 1972) are the major precursors of glucose.

Although there is limited data on the control of gluconeogenesis in the ruminants, insulin and glucagon probably have key roles in

homeostatic regulation. This topic was reviewed by Bassett (1974) who stated: "Insulin stimulates the uptake and utilization of glucose by many peripheral tissues, inhibits gluconeogenesis and glucose release from the liver, stimulates the uptake and incorporation of amino acids into protein, inhibits proteolysis, stimulates lipogenesis and inhibits lipolysis". Also he discussed the role of glucagon in promoting hepatic gluconeogenesis which accelerates glycogenolysis, thereby counteracting the effects of insulin.

It appears that regulation of insulin and glucagon in ruminants by plasma levels of glucose, amino acids, and VFAs, may be relatively unimportant but events such as secretion of gastro-intestional hormones may be important (Bassett, 1974).

In lactating cows the rate of entry of glucose into blood equals the rate of removal; 85 % of which is by the udder (Kronfeld, 1976). Thus plasma glucose concentration is regulated, and the controls include the rate of endogenous glucose production. This is retarded by the product, and accelerated by the substrates. As the rate of glucose production increases so do the rates of mammary glucose uptake and milk secretion. Thus increased food intake or highly glucogenic rations tend to promote milk production.

Since the milk secreted is isotonic with blood plasma, and lactose is the major osmotically active constituent synthesized by the mammary gland (Rook and Wood, 1959), the fall in lactose yield would lead to a decrease in the output of water and volume of milk secreted, and to a consequent increase in fat and protein contents of milk (Storry and Rook, 1962). However severe underfeeding may cause a slight depression in the lactose content of milk (Rook and Line, 1961). Rook (1976) pointed out that such effects depend on

associated affects on milk yield, and occur from the decrease in the relative contribution of lactose to the osmotic pressure of milk as the volume of milk is depressed.

2.3.2 Effect of diet and level of feeding on digestion

2.3.2.1 Effect of diet and feeding level on digestibility and metabolizability

(a) Feeding level

Marked falls in digestibility have been found with increased feeding levels for diets of mixed roughages and grain (Tyrrell and Moe, 1975). Experiments with sheep on all forage diets also showed a fall in digestibility with increased feeding levels (Blaxter, 1962). The falls were greater for diets of low apparent digestibility (60 % DM digestibility) although there were marked variations between diets. Rattray and Joyce (1969) and MacRae and Ulyatt (1974) found when sheep were fed pasture above 74 % DM digestibility, feeding level had little effect on DM digestibility. Further, when cows were fed pasture ranging in DM digestibility values of 65-78 %, differences in digestibility between different feeding levels were comparatively small (Hutton, 1962). Hutton (1962) has shown when pasture intake of lactating cows differed by 60 % the difference in the mean digestibility coefficient was only 1.5 % units. No comparable data are available for silage.

While the proportion of food energy lost as heat production and methane are known to decline as feeding level increases (Blaxter, 1962; Rattray and Joyce, 1969), the overall effect of feeding level on the metabolizability of pasture appears to be small

(Ulyatt, Dellow, Egan and Walker, 1973). The same appears true for silage (Smith, Wainman and Dewey, 1975). Hence, if feeding level of pasture has relatively small effects on ME value, then availability of nutrients for milk synthesis will closely be reflected by intake levels. However changes in milk quality with feeding levels may result if there are changes in the efficiency of nutrient use for synthesis of milk components as may be caused by changes in the site of digestion and the quality of digestion end-products.

(b) Diet

The digestibility of the dry matter and energy contents of pasture may vary from a maximum in spring to a minimum value in mid summer (Wallace, 1961; Hutton, 1962). This is largely due to advancing maturity which is associated with an increase in the indigestible lignin fraction in cell walls and a decrease in the proportion of digestible cell contents such as soluble carbohydrates and proteins. The digestibility of pasture may also be influenced by the pasture species present since at the same stage of growth there are differences in their digestibilities and in practice these differences may be largely due to differences in their maturity dates, (Minson, Raymond and Harris, 1960; Minson, Harris, Raymond and Milford, 1964). Because stage of growth and botanical composition are major factors affecting digestibility of pasture, grazing can greatly influence the digestibility of the pasture.

Changes in digestibility when pasture is directly ensiled have been shown to be small (Harris and Raymond, 1963; Demarquilly, 1973; Dulphy and Demarquilly, 1973; McDonald and Edwards, 1976). Although there are small increases in the concentration of structural carbohydrates, these are apparently compensated by an increase in

the digestibility of these fractions (Jackson and Anderson, 1968; Demarquilly, 1973).

Comparisons of silage and pasture have also shown these foods to have similar ME values (Jackson and Anderson, 1968; McDonald, Henderson and MacGregor, 1968; Jackson, 1971). Although silage has a higher gross energy content than pasture, similar ME values are due to increased urine energy losses as there appears to be little effect on methane production (McDonald and Edwards, 1976).

If silage does have specific effects on milk production then these effects must also be mediated by changes in the efficiency of utilisation of metabolizable energy for milk synthesis. This could arise from differences in the site of digestion and the quality of the end-products of digestion.

2.3.2.2 Effects of diet and level of feeding on rate and site of digestion

Diet and feeding level of forages have been shown to affect the rate and site of digestion. Ulyatt and MacRae (1974) found when different pasture species of similar apparent digestibility were offered at similar levels to sheep, the amounts of organic matter digested in the stomach region and in the small intestine were different. Also as the level of intake increased the relative amounts of each species digested in the stomach and small intestine differed. Flow rates through the rumen of digesta derived from these pasture species were different (Ulyatt, 1969) and all were rapid in comparison to value published for long, dried forages (Minson, 1966; Weston and Hogan, 1971).

Beever, Thomson, Pfeffer and Armstrong, (1971a) compared the sites of energy digestion of fresh grass and unwilted silage in sheep and found that slightly less apparent digestible energy disappeared in the stomach (57 % v. 63 %) and more in the small intestine for the silage ration although this difference was not significant.

The significance of such observations is that differences may arise in the quality and quantity of the end products of digestion. Moreover, any increase in the proportion of the diet that is digested in the small intestine is likely to lead to increased efficiency of utilisation, particularly of protein (Blaxter and Martin, 1962).

2.3.2.3 Effect of diet and feeding level on VFA production

Negligible amounts of soluble carbohydrate are digested post-ruminally on silage and pasture diets (Beever et al. 1971a; Ulyatt and MacRae, 1974), therefore VFA's are the major energy source for the animal and are the major end-products of rumen digestion of structural carbohydrate. Experiments with sheep fed on a wide range of forage diets have shown that VFA's comprise 60-80 % of the daily ME intake (Annison and Armstrong, 1970). On pasture diets Ulyatt et al. (1973) have shown that VFA's account for approximately 75 % of the energy digested in the stomach whereas Beever, Thomson and Harrison (1974) suggested that the value for silage may be as low as 55 %. There may be differences between pasture and silage in the efficiency of rumen fermentation. Beever et al. (1974) demonstrated the fraction of digested energy as rumen VFA's was improved when silage was treated with formaldehyde.

Ulyatt and MacRae (1974) found the efficiency of conversion of digested energy for VFA production was slightly higher when the feeding level of ryegrass and clover diets was reduced. However combining the data derived from clover and pasture diets they showed that VFA production rate was linearly related to the amount of energy digested in the stomach and closely resembled estimates based on Hungate's (1965) theoretical stoichiometric relationships for rumen fermentation of carbohydrate.

2.3.2.4 Effect of diet and feeding level on rumen fermentation pattern

Some mechanisms proposed for milk synthesis based on mixed concentrate and forage diets have indicated the relative proportions of rumen VFA may be important determinants. There is good evidence that in most situations the concentrations of each VFA is correlated with its production rate, and that the molar proportions closely reflect their relative rates of uptake (apart from some small interconversion of VFA) especially on forage diets (Leng, 1970b). However the extent to which the absorbed VFA contribute to milk synthesis will depend on overall body energy metabolism, and the relative contribution of endogenous sources of milk precursors. For instance in sheep the contribution of endogenous acetate to total entry rose from about 20 % in the fed animal to 50 % when fasted (Bergman and Wolff, 1971).

As indicated in the following two sections, there are few reports comparing various types of roughage as sole diets for lactating cows. Where comparisons have been made, detailed data on changes in concentrations and proportions of VFA were not given.

Interpretation of this limited data is further complicated since variations in pH and VFA values in the rumen of the dairy cow are caused by number of factors besides diet (Davey, 1965).

(a) Level of feeding on rumen VFA pattern

Bath and Rook (1963) noted a decrease in the proportion of acetic acid and an increase in propionic acid when increased amounts of a mixed hay and concentrate diet were offered. With hay alone little change occurred in the proportions of the acids with increased intake. Level of intake is without significant effect on the proportions of VFA in the rumen liquor of cows fed pasture (Davey, 1965) or sheep fed silage (Anderson and Jackson, 1971).

(b) Diet and rumen fermentation pattern

The effect of ration composition on the pattern of fermentation in the rumen is well documented for mixed grain and roughage diets. Severe changes in ration by increasing the amounts of grain and limiting the amount of roughage usually increase rumen propionate (Davis, Brown and Beitz, 1964; Schingoethe, Stake and Owens, 1973) reduce the acetate to propionate ratio (Davis, 1967; Armstrong and Prescott, 1971; Bauman, Davis and Bucholtz, 1971) and alter milk composition (Beitz and Davis, 1964; Yousef, Huber and Emery, 1970; Schingoethe et al. 1973). In contrast information on changes in the VFA pattern of animals fed all forage diets associated with milk composition changes is limited.

Bath and Rook (1965) have reported molar percentages for the individual VFA's in the rumen of cows fed on roughages. Hay, silages, and dried grass all gave high proportions of acetic acid and silages tended to give a lower proportion of butyric acid

and a higher percentage of branched chain acids. Davey (1965) examined data from several experiments with sheep and cattle and noted that despite differences in the pastures examined at different stages of maturity, the results were relatively uniform. The molar percentages for cattle varied from 61.9 - 73.6 for acetic acid, 15.5 - 23.2 for propionic acid, and 10.9 - 14.9 for butyric acid.

Comparisons of pasture and similar material after ensiling have shown the diets had no consistent effects on the relative major VFA proportions in sheep (Bryant and Lancaster, 1970; Anderson and Jackson, 1971) although there was a tendancy for silage to be associated with higher levels of valeric and branched chain fatty acids. In a similar study Donaldson and Edwards (1976) also found little difference in the relative proportions of VFA. The only change was a slightly higher level of acetic acid from pasture.

In those studies with concentrate and roughage diets, changes in milk composition generally occur when there are marked changes in the proportions of acetate and propionate in the rumen acids (Hinders and Owen, 1963; Colenbrander, Bartley, Morrill and Deyoe and Pfost 1967; Armstrong and Prescott, 1971). In addition changes in the molar proportions of VFA in the rumen are known to be associated with decreases in rumen protozoa and saliva secretion accompanying marked falls in rumen pH (Balch, 1958; Kaufmann and Rohr, 1966; Lawlor, Geisecke and Walser-Karst, 1966; Sutton and Johnson, 1969; Eadie, Hyldgaard-Jensen, Mann, Reid and Whitelaw, 1970; Harrison, Beever, Thomson and Osbourn, 1976). In contrast rumen pH appeared to be stable at about 6.5 - 7.0 for animals on pasture and silage diets (Anderson and Jackson, 1971; Donaldson and Edwards, 1976).

Thus in summary the limited evidence available suggests that VFA proportions in rumen liquor are similar when pasture and silage are fed and are not influenced by the level at which these materials are fed. It therefore appears that if VFA proportions reflect the pattern of fermentation, the latter is not a major factor contributing to the differences in the composition of the milk from cows fed pasture and silage.

2.3.2.5 Effect of diet and feeding level on digestion of lipids and on milk lipid synthesis

As indicated in section 2.2.4.2, VFA and B-hydroxy butyrate are major precursors of milk lipids and the dietary factors influencing their availability have been discussed in section 2.2.5.3.

Dietary fat contributes to the plasma triglycerides which are used for the synthesis of the long chain fatty acids in milk fat (Storry, 1970). The amounts of dietary fat absorbed from the small intestine of sheep have been shown to be affected by both level of feeding and the type of pasture fed (Ulyatt and MacRae, 1974. Moreoever cows supplemented with fat by encapsulation (Chandler, Robinson, Ripper and Fowler, 1973; Bartsh, Ellis, McLean, and Radcliffe, 1976) or by duodenal infusion (Bickerstaffe and Annison, 1971) increase milk fat yield. Scott and Cook (1975) suggested that the amount of fat absorbed by the animal has a direct bearing on energy use and milk fat synthesis.

However considering that relatively low levels of fat occur in pasture (Hutton, 1962) and silage (McDonald, Henderson and MacGregor, 1968) and that plasma triglycerides can also be synthesized de novo or be derived from body adipose reserves

(Rook, 1976) the importance of dietary fat in forage diets on milk composition appears to be low. Moreover the feeding of diets low in fat do not necessarily depress milk fat synthesis but result in a high proportion of the acids being synthesized in the mammary gland (Virtanen, 1966; MacLeod and Wood, 1972).

2.3.2.6 Effects of diet and feeding level on protein digestion

The main site of absorption of amino acids is the small intestine and the major factors influencing the absorption of amino acids from there are the flow of undigested protein from the rumen and the ruminal synthesis of microbial protein.

(a) The effects of feeding level on protein digestion

On pasture diets the level of food intake by sheep has been shown not to materially affect the apparent digestibility of dietary nitrogen (Rattray and Joyce, 1969; MacRae and Ulyatt, 1974). However the flow of amino N into the duodenum has been shown to be closely related to feeding level (MacRae and Ulyatt, 1974). Moreover the amounts of amino acids absorbed in the small intestine were mainly dependant on the amounts reaching the duodenum since digestibilities of amino acids in the small intestine were not affected by feeding level.

MacRae and Ulyatt (1974) showed that feeding level may alter the relative proportion of dietary nitrogen apparently digested in the stomach and in the small and large intestine, however these differences were small.

There is no evidence that the quality of the amino N absorbed from the small intestine is influenced by feeding level. Ulyatt,

MacRae, Clarke and Pearce (1975) showed that increasing the feeding level of perennial ryegrass reduced the relative proportion of dietary protein that was degraded in the stomach and increased the amount of bacterial protein synthesized per unit of food intake. No significant differences were detected between feeding levels in the contribution of bacterial protein nitrogen to total protein nitrogen entering the duodenum or in the amino acid composition of this digesta although the range in intake was small.

(b) Effect of diet on protein digestion

MacRae and Ulyatt (1974) measured significant differences between pasture diets, with a similar apparent nitrogen digestibility, in the sites of apparent nitrogen digestion in sheep. Considerable differences were measured in the relative amounts of amino acids digested in the stomach region and in the small intestine. By comparison 74 % of the total amino acids ingested were apparently absorbed from the small intestine by sheep fed Manawa ryegrass (Lolium perenne "Grasslands Manawa") whereas only 51 % and 41 % were absorbed of those ingested in perennial ryegrass and clover respectively. The main difference in protein digestion in the stomach was the reduced loss of nitrogen as ammonia and increased bacterial protein synthesis from Manawa (Ulyatt et al. 1975) suggesting the rumen micro-organisms digesting Manawa were able to synthesize protein from ammonia more efficiently. Since the percentage of dietary protein that was degraded in the stomach was approximately 70 % for all herbages, the extra microbial protein resulted in an increased flow of amino acids into the duodenum from Manawa.

Consequent qualitative differences in the protein quality

entering the duodenum were reflected in small differences in the digestibility of the amino acids in the small intestine which also contributed to quantitative differences in amino acid uptake at this site.

The quality of the amino acids absorbed appear to have a significant effect on the utilization of protein by the animal. For each additional unit of food nitrogen supplied as perennial ryegrass, Manawa or clover, 38, 43, and 42 % respectively were apparently absorbed from the small intestine as amino N and 6, 26, and 24 % respectively were retained by the sheep (MacRae and Ulyatt 1974).

These results demonstrate there can be marked differences between forage diets in the efficiency of nitrogen utilization by the animal depending upon the efficiency of microbial protein synthesis in the rumen and on the quantity and quality of dietary protein flowing into the duodenum.

These findings are supported by similar studies on nitrogen digestion of forage diets with sheep fitted with re-entrant cannulae (Hogan, 1965a; Clark, Ellinger and Phillipson, 1966; Hogan and Weston, 1970; Coelho da Silva, Seeley, Thomson, Beever and Armstrong, 1972a,b; MacRae et al. 1972; Harrison, Beever, Thomson and Osbourn, 1973; Beever, Thomson and Harrison, 1974) and by some limited studies with fistulated cows (van't Klooster and Boekholt, 1972; Tamminga, 1973; Haegmeister, Kaufmann and Pfeffer, 1976; Hvelplend and Moller, 1976).

Coelho da Silva et al.(1972a,b) found a significant correlation between the quantities of total amino acids presented at the duodenum and absorbed within the small intestine. This however

was not substantiated by Harrison et al. (1973) who suggested discrepancies could be due to differences in the protein quality of duodenal digesta of sheep on different diets. They found marked increases in the concentrations of essential amino acids, particularly lysine and methionine in duodenal digesta on some diets, and these essential amino acids were preferentially absorbed in the small intestine relative to the non essential amino acids. Purser (1970); MacRae et al. (1972) and MacRae and Ulyatt (1974) also observed that essential amino acids were generally of higher availability in the small intestine than non essential amino acids.

The main influence of microbial digestion is to slightly enrich the content of methionine and lysine of duodenal digesta on forage diets (Coelho da Silva et al. 1972a,b; Harrison et al. 1973). However MacRae and Ulyatt (1974) found little difference between fresh herbage and duodenal digesta in essential amino acid composition, due to the similarity in the composition of the bacteria and the herbage.

The preferential uptake of essential amino acids may explain why no relationship was found between the apparent absorption of total amino acids from the small intestine and nitrogen retention (Hogan and Weston, 1969; Hogan, Weston and Lindsay, 1969; MacRae and Ulyatt, 1974) since Nimrick, Hatfield, Kaminski and Owens (1970) have shown methionine and lysine were the most limiting essential amino acids for lambs fed urea as the sole N source. Obviously factors affecting the flow of essential amino acids through the duodenum are the major determinants of the efficiency of nitrogen utilization by the animal.

(c) Rumen protein synthesis

(i) Factors affecting rumen proteolysis

The extent to which dietary protein is modified by the action of the rumen has been the subject of many studies. Ulyatt et al. (1975) observed that about 30 per cent of dietary protein in fresh pasture diets is undegraded in the rumen and Beever et al. (1974) has indicated a comparable figure for silage may be about 22 per cent. However in absolute terms the comparative amounts of dietary protein of these two diets entering the duoednum may be markedly different due to the low protein content of silage arising from extensive proteolysis during ensiling (Donaldson and Edwards, 1976).

Protein solubility appears to be a major factor governing the rate of breakdown of dietary protein in the rumen (Chalmers and Synge, 1954; Annison, 1956) although other factors such as flow rate out of the rumen are also important (Sutherland, 1976). For instance, relatively insoluble proteins will be degraded if residence time is extended and conversely soluble proteins may escape degradation if flow rate is rapid. The solubility of proteins in most herbage species varies considerably with stage of growth and environmental conditions (Hume and Purser, 1974; Bryant and Newth, 1975). In fresh pasture 40 per cent of total nitrogen may be soluble and up to 70 per cent of soluble nitrogen may be protein, and 60 per cent of this protein can go immediately into solution with chewing (Bryant, 1964; Hogan, 1965b) indicating its highly soluble nature. Many of the processes of preserving herbage such as drying, freezing, and chemical treatment with formaldehyde before ensiling, significantly decrease the solubility of the proteins (MacRae, 1970; Beever, Thomson and Harrison, 1971b; Beever et al. 1974; Bryant

and Newth, 1975; Beever et al. 1976). These treatments reduce the rate of proteolysis in the rumen, resulting in lower concentrations of ammonia in rumen liquor and less wastage of dietary nitrogen anterior to the duodenum, thereby increasing the amounts of dietary protein and total protein reaching the small intestine. When sheep were given frozen red clover 105 % of the amino acids consumed reached the duodenum and of these 75 % were apparently digested in the small intestine whereas when sheep consumed the same quantity of amino acids as fresh clover only 63 % reached the small intestine and 40 % were apparently digested (Beever et al. 1971b). Similarly when silage was treated with formaldehyde the proportion of dietary amino acids undegraded in the rumen rose from 22 per cent to 93 per cent (Beever et al. 1974). Although the formaldehyde treatment reduced the amount of microbial protein synthesized in the rumen from 112 g/d to 36 g/d, it increased the total protein entering the duodenum by 25 per cent.

However when pasture is preserved by directly ensiling, the solubility of the total nitrogen may be increased due to proteclysis (Waldo, Keys and Gordon, 1973; Goering and Waldo, 1974; Castle and Watson, 1975; Donaldson and Edwards, 1976). More than 50 per cent of the protein may be degraded raising the soluble nitrogen content to 60-70 per cent with most of the soluble nitrogen in the form of non-protein nitrogen.

Further, Van Soest (1973) observed that as much as 50 per cent of the insoluble protein in silage is held in the lignin fraction which has been demonstrated to have a very low digestibility. Hence, it would appear that although the crude protein content of silage may be high, the amount of dietary protein available to the

animals for absorption is low due to the extensive proteolysis occurring both during ensiling and in the rumen, making the animal heavily dependant upon microbial protein for its amino acid supply. Beever et al. (1974) showed that of the total amino acids entering the duodenum of sheep fed silage, 71 per cent was microbial and 18 per cent was dietary, the balance being of endogenous origin.

(ii) Factors affecting rumen protein synthesis

Since the majority of rumen bacteria use ammonia as a nitrogen source (Nolan and Leng, 1972) the availability of ammonia is an important determinant of microbial protein production. It appears that maximum microbial synthesis occurs when the ammonia concentration is around 5 mg NH_3-N per 100 ml of rumen fluid (Satter and Roffler, 1975) although Miller (1973) has suggested a value 4 times as high. However rumen ammonia concentrations are not likely to limit microbial protein synthesis on pasture or silage diets since the levels are characteristically high, with peak values reaching 20 mg NH_3-N/100 ml for pasture and increasing with ensiling to 30-50 mg NH_3-N/100 ml (el-Shazly, 1952; Chalmers, 1963; Fatianoff, Durand, Zelter and Tisserand, 1966; Donaldson and Edwards, 1976).

Although high rumen ammonia concentrations apparently do not inhibit rumen protein synthesis (Satter and Slyter, 1974) they do increase the loss of ammonia from the rumen (Lewis and Buttery, 1973). The majority of ammonia passing through the rumen wall is transported to the liver, converted to urea, (Annison, Hill and Lewis, 1957) and 80 per cent may be excreted in urine (Blaxter, 1964), resulting in a net loss of nitrogen by the animal. The animal will also suffer a disadvantage arising from the extra energy costs involved in urea

synthesis and excretion (Martin and Blaxter, 1964), and the possibility that the high plasma ammonia levels may interfere with intermediary metabolism (Buttery, 1976).

The higher rumen ammonia levels of cows on silage rations compared with pasture (Donaldson and Edwards, 1976) may be associated with reduced efficiency of nitrogen utilisation by cows. Fatianoff et al. (1966) and Conrad, Hibbs, Pratt and Davis, (1961) found nitrogen retention was less in ruminants on diets of silage compared with fresh herbage, although in the latter case this was only true when the N content of the fresh herbage was above 24 g/kg DM. Armstrong (1974) calculated, from the data of Proud (1973) that the supply of amino N to the host on a silage diet was only about 76 % of that when fresh grass was given and suggested that the differences were due to differences in the rate of ammonia production or microbial protein synthesis, or both, in the rumen.

The concentration of ammonia in rumen contents reflects the balance achieved between the degradative and synthetic activities of rumen micro-organisms. The high concentrations and rapid changes indicate a lack of balance, with the synthetic processes in deficit. Because directly ensiled pasture contains negligible amounts of soluble carbohydrate relative to that in the parent crop (Donaldson and Edwards, 1976), the major factor limiting assimilation of rumen NH_3-N into bacterial protein may be inadequate readily fermentable energy for the rumen micro-organisms (Smith, 1969; Hogan, 1975).

Supplementation of silage with cereals reduces ammonia in the rumen (Castle and Thomas, 1976; Griffiths and Bath, 1973) and results in large improvements in nitrogen retention (Conrad et al. 1961; Thomson, 1968; Griffiths, 1969; Griffiths and Spillane, 1970;

Griffiths, Spillane and Bath, 1971). The addition of readily available carbohydrate reduces urinary nitrogen excretion (Griffiths and Spillane, 1971) and presumably leads to more efficient utilization of silage non-protein nitrogen for microbial synthesis in the rumen (Conrad and Hibbs, 1968).

Supplements of barley were found to increase the milk yields, SNF and milk protein concentrations of cows fed silage (Murdoch, 1962; Castle and Watson, 1975) but in these experiments the improvements may have been due to increased intakes or to an enhanced efficiency of nitrogen utilization in the rumen. Similarly Castle and Thomas (1976) found when sheep were given silage supplemented with barley the duodenal flow of nitrogen increased from 15.3 g/d to 17.3 g/d but again the barley was associated with improved intakes and the effect on the efficiency of microbial synthesis is uncertain. However Castle and Thomas (1976) suggested that supplying the micro-organisms with energy was the major factor influencing rumen protein synthesis, since when sheep were given a diet of barley plus silage and some of the barley was replaced with isocaloric amounts of groundnut meal or urea, duodenal flows of nitrogen remained the same even though nitrogen intakes were increased.

Although partial replacement of barley supplements by groundnut meal did not result in an increase of nitrogen entering the duodenum of sheep, consistent responses have been obtained in milk production when this treatment has been applied to lactating cows (Castle and Watson, 1969, 1974; Griffiths and Crowley, 1973). By comparison, increasing the amount of soyabean meal in a supplement to cows fed silage rations has resulted in an improvement in both milk yield and in milk protein concentration (Butler, 1973, 1974). This suggests

the nature of the protein supplement fed with silage may influence the response. That protein supplements may differ in their effect on microbial protein is suggested by the work of Hume (1974). He observed that only 39 % of soyabean meal was degraded in the rumen in comparison to 65 % for groundnut meal and the rumen ammonia levels with these supplements were 6.5 and 16 mg/100 ml respectively.

Protein supplements may also exert a direct effect on protein synthesis in the rumen though their contributing preformed peptides and amino acids (Wright and Hungate, 1967) and branched chain and higher VFA (Bryant and Doetsch, 1955; Hemsley and Moir, 1963; Hume, 1970; Oltjen, Slyter, William and Kern,1971).

There is also evidence that directly ensiled pasture silage may have a low methionine content since substantial increases in intake, wool and tissue growth were obtained by sheep on silage diets in response to supplements of methionine (Barry, Fennessy and Duncan, 1973).

(d) Protein digestion and milk protein synthesis

The efficiency of microbial protein synthesis determined in a number of studies was summarized by Egan and Walker (1973). They showed that the highest values obtained on forage diets were still inadequate to meet the requirements for milk production at a given energy level and calculated that 19-47 % of the protein requirements for milk protein synthesis in early lactation would need to be met from other sources such as herbage leaf protein and/or body reserves.

Since there is evidence that dietary protein is extensively degraded in the rumen for pasture rations (Ulyatt et al. 1975) and perhaps even to a greater extent on silage (Beever et al.1974) this

places a severe limit on the ability of these rations to meet the animals protein requirements in early lactation. This may be especially so for silage due to the lower initial true protein content caused during ensiling. Moreover factors limiting microbial protein synthesis would also reduce the flow of protein into the duodenum and therefore the availability of amino acids for milk protein synthesis. Since dietary carbohydrates appear to reduce the absorption of dietary nitrogen prior to the duodenum of sheep given herbages with a high nitrogen content (Hogan and Weston, 1969; Hogan et al. 1969; MacRae et al. 1972), then silage with very low levels of soluble carbohydrate would be at a serious disadvantage. The high levels of ammonia in the soluble nitrogen fraction of silage in the absence of readily available energy may be absorbed from the rumen and excreted, thereby reducing the efficiency of dietary nitrogen conversion into microbial protein.

Furthermore, calculated amino acid requirements for cows in early lactation, relative to estimated quantities absorbed from the small intestine, show that essential amino acids especially methionine may be limiting milk protein synthesis (Egan and Walker, 1973; Hogan, 1975). The possibility that relatively low amounts of protein may enter the duodenum of cows on silage rations due to decreased microbial protein synthesis and low levels of undegraded dietary protein flowing out of the stomach therefore suggests a greater likelihood that methionine could be limiting milk protein synthesis on silage rations.

The calculations of Hogan (1975) indicated that the total amounts of amino acids absorbed from the small intestine are likely to be less than the requirements in early lactation, inferring that

contrary to Flatt, Moore, Hooven and Plowman (1965) the requirements are met from tissue protein catabolism. This appears doubtful since apart from the first two weeks of lactation when protein is metabolized due to a change in hormone levels (Lenkeit, 1972) cows do not apparently mobilize much tissue protein except when they are in severe negative energy balance (Paquay, De Baere, and Lousse, 1972; van Es and Boekholt, 1976). Even under these conditions milk yield is lowered and milk protein concentrations may be depressed suggesting the contribution of tissue protein to milk protein synthesis is probably low.

In feeding experiments increases in dietary protein above those indicated by feeding standards have had little effect on milk yield or milk protein production (Rook and Line, 1961). The lack of an effect of excess dietary protein is not surprising as much of the excess is probably absorbed as ammonia from the rumen and excreted (Hogan and Weston, 1968) or recycled with an energy cost (Martin and Blaxter, 1964). Moreover at a constant energy intake, an increase in dietary protein implies a reduction in the non-protein energy content of the diet possibly reflecting a lower efficiency of utilization of the carbon chains of amino acids as an energy source relative to non-nitrogenous components (Tyrrell, Moe and Flatt, 1970).

Two experiments have reported responses by animals grazing pasture when supplemented with proteins with a high resistence to ruminal degradation. Lamb growth has been stimulated by drenching the animals with a slurry containing fish meal (Archer, Babwick, Kempton and Leng, 1976) and Wilson, (1970) obtained small responses in milk protein concentration by lactating cows drenched with formaldehyde protected casein.

There are several experiments in which increases in milk protein synthesis have been observed in response to abomasal infusions of casein in animals receiving mixed concentrate and roughage diets, supposedly having adequate energy and protein contents according to nutritional standards (Broderick et al. 1970; Hale et al. 1972; Spechter, 1972; Clark et al. 1973; Spires et al. 1973; Derrig et al. 1974; Vik-mo et al. 1974). Since casein was not serving primarily as an energy source these results have been taken to indicate that milk protein synthesis is limited by the supply of one or more amino acids. Moreover the responses to casein infusions were significantly greater from cows on diets with a high NPN content. The same may be true for silage.

2.4 CONCLUSION

Experimental evidence indicates that different feeding levels of forage rations appear to be associated with variations in the yield and composition of milk. However the effects of the type of forage diet on milk yield and composition are uncertain since interpretation of available evidence is confusing due to confounding with feeding level. Indirect evidence indicates that silage, a common diet of dairy cows in New Zealand, may have specific effects on the synthesis of milk and its constituents.

Mechanisms responsible for any effects on milk yield and composition when forage rations are fed to dairy cows are obscure. Changes in the nature of the rumen fermentation of cows on forage diets appear small and are likely to be relatively unimportant influences on milk composition, especially milk protein concentration. Differences between forages in the extent of rumen fermentation and site of digestion of nutrients are likely to be more important factors affecting changes in milk yield and composition. The amount of VFA produced in the rumen and the quantity and quality of protein entering the duodenum of cows would seem to be major determinants of variations in the synthesis of milk and its constituents by cows fed varying levels or types of forage rations.

Nutritional experiments with dairy cattle have not been designed to investigate the separate effects of feeding level and diet on milk yield and composition. To substantiate the evidence from the review that feeding level and diet type, notably silage, may have independent effects on the yield and composition of milk, experiments were conducted to look specifically at these factors.

Initially comparisons were made between pasture and silage on milk yield and composition. Subsequent experiments then examined how nutrient utilization of high moisture silage, offered to lactating cows, was modified by controlling the ensiling process or by supplementing cows on silage rations with either other forages (pasture or maize silage) or with protein concentrates.

Some of the mechanisms that may affect the synthesis of milk and its constituents by cows on silage rations were then investigated in a series of experiments by examining effects on milk yield and nutrient utilization when intra-abomasal infusions of nutrients were made to these cows.

3 EXPERIMENTAL

3.1 THE INFLUENCE OF LEVEL OF DIGESTIBLE ENERGY INTAKE ON THE YIELD AND COMPOSITION OF MILK OF COWS FED PASTURE OR UNWILTED PASTURE SILAGE

Introduction

Cows usually produce more milk containing more SNF and protein, and less fat, when increasing amounts of concentrates are offered with roughages (Burt, 1957; Huber and Bowman, 1966a,b; Kirchgessner, Friesecke and Koch, 1967; Broster, 1969; Kay, 1969; Armstrong and Prescott, 1971; Balch, 1972). These reports attribute the enhanced milk yield, SNF and protein content to an increased DEI, and the decline in milk fat content principally to a change in the concentrate to roughage ratio.

Information on the separate effects of diet and level of feeding on yield and composition of milk when roughage is the sole ration is limited. Reducing the level of pasture offered to cows by increasing grazing intensities has resulted in lower milk yields and milk protein contents, and higher milk fat contents (Hutton, 1973, 1975; T.E. Trigg unpublished). Low yields of milk and fat, and low contents of SNF when cows are offered pasture silage have been attributed to low voluntary intakes of silage (Wallace and Parker, 1966; Hutton *et al.* 1971; Lancaster *et al.* 1974). However when Fisher *et al.* (1975) restricted a ration of silage and pasture (1/1) from 100 % to 50 % of appetite, cows produced less milk of the same fat and protein content. Moreover Hutton (1975) found that when the proportion of silage fed with pasture to groups of cows was increased from $\frac{1}{3}$ to $\frac{2}{3}$ of the ration there was a decrease in

milk yield and milk protein content although the average DMI remained similar.

The effects of silage diets on milk yield and composition may therefore be explicable in terms of dietary factors rather than intake. This possibility was examined in the experiment reported here. Various amounts of pasture and silage were offered as sole rations and changes in the yield and composition of milk were examined.

Materials and Methods

Preparation of Foods - The pasture silage was made with a flail harvester in the spring of 1974 from a predominantly ryegrass (Lolium perenne) and white clover (Trifolium repens) pasture in the early flowering stage of maturity. Ensiled herbage was consolidated in a concrete bunker by rolling with a tractor and then covered with a weighted polythene film. Simultaneously, pasture herbage was harvested from the same crop with a rotary mower, quick frozen at -33 ^{o}C and stored at -18 ^{o}C (Hutton, Hughes, Bryant, Pluck and Taylor, 1975). The silage was removed from the bunker 2 months after ensiling and frozen in a similar manner to the pasture.

Design - Twenty four crossbred cows in early lactation were removed from grazing and individually fed in stalls on a mixture (1:1, DM basis) of pasture and silage for ten days (Period I). Food intake, milk yield and composition, and live weight during the last 7 days of Period I were used as a basis for stratifying the cows into eight similar treatment groups. For 10 days thereafter (Period II) groups were fed to appetite, four on pasture and four

on silage. The animals continued on the same food in Period III but at one of four levels of intake. The levels offered were (A) silage to appetite based on the voluntary intake measured over the last 7 days of Period II, (B) 90 %, (C) 80 %, (D) 70 % of (A) respectively and (E) pasture to appetite based on the voluntary intake measured over the last 7 days of Period II, (F) 80 %, (G) 60 %, and (H) 50 % of (E) respectively. After one week the restricted rations were adjusted by the same percentage that the intake of animals on unrestricted feeding had changed relative to Period II. Period III lasted 21 days. Two cows from each of the eight treatments were then transferred to metabolism stalls for 10 days to enable measurement of the apparent digestibilities of dietary nitrogen and energy at each level of feeding (Period IV).

Feeding, Milking and Weighing - Feeding was at 0800 to 1300 hours and from 1600 to 2100 hours. Sufficient amounts of the rations were removed from the freezer each afternoon and allowed to thaw overnight. The rations offered in the digestibility period were representative of those offered in the experimental period. The cows fed to appetite were offered 115 % of the previous day's intake with subsequent additions where necessary.

Except when in digestibility stalls the animals were confined in a loafing barn when not being fed or milked.

Cows were milked twice daily at 0700 and 1600 hours. Individual milk yields were recorded and aliquot samples were taken and bulked over two successive periods of 3 and 4 days in each week and preserved with Lactabs (commercial pellets weighing 160 mg and containing $K_2Cr_2O_7$ (96 %) - Tompson and Capper Ltd., Liverpool).

Additional milk samples were collected over the last 7 days of the digestibility period for each cow and preserved with 1 ml formalin. Cows were weighed each morning after milking during Periods I - III and on the final day of the digestibility period.

Sampling of Feeds and Residues - Each day a representative sample of each food offered, and aliquot portions of food refused by each animal were bulked and frozen, combined over each week and subsampled for analysis. In Period IV, food residues, faeces, and urine were collected daily, weighed and daily aliquots of each for each animal were aggregated over the last seven days. Equipment similar to that described by Hughes (1963) was used to separate and collect faeces and urine from these cattle. Urine was collected in 150 ml of 6N HCl and samples were refrigerated (1 ^{o}C). Faeces samples were frozen (-20 ^{o}C) until analysed.

Sampling of Blood and Rumen Liquor - Blood (15 ml) was collected from the jugular vein in heparinized vacutainer tubes (Becton-Dickinson, Rutherford, New Jersey) immediately prior to feeding and again 3 hours later on the last day of Period I and Period III. Immediately after withdrawal, packed cell volumes were determined, and plasma for analysis separated using a refrigerated centrifuge (4 ^{o}C, 2000r.p.m., for 15 minutes).

Rumen liquor (100 ml) was withdrawn by stomach tube 3 hours after feeding commenced on the last day of Period III. Saturated mercuric chloride solution was immediately added to the sample which was stored at -20 ^{o}C after filtering through cheese cloth.

Analytical Methods - Fat, protein (N x 6.38), and lactose contents of milk were determined, respectively, by the Gerber technique (BS.696,2), the standard Kjeldahl nitrogen digestion followed by steam distillation (AOAC, 1965) and by the ferricyanide method using the Technicon Autoanalyser (Technicon methodology N, 2a). The dry matter content of the pasture silages were determined by toluene distillation (Minson and Lancaster, 1963), and pasture by oven drying at 100 °C for 24 hours.

The gross energy content was determined by adiabatic bomb calorimetry on oven dried samples of foods and faeces, and freeze dried samples of urine. The loss of energy from the silage during drying was assumed to equal the calorific value of its VFA content as indicated by gas liquid chromatography (G.L.C.). VFA in food samples (40 g) were determined by adding 6N H_2SO_4 (60 g), extracted for 24h, then distilling 4 ml of extract in a Markham still. After making the distillate alkaline with N NaOH and drying, 0.5 ml phenol solution (2 g/l) was added, acidified with H_3PO_4 (85 % $^{W}/_{w}$), and samples (2 ul) were injected into the GLC (glass column, 1 x 3 mm I.D., packed with Chromosorb 101 (Johns-Manville, U.S.A.) (100-120 mesh) and operated at 190 °C with carrier gas (N_2) flow of 35 ml/minute). Concentrations of VFA were calculated by comparing peak areas with those of phenol, the internal standard.

Crude protein (N x 6.25) was determined on undried samples of foods, faeces, and urine by standard Kjeldahl digestion procedures followed by steam distillation (AOAC, 1965). Water soluble nitrogen was measured in 25 ml aliquots of supernatant obtained after 80 g silage in 400 ml of water was homogenized at high speed for 2 minutes and centrifuged at 5000 g for 10 minutes. Further aliquots (50 ml) of supernatant were treated with an equal volume

of 10 % TCA for determining NPN. Soluble protein nitrogen was calculated by difference. Water soluble carbohydrate in the supernatant was determined by an anthrone procedure (Bailey, 1958). Modified acid detergent (MAD) fibre was determined by the method of Clancy and Wilson (1966).

Packed cell volumes were determined using a micro haematocrit centrifuge. Plasma was analysed for α amino N by the ninhydrin method of Rosen (1957), and blood urea by micro-diffusion (Conway, 1957).

Strained rumen liquor samples (2 ml) were made alkaline with N NaOH, dried, and analysed for individual concentrations of VFA by G.L.C. as described above.

Statistical Analysis and Data Collection - Data compiled for each cow over the last week of Periods, I, II, IV and the last two weeks of Period III were used in the analysis. Live weight change was predicted by regression of live weight on time. The apparent digestibility coefficients determined in Period IV were used to derive digestible energy intakes.

The mean yields of milk and constituents, milk composition, and digestion data of each treatment group were calculated and the relationships between the variates and $DEI/LW^{0.75}$ were studied by multiple regression analysis using standard procedures (Snedecor, and Cochrane, 1967). Intersecting lines were fitted using the technique of Gujarati (1970).

The data were fitted to four mathematical models:

$$\begin{aligned}
&(i) && Y_{ij} = Bu_{ij} + b_i x_{ij} + c_i \\
&(ii) && Y_{ij} = Bu_{ij} + b_i x_{ij} + c \\
&(iii) && Y_{ij} = Bu_{ij} + b x_{ij} + c_i \\
&(iv) && Y_{ij} = Bu_{ij} + b x_{ij} + c
\end{aligned}$$

where u_{ij} = production data in Period I
Y_{ij} = production data in Period III
x_{ij} = $DEI/LW^{0.75}$ in Period III

The partial regression coefficient B allows for the inherent production of the individual cow for each milk variate and subsequent reference to regression coefficients is to the partial regression coefficient b, which describes the change in the milk variates per unit change in $DEI/LW^{0.75}$.

Significant differences in the regression coefficients and intercepts between the diets were tested by the differences in the residual sums of squares between the models at the appropriate degrees of freedom. Model (iii)-(i) tested different coefficients given different intercepts. Model (iv)-(iii) tested different intercepts given the same coefficient and Model (iv)-(ii) tested different slopes with the same intercept. The effects of the rations fed ad libitum were compared by analysis of covariance of Period II data using Period I data as the covariate.

Linear regressions of the energy contents of milk and live weight change (ΔLW) per kg $LW^{0.75}$ (calculated using respective conversion factors of 3.14 MJ/kg FCM and 20.95 MJ/kg ΔLW (Bath, Ronning, Myer and Lofgreen, 1965) on digestible energy intake per kg $LW^{0.75}$ were derived to provide estimates of maintenance requirements.

The metabolizability of the diets were calculated by estimating the methane production during Period IV using the relationship (A.R.C., 1965) CH_4 (MJ/100 MJ GEI) = 4.28 + 0.59 (apparent digestibility of energy).

Multiple regressions relating DEI as silage or pasture to F.C.M., Δ LW, and $LW^{0.75}$ were also derived.

The following symbols are used to indicate degrees of significance:

+ = $P < 0.10$, * = $P < 0.05$, ** = $P < 0.01$, *** = $P < 0.001$,
NS = not significant, $P > 0.10$.

Results

Chemical Composition (Table 5) - Pasture had a lower content of dry matter but a higher concentration of nitrogen and soluble carbohydrate than silage. Small amounts of lactic, acetic and butyric acids were detected in pasture compared with those in silage. The forms of nitrogen in the herbage were considerably altered by ensiling, as shown by changes in levels of ammonia, soluble nitrogen and soluble protein nitrogen.

Intake, Milk Production and Live Weight - When silage or pasture was offered to appetite in Period II, the voluntary intake of DE of silage was 35 % less than pasture (Table 6) and as a consequence the range in intake of silage in Period III was limited. For silage the digestibility of the nitrogen was higher, and that of dry matter, O.M., and energy, lower, than in pasture (Table 7). Altering the level of intake had no significant effect on the apparent digestibilities of dry matter, nitrogen and energy in the rations. The low intake of silage relative to pasture in Period II resulted

TABLE 5. Chemical composition of diets

Nutrient	Pasture	Silage
DM %	15.7 ± 0.37[a]	17.7 ± 0.37
Composition of 100 g DM		
OM (g)	87.8 ± 0.32	91.4 ± 0.21
MAD fibre (g)	29.7 ± 0.45	35.3 ± 1.23
GE (MJ)	1.81± 0.021	1.79± 0.010
Total N (g)	2.8 ± 0.09	2.5 ± 0.08
Soluble N (g)	0.8 ± 0.04	1.3 ± 0.03
Soluble protein N (g)	0.49± 0.03	0.0 ± 0.00
NH_3-N (g)	0.08± 0.005	0.25± 0.005
Soluble carbohydrate (g)	5.8 ± 0.69	1.4 ± 0.20
Lactic acid (g)	0.4 ± 0.25	8.6 ± 0.42
Acetic acid (g)	0.4 ± 0.03	1.7 ± 0.14
Propionic acid (g)	0.0 ± 0.00	0.1 ± 0.03
Butyric acid (g)	0.1 ± 0.00	0.9 ± 0.09
pH	5.4 ± 0.04	3.9 ± 0.02

[a] SEM, n = 6

TABLE 6. Intake of DM (g/kg $LW^{0.75}$/d) and DE (MJ/kg $LW^{0.75}$/d).

Treatment	Period I		Period II		Period III	
	$DMI/LW^{.75}$	$DEI/LW^{.75}$	$DMI/LW^{.75}$	$DEI/LW^{.75}$	$DMI/LW^{.75}$	$DEI/LW^{.75}$
Silage:						
Group A	145	1.874	112	1.409	115	1.413
B	145	1.880	123	1.545	114	1.399
C	146	1.897	124	1.575	107	1.341
D	140	1.802	118	1.488	94	1.158
Pasture:						
Group E	143	1.841	169	2.250	157	2.059
F	144	1.804	165	2.237	130	1.701
G	146	1.869	170	2.300	104	1.365
H	135	1.748	153	2.088	79	1.105
S.D.	8.1	0.106	8.3	0.108	6.2	0.069

TABLE 7 - Mean apparent digestibility coefficients of diets.

Item	Digestibility (%) Silage	Pasture	SED	Sign.
GE	69.7	74.3	± 0.64	***
DM	68.8	75.8	± 0.69	***
OM	73.0	77.7	± 0.58	***
N	68.3	65.1	± 0.98	**

n = 8

TABLE 8. Mean daily intake and yield and composition of milk of cows in Period II[1].

Item	Silage	Pasture	SED	Significance
Milk yield (kg)	11.7	13.6	± 0.42	***
Milk fat (g)	351	433	±14.1	***
Milk protein (g)	235	288	± 8.3	***
Milk lactose	421	489	±12.4	***
Milk fat %	4.27	4.58	± 0.071	***
Milk protein %	2.87	3.06	± 0.052	**
Milk lactose %	5.09	5.11	± 0.068	N.S.
DMI (kg)	9.59	13.68	± 0.234	***
DEI (MJ)	120.7	184.9	± 3.21	***

[1] (adjusted for variation in Period I, n=12)

in 14 % less milk, 19 % less fat and 18 % less protein (Table 8). Relative to pasture, silage reduced the fat and protein content of milk but lactose content was unaltered by the type of food consumed.

The mean yield and composition of milk and Δ LW in Period III are presented in Table 9 and the relationships between milk production and the intake of DE, (Model i) are summarized in Table 10. The partial regression coefficients for silage were not significant. However it was evident that a single regression equation could not be used to describe the pasture and silage data because there was a significant reduction in the residual sums of squares from Model (iv) to Models (ii) or (iii) (see Appendix 1). Except for milk fat percentage, Model (ii) was used to describe the data (Table 11, Figures 3-8), since it accounted for more of the variation than Model (iii). The regression coefficients were positive except for milk fat percentage, and significant except for lactose percentage. The differences in the coefficients between pasture and silage were 1.16 ± 0.31, 102.8 ± 16.4, 48.4 ± 12.1, 182.7 ± 40.6 for milk, fat, protein and lactose yields, respectively, and 0.14 ± 0.04 for milk protein percentage. It should be noted that while the magnitude of the partial regression coefficients for the silage group in Model (ii) (Table 11) were similar to those in the unconstrained Model (i) (Table 10) their significance was largely due to the constraint placed on the intercept.

These regression analyses indicate that at similar intakes of DE, cows fed pasture produced more milk, fat, protein and lactose, and had higher contents of fat and protein in milk than cows fed silage. Relationships between pasture intake and the yields of milk, fat, protein and lactose, and of protein percentage were

TABLE 9. Mean daily yield and composition of milk, and LW change in Period III.

(i) Actual Means

Item	Silage				Pasture				S.D.
	A	B	C	D	E	F	G	H	
Milk yield (kg)	9.6	9.7	9.1	7.0	13.7	11.6	10.8	6.9	3.13
Milk fat (g)	401	367	335	314	633	544	472	373	153
Milk protein (g)	278	259	242	186	442	349	304	195	85
Milk lactose (g)	487	483	427	335	676	577	576	326	158
Milk fat %	4.15	3.85	3.68	4.41	4.65	4.64	4.51	4.92	0.63
Milk protein %	2.88	2.69	2.65	2.64	3.25	3.01	2.86	2.87	0.19
Milk lactose %	5.04	4.99	4.70	4.77	4.91	4.93	5.46	4.55	0.52
Liveweight change (kg)	+0.16	-0.83	-0.70	-0.70	-1.08	-1.58	-1.56	-2.12	0.62
(ii) Adjusted Means *									R.S.D.
Milk yield (kg)	8.6	9.1	8.4	8.4	12.6	12.1	9.9	9.3	0.97
Milk fat (g)	323	353	359	347	556	553	487	447	53
Milk protein (g)	229	241,	233	222	406	365,	304	253	30
Milk lactose (g)	411	444	398	406	613	610	546	460	66
Milk fat %	3.98	3.90	4.12	4.15	4.45	4.57	4.78	4.85	0.27
Milk protein %	2.81	2.66	2.71	2.62	3.29	3.04	2.95	2.79	0.15
Milk lactose %	4.78	4.82	4.71	4.78	4.79	5.04	5.56	4.84	0.35

* (Adjusted for variation in Period I, n=3)

TABLE 10. Relationships between milk variates and DEI/$LW^{0.75}$ (MJ/kg $LW^{0.75}$/d) of silage (S) or pasture (P) in Period III (Model i).

Item	Mean	S.D.	$B\mu \pm SE$	Sign	Ration	$bx \pm SE$	Sign	C	R.S.D.	R
Milk Yield (kg/d)	9.82	3.41	0.800 ± 0.064	***	S	1.206 ± 2.436	NS	-3.597	0.96	0.97
					P	3.699 ± 0.802	***	-5.413		
Milk Fat (g/d)	428	167	0.843 ± 0.073	***	S	23.98 ± 123.62	NS	-173	50.4	0.96
					P	119.73 ± 42.32	*	-163		
Milk Protein (g/d)	282	107	0.749 ± 0.062	***	S	63.03 ± 68.70	NS	-155	27.3	0.97
					P	160.02 ± 22.87	***	-221		
Milk Lactose (g/d)	486	176	0.828 ± 0.089	***	S	31.51 ± 167.44	NS	-169	64.7	0.94
					P	146.44 ± 54.65	*	-213		
Milk Fat %	4.35	0.662	0.857 ± 0.093	***	S	-0.694 ± 0.629	NS	1,215	0.24	0.95
					P	-0.460 ± 0.192	*	1.639		
Milk Protein %	2.86	0.251	0.623 ± 0.152	***	S	0.293 ± 0.369	NS	0.415	0.15	0.85
					P	0.488 ± 0.122	***	0.363		
Milk Lactose %	4.92	0.503	1.562 ± 0.349	***	S	0.289 ± 0.965	NS	-2.537	0.38	0.73
					P	0.322 ± 0.312	NS	-2.120		

(i) μ denotes production in Period I

(ii) x denotes DEI/kg $LW^{0.75}$ in Period III

(iii) Mean DEI/kg $LW^{0.75}$ = 1.44 ± 0.30

TABLE 11. Relationships between milk variates and DEI/$LW^{0.75}$ (MJ/kg $LW^{0.75}$/d) of silage (S) or pasture (P) in Period III.

Item	Mean	S.D.	$B\mu \pm SE$	Sign	Ration	$bx \pm SE$	Sign	C	R.S.D.	R
Milk Yield (kg/d)	9.82	3.42	0.79 ± 0.06	***	S P	2.41 ± 0.93 3.57 ± 0.75	* **	-5.31	0.94	0.97
Milk Fat (g/d)	428	167	0.84 ± 0.07	***	S P	17.7 ± 47.46 120.5 ± 38.59	NS **	-164.5	49.1	0.96
Milk Protein (g/d)	282	107	0.74 ± 0.06	***	S P	106.3 ± 26.6 155.1 ± 21.4	*** ***	-211.1	27.0	0.97
Milk Lactose (g/d)	486	175	0.82 ± 0.08	***	S P	60.9 ± 63.82 143.6 ± 51.30	NS *	-206.8	63.1	0.94
Milk Fat %	4.35	0.66	0.87 ± 0.08	***	S P	-0.48 ± 0.178	*	0.88 1.62	0.23	0.92
Milk Protein %	2.86	0.25	0.62 ± 0.14	***	S P	0.34 ± 0.136 0.48 ± 0.113	* ***	0.36	0.14	0.80
Milk Lactose %	4.92	0.50	1.58 ± 0.34	***	S P	-0.56 ± 0.36 -0.29 ± 0.28	NS NS	-2.24	0.37	0.72

(i) μ denotes production in Period I

(ii) x denotes DEI/kg $LW^{0.75}$ in Period III

(iii) Mean DEI/kg $LW^{0.75}$ = 1.44 ± 0.30

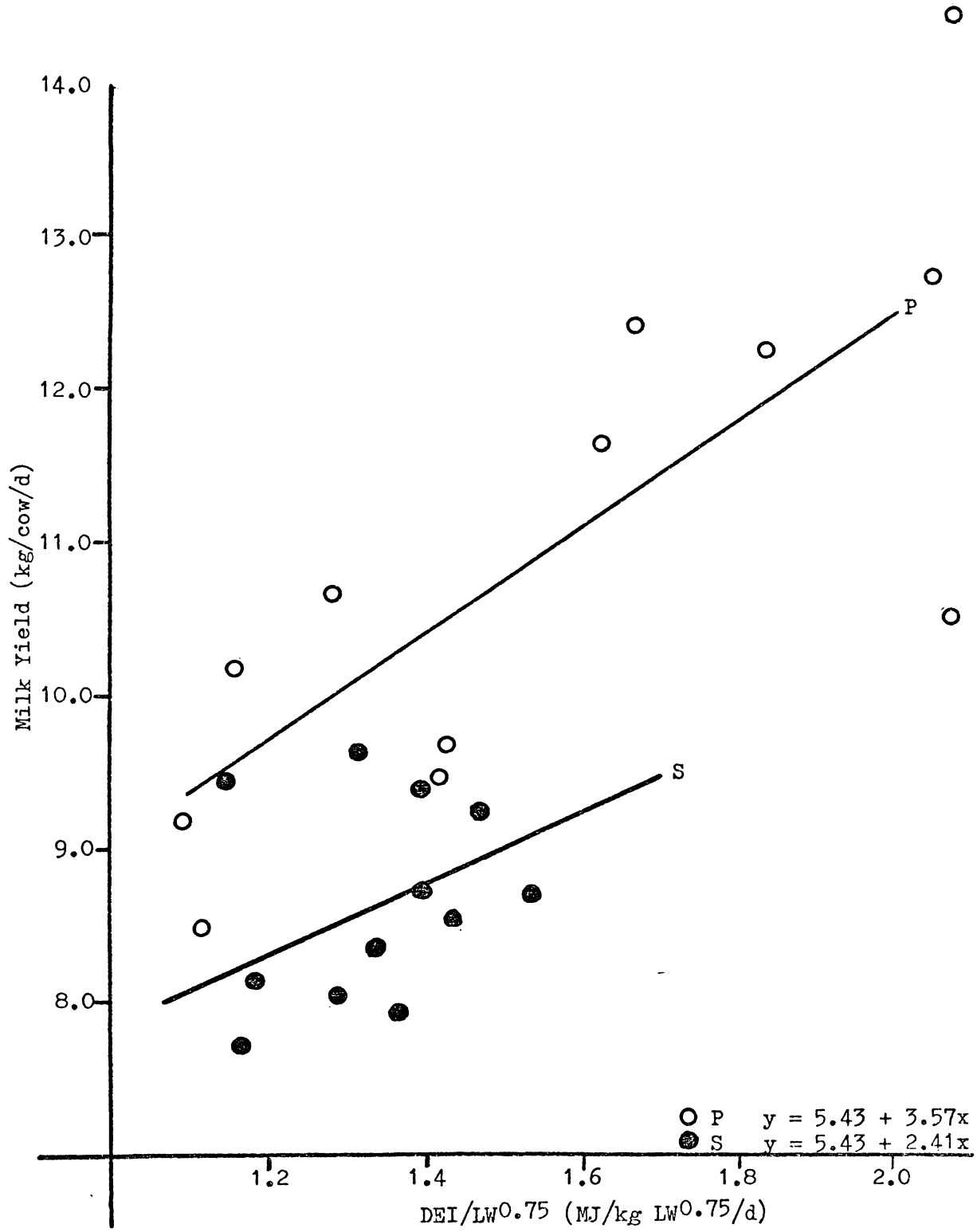


Fig.3 Relationship between adjusted milk yield and DEI/$LW^{0.75}$ of pasture (P) and of silage (S).

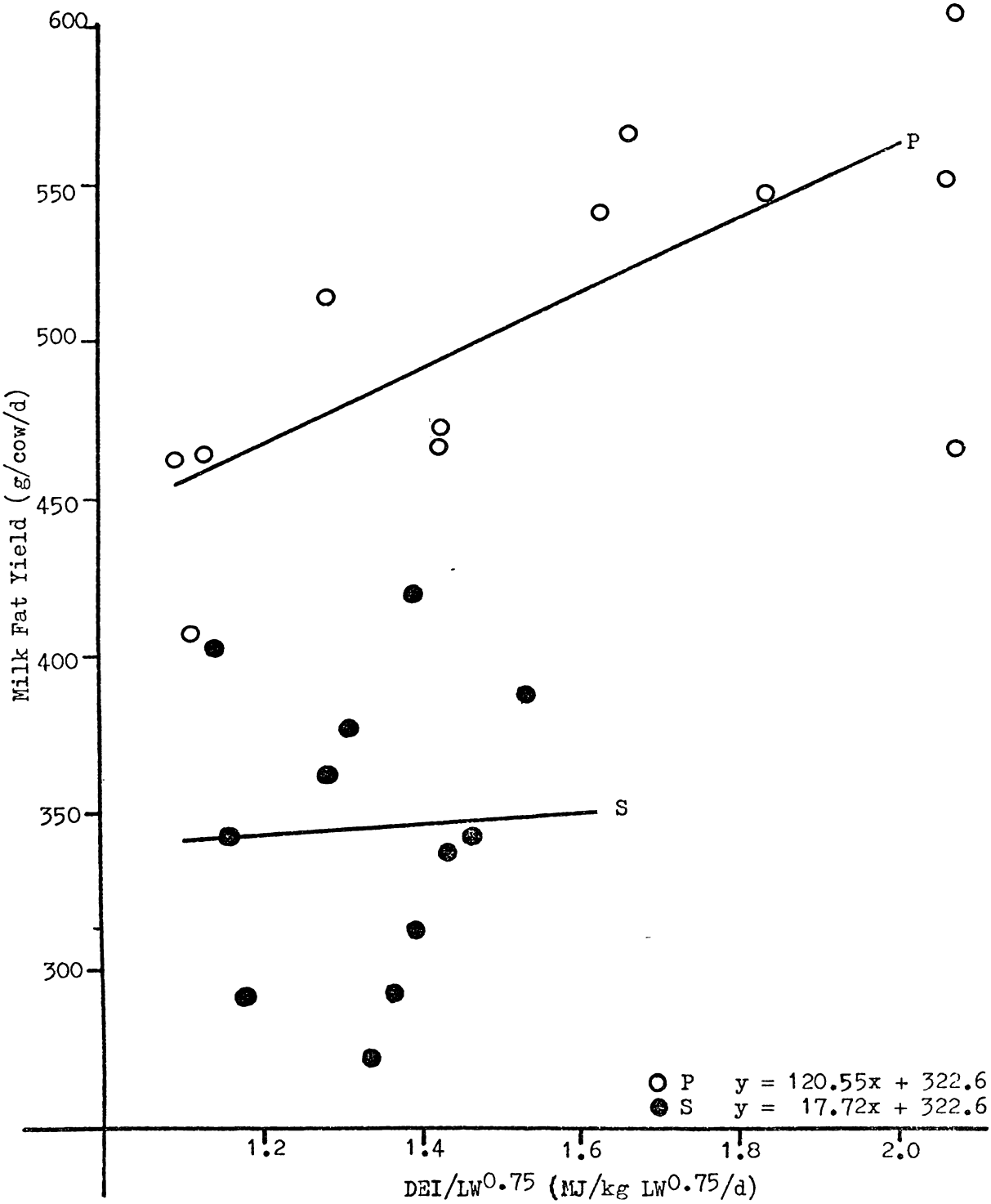


Fig.4. Relationship between adjusted milk fat yield and DEI/$LW^{0.75}$ of pasture (P) and silage (S).

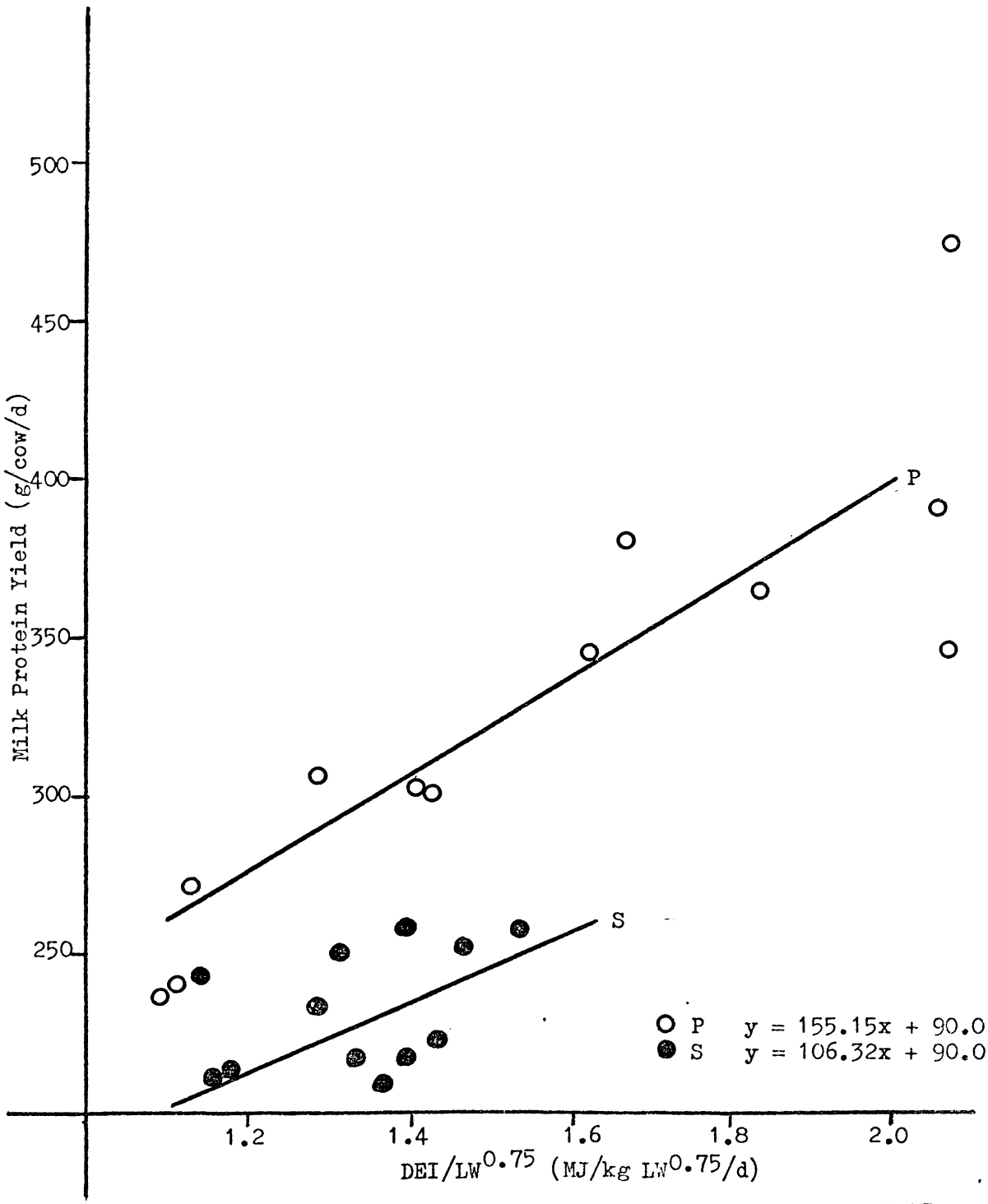


Fig. 5. Relationship between adjusted milk protein yield and DEI/$LW^{0.75}$ of pasture (P) and of silage (S).

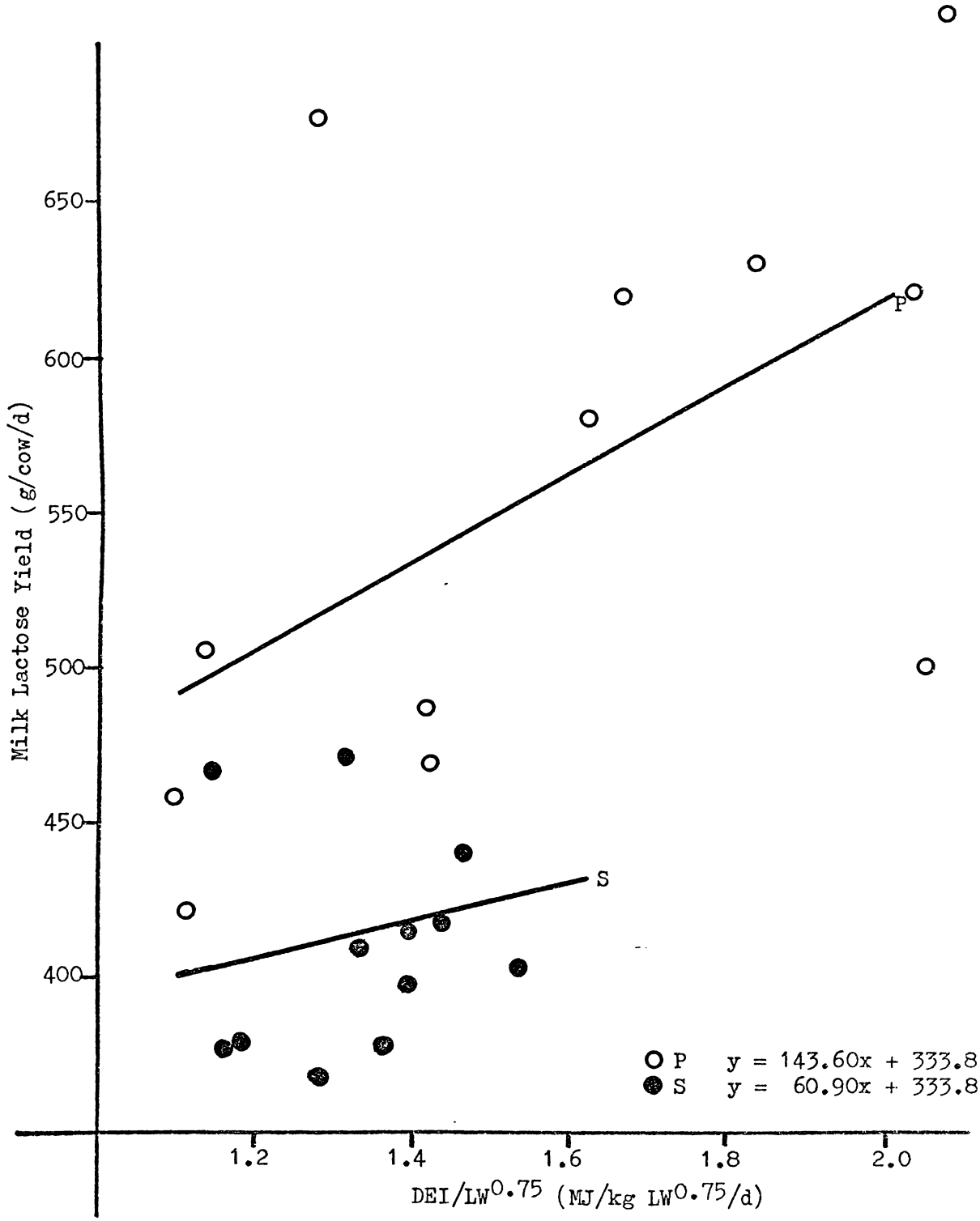


Fig. 6. Relationship between adjusted yield of milk lactose and DEI/$LW^{0.75}$ of pasture (P) and of silage (S).

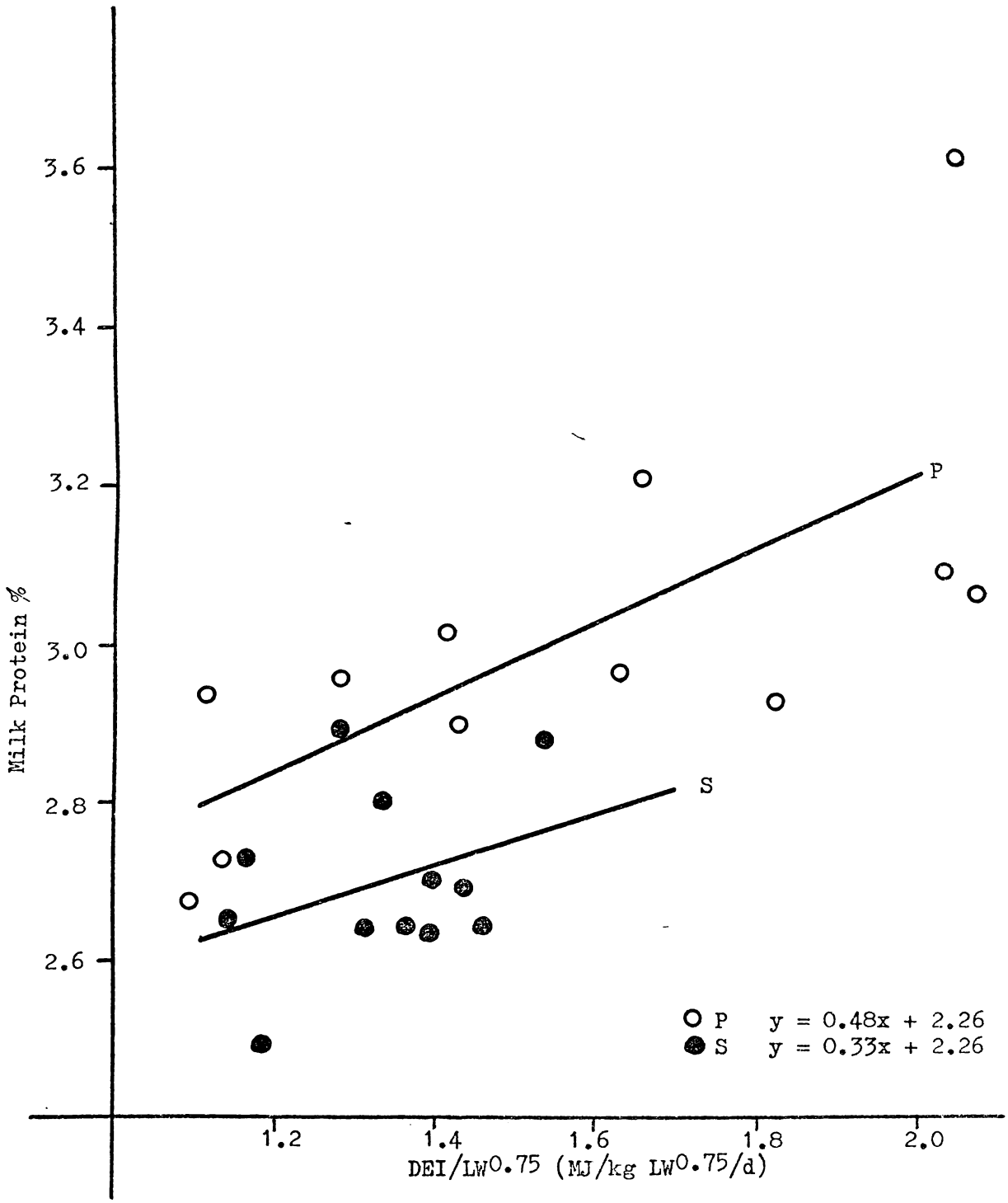


Fig. 7. Relationship between adusted milk protein concentration and DEI/$LW^{0.75}$ of pasture (P) and of silage (S).

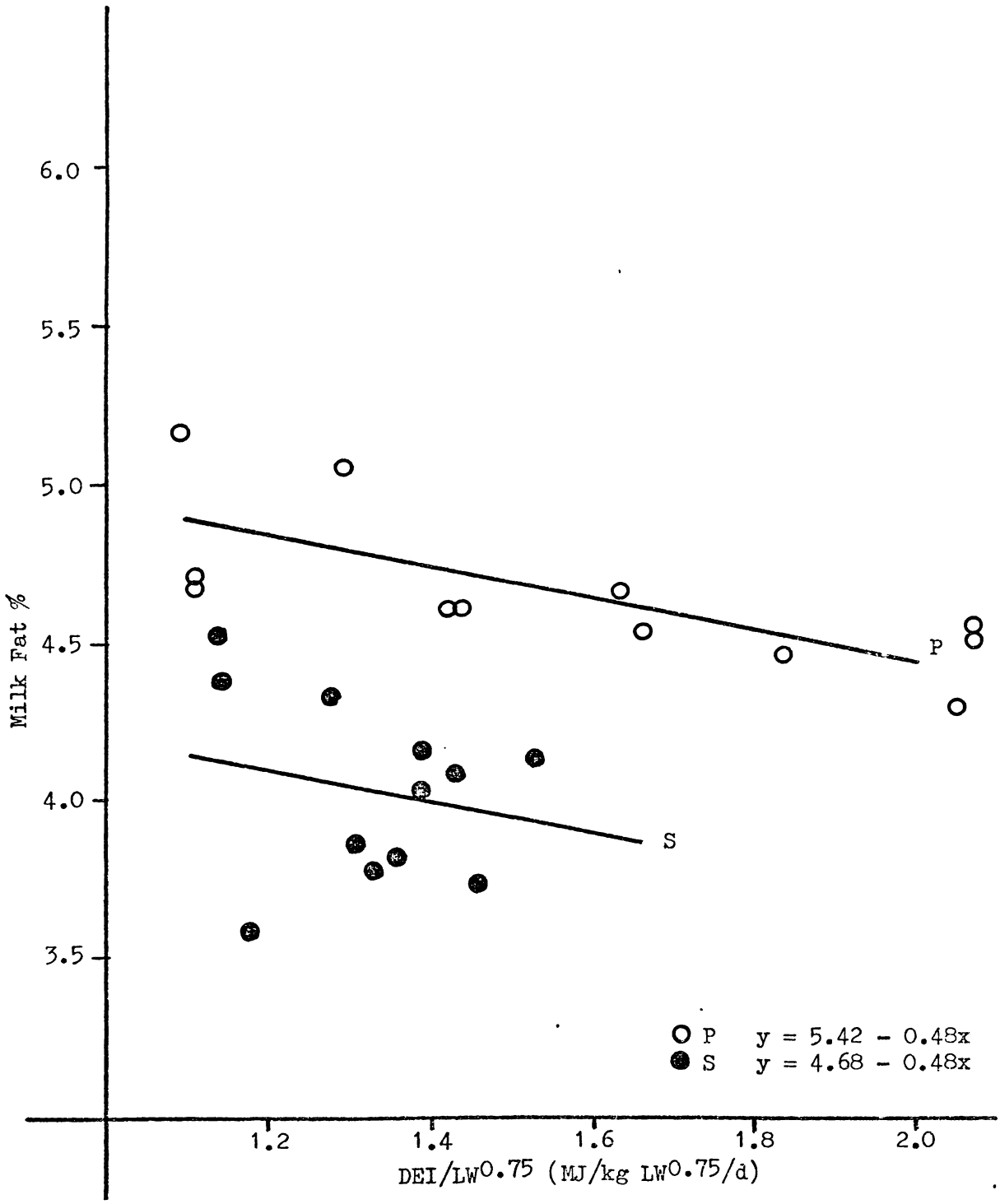


Fig. 8. Relationship between milk fat concentration (%) and DEI/$LW^{0.75}$ of pasture (P) and of silage (S).

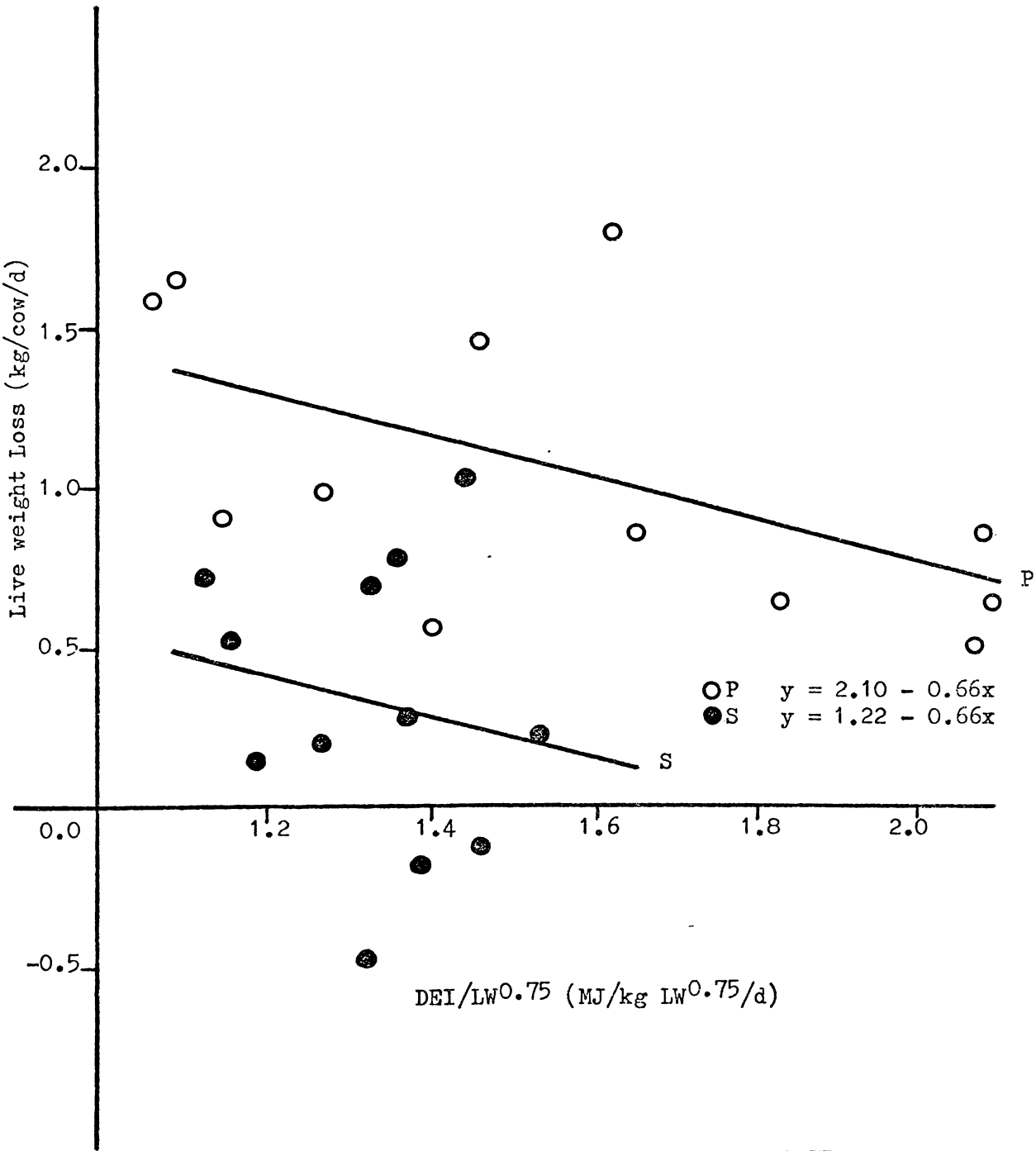


Fig. 9. Relationship between live weight change and DEI/$LW^{0.75}$ of pasture (P) and of silage (S).

positive, and for milk fat percentage were negative. However, although similar trends were found between silage intake and these milk variates, the responses were lower and were not significant.

Relative to intake, the live weight loss of the cows fed pasture was significantly higher than those fed silage (Figure 9 and Appendix 2).

Composition of Rumen Liquor and Blood - Molar percentages and concentrations of acetic and propionic acids were higher, and butyric acid was lower in rumen liquor of cows fed silage in comparison to cows fed pasture (Table 12). The molar ratio of acetate to propionate was less with silage. There was no significant effect of feeding level on molar percentage, molar ratio of acetate to propionate, or concentration of VFA in rumen liquor of cows on the silage ration. A significant relationship was obtained in the concentrations of total rumen VFA's, and in the molar percentages and molar ratio of acetate and propionate in rumen liquor with the feeding level of pasture, and the coefficients are shown in Table 12.

When cows were offered silage, the PCV % in blood increased with time from feeding whereas there was a small decrease in PCV % of cows eating pasture; the diet x time interaction being significant (Table 13). A positive relationship between PCV % and feeding level occurred for the cows on the silage ration. The levels of urea and α amino N in blood plasma increased with time from feeding on both rations. Positive interactions occurred between feeding level and plasma urea for cows on the silage ration, and with α amino-N concentration in plasma for cows on the

TABLE 12. Mean molar percentages and concentrations of individual and total VFA in the rumen liquor, and molar ratios of acetate to propionate.

	Concentration (meq/100 ml) of VFA				Molar % of Total VFA			HAc % / HPr %
	HAc	HPr	HBu	Total	HAc	HPr	HBu	
Silage:								
Group A	5.03	1.84	0.84	7.71	65.2	23.9	10.9	2.7
B	5.58	2.01	0.84	8.43	66.3	23.8	9.9	2.8
C	5.82	2.04	0.89	8.75	66.7	23.2	10.1	2.9
D	5.67	2.18	0.89	8.74	64.8	24.9	10.3	2.6
b ± SE	-0.73±0.92	-0.02±0.26	1.4±1.04	-0.68±0.44	2.5±3.8	-4.0±2.7	1.5±2.4	0.59±0.47
Pasture:								
E	4.42	1.19	1.09	6.68	66.4	17.4	16.2	3.8
F	4.96	1.60	1.44	8.01	61.9	20.0	18.0	3.8
G	5.28	1.75	1.49	8.52	61.9	20.6	17.5	3.0
H	4.90	1.60	1.40	7.90	61.9	20.3	17.8	3.0
b ± SE	-0.43±0.47	-0.25±0.17	-1.1±0.8	-0.48±0.18+	3.7±1.4*	-2.7±1.1*	-1.1±0.7	0.82±0.27*
S.D.	0.51	0.20	0.17	0.82	1.6	1.3	1.0	0.4

TABLE 13. Effect of diet on PCV %, urea and ∝ amino N concentrations in plasma immediately prior to feeding (0h) and 3h later (adjusted for Period I).

	PCV %		Plasma Urea (mg/100 ml)		Plasma Amino N (mg/l)	
	0 h	3 h	0 h	3 h	0 h	3 h
SILAGE						
Group A	33.1	36.7	10.6	15.9	25.0	29.2
B	31.2	32.9	10.0	15.3	20.6	26.2
C	27.8	32.8	13.4	15.8	23.9	29.6
D	30.8	32.5	13.9	15.8	26.0	32.1
Mean $\bar{S}$	30.8	33.7	11.9	15.7	23.9	29.0
Mean difference (3 h - 0 h) ± SED	2.9 ± 1.0 **		3.8 ± 0.4 ***		5.1 ± 0.9 ***	
PASTURE						
Group E	34.6	30.4	14.2	16.9	25.5	34.4
F	33.9	29.3	12.3	15.0	29.8	38.0
G	30.8	33.2	13.5	15.9	24.4	29.7
H	31.8	33.0	12.0	14.9	22.7	24.5
Mean $\bar{P}$	32.8	31.5	13.0	15.7	25.9	31.7
Mean difference (3 h - 0 h) ± SED	-1.3 ± 1.0		2.7 ± 0.4 ***		5.8 ± 0.9 ***	
Mean Difference of diets ($\bar{S}$ - $\bar{P}$)	-2.0	+2.2	1.1	0.0	-2.0	-2.7
SED	1.2		1.1		1.9	

TABLE 14. Effect of diet on nitrogen and gross energy partition.

Item	Silage	Pasture	SED	Significance
N, % of intake				
Faeces	31.6	34.8	± 1.21	**
Urine	42.4	48.6	± 2.6	+
Milk	17.8	24.1	± 1.6	**
Retention (NR)	8.2	-7.6	± 2.0	***
Milk + NR	26.0	16.4	± 2.5	**
N, % of digested				
Urine	62.1	74.8	± 3.5	*
Milk	26.0	36.9	± 2.4	**
Retention (NR)	11.9	-11.8	± 2.3	**
Milk + NR	37.9	25.1	± 3.5	*
GE, % of intake				
Faeces	30.3	25.6	± 0.7	***
Urine	4.8	4.3	± 0.3	NS
Methane (est)	8.4	8.7	± 0.03	***
Milk	18.6	21.3	± 2.0	NS
Residual (est)	37.9	40.1	± 1.8	NS
Milk and Residual	56.5	61.4	± 0.8	***
GE, % of digested				
Milk	26.6	28.5	± 2.8	NS
Milk and Residual	81.0	82.5	± 0.5	*
GE, % of ME				
Milk	32.8	34.6	± 3.3	NS
Metabolisable Energy (MJ/kg DM)	10.0	11.3	± .3	**

pasture (Table 13).

Partitioning of Dietary Nitrogen and Energy (Table 14) - Faecal, urinary, and milk nitrogen levels, expressed as a percentage of nitrogen intake, and urinary and milk nitrogen as a percentage of digested nitrogen, were significantly higher for cows on pasture than for cows on the silage ration. Nitrogen retention as a percentage of nitrogen intake, and digested nitrogen, was lower for the cows on the pasture ration. Increasing the level of feeding of silage or pasture increased nitrogen retention and decreased urinary nitrogen, as percentages of nitrogen intake and nitrogen digested (Appendix 3). In contrast feeding level did not significantly affect the partitioning of dietary energy intake on either ration. Diet had a marked effect, for there was a higher percentage of dietary energy in faeces, and a lower percentage in methane, and ME, for silage compared with the pasture diet. The mean ME contents of the silage and pasture diets were 10.0 $\pm$ 0.3 and 11.3 $\pm$ 0.7 MJ per kg DM respectively (Table 14). Despite gross differences in numerical values between estimates of the partitioning of energy derived from linear or multiple regression analyses, both methods indicate the animals on the silage ration had a lower maintenance requirement (Appendix 4).

Discussion

When fed to appetite, cows offered silage produced less milk, protein, fat and lactose than when pasture was offered. Fat and protein content of milk was also reduced. These effects were

associated with a decrease of 35 % in the intake of DE and are similar to those observed by other workers (Wallace and Parker, 1966; Hutton et al. 1971; Lancaster et al. 1974).

However in Period III of the present experiment it was clearly demonstrated that silage had specific effects on milk yield, and on the concentrations of protein and fat in milk. These results indicate that factors other than the amount of DE consumed may also affect milk synthesis of cows feeding solely on forage diets.

Effects of Diet on Milk Yield and Composition - Intra-ruminal infusions of acetate solutions have been shown to increase milk yield and milk fat percentage, while infusions of propionate solutions increase milk protein percentage (Rook and Balch, 1961; Rook et al. 1965; Wilson et al. 1967). Silage resulted in higher concentrations and molar proportions of acetate and propionate in rumen fluid than did pasture, yet milk yield and the concentrations of fat and protein in the milk were less. This suggests the molar proportions and concentrations of these rumen acids in cows fed silage may not be important determinants of milk yield, milk fat and milk protein contents.

Other studies (Armstrong and Prescott, 1971) indicate the molar ratio of acetate and propionate in rumen fluid may be an important determinant of milk fat percentage. In the present study the lower milk fat percentage of cows offered silage was associated with a decrease in the molar ratio of acetate to propionate. However the differences were small, suggesting that this factor was also unlikely to be of major importance. The lower milk fat synthesis by cows fed silage was also associated with a large reduction in the molar

percentage of rumen butyrate. Since butyrate is metabolized to B-hydroxybutyrate, a major precursor for milk fat synthesis (Smith et al. 1974), this factor may have contributed to the reduction in milk fat.

Diets inducing a decrease in milk fat percentage have also increased molar proportions of propionate in rumen fluid (McCullough, 1966; Armstrong and Prescott, 1971; Annison et al. 1974) and have reduced levels of milk fat precursors in plasma (Annison et al. 1974) presumably due to increased synthesis of tissue fat (Annison, 1976). Since cows fed silage tended to have higher molar proportions of rumen propionate and lost less body weight than cows fed pasture the lower milk fat synthesis of cows fed silage may have in part been caused by a lower supply of endogenous precursors.

Diet type had pronounced affects on the partitioning of dietary nitrogen. Cows offered pasture produced milk containing more nitrogen, expressed as a percentage of dietary nitrogen, than cows offered silage although they excreted higher proportions of this nitrogen in urine and faeces. Overall combined efficiencies of nitrogen utilization for milk plus tissue were less for pasture than silage. Conrad et al. (1961) found a similar result when these foods were compared at similar levels of intake.

Pasture reduced nitrogen retention principally as a result of its marked increase of urinary nitrogen excretion. The association of the higher live-weight loss and negative nitrogen retention for the pasture fed animals indicated they were mobilizing tissue protein. The resulting increased supply of endogenous amino acids may account for the increased quantities of nitrogen in the milk of the cows fed pasture although the use of this source of amino acids for milk protein synthesis is uncertain (van Es and Boekholt, 1976).

Alternatively the improved efficiency of utilization of digested pasture nitrogen for milk synthesis may have been due to differences in nitrogen metabolism in the rumen between the diets. The higher nitrogen solubility and NPN content of silage would tend to increase rumen ammonia concentrations (Sniffen, 1973; Donaldson and Edwards, 1976), and lower soluble carbohydrate in silage would impair assimilation of ammonia into microbial protein (Smith, 1969). This may promote increased ammonia absorption from the rumen (Armstrong, 1974) reducing the amount of dietary protein entering the duodenum (Hogan and Weston, 1969; Hogan et al. 1969; MacRae et al. 1972). However these possibilities cannot be substantiated from the data presented here and furthermore it is notable that no differences were observed in plasma levels of urea or α amino N for the rations.

Diet also affected partitioning of dietary energy. In contrast to nitrogen, the digestibility of dietary energy was less for silage than pasture. The higher ME content of pasture was a result mainly of a lower faecal energy output since differences between the rations in urinary energy excretion were small. Differences in ME/DE for the rations were small although significant in favour of pasture. This suggests differences in milk production obtained in Period III at a common DEI were largely due to increased efficiencies of utilization of ME for milk production. However milk energy/ME was not significantly higher for pasture than silage. This discrepancy arises since body tissue energy change and maintenance energy are not taken into account. Since the pasture fed cows incurred a greater live-weight loss then more tissue energy must have been used to supplement ME intake to support a higher

milk production or maintenance requirement.

Effect of Level of Feeding on Milk Yield and Composition - Increasing the feeding level of pasture increased the yields of milk and constituents, milk protein percentage, and decreased milk fat concentration. In comparison, the relationships for silage were of smaller magnitude than those of pasture, but they were not significant. The small range of silage intakes studied would have contributed to this result.

The lack of a relationship between milk protein percentage and silage intake is consistent with the inferences of Hutton et al. (1971) and Lancaster et al. (1974) and is in marked contrast to responses with pasture. Increases in milk protein yield with pasture intake were associated with increases in milk yield and milk protein percentage. This infers increases in pasture intake resulted in disproportionate increases in precursors for milk protein synthesis relative to silage.

Although milk fat yield increased with increased pasture intake, milk fat concentration decreased. These effects may be due to the variations in the rumen fermentation pattern since increasing pasture intake raised the molar percentage of acetate and decreased that of butyrate which are known to have specific effects on fat synthesis and milk fat content respectively (Rook and Balch, 1961). However the molar ratio of acetate and propionate increased with increased pasture intake and was therefore negatively related to trends in milk fat percentage. Since this is contrary to other findings (Armstrong and Prescott, 1971) this suggests this factor may be relatively unimportant in accounting

for the effects of pasture intake on milk fat percentage. A decrease in the availability of milk fat precursors could also arise if the decreased live-weight loss associated with increased pasture intake reflected lowered fat mobilization in adipose tissue.

The lack of variation in milk lactose percentage is in agreement with other studies (Holmes, Arnold and Provan, 1960; Castle, Drysdale and Waite, 1961; Rook and Line, 1961) and is consistent with the hypothesis that lactose is the major determinant of isotonicity between milk and blood (Rook and Wood, 1959). However other studies with silage and pasture respectively (Fisher et al. 1975; T.E. Trigg, unpublished) have shown milk lactose to vary with feeding level.

In this regard it is of interest that cows fed silage had a higher PCV % than cows offered pasture. A possible explanation could be that the higher osmotic pressure of silage (Bryant and Lancaster, 1970) may reduce extracellular fluid (Ternouth, 1967) and possibly affect mechanisms responsible for the regulation of water and nutrient content of milk.

In summary the experiment has demonstrated that diet type and feeding level can affect milk yield and composition. Silage was found to have specific effects on milk yield and composition which were independent of DEI. The next experiment examines the effects on milk yield and composition of silages varying in chemical composition in an attempt to define the characteristics of pasture silage limiting milk synthesis.

3.2 THE EFFECTS OF WILTED, FORMALIN TREATED, AND UNWILTED PASTURE SILAGES ON THE YIELD AND COMPOSITION OF MILK

Introduction

In Experiment 3.1 high moisture pasture silage was shown to have specific effects on milk yield and composition. At the same level of DEI, cows offered silage produced less milk of lower fat and protein contents than cows fed pasture.

The chemical characteristics of silage can be altered by wilting (Murdoch, 1960; Gordon, Derbyshire, Wiseman, Kane and Melin, 1961; Gordon, Derbyshire, Jacobson and Humphrey, 1965; McDonald, and Whittenbury, 1967; Jackson and Anderson, 1968; McDonald, Henderson and MacGregor, 1968; Castle and Watson, 1970; Jackson and Forbes, 1970; Donaldson and Edwards, 1976; Hinks, Edwards and Henderson, 1976) and by the addition of formalin (Barry and Fennessy, 1972; Brown and Valentine, 1972; Valentine and Brown, 1973; Wilkins, Wilson and Woolford, 1974; Valentine and Radcliffe, 1975).

In the present experiment use was made of these techniques to study their effects on milk yield and composition and to get some indication of the characteristics of high moisture pasture silage that might give rise to changes in milk yield and composition. The results of previous studies comparing these silages for milk production (Brown, 1960; Murdoch, 1962; Kormos, 1967; Castle and Watson, 1970; Fisher, Lessard and Lodge, 1971; Valentine and Radcliffe, 1975) have been confounded by level of feeding and supplementation with concentrates.

Pasture herbage was ensiled either immediately after harvesting

with and without the addition of formalin, or after being cut and allowed to wilt. The effects of these silages on the yield and composition of milk were subsequently determined.

Materials and Methods

Preparation of Silages - The silages were made in late spring 1974, from a common perennial ryegrass (Lolium perenne)/white clover (Trifolium repens) pasture mixture approaching ear emergence. The unwilted silage (C.S.) was cut with a single chop flail harvester. Harvesting of the formalin treated silage was similar with formalin (35 %^{w}/v HCHO) added (5 l/t wet weight) by an applicator attached to the harvester. The wilted silage (WS) was mown with a rotary mower, wilted for 24 hours and then fine chopped with a New Holland 717 harvester. Ensiled herbage was covered after consolidation with polythene film. Two months later the silages were removed from the bunkers, frozen, and stored at -20 ^{o}C. High moisture silage used during the uniformity period was made from a predominantly ryegrass/white clover pasture with a single chop flail harvester in October, 1974.

Design - Twenty seven crossbred Jersey cows in mid-lactation, ranging in age from 3-8 years and weighing 240-500 kg, were removed from grazing after being accustomed to silage and individually stall fed on high moisture silage for 10 days (Period I). Food intake, milk yield and composition, and live weight during the last 7 days of this period was used as a basis to assign the cows to nine equal groups. The three experimental silages were fed at three levels of intake for a period of 21 days (Period II). The levels imposed

were (1) to appetite based on intake during the Period I; (2) 75 %; (3) 50 % of (1) respectively. After one week the restricted rations were adjusted by the same percentage as the intake of animals on unrestricted feeding had changed relative to Period I. For 10 days following Period II, cows at 75 % level of feeding were confined in metabolism stalls for nitrogen and energy digestion trials (Period III).

Feeding, Sampling, Milking and Weighing - Feeding was at 0800 to 1300 hours and from 1600 to 2100 hours. Frozen silage was allowed to thaw before feeding. The cows fed to appetite were offered 115 % of expected intake with subsequent additions when necessary.

The animals were confined in a loafing barn when not being fed, milked, or when in digestibility stalls.

Milking, weighing, and sampling of offered and refused foods, milk, faeces, urine, blood, and rumen fluid were carried out as described in Experiment 3.1. Blood and rumen fluid were sampled from cows in the 100 % and 50 % feeding levels for each silage. Blood was collected on the last day of Periods I and II immediately prior to feeding (0h) and 3h later. Rumen fluid was samples at 3h and 12h on the last day of Period II.

Analytical Methods - Fat, protein, and lactose contents of milk were determined by infra-red analysis which was calibrated against milk samples that had been analysed by the standard procedures described in Experiment 3.1. Other procedures followed have also been outlined in Experiment 3.1.

Statistical Analysis and Data Collation - Data compiled for each cow over the last week of Period I and III and the last two weeks of Period II were used in the analysis. Procedures and regression models were as described in Experiment 3.1. The effects of treatments on milk yield and composition were also compared by analysis of variance (Snedecor and Cochrane, 1967).

Results

Chemical Composition, Digestibility and Intake (Tables 15, 16, Appendix 5) - There were significant differences between the rations in contents of DM, soluble nitrogen (SN), soluble protein nitrogen (SPN), ammonia nitrogen (NH_3-N), soluble carbohydrate, and MAD fibre (Table 15). Wilting and formalin treatment had no effect on nitrogen content but formalin reduced the solubility of nitrogen and both treatments reduced the amount of free ammonia and acids in the silage. The soluble carbohydrate content was higher in wilted silage than control silage with the amount in formalin silage being intermediate. MAD fibre content was lower in wilted and formalin silages.

Both wilting and formalin reduced the apparent digestibility of nitrogen, the effect being greater with formalin (Table 16). Wilting and formalin also reduced the digestibility of energy compared with the control but this was significant only for formalin. The mean intakes of DE, DM, and DN are presented in Appendix 5. Compared with control silage, the mean intakes of wilted and formalin treated silage were reduced by 6 and 15 % for DEI, and by 8 and 24 % for DNI, respectively.

TABLE 15. Chemical composition of diets.

Nutrient	C.S.	F.S.	W.S.
DM (%)	21.8 ± 0.03[a]	22.9 ± 0.03	56.7 ± 0.03
Composition of 100 g DM			
OM (g)	85.6 ± 0.47	85.5 ± 0.64	90.3 ± 0.12
MAD fibre (g)	40.0 ± 0.01	38.3 ± 0.26	35.7 ± 0.03
GE (MJ)	1.78± 0.003	1.75± 0.006	1.82± 0.010
Total N (g)	2.5 ± 0.07	2.5 ± 0.01	2.5 ± 0.01
Soluble N (g)	1.14± 0.008	0.72± 0.008	1.15± 0.006
Soluble Protein N (g)	0.01± 0.001	0.013± 0.001	0.017± 0.001
NH_3-N (g)	0.23± 0.001	0.108± 0.001	0.124± 0.001
Soluble Carbohydrate (g)	0.4 ± 0.04	1.96 ± 0.13	4.51 ± 0.16
Lactic acid (g)	5.7 ± 0.13	3.1 ± 0.09	6.8 ± 0.11
Acetic acid (g)	3.7 ± 0.04	1.5 ± 0.05	0.1 ± 0.05
Propionic acid (g)	0.6 ± 0.04	0.1 ± 0.04	0.0 ± 0.0
n-Butyric acid (g)	1.5 ± 0.05	0.0 ± 0.0	0.0 ± 0.0
Isobutyric acid (g)	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
n-Valeric acid (g)	0.0 ± 0.0	Trace	0.0 ± 0.0
Isovaleric acid (g)	Trace	Trace	0.0 ± 0.0
pH	4.3 ± 0.04	4.3 ± 0.06	5.0 ± 0.06

[a] SEM, n = 6

TABLE 16 - Mean apparent digestibility coefficients of diets.

Item	Digestibility (%) C.S.	F.S.	W.S.	SED	Sign.
GE	62.1	52.7	58.8	2.40	*
DM	60.0	53.6	60.8	2.42	+
OM	64.6	58.0	61.7	2.24	+
N	60.5	44.8	55.4	1.54	***

n = 3

Milk Yield and Composition, and Live-weight Change (Table 17).

Diet significantly affected the yields of milk, protein, lactose and milk fat percentage. Although the overall effect of diet on milk protein percentage was not significant, individual comparisons of wilted and formalin silages with control silage were significant. Levels of silage intake had no effect on milk protein percentage. There was no relationship between milk lactose percentage for either diet or feeding level.

The relationships between the milk variates and $DEI/LW^{0.75}$ (Model i) are summarized in Appendix 6. Since no significant differences were obtained between partial regression coefficients for each silage, coefficients were derived from the pooled data for each milk variate, together with the intercept for each silage (Model iii) Table 18). The significance of differences in the intercepts is indicated in Table 18. Pooled regression coefficients were significant, and positive for the yields of milk, fat, protein and lactose and negative for milk fat percentage. Regression coefficients for the percentages of protein and lactose were small and not significant.

In comparison to cows fed the control silage cows fed wilted silage at the same $DEI/LW^{0.75}$ had significantly higher yields of milk, fat, protein and lactose and a higher milk protein percentage (Table 18, Figures 10-15). Similarly at the same $DEI/LW^{0.75}$ cows fed formalin treated silage produced more fat and protein and had a higher milk fat percentage compared with cows on the control silage. No significant differences were observed in any of the milk variates between cows fed on wilted or formalin treated silages.

TABLE 17. Mean daily milk yield and milk composition (adjusted), and LW change in Period II.

Diet		Level	Milk (kg/d)	Protein (g/d)	Protein %	Fat (g/d)	Fat %	Lactose (g/d)	Lactose %	ΔLW (g/d)
C.S.		100	7.19	216	3.01	334	4.69	342	4.74	123
		75	6.40	189	3.01	292	4.70	307	4.78	103
		50	5.11	158	3.15	252	4.98	241	4.73	-597
F.S.		100	6.81	209	3.12	334	4.95	323	4.75	908
		75	5.36	173	3.23	282	5.28	250	4.65	-211
		50	4.88	161	3.21	264	5.27	231	4.67	-25
W.S.		100	7.54	242	3.27	366	4.95	354	4.69	1090
		75	5.90	185	3.24	278	4.94	278	4.70	258
		50	5.49	171	3.08	285	5.19	250	4.63	62
Diet Means	C.S.		6.23	188	3.05	293	4.79	297	4.75	-192
	F.S.		5.68	181	3.19	293	5.17	268	4.69	224
	W.S.		6.31	200	3.20	309	5.03	294	4.68	470
		SED	0.24	7.60	0.07	13.40	0.12	12.40	0.04	280
		Sign.	*	+	NS	NS	*	+	NS	+
Level Means		100	7.18	223	3.13	345	4.86	340	4.73	-707
		75	5.89	182	3.16	284	4.97	278	4.71	-19
		50	5.16	163	3.15	267	5.15	240	4.68	-187
		Sign.	***	***	NS	***	+	***	NS	*
Diets x Level		Sign.	NS	NS	NS	NS	NS	NS	NS	NS

TABLE 18. Relationships between the milk variates and DEI/$LW^{0.75}$ (MJ/kg $LW^{0.75}$/d) in Period II (Model iii).

Item	Mean	Bμ ± SE	bx ± SE	Intercepts S	Intercepts FS	Intercepts WS	R	RSD	Mean Prod. (a) S	Mean Prod. (a) FS	Mean Prod. (a) WS	Sign. of Diffs. SvFS	Sign. of Diffs. SvWS	Sign. of Diffs. FSvWS
Milk kg/d	6.08	0.81*** ± .062	2.68*** ± .380	-4.16	-3.82	-3.49	0.95	0.579	5.74	6.08	6.41	NS	*	NS
Fat g/d	298.5	0.82*** ± .073	104.7*** ± 19.92	-154.0	-120.5	-115.4	0.93	30.39	274.	308	313	*	*	NS
Protein g/d	189.4	0.73*** ± .062	81.2*** ± 11.56	-106.5	-87.2	-77.3	0.94	17.47	173	192	202	*	**	NS
Lactose g/d	286.2	0.81*** ± .066	131.6*** ± 19.39	-202.4	-186.7	-175.3	0.94	29.49	272	287	299	NS	+	NS
Fat %	4.99	0.99*** ± 0.86	-0,41* ± .150	0.90	1.12	1.03	0.93	0.23	4.88	5.10	5.01	+	NS	NS
Protein %	3.15	0.97 ± .131	-0.03 ± .097	0.15	0.26	0.28	0.84	0.15	3.06	3.18	3.20	NS	+	NS
Lactose %	4.70	0.91*** ± .089	0.07 ± .050	0.35	0.31	0.29	0.91	0.08	4.74	4.70	4.68	NS	NS	NS

(i) μ denotes production in Period I
(ii) x denotes DEI/kg $LW^{0.75}$ in Period II
(iii) overall mean DEI/kg $LW^{0.75}$ = 1.19 ± 0.33
(iv) a = adjusted mean production at overall mean DEI/$LW^{0.75}$

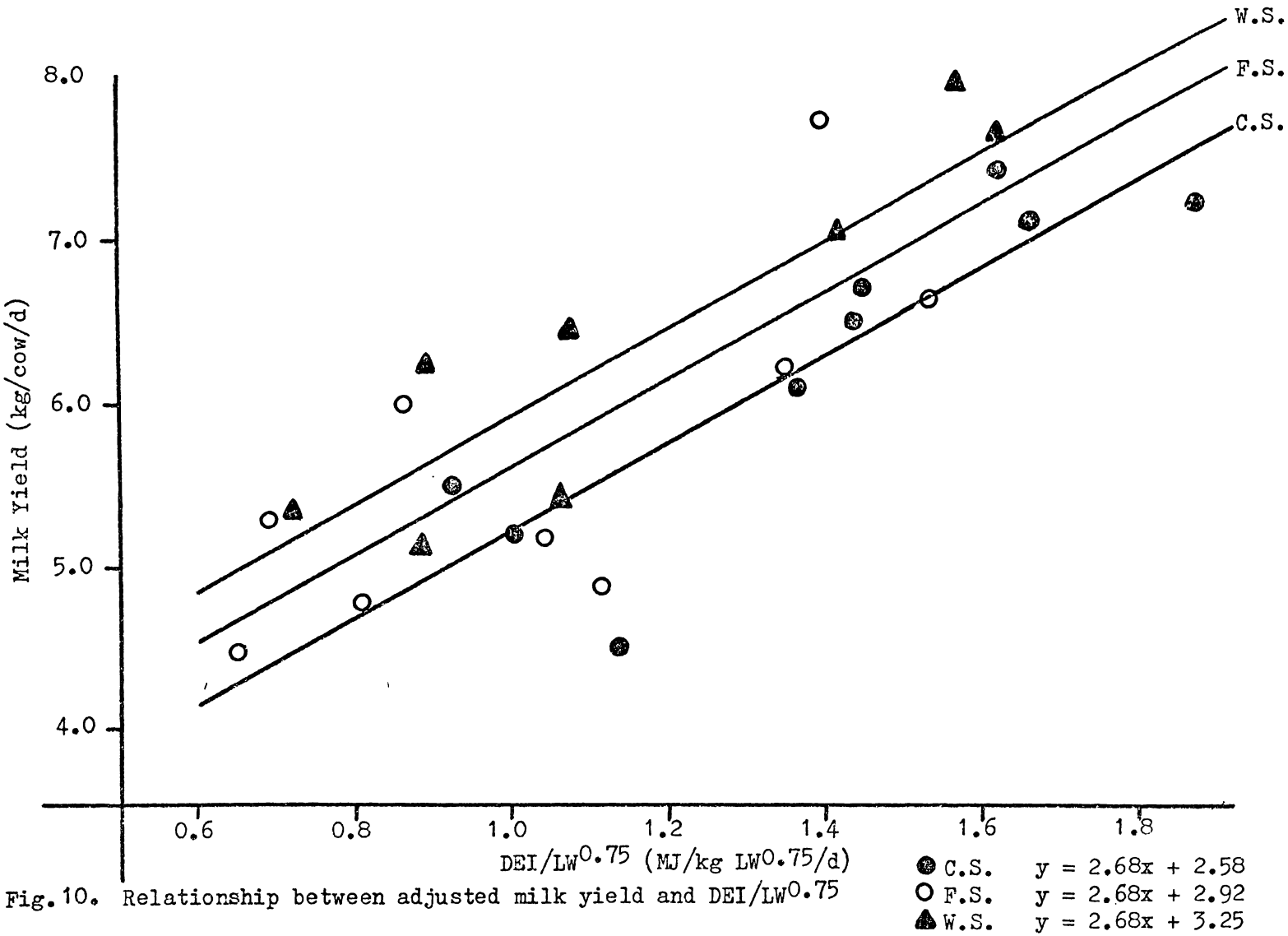


Fig. 10. Relationship between adjusted milk yield and DEI/LW$^{0.75}$

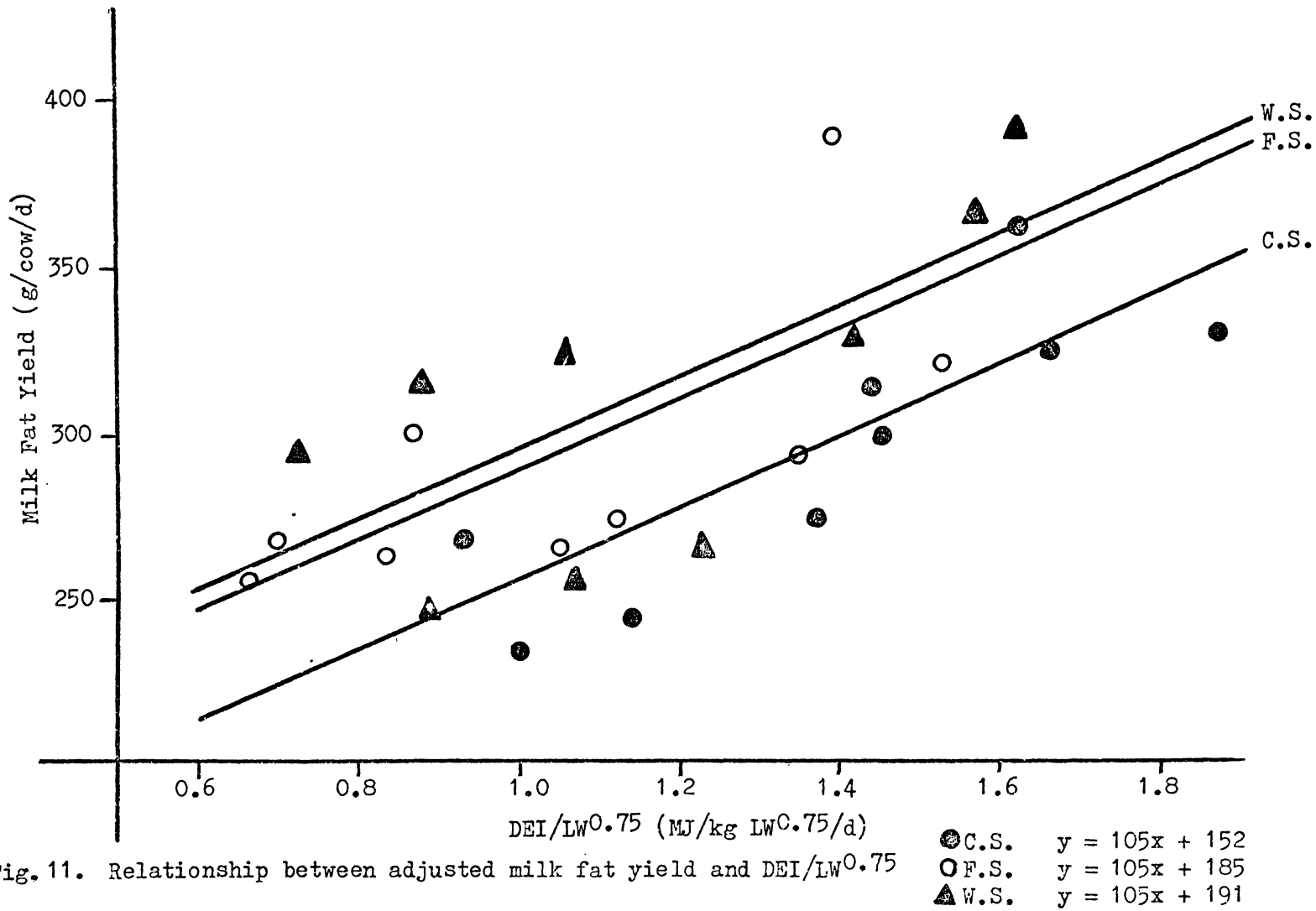


Fig. 11. Relationship between adjusted milk fat yield and DEI/LW$^{0.75}$

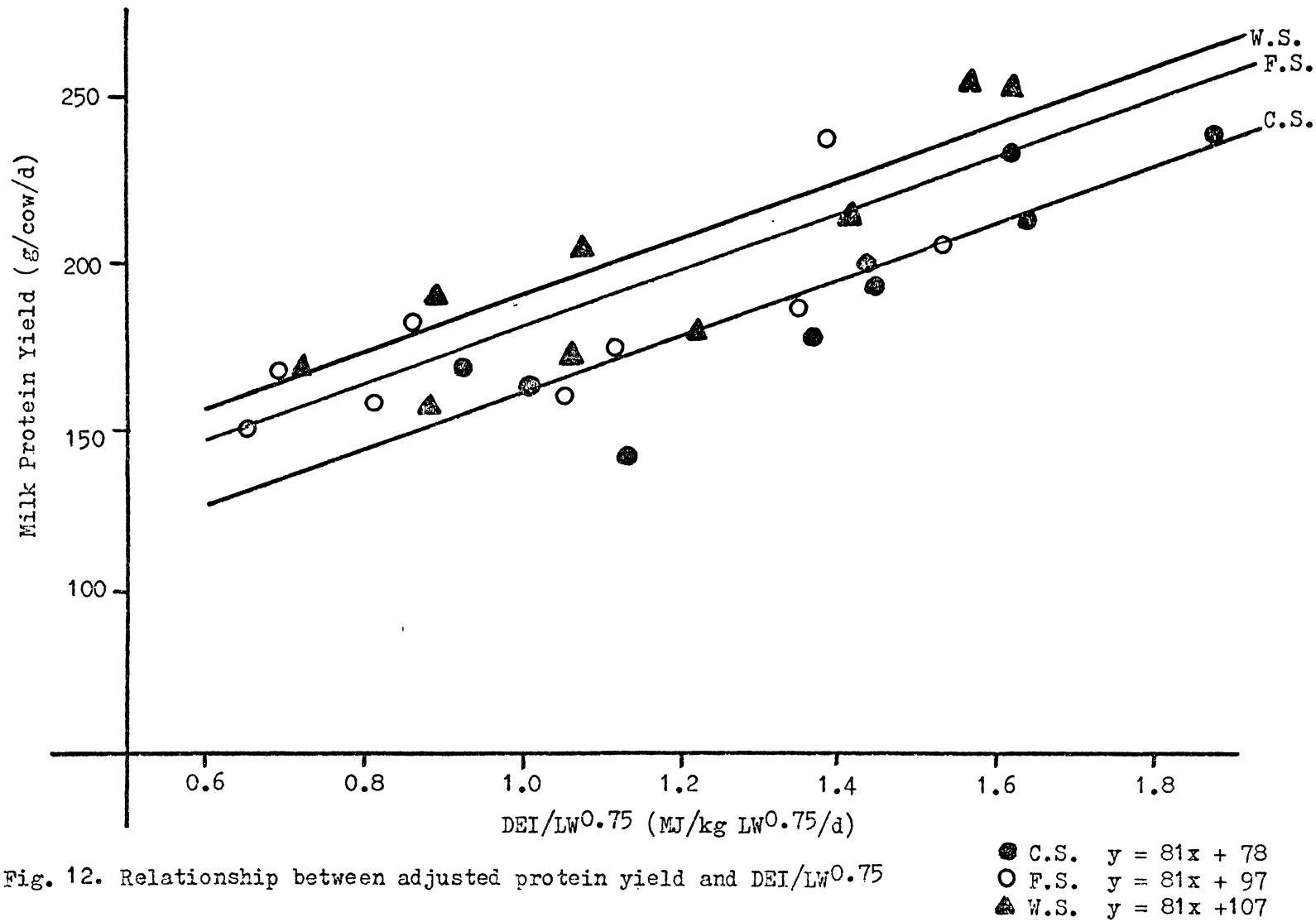


Fig. 12. Relationship between adjusted protein yield and DEI/LW$^{0.75}$

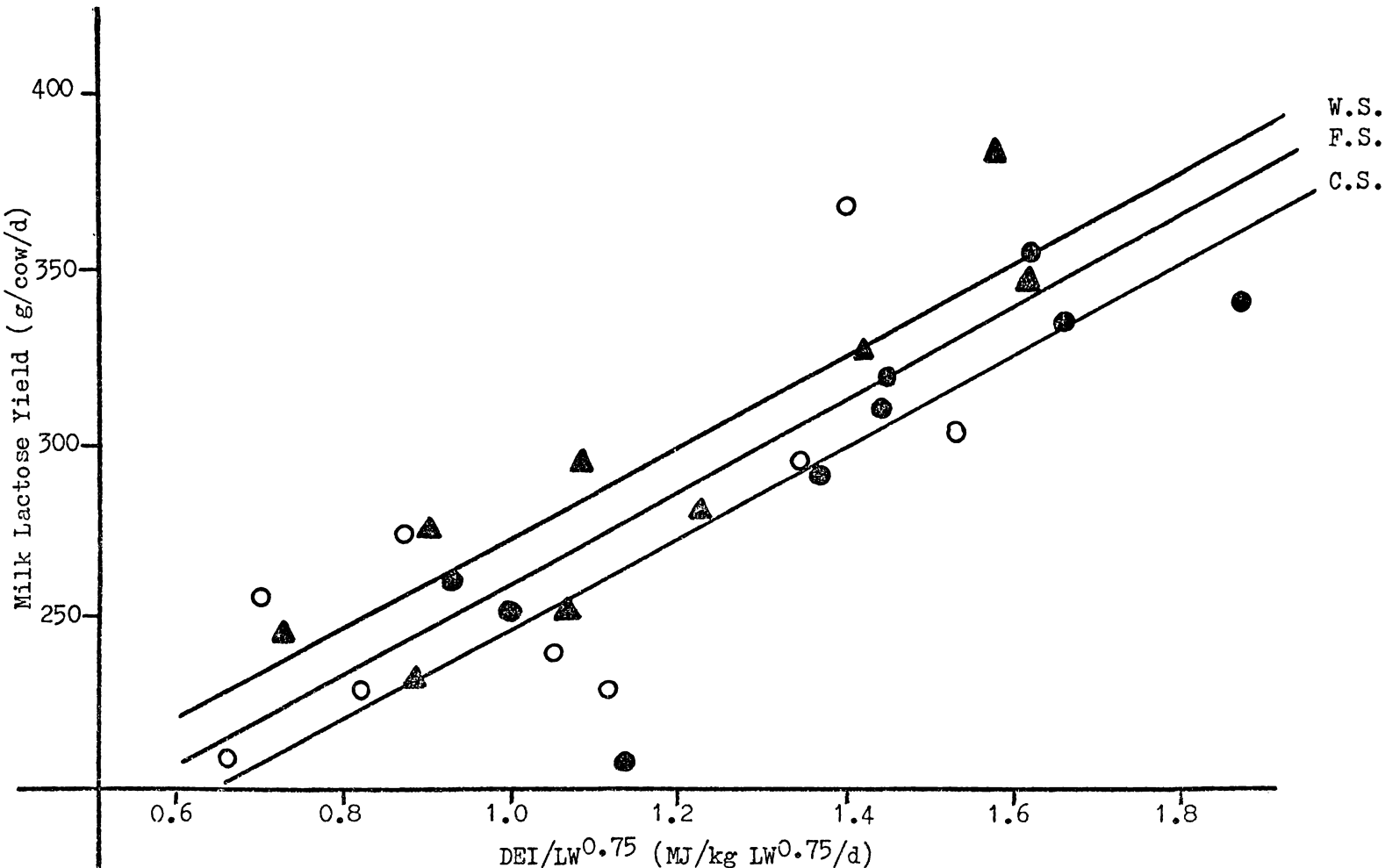


Fig. 13. Relationship between adjusted milk lactose yield and DEI/LW$^{0.75}$

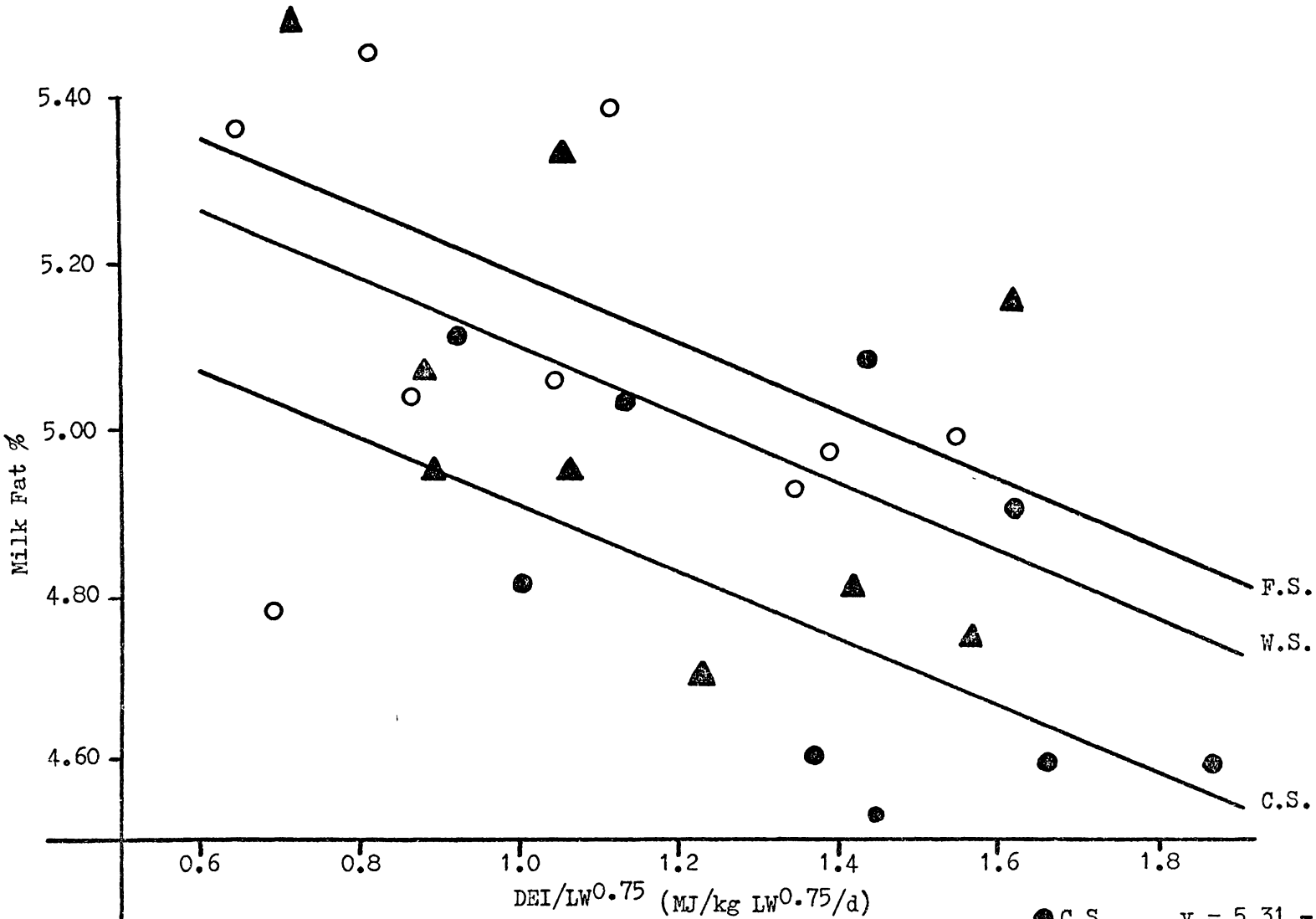


Fig. 14. Relationship between adjusted milk fat concentation and DEI/LW$^{0.75}$

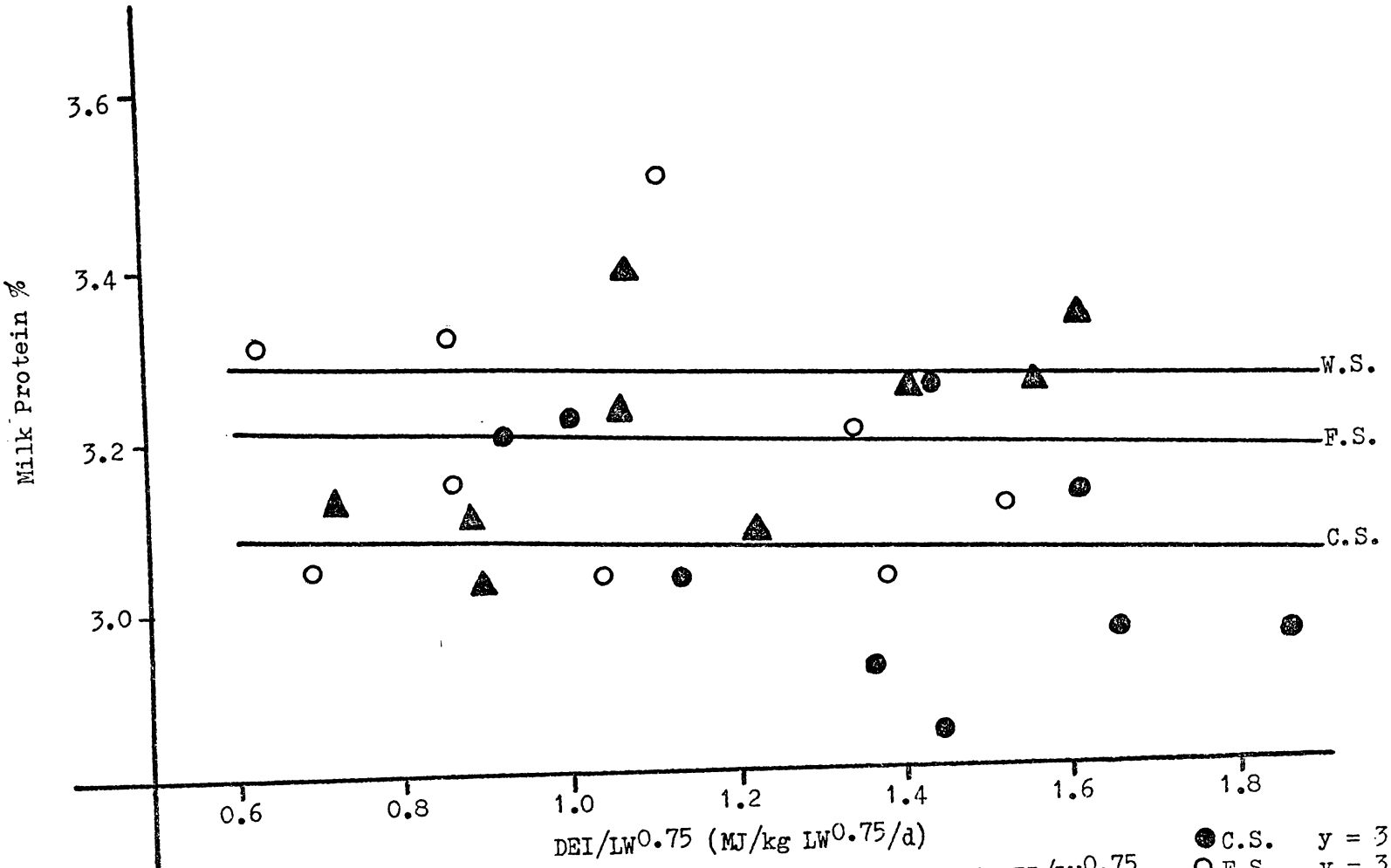


Fig. 15. Relationship between adjusted milk protein concentration and DEI/LW$^{0.75}$

Pooling the regression coefficients for the relationship between live-weight change and DEI/$LW^{0.75}$ (Appendix 7) indicated cows fed control silage lost more weight than animals on the other silages.

Rumen Volatile Fatty Acids (Table 19) - Concentrations of total and individual VFA in rumen fluid were less for cows feeding on formalin treated silage than the other rations at 3h after feeding. At 12h cows on the high level of feeding had significantly higher concentrations of VFA in rumen fluid.

Molar percentages of propionate and valerate were higher and butyrate was lower in rumen fluid of cows fed control silage compared with the other treatment rations. Higher levels of feeding were associated with increased molar proportions of propionate and valerate and decreased acetate 12h after feeding began.

Blood Metabolites (Table 20) - Diet had no effect on PCV % although 3h after feeding PCV % was higher at the lower feeding level. Before feeding, plasma urea levels were significantly higher for cows fed on control silage than wilted silage, and 3h later were higher than cows on both the other treatment rations. Although plasma levels of α amino N increased after feeding, there was no effect of diet or feeding level.

Partitioning of Dietary Nitrogen and Energy (Table 21) - Expressed as a percentage of nitrogen intake, cows fed formalin silage excreted more nitrogen in faeces, and less in urine, than animals fed control silage, with those on wilted silage being

TABLE 19. Summary of mean concentrations and molar proportions of rumen VFA at 3h and 12h after feeding commenced in Period II.

Diet/Level	Time		Conc (meq/100 ml) of VFA					Molar % of VFA			
			HAc	HPr	HBu	HVa	Total	HAc	HPr	HBu	HVa
C.S.	3 h		3.21	1.29	0.90	0.38	5.78	55.6	22.4	15.4	6.5
F.S.			2.86	1.02	0.90	0.22	5.01	57.2	20.4	17.8	4.4
W.S.			3.44	1.30	1.16	0.30	6.18	55.7	20.6	18.7	4.8
		SED	0.21	0.08	0.08	0.03	0.38	1.02	0.46	0.76	0.44
		Sign.	*	**	*	**	*	NS	**	**	**
Level 100 %			3.25	0.99	1.32	0.29	5.74	55.6	21.1	17.1	4.9
50 %			3.09	0.98	1.23	0.31	5.56	55.5	21.2	17.6	5.5
		SED	0.17	0.07	0.06	0.02	0.31	0.83	0.38	0.62	0.36
		Sign.	NS	NS	NS	NS	NS	NS	NS	NS	NS
C.S.	12 h		3.09	1.05	0.74	0.16	5.01	61.9	20.7	14.6	3.1
F.S.			3.22	1.01	0.86	0.11	5.19	62.0	19.4	16.4	2.1
W.S.			3.46	1.10	0.88	0.13	5.55	62.6	19.7	15.5	2.2
		SED	0.22	0.08	0.08	0.02	0.37	0.88	0.44	0.82	0.27
		Sign.	NS	NS	NS	*	NS	NS	*	*	**
Level 100 %			3.44	1.19	0.94	0.17	5.74	59.8	20.8	16.3	3.0
50 %			3.07	0.91	0.71	0.09	4.75	64.5	19.0	14.8	1.9
		SED	0.18	0.07	0.07	0.01	0.30	0.72	0.35	0.67	0.22
		Sign.	+	**	**	***	**	***	***	NS	***

TABLE 20. Effects of diet and level of feeding on PCV %, urea and ∝ amino N concentrations in plasma immediately prior to feeding (0h) and 3h later (adjusted for Period I).

Diet/Level		PCV %		Plasma Urea (mg/100 ml)		Plasma Amino N (mg/l)	
		0 h	3 h	0 h	3 h	0 h	3 h
C.S.		32.8	33.4	12.5	17.3	31.3	36.9
F.S.		33.4	32.9	10.8	14.1	33.9	37.2
W.S.		32.0	32.0	10.4	15.8	34.6	40.0
	SED	1.63	1.52	0.79	0.99	2.3	1.76
	Sign.	NS	NS	*	*	NS	NS
Level 100 %		31.7	30.7	9.2	15.9	32.7	38.8
50 %		33.8	34.9	11.6	15.6	33.8	37.2
	SED	1.25	0.97	0.66	0.82	1.98	1.46
	Sign.	NS	**	NS	NS	NS	NS

TABLE 21. Effect of diet on partitioning of nitrogen and gross energy.

Item	C.S.	F.S.	W.S.	SED	Sign.
N, % of intake					
Faeces	39.4	55.2	43.6	1.23	***
Urine	45.3	30.9	41.7	0.59	***
Milk	13.5	12.1	14.3	1.56	NS
Retention (NR)	1.8	1.8	0.5	1.95	NS
Milk + NR	15.3	13.7	14.7	1.11	NS
N, % of digested					
Urine	74.9	69.2	73.8	1.89	+
Milk	22.3	27.1	25.3	2.90	NS
Milk + NR	25.1	30.5	26.1	1.72	+
GE, % of intake					
Faeces	37.9	46.8	41.4	2.46	*
Urine	5.1	3.5	3.8	0.19	**
Methane (est)	7.8	7.5	7.8	0.18	NS
Milk	12.7	12.3	12.5	1.71	NS
Residual (est)	36.4	29.9	34.5	3.90	NS
Milk + Residual	49.2	42.2	46.9	2.54	+
GE, % of digested					
Urine	8.2	6.6	6.5	0.52	*
Milk	20.8	23.1	21.3	3.33	NS
Milk and Residual	79.1	79.3	80.0	1.03	NS
Metabolisable Energy (MJ/kg DM)	8.8	7.4	8.8	0.45	+

intermediate. Nitrogen retained in milk and tissue as a percentage of digested nitrogen was higher, whereas that in urine was lower, for the animals fed formalin treated silage compared with the other rations. Milk N as a percentage of DN, was higher for formalin and wilted silages compared with unwilted control silage, but was not significant.

Excretion of energy, as a percentage of energy intake, by cows fed formalin treated silage was higher in faeces and lower in urine than cows on the control silage, the wilted silage treatment being intermediate. Energy excreted in urine, as a percentage of digested energy was higher for the control ration than the other rations.

The overall ME content of the formalin ration was estimated to be 7.4 MJ/kg DM, which was less than the 8.8 MJ/kg DM of the other silages.

Discussion

In Experiment 3.1 it was shown that milk production of cows offered ensiled herbage was lower than on fresh herbage at similar intakes of DE. The present study unequivocally demonstrates that different silages differ in their effects on milk production. In comparison to cows fed on control unwilted silage at the same intake of DE, cows fed wilted silage produced more milk, fat, protein and lactose and had a higher protein concentration, whilst cows fed formalin treated silage produced similar quantities of milk but higher yields of fat and protein, and a higher milk fat concentration. Whether the effects observed with the wilted silage were due to the wilting or to the precision chopping is of

no importance in the present instance. As in the pasture silage comparison (Experiment 3.1) diet type had no effect on lactose concentration.

The level of DE intake of the silages was not related to protein percentage which is consistent with Experiment 3.1 and confirms the inferences of Hutton et al. (1971) and Lancaster et al. (1974). This result is in contrast to that achieved with increased pasture intakes which increased milk protein percentage (Experiment 3.1). Milk lactose percentage was not affected by the DEI of the silages but milk fat percentage was negatively related to the level of silage intake. These results are also consistent with those observed in Experiment 3.1 which were discussed previously on p.83-84.

Differences between the rations in the concentrations and molar proportions of VFA in rumen fluid may reflect possible quantitative and qualitative differences in energy absorption from the rumen (Leng, 1970b). The lower milk fat and milk protein contents of cows fed unwilted silage co incided with an increased molar percentage of propionate, decreased butyrate, and a decrease in the molar ratio of acetate to propionate. Again, these results are consistent with those observed in Experiment 3.1 and were discussed in that section (p.80). It is suggested that since butyrate is a precursor of B-hydroxy butyrate (Smith, McCarthy and Rook, 1974) the reduction in the proportion of butyrate may contribute to the lower milk fat synthesis. It is also apparent that factors other than the molar proportion of propionate may be more important determinants of milk protein synthesis.

Although the gross energy content of the silages were similar, differences occurred in the partitioning of energy between the silages. Cows fed formalin silage excreted more energy in faeces and less urine than cows fed control silage suggesting rumen digestion was more extensive for the latter. Higher levels of valerate in rumen liquor of these animals would also suggest a more extensive rumen degradation of the protein in control silage (el-Shazly, 1952). Increased urinary energy losses of these cows may therefore be due to increased losses of nitrogen from the rumen. The significantly higher solubility of nitrogen in control silage in comparison to formalin silage, and the higher levels of blood urea nitrogen for animals fed control silage would support this possibility. However this increase in both urinary energy excretion and blood urea nitrogen levels shown by the cows fed the control silage may be associated in part with an increased tissue loss since these animals showed the greatest loss in live weight.

Nitrogen contents of the rations were similar, but there were marked differences in the partitioning of dietary nitrogen by the animal. Formalin treatment reduced the digestibility of dietary nitrogen, caused a marked decrease in ammonia content, indicating reduced proteolysis during ensiling, and a fall in the solubility of the nitrogen. Formaldehyde apparently bonds with dietary protein inhibiting proteolysis both during ensiling and in the rumen (Barry, 1976; Wilkinson, Wilson and Barry, 1976). Beever et al. (1974) indicated that when silage is treated with formalin, substantial quantities of dietary protein escape rumen degradation to increase the amount of protein entering the duodenum. Such a mechanism may therefore account for the increase

in milk protein yields of cows on formalin silage. If present, this effect was probably much less for the wilted silage since the effect of wilting on nitrogen digestibility was less pronounced than formalin. Also, wilting did not alter the solubility of the nitrogen but it did reduce the ammonia level in the silage indicating that although proteolysis may have been as extensive as in the control silage, deamination was less.

Since microbial use of ammonia is limited by the availability of energy (Smith, 1969), the higher soluble carbohydrate content of wilted silage may enhance microbial protein synthesis and thereby reduce the loss of dietary nitrogen as ammonia absorbed from the rumen (Armstrong, 1974). Reduced plasma urea levels of cows fed wilted silage compared with the control animals support this possibility. An increased flow of protein from the rumen as a result of reduced losses of dietary nitrogen from the rumen and increased microbial protein synthesis may account for the increased yields of milk protein of cows fed wilted silage.

Despite the possibilities that formalin and wilted silages may result in increased quantities of protein entering the small intestine, increased uptakes of amino acids by animals on these diets could not be substantiated by plasma α amino N concentrations. PCV % was also not altered by the silage rations suggesting that changes in milk composition and PCV % were unrelated.

In summary this experiment has indicated that milk yield and composition can be changed by modifying the fermentation of silage. Degradation of dietary protein and/or soluble carbohydrates during ensiling appear to be major factors affecting nitrogen utilization

and milk protein synthesis. The next experiment therefore examines the effect on yield and composition of milk of cows fed unwilted silage and given protein or energy supplements.

3.3 THE EFFECTS OF FEEDING SUPPLEMENTS WITH PASTURE SILAGE ON MILK YIELD AND COMPOSITION

Introduction

In the previous experiment (3.2) attention was drawn to the possibility that the effects of unwilted silage on milk yield and composition may be associated with reduced amounts of protein entering the duodenum as a result of low dietary true protein in silage and reduced utilisation of ammonia for microbial protein synthesis.

Other experiments have shown that yield and composition of milk from cows can be altered by feeding supplements with silage such as pasture (Bryant and Donnelly, 1974; Hutton, 1975), energy concentrates (Murdoch, 1962; Griffiths and Crowley, 1973), or mixtures of energy and protein concentrates or dried grass (Murdoch, 1962; Castle and Watson, 1969, 1974, 1975; Butler, 1973, 1974; Griffiths and Crowley, 1973).

In many of these reports the specific effects of the supplements were confounded by changes in silage and total intake and/or changes in proportions and levels of concentrates fed. Furthermore the effects of the supplements on milk yield and composition have not always been compared with those from cows offered silage as a sole ration.

This section describes two experiments that were carried out to obtain more information on factors affecting yield and composition of milk of cows fed silage. In the first experiment, silage was supplemented with pasture or maize silage, and in the second experiment with protein concentrates.

Materials and Methods

Experiment 3.3.1

General Design - Twenty seven Jersey cross-bred cows approximately 5-7 weeks postpartum were removed from grazing and individually fed in stalls for 10 days on a mixture of pasture silage, maize silage and pasture (1/1/1) for a period of 10 days (Period I). Cows were assigned to nine equal groups on the basis of their food intake, LW, yield and composition of milk, during the last 7 days of Period I. Unwilted pasture silage, pasture silage plus maize silage (50/50) and pasture silage plus pasture (50/50) were each offered at three levels of intake. The feeding levels imposed were ad libitum or either 75 or 50 % of the amount eaten by the individual animals during the last 7 days of Period I. The treatment rations were offered over the next 21 days (Period II), after which two cows from each of the 75 and 100 % feeding levels of each ration were housed for 10 days in metabolism stalls for nitrogen and energy digestion trials.

Preparation of Foods - The pasture silage was made with a flail harvester from a predominantly ryegrass (Lolium perenne)/white clover (Trifolium repens) pasture in spring, 1974. The maize silage was harvested at the dent stage with a precision chop harvester in the autumn, 1975. Both silages were ensiled in bunkers, and covered with polythene film after consolidation. The pasture herbage, also predominantly ryegrass and white clover, was harvested in the autumn with a rotary mower and stored at -20 °C. The silages were removed from the stacks immediately prior to the experiment and stored at -20 °C.

Feeding, Milking and Weighing - Feeding was from 0800 to 1300 h and 1600 to 2100 h. Sufficient of the foods were removed from the freezer each afternoon and allowed to thaw over-night. During Period I the foods were fed separately to supply one third of the ration. All cows in Period I and those cows fully fed in Period II were offered 115 % of the previous day's estimated dry matter intake. The cows on mixed rations were offered the rations alternately every 2 hours. After the first week of Period II the amount of food offered to the animals on the restricted feeding levels was adjusted according to the percentage change in $DMI/LW^{0.75}$ of the fully fed cows for each ration in relation to intakes in Period I. The animals were confined in a loafing barn when not being fed or milked during Periods I and II.

In the digestion trial cows were offered the same quantity of food with the same feeding routine as in Period II.

Procedures for milking and weighing cows, and sampling milk were similar to Experiment 3.1 except that milk samples were bulked over three successive periods of 2, 3, and 2 days each week for analysis.

Sampling of Foods, Residues, Faeces, and Urine

Sampling procedures were as outlined in Experiment 3.1.

Blood and Rumen Fluid - Samples of blood and rumen fluid were obtained from the cows in the 100 % and 50 % feeding levels of each ration. Blood was collected on the last day of Period I and II immediately prior to feeding (0 h) and 4 h later. Rumen fluid was sampled at 0 h on the last day of Period I and at 0 h, 4 h, and 12 h, on the last day of Period II.

Analytical and Statistical Analyses - Procedures followed were as described in Experiment 3.1. Effects of treatments on milk yield and composition were also compared by analysis of variance (Snedecor and Cochrane, 1967).

Experiment 3.3.2

Treatments - Pasture silage fed as a sole ration was contrasted with two rations of pasture silage supplemented with a protein concentrate to provide either 8.5 % (PC_1) or 17 % (PC_2) of the total dry matter offered. Each of the three rations were offered at three levels of intake; ad libitum or 75 % and 50 % of the individual animal's DM intake of silage over the 7 days immediately prior to the treatment period.

Foods - The silage was cut from a predominantly ryegrass (Lolium perenne) / white clover (Trifolium repens) pasture with a forage harvester in spring, 1975, and ensiled in a bunker and covered with polythene film. The protein concentrate consisted on a dry matter basis of 60 % soyabean meal (Solvent extracted), 15 % meat meal (dehydrated), 15 % fish meal (dehydrated) and 10 % linseed meal (pressed anddehydrated).

Design - Twenty seven spring calving Jersey crossbred cows approximately 5-7 weeks post-partum and accustomed to silage, were removed from grazing and individually fed in stalls on silage for 10 days (Period I). The cows were assigned to nine equal groups on the basis of their food intake, milk yield and composition, and live weight during the last 7 days of Period I. The treatment

rations were offered over the next 21 days (Period II), after which two cows from the 100 and 75 % feeding levels of each ration were housed in metabolism stalls for 10 days for nitrogen and energy digestion trials.

Feeding, Milking and Weighing - Feeding routines were similar to Experiment 3.1. Each morning silage was collected from the bunker and fed fresh. The appropriate quantities of concentrate mixture were mixed with the silage each morning before being offered.

Milking and weighing procedures were as for Experiment 3.1.

Sampling of Feeds and Residues - Sampling procedures followed were as in Experiment 3.1 except that in the experimental period food residues of the supplemented rations within feeding levels were aggregated.

Sampling of Blood and Rumen Liquor - Samples of blood and rumen liquor were obtained from the animals in the highest and lowest feeding levels on each treatment ration on the last day of Period I and Period II. Blood and rumen liquor collection and treatment were the same as for Experiment 3.1.

Analytical and Statistical Analysis - The procedures followed were as described for Experiment 3.3.1.

Results

Experiment 3.3.1

Diet Composition (Table 22) - Relative to maize silage, the autumn pasture was characterized by high levels of nitrogen and soluble carbohydrates. In comparison, pasture silage had low amounts of soluble carbohydrate and high concentrations of soluble nitrogen and free ammonia.

Digestibility and Intake (Table 23, Appendix 8) - Supplementing pasture silage with maize silage significantly reduced the apparent digestibilities of gross energy and nitrogen, whereas supplementing with pasture had no effect on the digestibility of gross energy and only slightly increased the digestibility of nitrogen (Table 23). Digestibility was not significantly affected by feeding level. The mean intakes of DE, DM, and DN are presented in Appendix 8. Compared with silage, the mean intake of DE and DN of silage plus pasture was increased by 18 and 48 %, respectively, whereas the mean intake of silage plus maize silage was decreased by 10 % for DE and by 40 % for DN.

Milk Yield and Composition, and Live-weight Change (Table 24) - Feeding level significantly affected the yields and composition of all milk constituents and significant differences between diets were obtained for all milk variates excepting lactose and milk fat concentrations.

The relationships between the milk variates and $DEI/LW^{0.75}$ (Model i) are summarized in Appendix 9. Differences in the partial regression coefficients (b), were not significant. The same

TABLE 22. Chemical composition of forages.

Nutrient	S	P	M.S.
DM (%)	18.2 ± 0.38[a]	15.5 ± 0.85	35.6 ± 0.36
Composition of 100 g DM			
OM (g)	86.9 ± 1.26	90.3 ± 0.72	95.3 ± 0.08
MAD fibre (g)	53.9 ± 1.83	45.0 ± 0.56	52.0 ± 2.60
GE (MJ)	1.86± 0.014	1.84± 0.014	1.82± 0.004
Total N (g)	2.9 ± 0.12	3.7 ± 0.08	1.1 ± 0.02
Soluble N (g)	1.5 ± 0.08	1.4 ± 0.02	0.5 ± 0.01
Soluble Protein N (g)	0.01± 0.008	0.01± 0.001	0.006± 0.001
NH_3-N (g)	0.49± 0.016	0.10± 0.01	0.19± 0.008
Soluble Carbohydrate (g)	1.4 ± 0.04	18.7 ± 0.34	0.9 ± 0.04
Lactic acid (g)	1.0 ± 0.37	Trace	2.7 ± 0.12
Acetic acid (g)	3.2 ± 0.09	Trace	1.2 ± 0.13
Propionic acid (g)	0.5 ± 0.03	0.0 ± 0.0	0.05± 0.02
n-Butyric acid (g)	1.0 ± 0.05	0.0 ± 0.0	Trace
Isobutyric acid (g)	0.3 ± 0.09	Trace	Trace
n-Valeric acid (g)	0.2 ± 0.05	0.0 ± 0.0	Trace
Isovaleric acid (g)	0.3 ± 0.02	0.0 ± 0.0	Trace
pH	4.90 ± 0.08	5.80± 0.08	4.20± 0.04

[a] SEM, n = 6

TABLE 23 - Mean apparent digestibility coefficients of diets.

Item	Digestibility (%)			SED	Sign.
	S	S + P	S + MS		
GE	72.3	73.4	67.1	0.97	**
DM	72.2	73.8	67.3	0.90	***
OM	71.8	72.9	67.7	0.99	**
N	65.6	68.8	57.5	0.85	***

n = 4

TABLE 24. Mean daily milk yield and composition (adjusted), and LW change in Period II.

Diet	Level	Milk (kg/d)	Protein (g/d)	Protein %	Fat (g/d)	Fat %	Lactose (g/d)	Lactose %	ΔLW (g/d)
S	100	10.2	272	2.73	417	4.07	503	4.92	-0.431
	75	9.8	250	2.59	415	4.08	477	4.88	-1.478
	50	7.0	184	2.65	334	4.87	333	4.77	-1.979
S + P	100	11.8	350	2.96	497	3.85	578	4.97	-0.318
	75	10.7	308	2.97	481	4.73	523	4.89	-0.309
	50	8.9	246	2.80	440	5.11	432	4.83	-1.291
S + MS	100	9.8	271	2.86	441	4.81	476	4.86	-0.274
	75	9.0	236	2.65	371	4.12	448	4.97	-1.175
	50	7.8	199	2.61	354	4.80	368	4.75	-1.644
Diet Means	S	9.0	236	2.65	388	4.34	438	4.86	-1.296
	S+P	10.5	301	2.91	473	4.56	511	4.90	-0.640
	S+MS	8.9	235	2.70	389	4.58	431	4.86	-1.031
	SED	0.32	7.8	0.048	18.6	0.224	17.2	0.047	0.2575
	Sign.	***	***	**	***	NS	***	NS	+
Level Means	100	10.6	298	2.85	452	4.24	519	4.92	-0.341
	75	9.8	265	2.74	422	4.31	483	4.92	-0.988
	50	7.9	210	2.69	376	4.93	378	4.79	-1.638
	Sign.	***	***	**	**	*	***	**	***
Diets x Levels	Sign.	NS	NS	NS	NS	+	NS	NS	NS

regression (Model iv) could not be used to describe the data since there was a significant reduction in the residual sums of squares from Model (iv) to Models (ii) or (iii) (see Appendix 10). Model (iii) was used to describe the data (Table 25, Figures 16-21), since it accounted for more of the variation than Model (ii). The significance of the intercepts is indicated in Table 25. Pooled regression coefficients were significant for all milk variates and except for milk fat percentage were positive.

The regression analyses indicate that at the same $DEI/LW^{0.75}$ supplementing pasture silage with pasture increased the yields of milk, fat, protein and lactose, and the fat and protein concentrations in milk, whereas supplementing silage with maize silage had no effect. Neither supplement had any effect on milk lactose percentage. These results largely agree with those of the analysis of variance (Table 24).

Relative to intake, no significant differences were observed in live-weight change of cows on the rations (Appendix 11).

Volatile Fatty Acids in Rumen Fluid - Since the mean effects of diet and feeding level were generally similar at each sampling period the results (Appendix 12) have been bulked and presented in Table 26. Differences in the molar percentages of VFA in rumen fluid of cows on the treatment diets were small and largely non-significant. Addition of pasture to silage increased the molar percentage of n-butyrate in rumen fluid. Reducing the feeding level significantly increased the molar concentrations of iso-butyrate and iso and n-valerate.

TABLE 25. Relationships between milk variates and DEI/$LW^{0.75}$ (MJ/kg $LW^{0.75}$/d) in Period II (Model iii).

Item	Mean	Bμ ± SE	bx ± SE	Intercepts S	Intercepts S+P	Intercepts S+MS	R	RSD	Mean Prod. (a) S	Mean Prod. (a) S+P	Mean Prod. (a) S+MS	Sign. of Diffs. S v. S+P	Sign. of Diffs. Sv. S+MS	Sign. of Diffs. S +Pv. S+MS
Milk kg/d	9.44	0.65*** ± 0.04	3.85*** ± 0.45	-2.58	-1.88	-2.37	0.96	0.67	9.14	9.83	9.35	*	NS	NS
Fat g/d	417	0.54*** ± 0.07	102.0** ± 27.7	-0.1	63.0	8.8	0.88	39.55	392	456	401	**	NS	*
Protein g/d	257	0.52*** ± 0.05	131.1*** ± 11.5	-88.1	-49.9	-76.0	0.96	16.62	241	279	253	***	NS	**
Lactose g/d	460	0.64*** ± 0.05	197.1*** ± 24.4	-130.1	-98.2	-118.4	0.95	36.46	445	477	457	+	NS	NS
Fat %	4.49	1.07*** ± 0.17	-0.97* ± 0.34	0.42	0.84	0.52	0.82	0.48	4.32	4.74	4.42	+	NS	NS
Protein %	2.76	0.83*** ± 0.07	0.23** ± 0.07	-0.23	-0.01	-0.15	0.94	0.10	2.66	2.87	2.74	***	NS	*
Lactose %	4.87	0.97*** ± 0.11	0.16* ± 0.07	-0.03	-0.02	-0.01	0.89	0.10	4.86	4.87	4.88	NS	NS	NS

(i) μ denotes production in Period I
(ii) x denotes DEI/kg $LW^{0.75}$ in Period II
(iii) overall mean DEI/kg $LW^{0.75}$ = 1.08 ± 0.32
(iv) a = adjusted mean production at overall mean DEI/$LW^{0.75}$

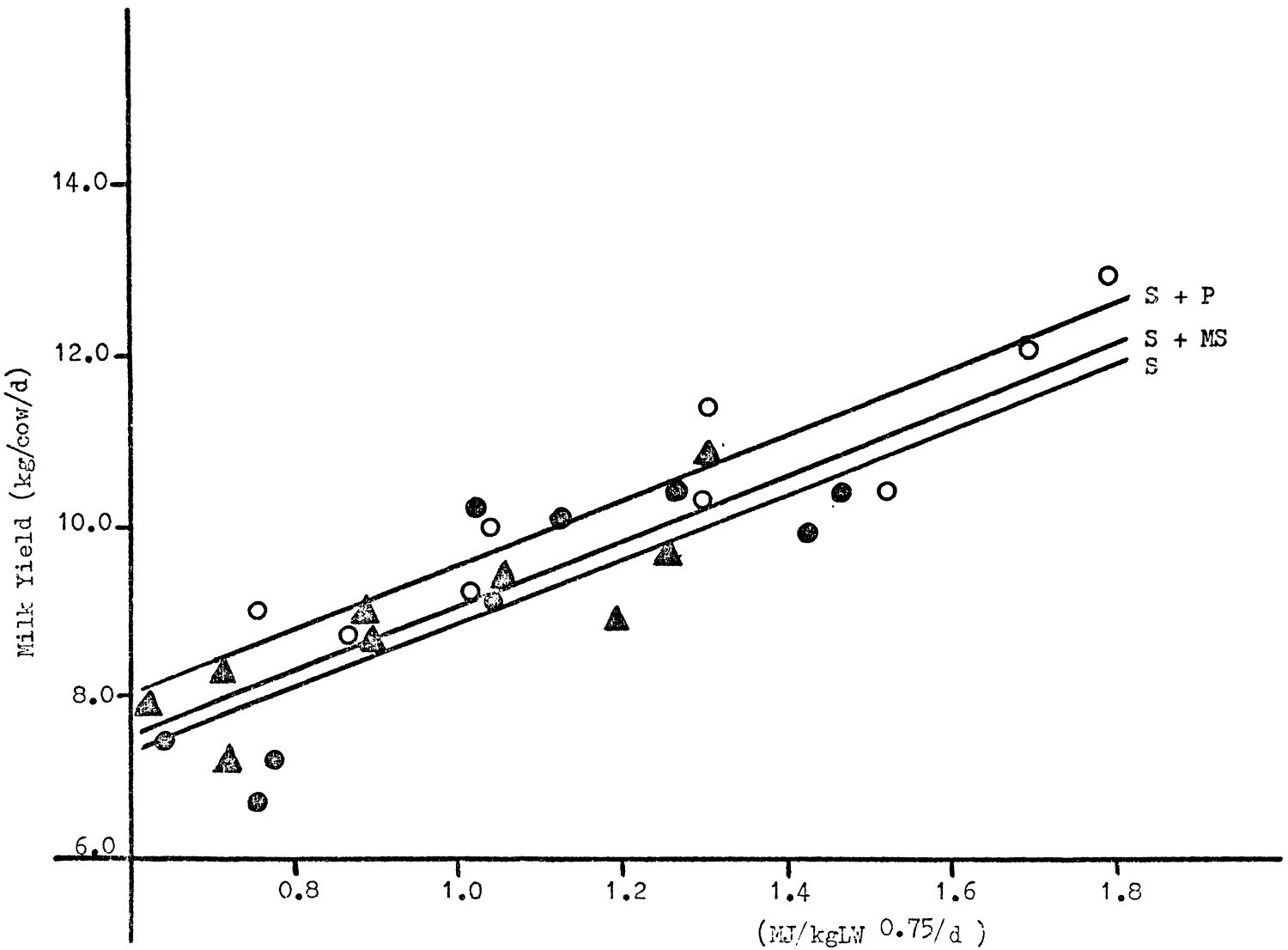


Fig. 16 Relationship between adjusted milk yield and DEI/LW$^{0.75}$

● S
○ S + P
▲ S + MS

y = 3.85x + 4.96
y = 3.85x + 5.66
y = 3.85x + 5.17

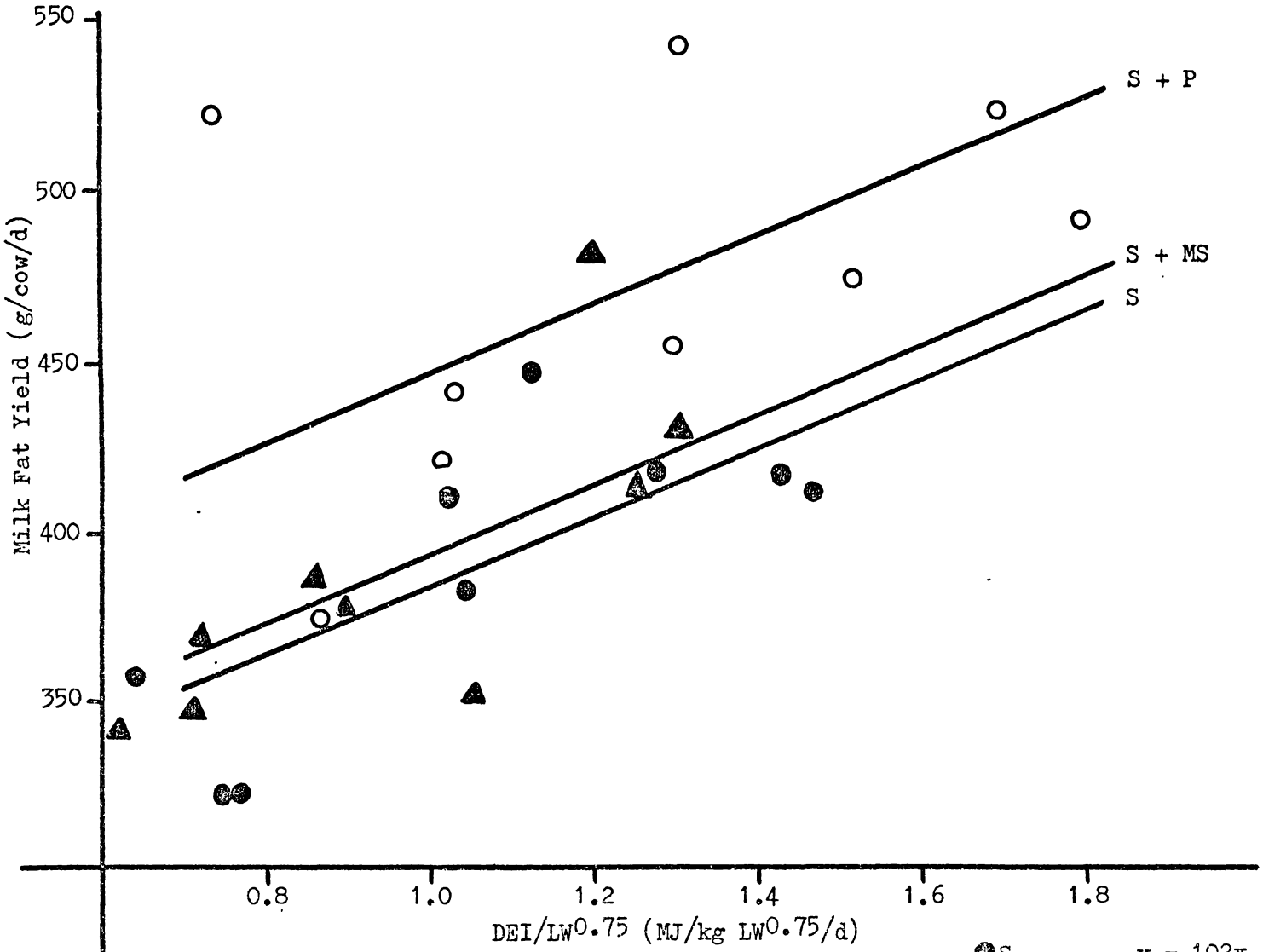


Fig. 17 Relationship between adjusted milk fat yield and DEI/LW$^{0.75}$

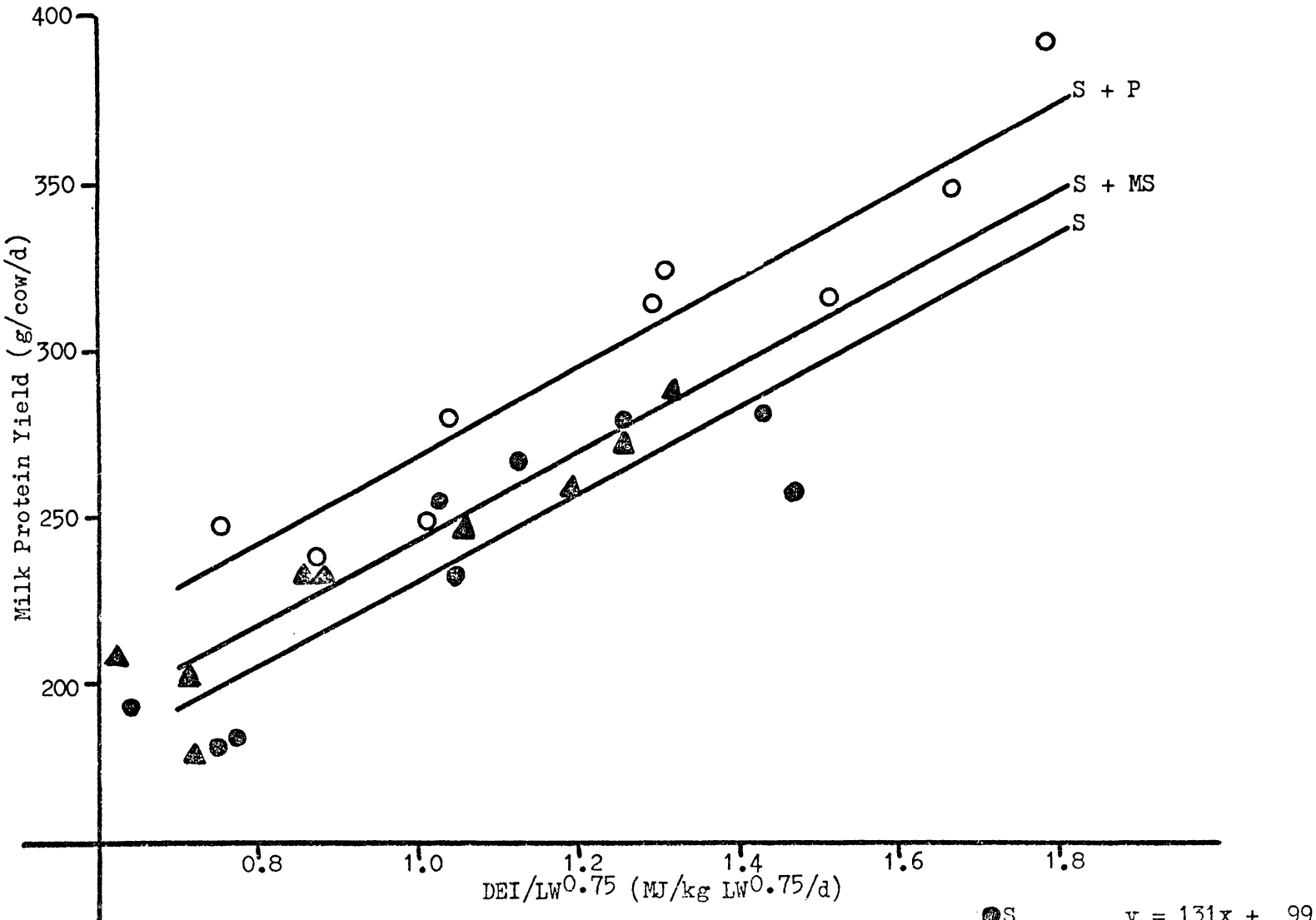


Fig.18 Relationship between adjusted milk protein yield and DEI/$LW^{0.75}$

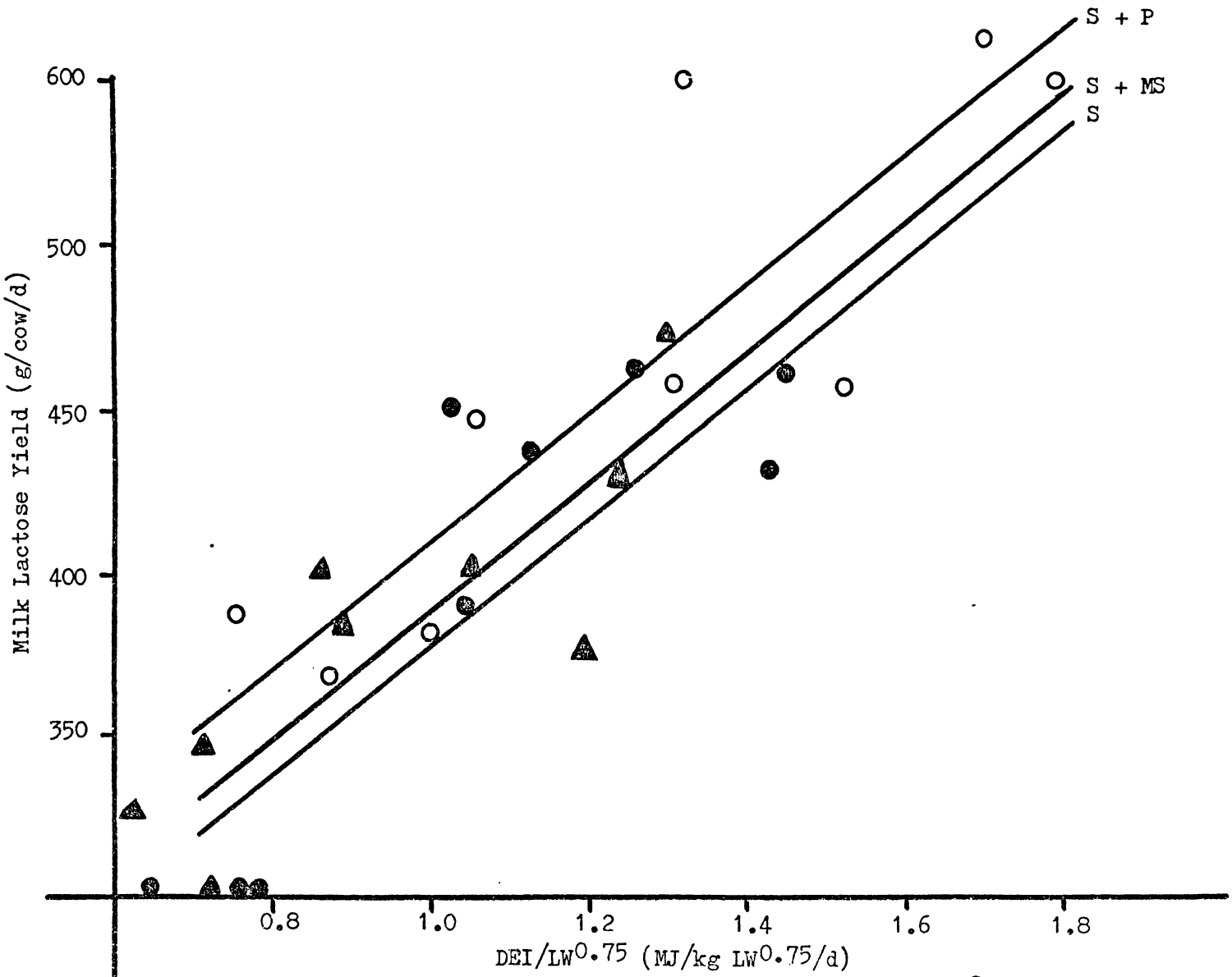


Fig. 19. Relationship between adjusted milk lactose yield and DEI/LW$^{0.75}$

S	y = 197x + 232
S + P	y = 197x + 263
S + MS	y = 197x + 243

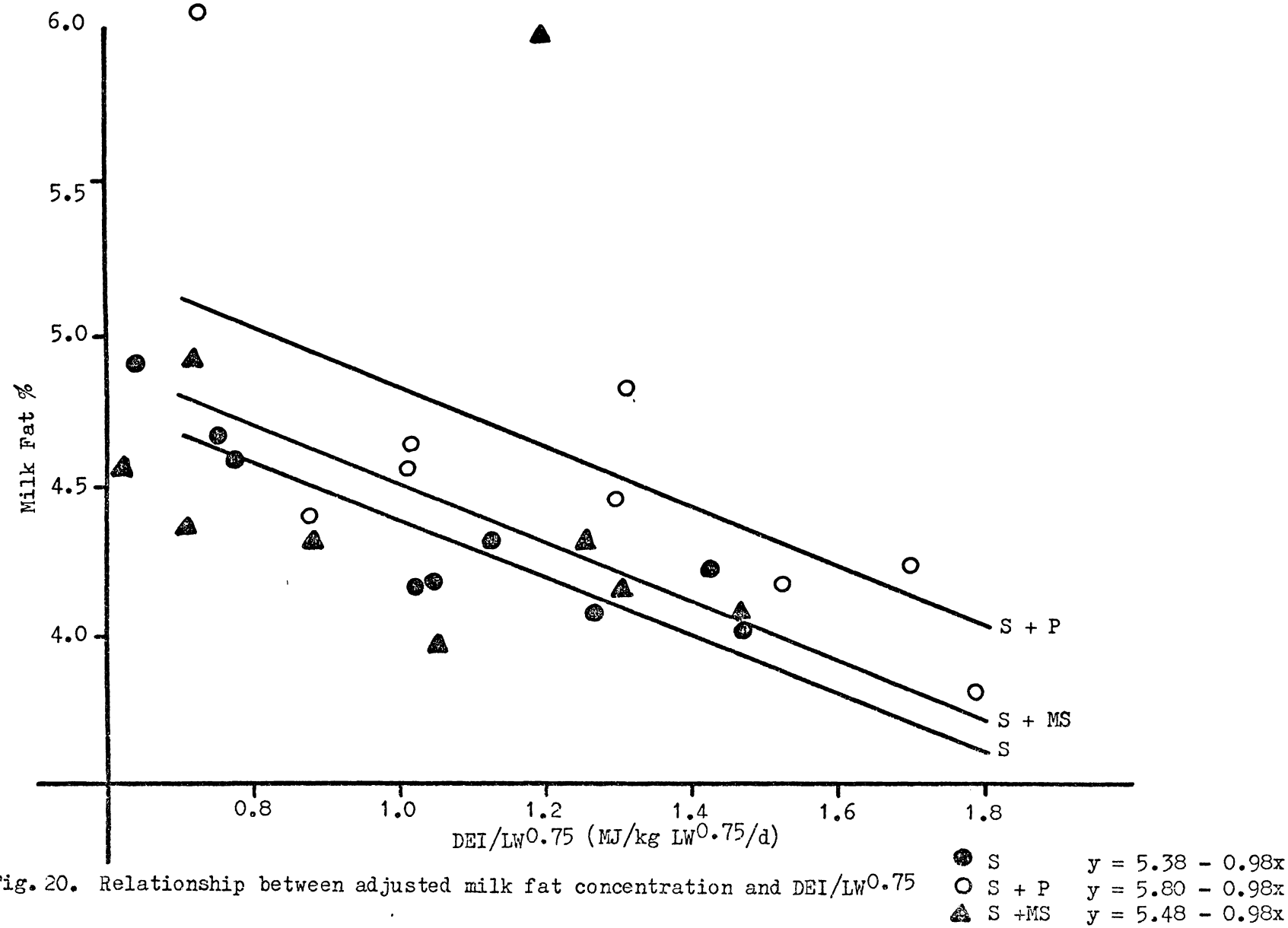


Fig. 20. Relationship between adjusted milk fat concentration and DEI/LW$^{0.75}$

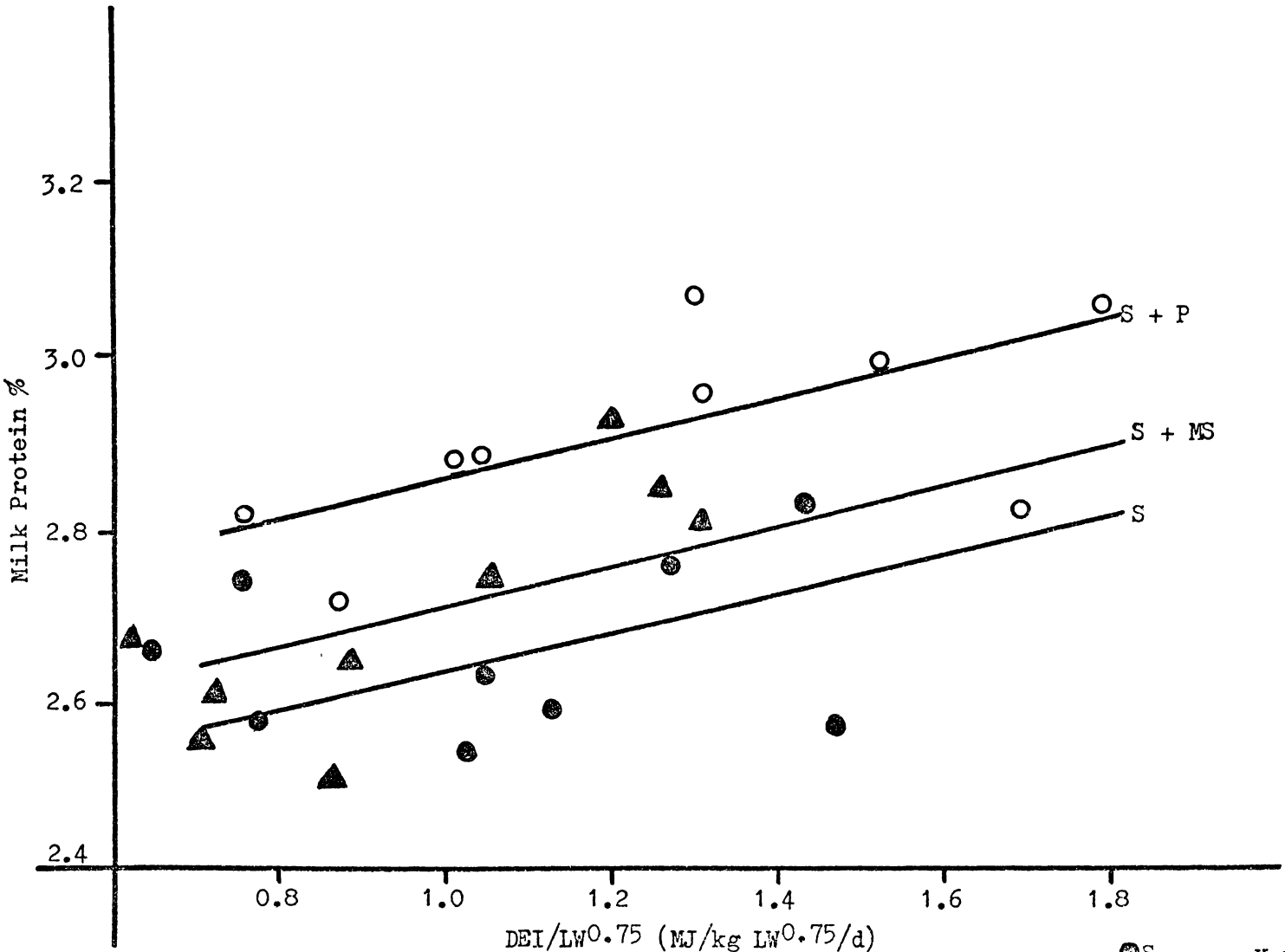


Fig. 21. Relationship between adjusted milk protein concentration and DEI/$LW^{0.75}$

TABLE 26. Effects of diet and level of feeding on mean concentrations and molar proportions of VFA in rumen fluid (adjusted for Period I).

Diet/Level	Concentration (meq/100 ml) of VFA							Molar % of Total VFA					
	HAc	HPr	IHBu	nHBu	IHVa	nHVa	Total	HAc	HPr	IHBu	nHBu	IHVa	nHVa
S	4.65	1.28	0.09	0.71	0.12	0.13	6.92	67.3	18.1	1.3	9.8	1.6	1.8
S + P	4.32	1.09	0.08	0.77	0.11	0.12	6.58	66.0	17.6	1.2	11.7	1.7	1.8
S + MS	4.36	1.22	0.07	0.68	0.11	0.11	6.53	66.9	18.2	1.1	10.4	1.8	1.7
SED	0.583	0.152	0.010	0.098	0.018	0.015	0.827	0.82	0.87	0.11	0.74	0.16	0.10
Sign.	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	+	NS	NS
Level 100 %	4.57	1.22	0.07	0.73	0.10	0.12	6.82	67.1	18.0	1.0	10.6	1.5	1.7
50 %	4.31	1.17	0.08	0.70	0.12	0.12	6.53	66.4	17.9	1.3	10.7	1.8	1.9
SED	0.476	0.124	0.008	0.080	0.015	0.012	0.675	0.66	0.71	0.09	0.60	0.13	0.08
Sign.	NS	NS	NS	NS	NS	NS	NS	NS	NS	*	NS	*	*

Blood Metabolites (Table 27) - Plasma urea levels were significantly less for animals on the silage plus maize silage ration compared with animals on the other treatment diets at each sampling time. The plasma urea levels of cows on all diets increased with time from feeding (from 0h to 4h) and were slightly higher at the 50 % feeding level compared with ad libitum feeding.

Plasma ∝ amino nitrogen levels decreased for cows on all diets with time from feeding (from 0h to 4h) and were significantly higher for cows on the silage plus pasture ration in comparison with the other treatments immediately prior to feeding.

Partitioning of Dietary Nitrogen and Energy (Table 28) - Urinary nitrogen as a percentage of dietary nitrogen and DNI was significantly reduced by supplementing pasture silage with maize silage, and the effect of the pasture supplement was intermediate. Milk nitrogen as a percentage of dietary and DNI was significantly lower for the silage ration than the maize silage plus silage ration, but higher than the pasture supplemented ration. Nitrogen retention was improved by the pasture supplement, and productive nitrogen (milk N + NR) as a percentage of DNI was significantly greater for the supplemented rations than the control silage ration.

Faecal energy output as a percentage of GEI was significantly increased by supplementing pasture silage with maize silage, and both supplements reduced urinary energy output as a percentage of DEI.

Experiment 3.3.2

Diet Composition, Digestibility and Intake (Tables 29, 30, Appendix 13) - Supplementing silage with 17% protein concentrate

TABLE 27. Effects of diet and level of feeding on PCV %, urea and ∝ amino N concentrations in plasma immediately before feeding (0h), 4 and 12h later (adjusted for Period I).

Diet/Level	PCV %			Plasma Urea (mg/100 ml)			Plasma Amino N (mg/l)		
	0h	4h	12h	0h	4h	12h	0h	4h	12h
S	33.6	33.6	31.7	5.9	12.2	11.1	44.2	35.8	34.8
S + MS	32.9	33.5	32.3	3.7	8.1	5.3	46.5	36.1	38.0
S + P	34.0	34.8	35.3	6.9	14.6	11.6	51.8	37.0	37.6
SED	2.08	1.71	2.26	0.47	1.13	1.11	2.26	1.77	1.86
Sign.	NS	NS	NS	***	***	***	*	NS	NS
Level 100 %	32.0	33.5	32.2	5.0	10.8	8.3	49.6	35.4	38.6
50 %	35.0	34.4	33.3	6.0	12.6	10.3	45.4	37.2	35.0
SED	1.70	1.39	1.85	0.38	0.92	0.91	1.85	1.45	1.52
Sign.	+	NS	NS	*	+	*	*	NS	*

TABLE 28. Effect of diet on partitioning of nitrogen and gross energy.

Item	S	S + P	S + MS	SED	Sign.
N, % of intake					
Faeces	34.3	31.2	42.4	0.89	***
Urine	50.5	44.5	30.9	3.96	**
Milk	23.8	20.3	35.1	0.87	***
Retention (NR)	-8.2	4.3	-8.4	4.38	*
Milk + NR	15.6	24.6	26.7	3.83	*
N, % of digested					
Urine	76.2	64.1	53.1	5.12	*
Milk	36.3	29.5	61.0	1.16	***
NR	-12.5	6.4	-14.1	6.12	*
Milk + NR	23.8	35.9	46.9	5.37	*
GE, % of intake					
Faeces	27.7	26.8	33.0	0.91	***
Urine	5.9	5.0	3.8	0.30	**
Methane (est)	8.5	8.6	8.2	0.05	**
Milk	23.9	23.6	24.6	1.55	NS
Residual (est)	33.8	35.9	30.2	1.84	*
Milk and Residual	57.7	59.5	54.9	0.65	***
DE, % of digested					
Urine	8.3	6.9	5.7	0.34	***
Milk	33.1	32.3	36.8	2.27	NS
Milk and Residual	79.9	81.3	81.9	0.30	**
Metabolisable Energy (MJ/kg DM)	10.79	11.09	10.11	0.12	***

TABLE 29. Chemical composition of feedstuffs.

Nutrient	Silage	Protein Concentrate
DM (%)	17.5 ± 0.31[a]	89.9 ± 0.10
Composition of 100 g DM		
O.M. (g)	86.9 ± 0.25	89.2 ± 0.10
GE (MJ)	1.82± 0.030	2.08± 0.033
Total N (g)	3.2 ± 0.14	8.9 ± 0.12
Soluble N (g)	1.4 ± 0.05	1.2 ± 0.06
Soluble Protein N (g)	0.03± 0.001	0.31± 0.026
NH_3-N (g)	0.25± 0.001	0.0 ± 0.0
Soluble Carbohydrate (g)	1.35 ± 0.15	7.79± 0.31

[a] SEM, n = 6

TABLE 30 - Mean apparent digestibility coefficients of diets.

Item	Digestibility (%)			SED	Sign.
	S	S + PC_1	S + PC_2		
GE	68.8	70.7	71.5	0.57	**
DM	68.1	69.3	69.3	0.55	NS
OM	72.9	74.4	74.5	0.40	*
N	71.6	74.9	76.5	0.65	***

n = 4

raised the crude protein content from 20.2 to 25.4 per cent and reduced the soluble N and NPN fractions by 10 % to 34 and 32 % of total nitrogen respectively.

Protein concentrates significantly increased the apparent digestibility coefficients of nitrogen, OM and gross energy (Table 30). Although mean DEI increased 20 per cent by supplementing silage with the high level of protein concentrate, the corresponding increase in DNI was more than 50 per cent (Appendix 13).

Milk Yield and Composition, and Live-Weight Change (Table 31) Diet significantly affected all milk variates apart from the concentrations of fat and lactose. Feeding level significantly affected all milk variates and significant interactions of diet x feeding level were observed for protein and milk fat yields.

The relationships between DEI/$LW^{0.75}$ (Model i) and the yield and composition of milk, are summarized in Appendix 14. The same regression equation (Model iv) could not be used to describe the data since there was a significant reduction in the residual sums of squares from Model (iv) to Models (ii) and (iii) (see Appendix 15). Since intersecting lines (Model ii) accounted for more of the variation than parallel lines (Model iii), Model (ii) was used to describe the data (Table 32, Figures 22-28). This gave similar residual sums of squares to Model (i). The regression coefficients for all milk variates were positive except for milk fat concentration, and significant apart from fat yield for the silage ration. The high level of protein supplementation significantly increased the yield of milk, fat, protein and lactose, and the concentrations of fat

TABLE 31. Mean daily milk yield and milk composition (adjusted), and LW change in Period II.

Diet	Level	Milk (kg/d)	Protein (g/d)	Protein %	Fat (g/d)	Fat %	Lactose (g/d)	Lactose %	ΔLW (g/d)
S	100	10.0	272	2.72	393	3.92	492	4.92	-242
	75	9.6	253	2.67	405	4.27	465	4.83	-703
	50	8.0	211	2.64	388	4.92	384	4.80	-1588
S + PC_1	100	11.6	340	2.92	484	4.19	569	4.91	233
	75	10.9	295	2.74	493	4.55	526	4.82	-466
	50	8.7	227	2.61	408	4.77	414	4.81	-1035
S + PC_2	100	12.4	371	3.01	560	4.53	604	4.85	708
	75	11.2	309	2.81	487	4.47	531	4.75	-171
	50	8.7	233	2.69	420	4.86	414	4.76	-1070
Diet Means S		9.2	245	2.68	395	4.37	447	4.85	-844
S + PC_1		10.4	287	2.76	462	4.50	503	4.85	-423
S + PC_2		10.8	304	2.84	489	4.62	516	4.79	-178
	SED	0.69	18.0	0.12	34.5	0.32	35.2	0.11	200
	Sign.	***	***	*	***	NS	***	NS	**
Level Means	100	11.3	327	2,88	479	4.21	555	4.89	233
	75	10.6	286	2.74	462	4.43	507	4.80	-447
	50	8.5	224	2.65	405	4.85	404	4.79	-1231
	Sign.	***	***	**	***	**	***	+	***
Diets x Level	Sign.	NS	*	NS	*	NS	NS	NS	NS
Linear Coeff.	Sign.	***	***	**	***	NS	***	NS	**

TABLE 32. Relationships between milk variates and DEI/$LW^{0.75}$ (MJ/kg $LW^{0.75}$/d) in Period II (Model ii).

Item	Mean	S.D.	$B_\mu \pm$ SE	Sign.	Ration	$b_x \pm$ SE	Sign.	C	R.S.D.	R
Milk Yield (kg/d)	10.11	2.22	0.70 ± 0.09	***	S	2.77 ± 0.61	*	-1.98	0.92	0.93
					$S+PC_1$	3.50 ± 0.59	***			
					$S+PC_2$	3.50 ± 0.53	***			
Milk Fat (g/d)	443	87.1	0.66 ± 0.07	***	S	36.4 ± 28.87	NS	-20.7	37.8	0.91
					$S+PC_1$	96.6 ± 24.36	***			
					$S+PC_2$	111.7 ± 21.77	***			
Milk Protein (g/d)	278	66.6	0.70 ± 0.10	***	S	101.3 ± 17.46	***	-102.1	24.4	0.94
					$S+PC_1$	129.9 ± 16.62	***			
					$S+PC_2$	131.7 ± 14.85	***			
Milk Lactose (g/d)	489	109.1	0.65 ± 0.09	***	S	154.5 ± 29.63	***	-116.9	43.3	0.93
					$S+PC_1$	188.6 ± 29.12	***			
					$S+PC_2$	182.4 ± 26.02	***			
Milk Fat %	4.44	0.71	0.85 ± 0.09	***	S	-1.01 ± 0.228	***	+1.42	0.34	0.89
					$S+PC_1$	-0.71 ± 0.220	**			
					$S+PC_2$	-0.53 ± 0.200	*			
Milk Protein %	2.75	0.24	0.90 ± 0.09	***	S	0.26 ± 0.072	**	-0.17	0.11	0.92
					$S+PC_1$	0.32 ± 0.069	***			
					$S+PC_2$	0.35 ± 0.062	***			
Milk Lactose %	4.83	0.17	0.76 ± 0.09	***	S	0.18 ± 0.064	**	0.86	0.09	0.86
					$S+PC_1$	0.16 ± 0.062	*			
					$S+PC_2$	0.09 ± 0.056	+			

(i) μ denotes production in Period II
(ii) x denotes DEI/kg $LW^{0.75}$ in Period II
(iii) overall mean DEI/kg $LW^{0.75}$ = 1.17 ± 0.33

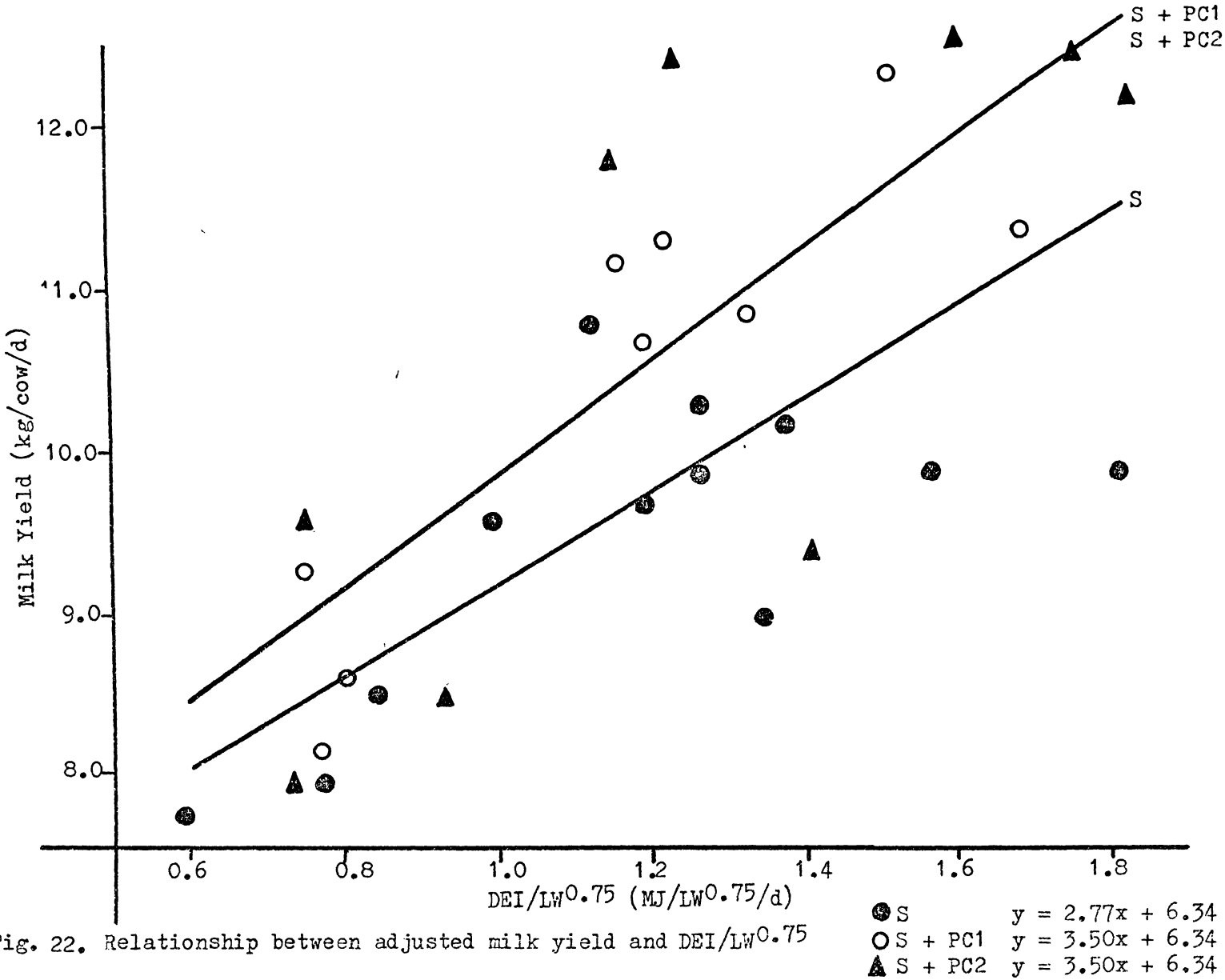


Fig. 22. Relationship between adjusted milk yield and DEI/LW$^{0.75}$

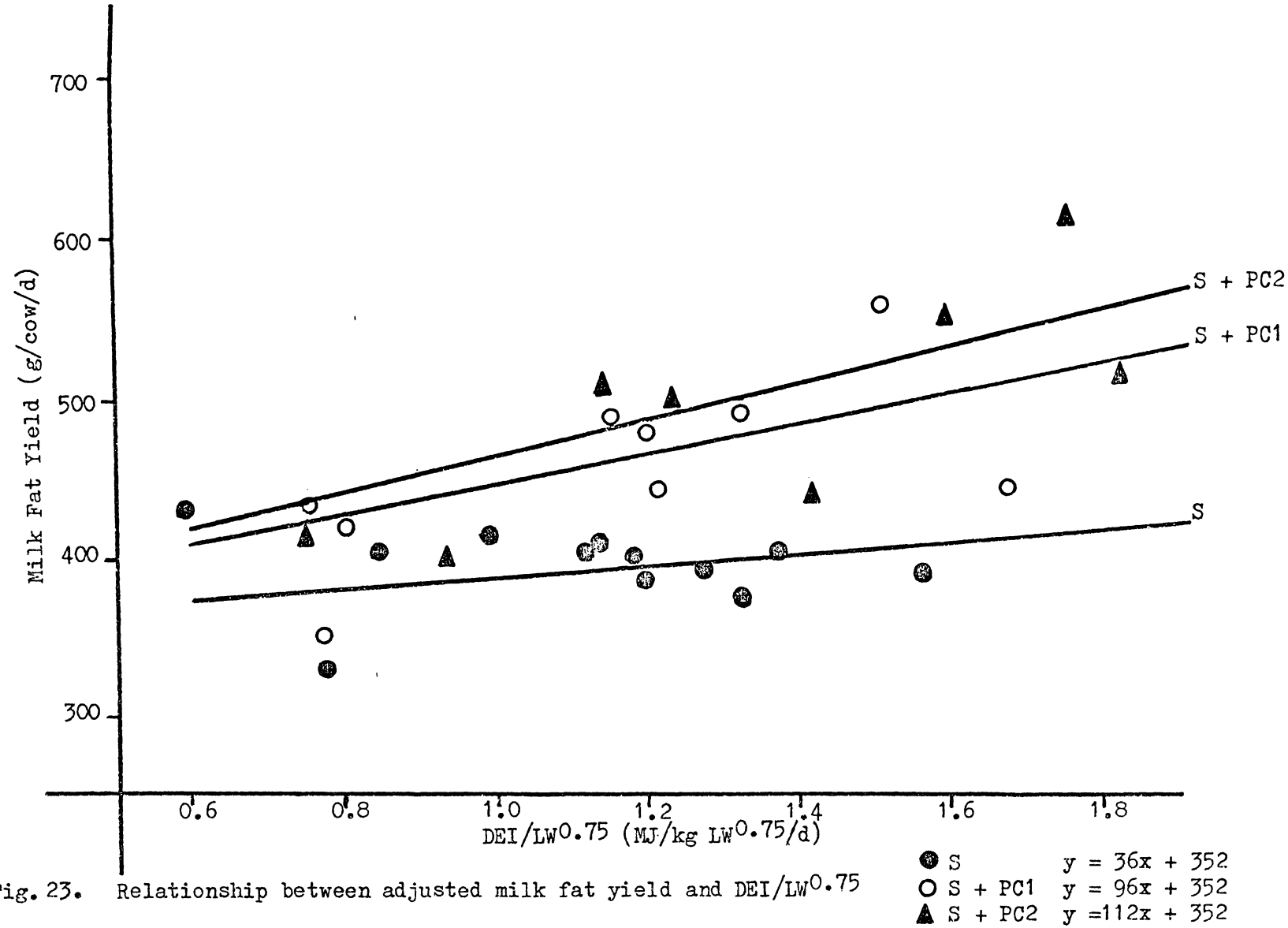


Fig. 23. Relationship between adjusted milk fat yield and DEI/LW$^{0.75}$

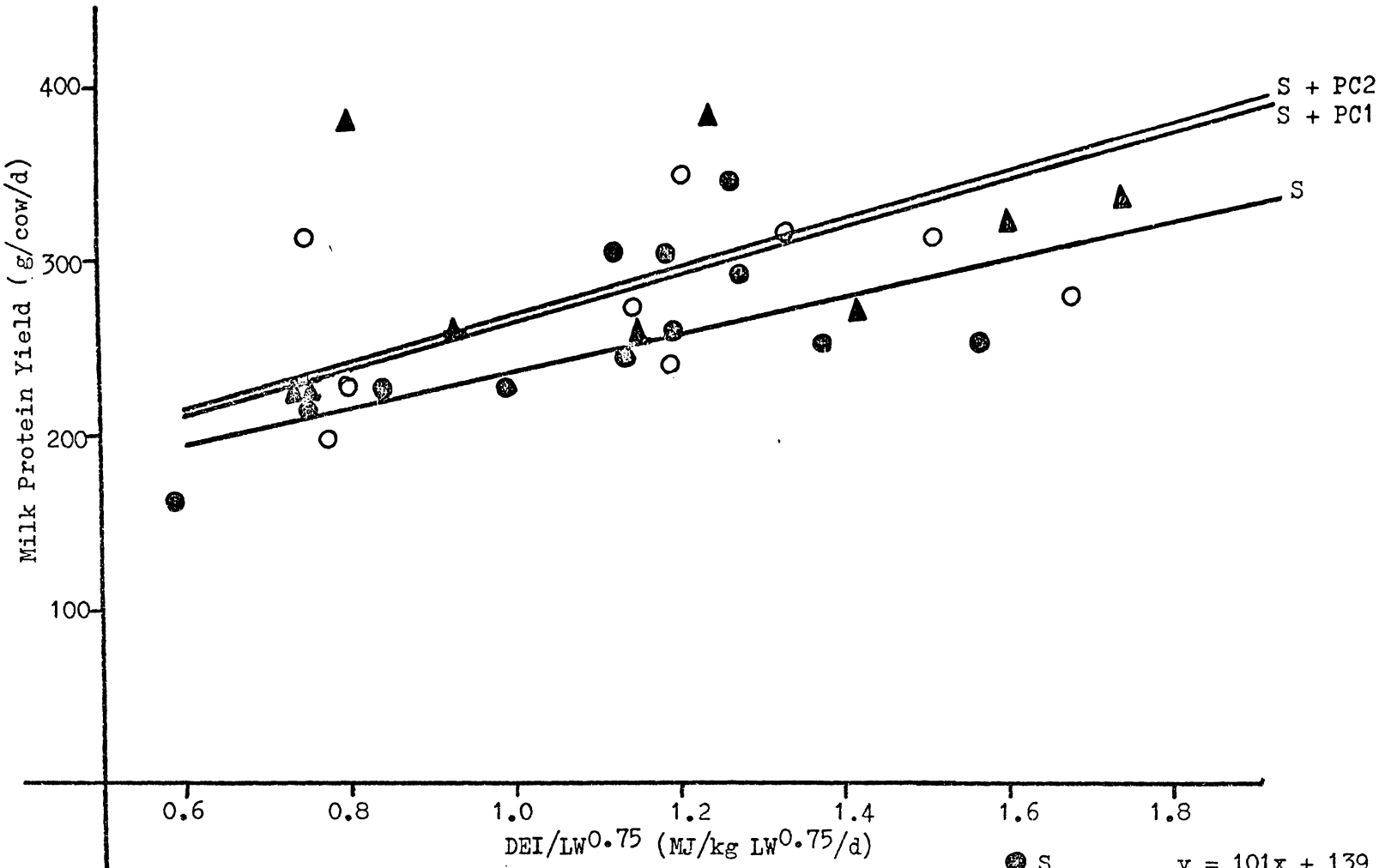


Fig. 24. Relationship between adjusted milk protein yield and DEI/$LW^{0.75}$

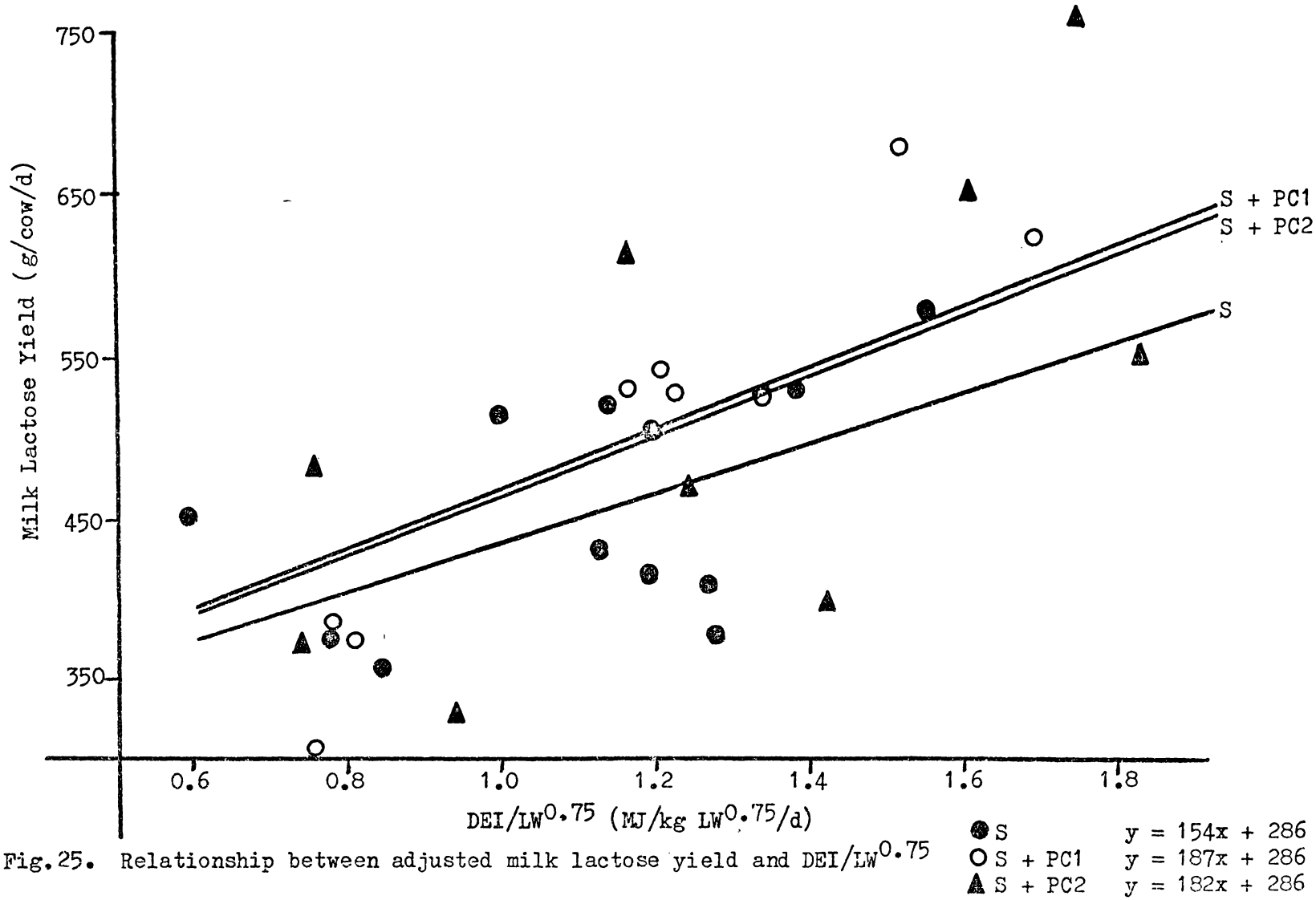


Fig.25. Relationship between adjusted milk lactose yield and DEI/$LW^{0.75}$

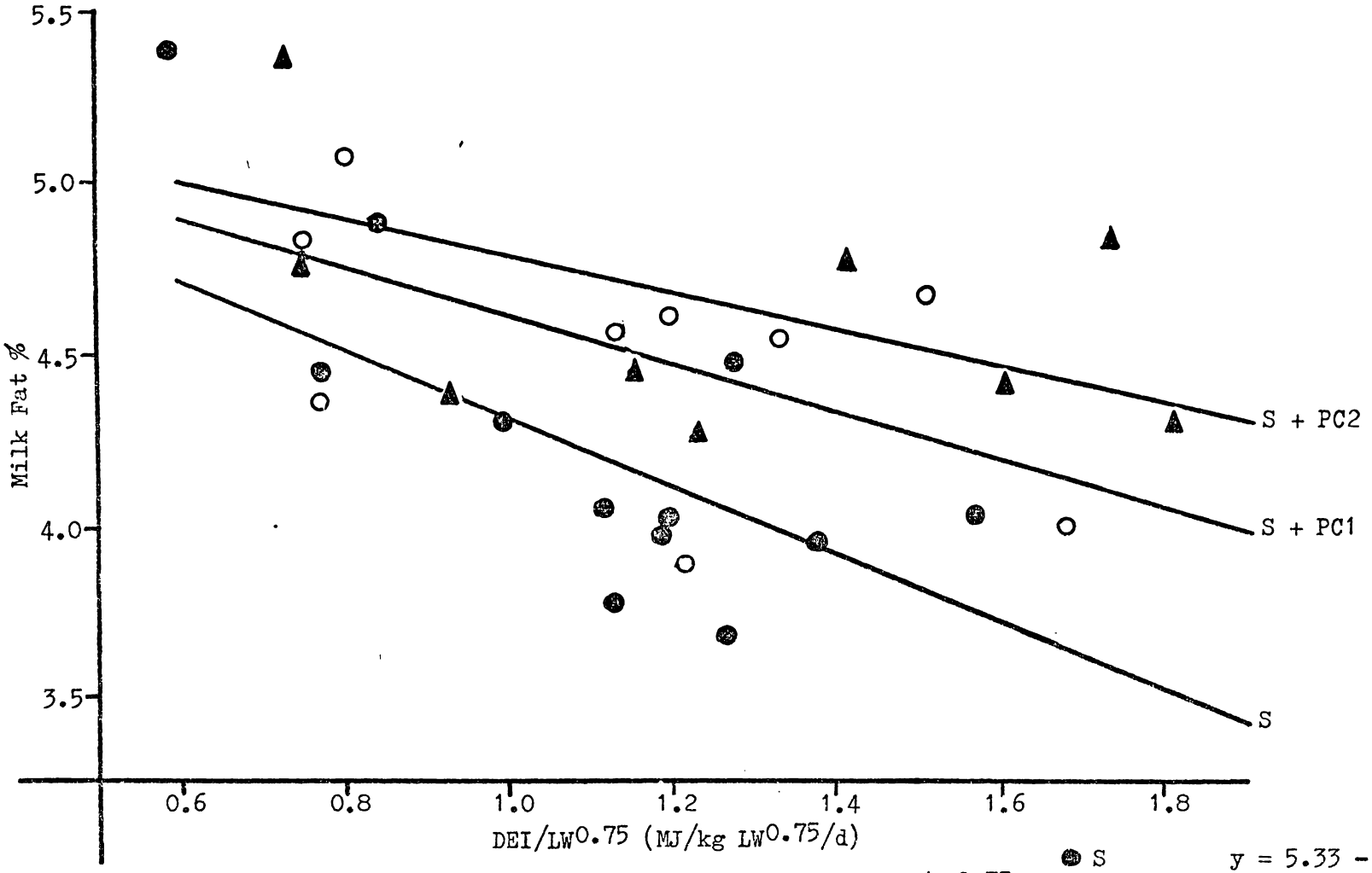


Fig.26. Relationship between adjusted milk fat concentration and DEI/LW$^{0.75}$

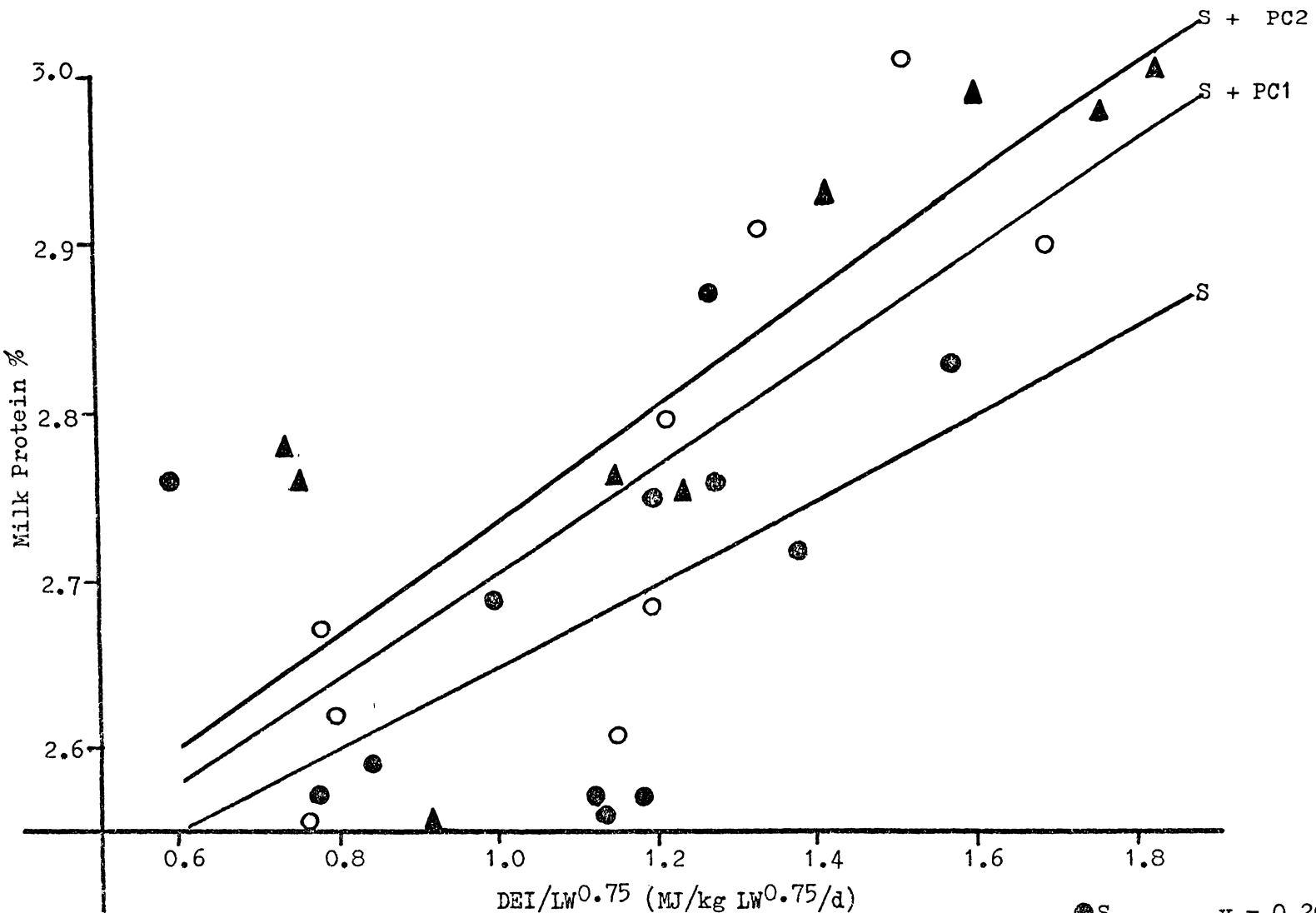


Fig.27. Relationship between adjusted milk protein concentration and DEI/$LW^{0.75}$

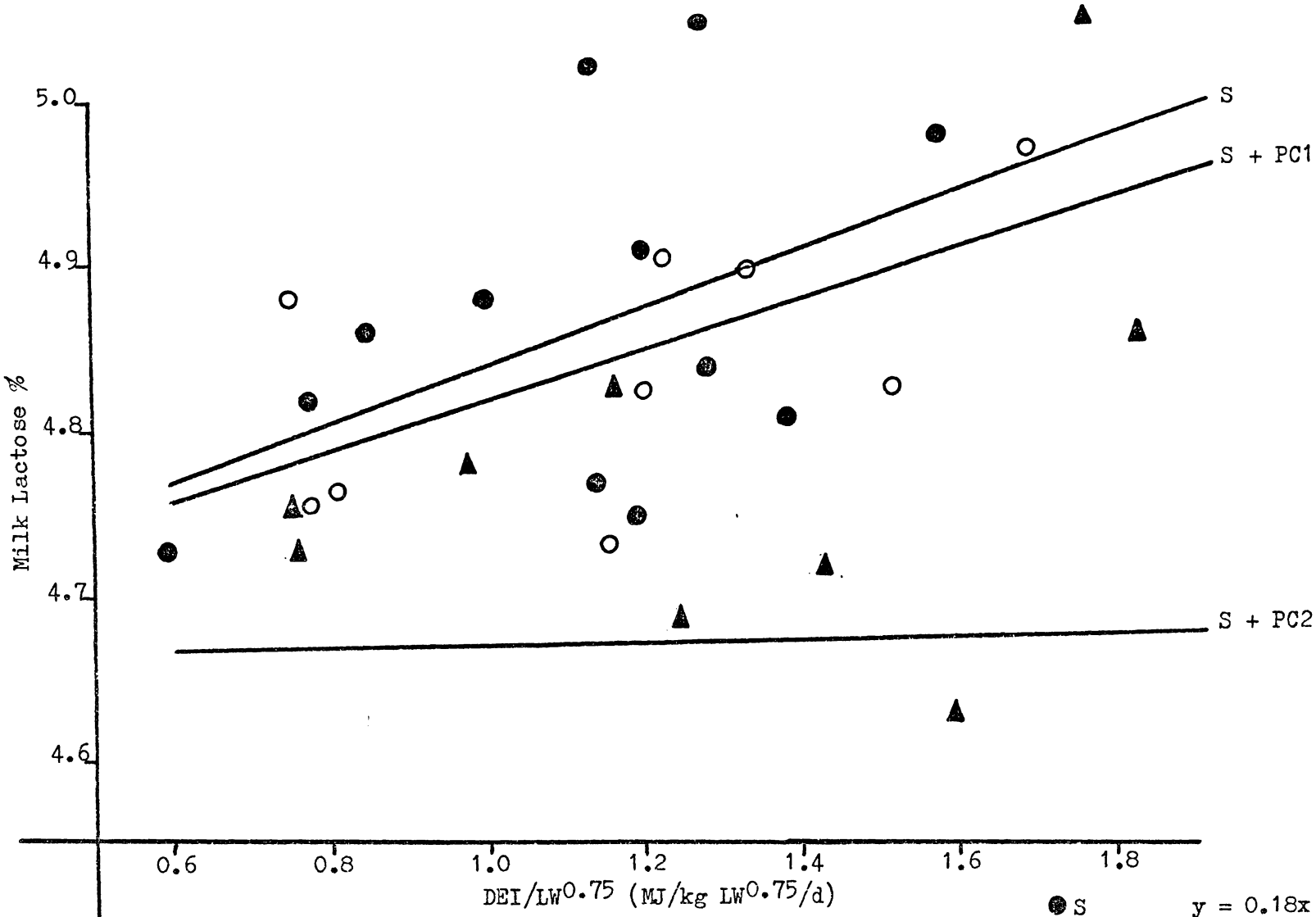


Fig. 28. Relationship between adjusted milk lactose concentration and DEI/$LW^{0.75}$

and protein in milk, and reduced lactose percentage compared with that produced by the unsupplemented cows. The effects of the lower level of protein supplementation on milk composition were intermediate, although responses in milk yield were similar to those obtained with the higher level of concentrates.

Relative to intake, no significant differences were observed in live-weight change of cows on the rations (Appendix 15).

Composition of Blood and Rumen Fluid (Tables 33, 34) - Plasma urea levels were increased by supplementing silage with the high level of protein concentrate (Table 33). Although the plasma urea levels associated with the lower level of protein supplements were not significantly different from those of the silage treatment the means were intermediate.

Molar concentrations of volatile fatty acids in rumen fluid were not altered by supplementing silage with protein concentrates either before, or 4 hours after feeding (Appendix 16). The mean results are shown in Table 34. The mean total concentrations of volatile acids in rumen fluid were significantly increased by adding protein concentrates to silage.

Partition of Dietary Energy and Nitrogen (Table 35) - Supplementing silage with protein concentrates had no effect on the apparent partitioning of dietary or DN into milk, urine and nitrogen retention. Similarly no change resulted in the partition of dietary energy or digested energy into milk, urine and residual energy.

TABLE 33. Effects of diet and level of feeding on PCV %, urea and ∝ amino N concentrations in plasma immediately before feeding (0h) and 4h later (adjusted for Period I).

Diet/Level	PCV %		Plasma Urea (mg/100 ml)		Plasma Amino N (mg/l)	
	0h	4h	0h	4h	0h	4h
S	32.9	31.1	13.2	19.0	35.0	27.9
S + PC_1	31.3	32.9	15.3	19.9	35.4	30.6
S + PC_2	32.3	32.1	18.3	25.1	35.7	28.0
SED	1.84	2.56	1.16	1.52	3.62	2.32
Sign.	NS	NS	**	**	NS	NS
Level 100 %	31.9	31.5	16.3	22.2	39.0	27.5
50 %	32.4	32.5	14.9	20.4	31.7	30.1
Sign.	NS	NS	NS	NS	NS	NS
Diets and Level	NS	NS	NS	NS	NS	NS

TABLE 34. Effects of diet and level of feeding on mean concentrations and molar proportions of VFA in rumen fluid (adjusted for Period I).

Diet/Level	Concentration (meg/100ml) of VFA							Molar % of Total VFA					
	HAc	HPr	IHBu	nHBu	IHVa	nHVa	Total	HAc	HPr	IHBu	nHBu	IHVa	nHVa
S	5.01	1.35	0.10	0.72	0.14	0.12	7.42	67.9	17.9	1.2	9.9	1.7	1.5
S + PC_1	5.81	1.59	0.12	0.81	0.16	0.12	8.59	68.1	18.2	1.3	9.6	1.8	1.2
S + PC_2	6.79	1.71	0.12	0.95	0.17	0.16	9.89	68.8	17.3	1.3	9.7	1.7	1.4
SED	0.483	0.126	0.008	0.072	0.011	0.010	0.637	1.05	0.91	0.11	0.43	0.11	0.13
Sign.	*	*	*	*	*	*	*	NS	NS	NS	NS	NS	NS
Level 100 %	6.32	1.61	0.12	0.87	0.16	0.14	9.21	68.9	17.4	1.2	9.6	1.7	1.3
50 %	5.41	1.48	0.11	0.78	0.15	0.13	8.05	67.6	18.1	1.3	9.8	1.8	1.4
SED	0.394	0.103	0.006	0.059	0.009	0.008	0.520	0.86	0.74	0.09	0.35	0.009	0.10
Sign.	*	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS

TABLE 35. Effect of diet on the partitioning of nitrogen and gross energy.

Item	S	S + PC_1	S + PC_2	SED	Sign.
N, % of intake					
Faeces	28.4	25.2	23.5	0.66	***
Urine	47.2	49.7	50.9	1.60	NS
Milk	16.4	15.7	16.4	1.66	NS
Retention (NR)	8.0	9.3	8.8	1.53	NS
Milk + NR	24.4	25.0	25.2	1.80	NS
N, % of digested					
Urine	66.1	66.5	66.7	2.22	NS
Milk + NR	34.0	33.4	33.0	2.45	NS
Milk	22.9	21.0	21.4	2.26	NS
NR	11.1	12.4	11.5	2.02	NS
GE, % in intake					
Faeces	30.9	29.3	28.7	0.63	*
Urine	6.9	7.8	7.9	0.49	NS
Methane (est)	8.4	8.4	8.5	0.20	+
Milk	21.4	23.4	25.2	3.01	NS
Residual (est)	32.4	31.0	29.7	2.72	NS
Milk and Residual	53.8	54.4	54.9	0.68	NS
GE, % of digested					
Urine	10.0	11.1	11.2	0.69	NS
Milk	30.9	33.1	35.7	3.88	NS
Milk + Residual	77.9	77.0	78.0	1.21	NS
Metabolisable Energy (MJ/kg DM)	9.5	9.6	9.8	0.13	+

Discussion

In the previous experiment (3.2) it was indicated the low levels of true protein and soluble carbohydrate in silage may be important factors affecting milk yield and composition. In the present experiments cows offered silage rations supplemented with pasture or protein concentrates produced more milk with higher concentrations of fat and protein in milk in comparison to cows fed exclusively on silage at similar intakes of DE. In contrast when maize silage was the supplement there was no effect on milk yield or composition. These results support the suggestion that level of true protein in silage was a major factor responsible for the low efficiency of utilization of DE for milk synthesis.

It has been reported elsewhere that individual VFA's may have specific effects on milk yield and composition (Rook and Balch, 1961; Rook et al. 1965; Wilson et al. 1967) and that the molar proportions of these acids in the rumen may be associated with changes in yield and composition of milk (Huber and Bowman, 1966a; Armstrong and Prescott, 1971). In the present study the effects of the rations on the molar proportions of rumen VFA were small and not significant and they did not appear to be associated with observed changes in milk yield and composition. An exception was a small increase in the molar percentage of rumen butyrate with the pasture supplement, which may have contributed to the rise in milk fat percentage (Smith, McCarthy and Rook, 1974).

Pasture and protein concentrates as supplements to silage increased both the nitrogen content and the digestibility of the nitrogen in the ration, with an associated increase in DN intake of more than 40 per cent. Moreover the low amounts of free

ammonia and the higher levels of insoluble nitrogen in these supplements indicate that a considerable proportion of this increased DN intake was in the form of digestible true protein. In contrast maize silage lowered the nitrogen content and the digestibility of the ration resulting in a decrease in DN intake by approximately 40 per cent.

This increase in digestible true protein intake by cows when supplements of pasture or protein concentrates were offered with silage was associated with increases in milk protein yield. However the apparent partitioning of DN was different for these supplements.

Protein concentrates did not alter the proportion of DN retained or excreted in milk or urine indicating that milk protein synthesis was a simple response to increased DN intake. However when supplements of pasture were given, the nitrogen in urine and milk as a proportion of DNI decreased, and the proportion retained increased. This suggests that the efficiency of utilization of dietary nitrogen for milk synthesis was reduced and that the increased milk protein yield must have been again due to an increased intake of true protein. The decrease in urinary nitrogen as a proportion of DN with the pasture supplement may have been due to the increase in soluble carbohydrate level of the ration stimulating assimilation of ammonia into microbial protein (Smith, 1969), and reducing the absorption of ammonia from the rumen (Armstrong, 1974). In contrast urinary nitrogen as a proportion of DNI was unaltered by the supplement of protein concentrates, possibly because the additional soluble carbohydrate provided by the supplement only marginally (2 %) raised the level in the ration. Alternatively

the low solubility of these protein concentrates (Miller, 1973; Hume, 1974) may reduce both the proportion of dietary nitrogen absorbed from the rumen and urinary nitrogen excretion, but this may then be compensated by an increased hepatic deamination and nitrogen excretion as a result of increased post-ruminal absorption of dietary nitrogen.

Although differences in the effects of the supplements on plasma urea levels were observed they do not necessarily indicate the relative amounts of dietary nitrogen absorbed from the rumen since differences in plasma urea levels may have also arisen from differences in protein intake or hepatic deamination of protein.

The increases in milk protein synthesis by cows on silage supplemented with pasture and protein concentrates support the results obtained in the previous experiment (3.1) and are consistent with the view that true protein intake is an important factor limiting milk protein synthesis of cows offered silage. However inspite of the possibilities considered above, the reasons for the differences in the partition of dietary nitrogen between the pasture and protein concentrate supplemented rations in this experiment, and that in experiment 3.1 are not clear.

When maize silage was given as a supplement with pasture silage, intake of true protein was decreased. However milk N as a proportion of the DN increased, which indicates that the efficiency of nitrogen utilization for milk synthesis was increased, with the result that milk protein production did not alter. This increased efficiency of DN utilization could be attributed to an increase in microbial protein synthesis due to the high level of readily fermentable carbohydrate provided by maize silage in the form of grain (Bryant, 1976). Alternatively, or in addition, the maize

silage supplement may have increased the efficiency of utilization of DN for milk protein by reducing proteolysis of dietary protein in the rumen (McDonald, 1952; Hume, 1974) and therefore the losses of dietary N from the rumen; thereby increasing the proportion of dietary protein (Hume, 1974) and the range of amino acids entering the duodenum. These effects of the maize silage supplement are similar to those observed with formalin treated silage in Experiment 3.2 where marked decreases in N digestibility were also associated with increased efficiency of DN for milk production. However unlike the formalin silage treatment, milk protein was not increased and it is suggested that the low N content and the low digestibility of the maize silage appeared to balance the increase in efficiency of N use for milk production.

Despite the increases in milk protein synthesis when supplements of pasture or protein concentrates were given to cows offered pasture silage, no differences were observed in the plasma levels of α amino N between the diets, apart from a small increase before feeding the pasture supplemented diet. These results are consistent with the findings in Experiment 3.1 and 3.2 and show that the concentration of amino N in plasma is not limiting milk protein synthesis of cows on silage diets. PCV % was also unaffected by diet. Although this is in contrast to the results observed in Experiment 3.1 it agrees with the findings in Experiment 3.2 and PCV % would appear to be of little consequence on factors involved in milk synthesis.

In contrast to maize silage the effects of pasture and protein concentrates on the partitioning of dietary energy were generally small. However addition of protein concentrates to silage

significantly increased the total VFA concentration in rumen fluid and confirms similar observations by Griffiths and Spillane (1970). Presumably if rumen VFA concentrations reflect production and absorption rates (Leng, 1970b) this may account in part for the increase in milk synthesis.

In summary these experiments have shown that supplementing pasture silage with pasture and protein concentrates increased milk synthesis. It is suggested that milk protein synthesis is limited by the amount of protein entering the duodenum; a consequence of the low true protein content of silage. The next series of experiments examines the effects of intra-abomasal infusions of protein on milk protein synthesis by cows offered pasture silage.

3.4 RESPONSE OF LACTATING COWS TO ABOMASAL INFUSIONS OF CASEIN, METHIONINE AND GLUCOSE

Introduction

The previous studies comparing pasture and high moisture silage made from the same herbage established that cows fed pasture produced more milk with a higher concentration of fat and protein than cows fed silage at equal intakes of digestible energy. Treatments which either reduced fermentation during ensiling or provided pasture or protein concentrates as supplements to silage improved the yield of milk and the percentages of fat and protein in milk. It was suggested that these responses were associated with increased intakes of true protein.

Because of the extensive degradation of dietary protein in the rumen, the possible inadequacy of protein or specific amino acids in silage rations can be best examined by post-ruminal supplementation of the diet.

Broderick, Kowalczyk and Satter (1970); Spechter, (1972); Derrig, Clark and Davis (1974) and Vik-mo, Emery and Huber (1974) have infused casein or mixtures of casein and methionine into the abomasum of cows fed diets made up of maize silage, urea and concentrates and containing adequate levels of energy and crude protein to meet production requirements according to NRC (1971) standards. Responses of 4.9-13 % and 2-16 % in the yields of milk and milk protein respectively have been observed in cows producing 16-30 kg milk daily. Substantially higher increments were recorded in higher producing cows indicating that a dietary protein deficiency was greater when the requirements were greater. Moreover

Spechter (1972) observed greater responses when cows were fed high NPN rations in early lactation.

Vik-mo and Huber (1971) and Vik-mo, Emery and Huber (1974) found that infusions of glucose also resulted in increased yields of milk and protein which suggests that the response to casein may have at least partly been due to gluconeogenesis rather than the provision of essential amino acids.

Armstrong (1974) indicated that pasture silage may be low in sulphur amino acids relative to dried pasture and Barry et al. (1973) and Lancaster (1976) obtained significant responses in wool growth and intake of sheep fed pasture silage after intraperitoneal infusions of methionine. The possibility that methionine may be limiting milk synthesis of cows fed corn and urea diets is indicated by the work of McCarthy, Patton and Griel (1970) who reported ruminal supplementation of methionine increased milk fat content, and by Fisher (1972) who observed a response in milk protein concentration to intravenous infusions of methionine.

The initial objectives of the present experiments described were to investigate the effects of abomasal infusions of casein, methionine and glucose on the yield and composition of milk from cows fed pasture silage. Subsequently the effect of diets were investigated by infusing casein into the abomasum of cows offered either pasture herbage or silage.

Materials and Methods

Foods - The pasture silage was made from predominantly ryegrass (Lolium perenne)/white clover (Trifolium repens) pasture in October, 1974, with a flail harvester. At the same time pasture from a

similar sward was cut with a rotary mower and stored at -20 °C.

Animals - Two sets of monozygotic twin Jersey cows producing 8-12 kg milk daily, and surgically prepared with abomasal cannulae immediately after calving in September, 1975, were used in the first four experiments described. Another five unrelated Friesian cows were fitted with abomasal canmulae soon after calving in April, 1976. These cows were producing 14-20 kg milk daily ana were used in the last experiment outlined below.

Cannulae - Each cannula was constructed from 100 cm of silastic medical grade tubing (Dow Corning - I.D., 1.50 mm and O.D. 2.50 mm). Two flanges, 2 and 5 cm in diameter were attached to the cannula approximately 1 and 3 cm from one end with silastic adhesive (see Plate 1).

Surgical Procedures - After withholding food and water for 24 h, each cow was initially tranquillized with 30 mg acetyl promazine injected intramuscularly. Prior to entering the surgery 15 minutes later, initial anaesthesia was induced by intravenous injection of 3 g 'Intraval' (sodium thiopentone).

A cuffed Magil endotracheal tube was inserted into the trachea and anaesthesia was maintained with a mixture of nitrous oxide (900 cc/min), oxygen (1400 cc/min) and fluothane (250 cc/min) in a closed circuit anaesthetic system.

The area on the right flank of the cow extending from the last rib to the hind leg and to the backbone was clipped, shaved and rinsed with 1 % iodine solution.

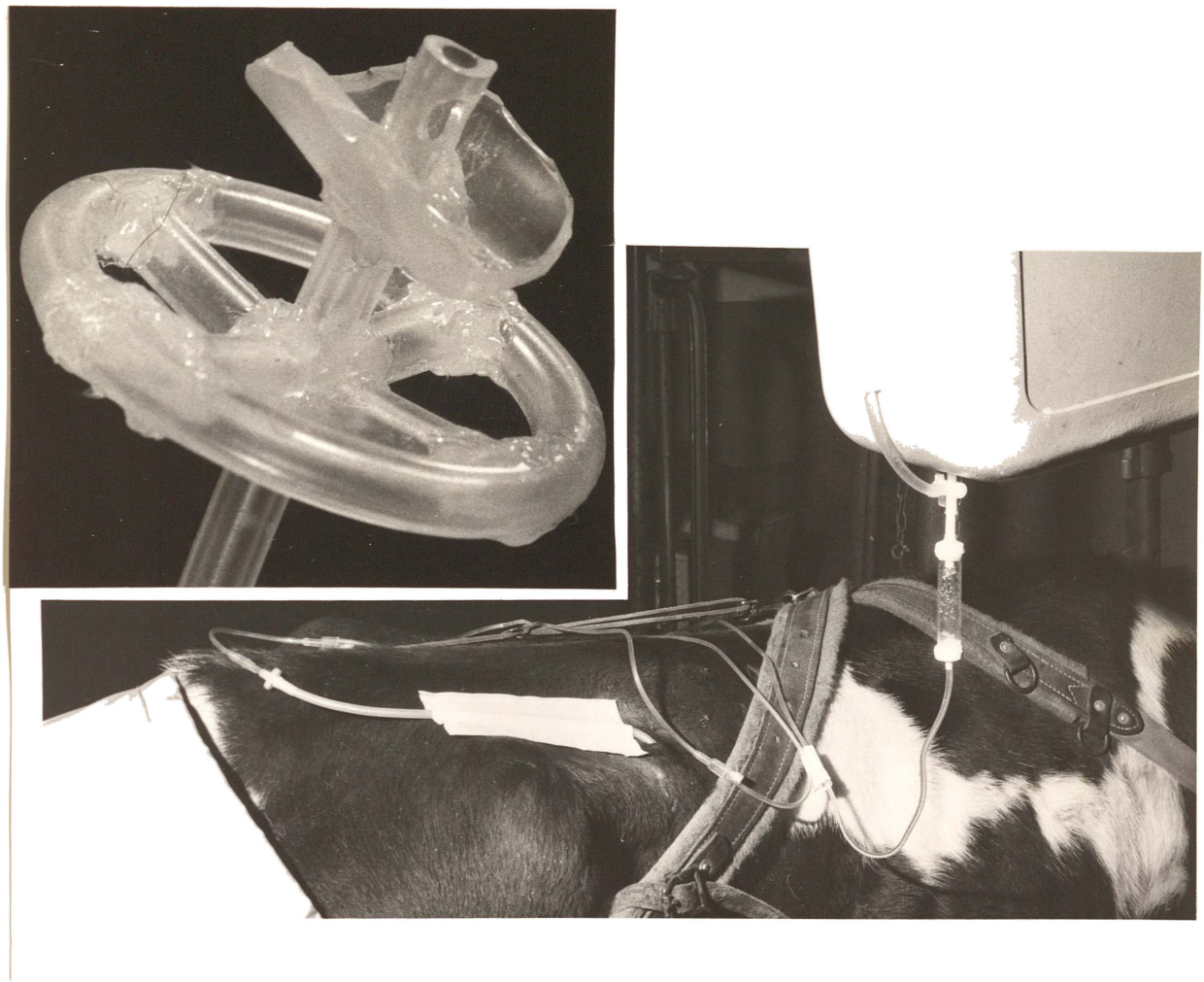


Plate 1. Infusion equipment showing detail of cannula flanges (Inset).

Plate 2. Cows in metabolism stalls for collection of milk, urine and faeces during infusion studies.

A vertical incision beginning adjacent to the mammary vein extended 22-24 cm dorsally and was maintained at a distance of approximately 10 cm from the last rib. After enteriorizing the abomasum a small incision (1 cm) was made in the greater curvature approximately 5 cm from the pylorus. The cannula was inserted so that the proximal flange was inside the lumen and fixed with purse string sutures. The distal flange was then sutured to the abomasal wall.

The cannula was exteriorized through a stab wound approximately 10 cm dorsal to the original incision, a trochar was then used to pass the tubing under the skin and exit near the spinal processes. The abominal muscle layers were continuously sutured with catgut and the skin was closed with braided silk using interrupted sutures. The animals were left with food in a warm recovery pen overnight and returned to grazing the following day.

Intramuscular injections of 10 ml streptomycin were administered for 4 days following the operation and antibiotic powder (chlortetracycline HCl and benzocaine) was applied twice daily to the wound area.

Skin sutures were removed 14 days after the operation. All cows remained in good health although in three instances hernia occurred at the site of the original incision.

Experimental Design

Experiment 3.4.1 - Six series of infusions were conducted with two sets of twin cows. Sodium caseinate was infused at a concentration of 30 g/l to provide 150 g/d (L) or 300 g/d (H) interspersed with infusions of normal saline at a volume equal to

casein infusion. The sequence of infusions was: Saline (L), Casein (L), Saline (L), Saline (H), Casein (H), Saline (H) or the reverse for the opposite twin in each set. Individual cows were offered constant amounts of silage each day. Infusion periods were of 5 days and treatment effects were assessed over the last 3 days.

Experiment 3.4.2 - Treatment infusions were 300 g/d sodium caseinate at a concentration of 30 g/l with (CM) and without (C) the addition of 12 g/d of L-methionine. Treatment infusions were randomized amongst the twins and interspersed with control infusions of 10 l/d of normal saline (S). Individual cows were offered a constant amount of silage each day. Infusion periods were of 7 days and treatment effects were assessed over the last 5 days.

Experiment 3.4.3 - Individual cows were offered constant amounts of silage and the effects on milk yield and composition, and nitrogen utilisation, were compared when infusions were made of (i) 300 g/d sodium caseinate, (ii) 290 g/d glucose, (iii) 12 g/d L-methionine, (iv) normal saline. Infusions were administered according to a 4 x 4 Latin Square arrangement. The casein and glucose furnished equal energy to cows based on 19.3 and 15.9 MJ ME/kg DM for the respective materials and a utilisation of 80 % for casein and 100 % for glucose (Blaxter, 1962). Infusion periods were of 7 days and treatment effects were assessed over the last 5 days.

Experiment 3.4.4 - Effects on milk yield and composition were compared when individual cows offered constant amounts of silage received abomasal infusions of (i) 300 g/d sodium caseinate, (ii) 290 g/d glucose, (iii) (i) + (ii), (iv) normal saline. Infusions were administered according to a 4 x 4 Latin Square design. Infusion periods lasted 7 days and treatment effects were assessed over the last 5 days.

Experiment 3.4.5 - Comparisons of the effects on milk yield and composition, and nitrogen utilization, were made when individual cows receiving abomasal infusions of casein, were offered equal amounts of DE as pasture or silage. The experimental design is illustrated below:

Ration	Silage/Pasture			
Days	10	7	7	7
Infusate		S	C	S

The infusions (10 l/d) of sodium caseinate (c) (300 g/d) were interspersed with infusions of normal saline (s). Cows were initially allocated to rations on a random basis. Prior to infusions, cows were given 10 days to adjust to the ration. Infusion periods were 7 days and the last 5 were used to assess the effects of the treatments.

Infusates - Infusates were prepared each morning using de-ionized water. Casein (premium grade sodium caseinate, (Rangitaiki Plains Dairy Company, Edgecumbe, New Zealand) and glucose (dextrose monohydrate Tokai Togyo Co. Ltd., Yokkachi City, Japan) were infused at concentrations of 30 g/l and 18 g/l respectively.

These concentrations resulted in solutions similar in osmolality to that of abomasal contents of the cows fed silage. In Experiment 3.4.3 the vehicle for L-methionine (Sigma grade L-methionine, Sigma Chemical Co., St Louis, MO., U.S.A.) was normal saline. Infusion was by gravity, carried out while the cows were feeding.

Feeding and Milking - Approximately one month after calving the cows were removed from grazing and offered high moisture silage ad libitum for 10 days in individual feeding stalls. Experimental rations were then offered at 80 % of their mean daily DMI during the last 5 days of the preliminary period. In subsequent experiments the food offered was fixed according to food refusals during a 7 day preliminary period so that food intake remained constant. The silage, removed from the stack each morning, was offered at 0800-1300 h and 1600-2100 h. The fresh weight of food offered each day was based on the oven DM % of the previous days food. Between feeding periods the cows were confined to a loafing barn with access to water. In Experiments 3.4.3 and 3.4.5 the cows were continuously housed in metabolism stalls for the last 5 days of each 7 day period (see Plate 2).

Milking and milk sampling procedures were carried out as described for Experiment 3.1. Milk samples were collected over three successive periods of 2, 3, and 2 days in each week.

Sampling of Foods and Residues - Representative samples of foods and residues over the last 5 days of each period (3 days in Experiment 3.4.1) were collected and subsampled as described in

Experiment 3.1. In Experiments 3.4.3 and 3.4.5 separate collections of faeces and urine were made as in Experiment 3.1.

Sampling of Blood and Rumen Liquor - Blood samples were collected on the last day of each infusion period immediately prior to the start of feeding (0 h) and 4 h later.

In Experiment 3.4.5 samples of rumen liquor were also collected at 0 and 4 h for the determination of pH and ammonia concentration. Sampling procedures were as described for Experiment 3.1.

Analytical Procedures - Chemical analyses of foods, residues, faeces, urine and blood were as described in Experiment 3.1.

In Experiment 3.4.2, plasma was deproteinized with sulphosalicylic acid and plasma filtrates were analysed for valine, isoleucine, leucine, phenylalanine, methionine, cysteine, tyrosine, α aminon-butyric acid, and ammonia with a model NCl Technicon amino acid analyser (Technicon Co., New York) using nor-leucine as an internal standard. In Experiment 3.4.5 rumen ammonia concentrations were determined by micro-diffusion (Conway, 1957).

Statistical Analysis - In Experiments 3.4.1, 3.4.2, and 3.4.5 responses of cows to casein infusions were determined by subtracting the mean of the pre and post control saline infusion values from the value obtained during casein infusion. The mean responses were evaluated by students 't' test. Differences in the effects of treatment infusions were tested by subtracting the responses of cows to the treatment infusions. The data from Experiment 3.4.3 and 3.4.4 were subjected to analysis of variance, and differences

in the effects of the infusates were evaluated by Duncan's New Multiple Range Test.

Results

The average composition of the rations during this series of investigations is shown in Table 36.

Experiment 3.4.1 - The effects of abomasal infusions of different levels of sodium caseinate on the yield and composition of milk are summarized in Table 37. Casein infused at 300 g/d significantly increased the yields of milk and protein, and milk protein concentration. In comparison, the responses to infusions of 150 g/d of casein were less and only protein yield was significant. Neither treatment affected lactose percentage but both treatments reduced milk fat percentage although only the high casein treatment attained significance.

Despite attempts to maintain silage intake constant, infusions of casein at both levels were associated with increases in silage intake.

Casein infusates had no signficant effect on the blood constituents measured (Appendix 18).

Experiment 3.4.2 - Responses to abomasal infusions of a mixture of casein fortified with L-methionine in comparison to casein are summarised in Table 38. Addition of methionine to the casein infusate tended to increase the response in all milk variates except lactose percentage, but differences in the responses to the treatment infusions were only significant for milk fat yield.

TABLE 36. Chemical composition of diets.

Nutrient	Silage	Pasture
Dry Matter %	16.9 ± 0.28[a]	17.1 ± 0.30
Gross Energy (MJ/kg DM)	17.9 ± 0.20	17.4 ± 0.20
Total Nitrogen, % DM	2.9 ± 0.20	2.4 ± 0.14

a SED, n = 6

TABLE 37. Mean DMI and yield and composition of milk of cows on silage rations receiving abomasal infusions of 150 (LC) and 300 g/d sodium caseinate (HC), or 5 (LS) and 10 l/d (HS) of normal saline.

Variable	HC	HC-HS Diff ± SED	LC	LC-LS Diff ± SED	(HC-HS)-(LC-LS) Diff ± SED
DM intake, kg/d silage	8.27	0.98 ± 0.17*	7.91	1.00 ± 0.29	-0.02 ± 0.44
Milk, kg/d	10.16	1.38 ± 0.19**	8.60	0.31 ± 0.25	1.07 ± 0.29
Milk Fat, g/d	404	22 ± 15.8	344	-18 ± 45.1	41 ± 31
Milk Protein, g/d	306	53 ± 8.0**	252	15 ± 6.3 +	38 ± 7.6*
Milk Lactose, g/d	495	66 ± 8.1	421	17 ± 12.1	49 ± 10.5*
Milk Fat %	3.99	-0.35 ± 0.13+	4.00	-0.36 ± 0.40	0.005 ± 0.30
Milk Protein %	3.02	0.14 ± 0.03*	2.94	0.09 ± 0.05	0.06 ± 0.07
Milk Lactose %	4.86	-0.02 ± 0.05	4.90	0.03 ± 0.02	0.00 ± 0.07

TABLE 38. Mean DMI and yield and composition of milk of cows on silage rations receiving abomasal infusions of 300 g/d sodium caseinate (C), 300 g/d sodium caseinate plus 12 g/d L-methionine (CM) or 10 l/d saline (S).

Variable	C	C-S Diff ± SED	CM	CM-S Diff ± SED	(CM-S)-(C-S) Diff ± SED
DM intake, kg/d silage	6.48	0.30 ± 0.11	6.46	0.34 ± 0.007*	0.05 ± 0.15
Milk kg/d	7.90	1.18 ± 0.18**	8.37	1.52 ± 0.24	0.34 ± 0.23
Milk Fat g/d	354	69 ± 11.5**	394	109 ± 10.9**	40 ± 12.4*
Milk Protein g/d	229	43 ± 8.7	248	56 ± 6.9**	13 ± 11.4
Milk Lactose g/d	379	55 ± 6.2**	390	60 ± 14.0*	5 ± 13.8
Milk Fat %	4.50	0.28 ± 0.24	4.71	0.55 ± 0.11*	0.28 ± 0.27
Milk Protein %	2.90	0.13 ± 0.05+	2.97	0.17 ± 0.04*	0.04 ± 0.08
Milk Lactose %	4.80	-0.02 ± 0.06	4.69	-0.11 ± 0.08	-0.09 ± 0.13

Compared to the adjacent control, both treatment infusions increased milk yield, milk protein yield and milk protein concentration. However these responses were both associated with small but significant increases in silage intake.

Both treatments raised plasma ∝ amino N concentrations (Appendix 18) and the responses were significantly greater for the methionine - casein infusate. In comparison to the control saline, the methionine-casein infusate significantly increased the plasma concentrations of valine, methionine, isoleucine, leucine, tyrosine, and ∝ amino-n-butyric acid, whereas the casein infusate increased the levels of valine, isoleucine, leucine, and tyrosine (Figure 29, Appendix 19). The mean responses in plasma amino acid concentrations were greater after 4h infusion of casein plus methionine compared with casein alone although the differences in these responses were not significant.

Experiment 3.4.3 - Milk yield and composition from cows infused with casein, glucose, methionine, or control saline are shown in Table 39. Glucose had no effect on milk production in comparison to control saline whereas both casein and methionine significantly increased milk protein percentage and milk protein yield when compared with the control and glucose infusions. The magnitude of the responses to infusions of casein and methionine were similar.

Casein increased N intake, N digestibility, N absorption and N retention compared with the other infusates (Table 40). Casein and methionine both increased milk N output. Casein also altered the partition of dietary N, by decreasing urinary nitrogen, and increasing nitrogen retained in tissue (NR) and in productive nitrogen (NR + milk N) as a percentage of DNI. Methionine increased

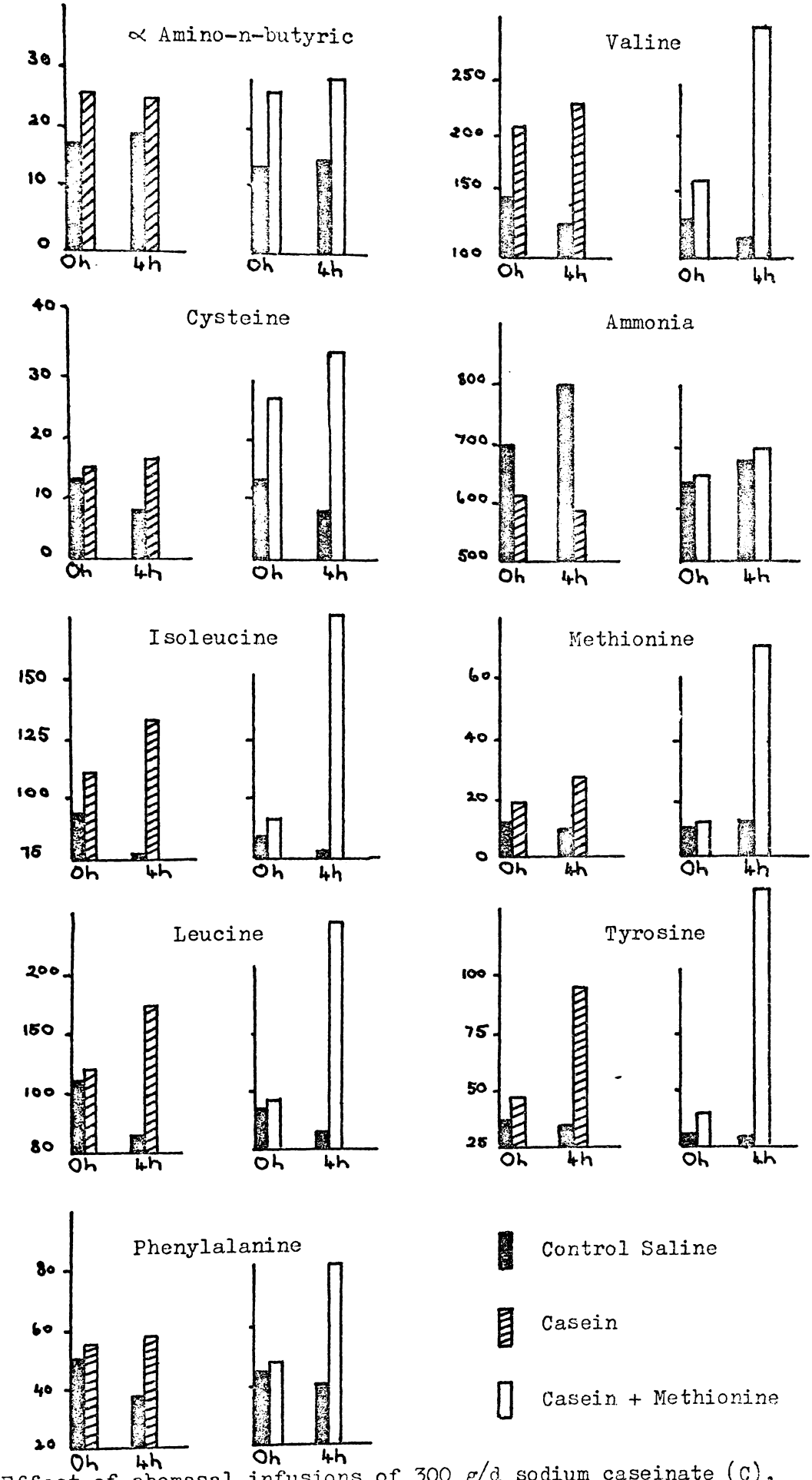


Fig. 29. Effect of abomasal infusions of 300 g/d sodium caseinate (C), or (C) plus 12 g/d L-methionine (CM) in comparison to saline infusions (S) on the concentration of plasma amino acids (u moles/l) of cows on silage rations immediately before feeding (Oh) and 4h later.

TABLE 39. Mean DMI, milk production and milk composition of cows receiving abomasal infusions of sodium caseinate (300 g/d), glucose (290 g/d), L-methionine (12 g/d), or saline (10 l/d).

Variable	Infusate				SED	Sign.
	Casein	Glucose	Methionine	Saline		
DM intake (kg/d)						
Silage	5.850a	5.903a	5.947a	5.973a	±0.0546	NS
Infusate	0.291	0.284	0.012	0.090		
TOTAL	6.141a	6.187a	5.959b	6.063ab	±0.0545	*
DM Digestibility %	67.9a	68.2a	67.5a	67.8a	±0.64	NS
Milk (kg/d)	5.47a	5.01a	5.23a	4.88a	±0.253	NS
Milk fat (g/d)	261a	240a	253a	236a	±23.4	NS
Milk protein (g/d)	179a	152b	172a	150b	±7.4	*
Milk lactose (g/d)	266a	244a	255a	237a	±13.0	NS
Milk fat %	4.98a	4.79a	4.85a	4.79a	±0.294	NS
Milk protein %	3.27a	3.02b	3.29a	3.08b	±0.035	***
Milk lactose %	4.85a	4.87a	4.88a	4.86a	±0.037	NS

Means not containing common letters differ significantly ($P<0.05$).

TABLE 40. Nitrogen (N) intake and utilization by cows on silage rations and receiving abomasal infusions of sodium caseinate (300 g/d), glucose (290 g/d), L-methionine (12 g/d), or saline (10 l/d).

Variable	Infusate				SED	Sign.
	Casein	Glucose	Methionine	Saline		
N Intake (g/d)						
Consumed	191a	193a	195a	195a	± 1.8	NS
Infused	42	-	1	-		
TOTAL	233a	193b	196b	195b	± 1.7	***
N Digestibility %	73.6a	70.3b	70.6b	69.9b	± 1.12	+
N Content (%) in:						
Milk	0.51a	0.47b	0.52a	0.48b	± 0.005	***
Faeces	3.13a	2.92a	2.96a	3.01a	± 0.083	NS
Urine	0.61a	0.51b	0.42c	0.45bc	± 0.030	**
N Excreted (g/d) in:						
Milk	28a	24b	28a	24b	± 1.3	*
Faeces	61a	57a	58a	59a	± 2.6	NS
Urine	125a	111b	108b	109b	± 4.0	*
N Absorbed (g/d)	172a	136b	138b	137b	± 2.5	***
N.Retained (g/d)	19a	1b	3b	4b	± 4.6	*
Productive N (g/d) (Milk + NR)	48a	25b	31b	28b	± 4.7	*
N % Intake:						
Faeces	26.4b	29.7ab	29.5ab	30.1a	± 1.12	+
Urine	53.8a	57.6a	55.3a	56.0a	± 1.53	NS
Milk	12.1b	12.6ab	14.0a	12.1b	± 0.64	+
Retained	7.7a	0.1b	1.4b	1.8b	± 1.92	*
Productive	19.8a	12.7b	15.3ab	14.1b	± 2.00	+
N % Absorbed						
Urine	73.4b	82.0a	78.3ab	80.4a	± 2.69	+
Milk	16.5b	18.6ab	19.9a	17.4ab	± 1.04	+
Retained	10.1a	0.1b.	1.9b	2.3b	± 2.66	*
Productive	26.6a	18.0b	21.6ab	19.9b	± 2.59	+

Means not containing commen letters differ significantly ($P<0.05$)

nitrogen in milk as a percentage of DNI, whereas glucose had no effect on N partition. The effects of the infusates on energy partition were small (Appendix 20).

Casein infusion tended to increase plasma urea concentration although this was only significantly different when compared with the methionine treatment at 4h (Appendix 21). Infusing casein increased plasma levels of α amino N whereas the other infusates tended to reduce α amino N, the difference being significant.

Experiment 3.4.4 - Responses in milk yield and milk composition of cows receiving infusions of casein, glucose, and casein plus glucose, relative to control saline infusions are summarised in Table 41. Relative to infusions of saline or glucose, casein and casein plus glucose infusions increased milk yield, milk protein yield, and milk protein percentage and the interaction of glucose plus casein on milk protein percentage was significant. Silage intakes were slightly but significantly less for those animals infused with glucose compared with saline or the other infusates.

The casein infusion significantly increased plasma α amino N concentration in comparison to saline or glucose infusions, with the casein plus glucose infusions having an intermediate effect (Appendix 22). Plasma urea tended to be higher in animals infused with casein compared with other infusates but the increase was not significant.

Experiment 3.4.5 - Effects of abomasal infusions of casein on milk yield and milk composition of cows consuming similar quantities of DE as pasture or silage are summarized in Table 42. When cows were infused with control saline, the yield of milk protein and milk protein concentration was significantly higher for cows fed

TABLE 41. Mean DMI and yield and composition of milk of cows on silage rations and receiving abomasal infusions of sodium caseinate (C, 300 g/d), glucose (G, 290 g/d), glucose plus sodium caseinate (G+C), or saline (S, 10 l/d).

Variable	Infusate					Significances		
	C	G	G+C	S	SED	C	G	CxG
DM Intake (kg/d)								
Silage	6.45	5.92	6.31	6.18	±0.12	*	+	NS
Infusate	0.29	0.28	0.57	0.09				
TOTAL	6.74	6.20	6.88	6.27				
Milk Yield (kg/d)	4.24	3.55	4.19	3.61	±0.15	**	NS	NS
Fat Yield (g/d)	203	192	203	193	±14.0	NS	NS	NS
Protein Yield (g/d)	147	120	154	121	± 4.8	***	NS	NS
Lactose Yield (g/d)	196	163	190	165	± 7.6	**	NS	NS
Fat %	4.74	5.27	4.80	5.27	± 0.24	*	NS	NS
Protein %	3.46	3.34	3.68	3.34	± 0.05	**	*	*
Lactose %	4.62	4.55	4.53	4.58	± 0.04	NS	NS	NS

TABLE 42. Mean DEI and yield and composition of milk by cows fed pasture (P) or silage (S) rations and receiving abomasal infusions of sodium caseinate (c, 300 g/d) or saline (s, 10 l/d).

Variable	Treatments		Pc-Ps		Sc-Ss		(Pc-Ps)-(Sc-Ss)		Ps-Ss	
	Pc	Sc	Diff ± SED	Sign.	Diff ± SED	Sign.	Diff ± SED	Sign.	Diff ± SED	Sign.
DEI (MJ/d)	89.1	96.1	5.14 ± 1.64	*	12.18 ± 3.13	*	7.04 ± 3.58	NS	0.4 ± 2.92	NS
Milk (kg/d)	11.23	11.54	0.33 ± 0.33	NS	1.64 ± 0.22	**	1.31 ± 0.23	**	0.85 ± 0.381	NS
Milk Fat (g/d)	563	510	34.2 ± 12.7	+	77.6 ± 19.5	*	43.4 ± 45.5	NS	82.1 ± 15.82	*
Milk Protein (g/d)	334	320	18.6 ± 7.5	+	63.6 ± 7.5	**	45.0 ± 17.5	+	55.1 ± 8.2	**
Milk Lactose (g/d)	551	567	15.8 ± 16.3	NS	79.0 ± 7.87	***	63.2 ± 31.1	NS	39.7 ± 22.4	NS
Milk Fat %	5.01	4.43	0.11 ± 0.098	NS	0.02 ± 0.122	NS	-0.08 ± 0.23	NS	0.40 ± 0.18	NS
Milk Protein %	3.00	2.79	0.08 ± 0.028	*	0.18 ± 0.026	**	0.09 ± 0.058	NS	0.32 ± 0.052	*
Milk Lactose %	4.92	4.94	0.002 ± 0.013	NS	-0.02 ± 0.035	NS	-0.02 ± 0.042	NS	-0.04 ± 0.04	NS

pasture than cows fed silage. Abomasal infusions of casein to pasture fed cows resulted in small but significant increases in milk protein percentage and milk protein yield. In comparison casein infusions resulted in large increases in the yields of milk, protein and milk protein percentage when silage was fed. Differences between the rations in the responses of cows to casein infusions were significant for the yields of milk and protein. Although the difference in the response to milk protein percentage was not significant, four of the five cows had higher concentrations of protein in milk when infused with casein on the silage ration compared with their responses on the pasture ration.

Infusions of casein increased total DE intakes of both rations but the significantly higher DE intake on the silage ration was due to a small increase (6 %) in silage intake when casein was infused.

The relative effects of casein infusion on nitrogen utilization of cows on pasture or silage rations are shown in Table 43. Pasture contained less nitrogen, of lower digestibility than silage. Although pasture fed cows had lower nitrogen intakes they secreted more nitrogen in milk in comparison to the silage fed cows when both were infused with saline. As percentages of absorbed nitrogen, milk nitrogen and urinary nitrogen were higher, and nitrogen retention less, when the animals were fed pasture compared with silage.

Expressed as a percentage of dietary nitrogen, abomasal infusions of casein increased nitrogen retention and reduced nitrogen in milk to a greater extent when the animals were fed pasture in comparison to silage.

The effects of the infusions on the blood and rumen parameters are summarized in Appendix 23. When saline was infused, plasma urea levels were higher in the animals fed silage although rumen ammonia levels tended to be slightly higher in animals fed pasture.

TABLE 43. Nitrogen (N) intake and utilisation by cows fed pasture (P) or silage (S) rations receiving abomasal infusions of sodium caseinate (c 300 g/d) or saline (s, 10 l/d).

Variable	Treatments		Pc-Ps		Sc-Ss		(Pc-Ps)-(Sc-Ss)		Ps-Ss	
	Pc	Sc	Diff ± SED	Sign.	Diff ± SED	Sign.	Diff ± SED	Sign.	Diff ± SED	Sign.
N Intake (g/d)										
Consumed	169.4	210.2	1.1 ± 1.9	NS	5.4 ± 5.03	NS	4.3 ± 8.1	NS	-46.5 ± 7.6	**
Infused	42.0	42.0	42.0 ± 0.0	***	42.0 ± 0.0	***	0.0 ± 0.0	NS	- -	-
TOTAL	211.4	252.2	43.1 ± 1.9	***	47.4 ± 5.0	***	4.3 ± 8.1	NS	-46.5 ± 7.6	**
N Digestibility %	69.3	71.4	7.89 ± 1.07	**	3.14 ± 0.69	*	-5.41 ± 1.56	*	-7.74 ± 1.92	+
N Content (%) in:										
Milk	.453	.437	0.02 ± 0.007	*	0.028 ± 0.004	**	0.008 ± 0.014	NS	.042 ± .007	**
Faeces	3.04	2.96	-0.34 ± 0.375	NS	0.12 ± 0.065	NS	0.495 ± 0.498	NS	.55 ± .007	*
Urine	.378	0.550	0.04 ± 0.019	NS	0.14 ± 0.021	**	0.106 ± 0.042	*	-0.19 ± .019	NS
Food	2.39	2.84	0.03 ± 0.044	NS	-0.12 ± 0.012	***	-0.152 ± 0.045	*	-.60 ± .01	**
N Excreted (g/d) in:										
Milk	52.4	50.0	3.8 ± 1.2	*	9.8 ± 1.2	**	6.0 ± 2.8	+	8.4 ± 1.5	*
Faeces	64.2	71.9	0.13 ± 3.54	NS	7.3 ± 2.1	*	9.0 ± 5.8	NS	1.25 ± 3.02	NS
Urine	98.2	124.4	10.6 ± 3.3	*	17.6 ± 4.4	*	8.3 ± 4.7	+	-21 ± 8.6	NS
N Absorbed (g/d)	146.5	180.4	43.4 ± 2.7	***	40.2 ± 3.1	***	-1.5 ± 8.5	NS	-43.5 ± 6.7	*
N Retained (g/d)	-5.9	6.0	29.3 ± 3.1	**	12.8 ± 4.7	+	-17.1 ± 7.1	*	-31.1 ± 5.7	*
Productive N (g/d)	48.3	56.0	32.8 ± 4.0	**	22.2 ± 4.5	**	-10.3 ± 9.1	NS	-23.0 ± 3.6	*

Cont.

TABLE 43. (Cont.)

Variable	Treatments		Pc-Ps		Sc-Ss		(Pc-Ps)-(Sc-Ss)		Ps-Ss	
	Pc	Sc	Diff ± SED	Sign.	Diff ± SED	Sign.	Diff ± SED	Sign.	Diff ± SED	Sign.
N % Intake										
Faeces	30.6	28.4	-7.86 ± 1.05	**	-3.31 ± 0.66	**	5.18 ± 1.77	+	7.7 ± 1.9	+
Urine	46.4	49.5	-6.33 ± 3.00	NS	-3.09 ± 1.94	NS	3.61 ± 1.92	NS	1.14 ± 2.94	NS
Milk	25.7	19.8	-4.70 ± 0.89	*	0.07 ± 0.39	NS	4.86 ± 0.74	**	10.24 ± 1.04	**
Retained	2.7	2.3	18.91 ± 3.31	*	6.35 ± 2.48	+	-13.2 ± 2.97	*	-19.0 ± 2.06	**
Productive	23.0	22.1	14.21 ± 3.21	*	6.23 ± 2.32	+	-8.60 ± 3.73	NS	-9.08 ± 2.57	+
N % Absorbed										
Urine	66.7	66.7	-18.98 ± 5.03	*	-8.02 ± 2.94	+	11.75 ± 4.88	+	11.0 ± 3.97	NS
Milk	37.1	26.2	-9.83 ± 2.36	*	-3.26 ± 1.32	+	6.45 ± 4.09	NS	15.3 ± 3.8	+
Retained	3.8	5.2	32.19 ± 5.36	**	9.28 ± 3.48	+	-24.10 ± 4.72	*	-32.2 ± 2.8	**
Productive	33.3	33.7	18.19 ± 5.03	*	8.42 ± 2.93	*	-11.26 ± 5.19	NS	-10.5 ± 3.9	NS

Casein infusions had no effect on plasma urea concentrations in animals on either ration but plasma α amino N concentrations were increased to a similar extent on each ration. The effects of diet or casein infusions on rumen pH and plasma ammonia concentration were small.

Discussion

Effect of Casein on Milk Protein Synthesis - Cows fed pasture silage consistently responded to abomasal infusions of casein by producing more milk with a higher protein percentage. These cows, producing 3.6-10.9 kg milk daily and receiving 300 g/d casein, increased milk yield by 1.08 kg/d, representing an average increase in milk yield of 15.8 %. Milk protein percentage was increased by an average 0.15 percentage units and milk protein yield by 43 g/d or 21.8 %.

In other studies post-ruminal administration of casein to cows fed diets of concentrates with various roughages, to meet NRC (1971) standards for energy and protein, have improved the yields of milk by 5-13 % and milk protein by 9-16 % (Broderick et al. 1970; Hale et al. 1972; Clark et al. 1973; Spires et al. 1973; Derrig et al. 1974; Vik-mo et al. 1974). The magnitude of responses was greater for high producing cows (Vik-mo et al. 1974; Clark, 1975).

In constrast, in the present experiment, cows at low levels of production responded to abomasal infusions of casein. Although food intakes were also low according to NRC (1971) standards, they contained adequate energy and an excess of DCP for the levels of production observed. Even in Experiment 3.4.4 when the cows were in mid lactation and consuming similar quantities of silage, as in the earlier experiments, but producing approximately half the

quantity of milk, responses in milk protein yield to casein infusions were still obtained.

Level of Casein on Milk Protein Synthesis - Ørskov and Grubb (1977) obtained consistent increases in milk protein yield when increments of casein were abomasally infused in conjunction with glucose to cows on low protein(13.1 % CP) diets, whereas Spechter (1972) obtained increases in protein yield of 140 g/d, 125 g/d and 180 g/d when 450, 785, and 810 g/d casein was infused into the abomasum of cows on corn plus urea diets.

In Experiment 3.4.1, increasing the amount of casein infused into the abomasum from 150 g/d to 300 g/d resulted in an average increase in milk protein yield from 15 g/d to 53 g/d representing increases of 6.3 % and 20.9 % respectively. Moreover in Experiment 3.4.2 addition of methionine to casein increased milk protein yield from 23 % to 30 % above the adjacent control infusions. These results suggest an increasing response in milk yield to additional amino acids supplied post-ruminally to cows on silage rations.

Effect of Casein Infusions on Intake - Several investigations have reported that abomasal supplementation of 300 to 600 g/d casein had no effect on feed intake (Hale et al. 1972; Clark et al. 1973; Spires et al. 1973; Derrig et al. 1974; Vik-mo et al. 1974). However in Experiments 3.4.1, 3.4.2, and 3.4.5 when the animals were switched from casein onto saline infusions their intake of silage dropped by 12 %, 5 %, and 6 % respectively. Since milk yield and milk protein concentration have been associated with level

of energy intake (Huber and Bowman, 1966a; Balch, 1972), the responses obtained may therefore be explained in terms of extra energy from increased silage intakes and from the extra gluconeogenic amino acids supplied rather than increasing the supply of amino acids per se for milk synthesis. However previous Experiments (3.1 - 3.3) have shown the response between silage digestible energy intake and milk protein yields to be low. Moreover in Experiment 3.4.1 both levels of infusion increased silage intake by approximately 12 % and since 300 g/d casein accounts for less than 5 % of the total energy ingested the marked differences in protein yield response between the treatments were apparently not due to differences in energy intake. This is supported by Experiment 3.4.3 when marked responses were observed for casein infusions and silage intakes were the same for the control and treatment infusions.

Effect of Glucose Infusion - Experiments 3.4.3 and 3.4.4 indicated that infusions of glucose to provide similar quantities of metabolizable energy as casein was without effect on milk protein synthesis. This validates that the response to casein was mediated by an improved supply of amino acids to the mammary gland rather than glucose derived from the amino acids supplied in the casein. Spechter (1972) and Clark, Spires and Derrig, (1973) also showed that when cows were fed on concentrate-roughage rations abomasal infusions of casein but not glucose increased milk and protein yields. However, Vik-mo et al. (1974) did observe responses to glucose administered post-ruminally but the magnitude of the responses were much lower than those obtained with isocaloric casein infusions.

Some post-ruminal infusion studies (Ørskov et al. 1971;

Tyrrell et al. 1972) have indicated that intestinal absorption of glucose by ruminants may be poor. In the current study only small amounts of glucose were infused. There was no evidence of increased faecal energy excretion and consistency of faeces was unchanged.

The lack of a response to glucose in the present trial could be due to the low amount of energy supplied by the glucose supplement (5 %) relative to the total intake of DE. Even so the glucose served as an energy control and the results indicate that amino acids per se were limiting milk protein synthesis of cows on silage rations rather than glucose, despite the major role of glucose in mammary metabolism (Kleiber, Black, Brown, Blaxter, Luick and Stadtman, 1957; Bickerstaffe, Annison and Linzell, 1974).

Effects of Casein Plus Glucose Infusions - Vik-mo et al. (1974) and Ørskov and Grubb (1976) did not observe any synergistic effect of casein and glucose on milk protein synthesis. Whether there was a synergistic effect between glucose and casein infusates on milk protein synthesis in the present studies on silage diets is inconclusive. Although glucose and casein increased milk protein concentration, milk yield was slightly less than that obtained with the casein infusate and overall milk protein yields were similar.

Effects of Methionine Infusions - The recovery of infused casein in the present experiments was relatively low but tended to be higher in Experiments 3.4.1 and 3.4.5 when the cows were in early lactation and producing the highest levels of milk protein. Hogan (1975) calculated that an amino acid deficiency, especially methionine, is more likely to occur in early lactation. In

Experiment 3.4.2, 300 g/d of casein resulted in an extra 43 g/d milk protein synthesized but addition of 12 g/d methionine to the casein increased recovery of protein to 56 g/d. Moreoever in Experiment 3.4.3, 12 g/d methionine increased milk yield, milk protein percentage and milk protein yield to the same extent as 300 g/d casein. These results suggest that methionine may be the essential amino acid that is limiting milk protein synthesis on silage rations. Barry et al. (1973) also concluded that methionine might be limiting in silage since marked increases in intake and wool growth of sheep on silage was demonstrated by intra-peritoneal administration of methionine.

Other direct evidence of responses in milk protein production to methionine supplements is limited to the one study of Fisher (1972), although it is noted that the response was confounded by intake. Based on examination of plasma amino acid concentrations relative to mammary uptake, it has been predicted (Chandler and Polan, 1972; Derrig et al. 1974) that methionine is limiting for cows on concentrate-roughage rations. However intravenous infusions of methionine (Fisher, 1969; Teichman et al. 1969), or feeding encapsulated methionine (Broderick et al. 1970; Williams et al. (1970), or abomasal infusions of methionine (Schwab et al. 1976), have not stimulated milk protein synthesis.

Effects of Casein Infusions with Different Diets - The differences during saline infusions in the yield and concentration of milk protein between the rations of pasture and silage when fed to provide the same levels of DEI (Experiment 3.4.5) agree with the results obtained in Experiment 3.1.

Abomasal infusions of casein increased the digestible energy intake of both rations but the higher DE intake on the silage ration was due to a small increase in silage intake. Since responses were obtained to casein infusions in Experiment 3.4.3 when silage intakes were constant and glucose infusions had no effect in Experiments 3.4.3 and 3.4.4, the response to casein by the silage fed animals in this experiment is presumably due to increased availability of amino acids for milk protein synthesis. The results suggest the differences in the responses of cows on the silage and pasture rations to casein infusion was due to an inadequate supply of essential amino acids available for digestion in the lower gut of animals on the silage ration. The small response of cows on the pasture ration to casein infusions could be due either to an increased supply of glucose or amino acids for milk protein synthesis.

Nitrogen Utilization - Abomasal infusions of casein to cows on silage diets improved the efficiency of nitrogen utilization by reducing the proportion of nitrogen intake excreted in faeces and urine. The lower percentage of digested nitrogen in urine was presumably due to the decrease in the proportion of nitrogen absorbed as ammonia and improved nitrogen retention, as a result of the increase in quantity and quality of the amino acids provided by the protein supplement. Abomasal infusion of casein increased the amount of milk nitrogen secreted but tended to decrease the efficiency of utilization of absorbed nitrogen for milk protein synthesis. In comparison methionine improved milk nitrogen output and the efficiency of utilization of nitrogen for milk protein synthesis.

In Experiment 3.4.5, the proportion of dietary nitrogen retained by cows was significantly less on pasture than silage when cows were infused with saline, reinforcing the results obtained in Experiment 3.1. The tendency for the animals to excrete more nitrogen in urine when feeding on pasture compared with silage is presumably the result of increased catabolism of tissue protein. Although the amount of nitrogen absorbed by cows fed pasture was less than on silage, milk nitrogen excretion was higher for the same level of digestible energy intake. The increased milk protein synthesis on pasture rations was presumably due to a more adequate supply or balance of amino acids to meet the requirements of milk protein synthesis derived from the diet or from tissue protein catabolism. In a recent experiment, Ørskov and Grubb (1977) infused casein into the abomasum of cows on a low protein diet and observed an increase in milk production. The authors calculated that the higher production was associated with an increased dietary energy deficit and proposed the effect of protein was mediated by stimulating mobilisation of body energy reserves. Similar observations were made in Experiment 3.1 where cows fed pasture produced more milk but lost more weight than cows fed silage at the same digestible energy intake.

In Experiment 3.4.5. abomasal infusions of casein to pasture fed cows resulted in only a marginal increase in milk and milk protein synthesis. In comparison to silage, the infusion of casein with pasture had a much greater effect on nitrogen retention and a lower effect on milk nitrogen suggesting the response on pasture was due to a correction of a minor imbalance of amino acids or to a small increase in energy provided from the gluconeogenic amino acids since

approximately 25 % of the nitrogen infused was excreted in urine.

Effect of Infusions on Blood Constituents - Abomasal infusions of casein did not appear to change plasma volume as no consistent trends in packed cell volume were observed. However infusions of casein tended to increase blood urea nitrogen levels in Experiments 3.4.3 and 3.4.4 suggesting greater deamination and increased urinary nitrogen loss, reducing the efficiency of dietary protein conversion to milk protein. Broderick et al (1974) and Vik-mo et al. (1974) also observed similar effects on blood urea levels with post-ruminal protein supplementation.

Since concentrations of free amino acids in plasma reflect an equilibrium between intestinal absorption, endogenous synthesis and utilization, changes in concentrations during treatment relative to control have been used as an index for predicting the adequacy of the diet. Rook and Line (1961) observed increases in α amino N levels in plasma were associated with the plane of nutrition and milk protein yields. In the present experiments (except 3.4.1) abomasal infusions of casein consistently increased the plasma α amino N concentrations, and casein plus methionine had a much larger effect than casein indicating the amount of amino acids being made available for milk synthesis was increased. These results however are in marked contrast with those of the previous feeding experiments (3.1 - 3.3) where no relationship was found between α amino N concentrations and milk protein synthesis.

Generally, increases in essential amino acids relative to non-essential amino acids have been observed during abomasal infusions of casein with the most prominent changes being an increase in the

branched chain amino acids (Clark, 1975). In Experiment 3.4.2 all the essential amino acids measured in plasma increased with abomasal infusion of casein. The magnitude of the effects were greater with casein plus methionine infusions, and since milk yield, milk protein and milk protein percentage followed similar trends, then the levels of essential amino acids in plasma would appear to be associated with milk protein synthesis.

The effect of methionine supplementation on the increased plasma methionine levels has also been observed by Reis and Tunks (1971) and by Reis et al. (1973). The associated increases in cysteine and ∝ amino-n-butyric acid are presumably because these are products of methionine metabolism (Finklestein, 1974). The increases in the other essential amino acids during casein-methionine infusion compared with the casein infusion indicate that besides being a major amino acid for milk protein synthesis, methionine may also have a general metabolic role such as transport of amino acids (Christensen, 1963, 1964) or in protein metabolism (Girard-Globa, Robin and Forester, 1972; Munro, 1976).

Effect of Infusions on Milk Fat Synthesis - Generally abomasal infusions of casein cause a reduction in milk fat percentage (Clark et al. 1973; Spires et al. 1973; Derrig et al. 1974; Vik-mo et al. 1974). This effect is apparently not due to a decrease in milk fat synthesis as fat yields tend to be slightly increased but is a result of a proportionately larger increase in milk yield. In the present experiments abomasal infusions of casein had no consistent effects on milk fat percentage although fat yield was significantly increased in two experiments.

Methionine significantly increased fat percentage and fat yield in Experiment 3.4.2 when infused with casein but in Experiment 3.4.3 methionine infused alone had no effect in comparison to the control saline infusion. Ruminal supplementation with methionine hydroxy analogue has been shown to increase fat percentage and fat yield in lactating cows but the mechanism is unknown, although rumen lipid synthesis (Patton, McCarthy and Griel, 1968, 1970; McCarthy, Patton and Griel, 1970) and liver metabolism have been implicated (McCarthy, Porter and Griel, 1968).

Effect of Infusions on Milk Lactose Synthesis - Abomasal infusions of casein or methionine had no effect on the lactose concentration in milk. This is possibly because milk secretion is regulated by osmotic pressure exerted mainly by lactose (Rook, 1976).

Lactose yield tended to be increased by abomasal infusions of casein. Provision of additional amino acids may meet the extra requirement for a minor milk protein, α lactalbumin, which is also a component of a rate-limiting enzyme involved in lactose synthesis (Brew, 1970). These factors may explain the response in milk yield to post ruminal protein supplements.

This study has indicated that increased yields of milk and milk protein and milk protein concentrations have been obtained from cows fed silage and receiving intra-abomasal infusions of casein. The implication of these experiments is that methionine may be the major amino acid limiting milk protein synthesis of cows on silage diets. Responses to casein infusions of silage diets were markedly increased in comparison to pasture. The small response of post-ruminal protein supplements to pasture requires further investigation

to determine if the quantity and quality of available protein on pasture diets is limiting milk protein synthesis.

4. GENERAL DISCUSSION

Although cows are generally fed exclusively on forage diets in N.Z. there is little information on the extent that diet type *per se*, and level of feeding affect milk yield and composition. Most published reports do not permit the separation of the effects of type of diet from the effects of level of feeding (Wilson and McDowell, 1966; Hutton and Parker, 1966; Wilson and Dolby, 1967, 1969; Hutton *et al*. 1971; Bryant and Donnelly, 1974; Lancaster *et al*. 1974; Hutton, 1975). Separation of these effects was a major consideration in designing the experiments reported in this thesis.

A regression approach was used so that comparisons between diets could be made at equal intakes of DE. With this approach it was established that the relationships between DEI and milk components were significant for the pasture diet. This was not so for the silage ration, possibly because of the small range of intakes due to the low voluntary intake of silage, and to the variation in milk production responses within that range of intakes. In using the regression technique to make comparisons between the treatments it was necessary to examine whether one relationship could describe the data. If not, alternative models with either the slope or intercept differing between treatment rations were used implying that at the same level of feeding different productions were obtained from the rations. Experiments 3.2 and 3.3 were designed so that the treatment effects could also be assessed by analysis of variance. The results of these analyses largely substantiated those of the regression analyses.

The studies have clearly demonstrated that cows fed on high

moisture silage produced less milk with a lower concentration of fat and protein than cows fed pasture at similar intakes of DE.

Subsequent experiments were aimed at obtaining evidence of the mechanisms involved. The effects of treatments that either reduced protein degradation during ensiling, provided pasture or protein concentrates as supplements to silage, or casein directly to the abomasum are consistent with the view that the amount of protein reaching the duodenum is a major factor involved. Furthermore because abomasal infusions of methionine in Experiment 3.4.3 had effects similar to casein it is likely that methionine may be the major essential amino acid limiting milk protein synthesis by cows fed silage. It is noted however that methionine may also have a general role in controlling protein metabolism (Christensen, 1963, 1964; Girard-Globa et al. 1972; Munro, 1976).

The evidence obtained during the course of this work does not define the reasons why the quantity and/or the quality of protein reaching the duodenum of cows on silage rations was limiting. The low true protein content coupled with the high solubility of the crude protein and the low content of readily available fermentable energy in silage may result in insufficient dietary or microbial protein reaching the duodenum. To distinguish between these possibilities would require a more detailed approach than was possible or desirable in this instance. Quantitative studies of the amounts of dietary and microbial protein entering and absorbed from the duodenum together with their amino acid compositions would appear necessary. Clearly the information obtained on blood parameters were of limited usefulness in distinguishing between

these possibilities.

The failure to improve milk yield and composition by dietary additions of maize silage suggests that if the synthesis of microbial protein was limiting, it was not because of the availability of fermentable energy (Smith , 1969; Hogan and Weston, 1969).

Of particular interest is the comparison of the effects of abomasal infusions of casein to cows fed either pasture or silage. Responses in milk yield and milk protein to infusions of casein were obtained on both diets but the magnitude of the response was greater for the silage diet. No other studies have been carried out to support this finding and it cannot be concluded that milk protein synthesis by the pasture fed cow is not limited by protein entering the duodenum. Further studies are required to verify if the present findings were due to a deficiency of amino acid(s) or energy, and to investigate effects on milk protein synthesis of post ruminal supplements of protein to cows fully fed on pasture.

Where changes in the levels of blood parameters, ruminal VFA concentrations and proportions were measured, a consistent feature was that the data obtained were of limited value in interpreting the mechanisms involved in milk composition changes. This was so even though changes in rumen fermentation patterns (Armstrong and Prescott, 1971; Balch, 1972) and intra-ruminal infusions of VFA (Rook and Balch, 1961; Rook et al. 1965; Wilson et al. 1967) have been associated with consistent changes in milk yield and composition. In the present studies the effects of ration type on the molar proportions and ratios of acetate and propionate were generally small and appeared to be of little consequence on milk yield and composition.

However the molar percentage of rumen butyrate was reduced by silage in three experiments and if this reflects the relative absorption rates (Leng, 1970b), this may have contributed to the lower milk fat synthesis on these diets, since butyrate is metabolized to B-hydroxybutyrate, a precursor of milk fat (Smith et al. 1974).

The fall in milk fat synthesis may also be caused by a lower supply of endogenous fat precursors if the lower live-weight loss of cows fed silage in Experiment 3.1 reflects a lower mobilization of body fat. Diets causing low milk fat have been associated with increased rumen propionate levels (McCullough, 1966; Armstrong and Prescott, 1971; Annison et al. 1974), and reduced plasma triglycerides and acetate (Annison et al. 1974). Possibly glucogenic materials increase plasma insulin levels promoting lipogenesis thereby reducing the supply of milk fat precursors (McClymont and Vallence, 1962; Annison, 1976). The responses of cows fed silage in Experiment 3.1 were also associated with increased proportions of rumen propionate although the relative changes were small. However the low fat production of cows on silage in Experiments 3.2 and 3.3 were not consistently related to changes in rumen propionate percentage or related to live-weight change. Since changes in live-weight in the short term may not be a true indication of relative changes in tissue more definitive studies would be required to establish if this is a contributing factor to low milk fat synthesis on silage diets.

Generally abomasal infusions of casein and methionine tended to increase milk fat synthesis although only on two occasions were the increases significant. Although this finding is in agreement with other studies (Clarke et al. 1973; Spires et al. 1973;

Broderick et al. 1970), apparent mechanisms to explain possible effects of dietary protein on milk fat synthesis are obscure.

Decreased synthesis of milk and lactose on silage rations may be due to lower amounts of protein reaching the duodenum since abomasal infusions of casein in contrast to glucose consistently increased the yields of milk and lactose. A possible explanation of the specific effects of amino acids may be due to increased synthesis of α lactalbumin, a minor milk protein, and a component of a rate-limiting enzyme for lactose synthesis (Brew, 1970). Since lactose is a major osmotic regulator of milk (Rook, 1976), increased lactose would increase milk volume.

The present study has also shown that the level of feeding of forage rations affect milk yield and composition. Increasing pasture intake (Experiment 3.1) increased the yield of milk and constituents, milk protein percentage, and decreased milk fat concentration, in agreement with other studies (Hutton, 1974; 1975; T.E. Trigg, unpublished).

When results of Experiments 3.1, 3.2 and 3.3 (Table 10, Appendices 6, 9, 14) were compared, the relationships between intake of silage and the yields of milk, fat, protein and lactose were all positive and for milk fat percentage were negative. Although these relationships were consistent they were not always significant. The relationships between silage intake and the percentages of protein and lactose in milk were small and non significant. When these data from Experiments 3.1, 3.2 & 3.3, for cows fed exclusively on silage rations are combined (data from 57 animals, for 6 different silages) the pooled regression coefficients for the relationships between silage intake and the yields of milk, fat, protein and lactose, and milk fat

percentage become highly significant (Table 44). However the pooled regression coefficients for the percentages of protein and lactose remain non-significant.

Table 44. Relationship between daily milk production and silage intake ($DEI/LW^{0.75}$)

Item	bx ± SE	Sign.
Milk (kg)	3.08 ± 0.48	***
Fat (g)	135.4 ± 55.0	***
Protein (g)	86.7 ± 10.9	**
Lactose (g)	170.4 ± 50.3	**
Fat %	-0.60 ± 0.15	**
Protein %	0.04 ± 0.07	NS
Lactose %	0.11 ± 0.07	NS

This finding, that no relationship exists between milk protein percentage and feeding level for the silage ration, is in contrast to pasture and confirms the inferences of Hutton et al. (1971) and Lancaster et al. (1974). It is suggested the increase in milk protein percentage of cows fed increased amounts of pasture is due to increased amounts of essential amino acids being available for milk protein synthesis. Possible reasons for a disproportionate increase in the quantity of essential amino acids reaching the small intestine with increased pasture feeding level can only be speculative but may be due to a reduction in the loss of dietary N as ammonia from the rumen as a result of possible decreased retention time (Milne and Campling, 1972). Since in comparison to pasture the level of true protein in silage is low and the level of soluble non-protein N is high, any increase in feeding level is likely to have a much smaller effect on the amounts of dietary protein escaping rumen proteolysis, and on the levels of ammonia absorbed from the rumen than pasture. Further investigations would be

necessary to explain the reasons for the effects of feeding levels of these diets on milk protein synthesis.

The feeding experiments have shown the low production of cows on silage rations can be largely overcome by either adding formalin to the crop during harvesting, or by wilting. These processes increased milk protein synthesis by 10 and 15 %, and improved milk protein percentage by 0.12 and 0.14 percentage units respectively. In practice when these rations are fed as sole rations to milking cows larger responses may be expected, since these treatments can also increase voluntary intake of silage (Wilkinson, Wilson and Barry, 1976). More studies are now required to define optimal conditions for these treatments in terms of milk production when fed to appetite.

The experiments have also shown that protein supplements can be fed with silage to improve the performance of dairy cows. Supplementing cows with protein concentrates (17 % of the DM) resulted in a 14 % increase in milk protein synthesis and a rise of 0.16 percentage units in milk protein concentration. However these concentrates are expensive in N.Z. and unlikely to be profitable under most local conditions.

Of greater importance is the value of using silage to supplement cows grazing limited pasture. This study has shown that when silage and pasture are fed in equal quantities (1:1 on a DM basis) milk protein synthesis is improved by 16 % and milk protein concentration is increased by 0.21 percentage units in comparison to cows offered silage as a sole ration at the same intake of DE. Hutton (1975) reported that when the proportion of silage fed with pasture decreased from two thirds to one third of the ration, milk

protein percentage increased by 0.2 percentage units and milk protein yield by approximately 24 %. In view of the difference obtained between pasture and silage in Experiment 3.1 at equal intakes of DE, these experiments using silage as a supplement to pasture indicated that if silage is below 50 % of the total ration, effects on milk protein synthesis apart from those of intake, are likely to be small. More work is required to define the milk production responses of cows fed different proportions of silage in a pasture ration in the two critical periods of winter and summer when normal pasture supplies are likely to be limiting. Moreover comparisons of direct cut high moisture pasture silage and wilted and formaldehyde treated silage are required when they are fed as supplements to pasture to examine if the benefits of these treatments during ensiling are reflected in milk yield and composition.

The effects of pasture and pasture silage on live-weight change observed during the course of this work are of considerable interest. The indications were that cows offered pasture in early lactation mobilized more tissue than cows offered silage at the same intake of DE. Perhaps this is a characteristic of pasture and is in part responsible for the high efficiency with which cows convert pasture to milk (Hutton, 1971). An extension of this argument is that pasture may not be the most suitable ration for improving the condition of cows in late lactation or during the dry period. Supplements like pasture silage may have this characteristic. The possibility that cows fed silage may have a lower maintenance requirement than those fed pasture (Appendix 4 and Hutton et al. 1971) may also be important in this regard. The important practical

implications of these possibilities strongly indicate that more work involving calorimetric and field investigations in this general field is essential.

5 BIBLIOGRAPHY

AGRICULTURAL RESEARCH COUNCIL 1965. Nutrient requirements of farm livestock.. No.2 Ruminants. Technical reviews and summaries. London; HMSO

ANDERSON, B.K. and JACKSON, N. 1971. Volatile fatty acids in the rumen of sheep fed grass, unwilted silage and barn dried hay. J. agric. Sci., Camb., 72, 453

ANNISON, E.F. 1956. Nitrogen metabolism in the sheep. Protein digestion in the rumen. Biochem. J., 64, 705

ANNISON, E.F. 1970. The mammary metabolism of glucose, acetate and ketone bodies. In Lactation, p.281. Ed. I.R. Falconer. London; Butterworths

ANNISON, E.F. and ARMSTRONG, D.G. 1970. Volatile fatty acid metabolism and energy supply. In Physiology of Digestion and Metabolism in the Ruminant, p.422. Ed. H.T. Phillipson. Newcastle-upon-Tyne; Oriel Press

ANNISON, E.F., BICKERSTAFFE, R. and LINZELL, J.L. 1974. Glucose and fatty acid metabolism in cows producing milk of low fat content. J. agric. Sci., Camb., 82, 87.

ANNISON, E.F. 1976. Energy metabolism in the body. In Principles of Cattle Production, p.169. Ed. H. Swan and W.H. Broster. London; Butterworths

ANON., 1969. 45th Farm Prod. Rep., 1968-69 Season, N.Z. Dairy Bd. Farm Prod. Div.

ANON., 1975. 51st Farm Prod. Rep., 1974-75 Season, N.Z. Dairy Bd. Farm Prod. Div.

ARCHER, K., BARWICK, S., KEMPTON, T.J. and LENG, R.A. 1976. Unpublished data cited by KEMPTON, T.J., NOLAN, J.V. and LENG, R.A. 1977. Principles for the use of non-protein-nitrogen and protected proteins in diets of ruminants. Department of Biochemistry, University of New England. (In preparation).

ARMSTRONG, D.G. and PRESCOTT, J.H.D. 1971. Amount, physical form and composition of feed and milk secretion in the dairy cow. In Lactation, p.349. Ed. I.R. Falconer. London; Butterworths

ARMSTRONG, D.G. 1974. Factors affecting the protein value of fresh and conserved feeds. Proc. 5th Gen. Mtg. Eur. Grassld. Fedn, Uppsala, p.114

ASSOCIATION OF OFFICIAL AGRICULTURAL CHEMISTS (AOAC) 1965 Official methods of analysis. 10th ed. Publ. AOAC, Washington, D.C.

BAILEY, R.W. 1958. The reaction of pentoses with anthrone. Biochem. J. 58, 669

BALCH, C.C. 1958. Observations on the act of eating in cattle. Br. J. Nutr., 12, 330.

BALCH, C.C. 1972. Milk composition. In Handbuch der Tierernahrung 2, 259. Ed. W. Lenkeit, K. Breirem and D. Crasemann. Hamburg; Paul Parey

BALDWIN, R.L., LIM, H.J., CHENG, W., CABRERA, R. and RONNING, M. 1969. Enzyme and metabolite levels in mammary and abdominal adipose tissue of lactating dairy cows. J. Dairy Sci., 52, 183

BARRY, J.M. and ROWLAND, S.J. 1953. Variations in the ionic and lactose concentrations in milk. Biochem. J., 54, 573

BARRY, T.N. and FENNESSY, P.F. 1972. The effect of formaldehyde treatment on the chemical composition and nutritive value of silage. I. Chemical composition. N.Z. Jl agric. Res., 15, 712

BARRY, T.N., FENNESSY, P.F. and DUNCAN, S.J. 1973. Effect of formaldehyde treatment on the chemical composition and nutritive value of silage. III. Voluntary intake, liveweight change, and wool growth in sheep fed the silages with and without intraperitoneal supplementation with D-L methionine. N.Z. Jl agric. Res., 16, 64.

BARRY, T.N. 1976. The effectiveness of formaldehyde treatment in protecting dietary protein from microbial degredation. Proc. Nutr. Soc., 35, 221

BARTSCH, B.D., ELLIS, N.J.S., McLEAN, D.M. and RADCLIFFE, J.C. 1976. Production, composition and manufacturing properties of milk from grazing dairy cows fed on formaldehyde treated sunflower seed supplement. Aust. J. agric. Res., 27, 917

BASSETT, J.M. 1974. Dietary and gastro-intestinal control of hormones regulating carbohydrate metabolism in ruminants. In Digestion and Metabolism in the Ruminant, p.383. Ed. I.W. McDonald and A.C.I. Warner. Univ. New England Publ. Unit. Sydney. Australia

BATH, I.H. and ROOK, J.A.F. 1963. The evaluation of cattle foods and diets in terms of the ruminal concentration of volatile fatty acids. I. The effects of level of intake, frequency of feeding, the ratio of hay to concentrates in the diet, and of supplementary feeds. J. agric. Sci. Camb., 61, 341

BATH, I.H. and ROOK, J.A.F. 1965., The evaluation of cattle foods in terms of the ruminal concentration of the volatile fatty acids. II. Roughages and succulents. J. agric. Sci., Camb. 64, 67

BATH, D.L., RONNING, M., MEYER, J.H. and LOFGREEN, G.P. 1965. Calorific equivalent of live-weight loss of dariy cattle. J. Dairy Sci., 48, 374

BAUMAN, D.E., DAVIS, C.L. and BUCHOLTZ, H.F. 1971. Propionate production in the rumen of cows fed either a control or high grain, low fibre diet. J. Dairy Sci., 54, 1282.

BEEVER, D.E., THOMSON, D.J., PFEFFER, E. and ARMSTRONG, D.G. 1971a. The effect of drying and ensiling grass on its digestion in sheep. Sites of energy and carbohydrate digestion. Br. J. Nutr., 26, 23.

BEEVER, D.E., THOMSON, D.J. and HARRISON, D.G. 1971b. The effects of drying and comminution of red clover on its subsequent digestion by sheep. Proc. Nutr. Soc., 30, 86A

BEEVER, D.E., THOMSON, D.J. and HARRISON, D.G. 1974. Energy and protein transformations in the rumen and the absorption of nutrients, by sheep fed forage diets. Proc. 12th int. Grassld Congr., 3, 56

BEEVER, D.E., THOMSON, D.J. and CAMELL, S.B. 1976. The digestion of frozen and dried grass by sheep. J. agric. Sci., Camb., 86, 443

BERGMAN, E.N., ROWE, W.E. and KON, K. 1966. Quantitative aspects of propionate metabolism and gluconeogenesis in sheep. Am. J. Physiol., 211, 793

BERGMAN, E.N. and WOLFF, J.E. 1971. Metabolism of volatile fatty acids by liver and portal drained viscera of sheep. Am. J. Physiol., 221, 586

BEITZ, D.C. and DAVIS, C.L. 1964. Relationship of certain milk fat depressing diets to changes in the proportions of the volatile fatty acids produced in the rumen. J. Dairy Sci., 47, 1213.

BICKERSTAFFE, R. 1971. Uptake and metabolism of fat in the lactating mammary gland. In Lactation, p.317. Ed. I.R. Falconer, London; Butterworth

BICKERSTAFFE, R. and ANNISON, E.F. 1971. The effects of duodenal infusions of sunflower oil on the yield and fatty acid composition of milk fat in the cow. Proc. Nutr. Soc., 30, 28A

BICKERSTAFFE, R., ANNISON, E.F. and LINZELL, J.L. 1974. The metabolism of glucose acetate, lipids and amino acids in lactating dairy cows. J. agric. Sci., Camb., 82, 71

BLAXTER, K.L. 1962. In The Energy Metabolism of Ruminants. Hutchinson; London

BLAXTER, K.L. and MARTIN, A.K. 1962. The utilization of protein as a source of energy in fattening sheep. Br. J. Nutr., 16, 397

BLAXTER, K. L. 1964. Utilization of the metabolizable energy of grass. Proc. Nutr. Soc., 23, 62.

BREW, K. 1970. Lactose synthetase : evolutionary origins, structure and control. Essays in Biochemistry, 6, 93

BRODERICK, G.A., KOWALCZYK, T. and SATTER, L.D. 1970. Milk production response to supplementation with encapsulated methionine per os or casein per abomasum. J. Dairy Sci., 53, 714.

BRODERICK, G.A., SATTER, L.D. and HARPER, A.E. 1974. Use of plasma amino acid concentration to identify limiting amino acids for milk production. J. Dairy Sci., 57, 1015

BROSTER, W.H. 1969. Recent research into levels of feeding for dairy cows. In Proc. 3rd Nutr. Conf. Feed Mfrs. Univ. Nott., p.53. Ed. H. Swan and D. Lewis. London; Churchill

BROSTER, W.H. 1971. The effect on milk yield of the cow of the level of feeding before calving. Dairy Sci. Abstr., 33, 253

BROSTER, W.H. 1972. Effect on milk yield of the cow of the level of feeding during lactation. Dairy Sci. Abstr., 34, 265

BROWN, S.M. 1960. Silage feeding of the dairy cow and its effect on milk yield and composition. III. A comparison of the ad libitum feeding of high dry matter, lacerated, and low dry matter, unlacerated, grass silages. Res. Exp. Rec. Min. Agric. N.I., 10, 9

BROWN, D.C. and VALENTINE, S.C. 1972. Formaldehyde as a silage additive. I. The chemical composition and nutritive value of frozen lucerne, lucerne silage and formaldehyde-treated lucerne silage. Aust. J. agric. Res. 23, 1093

BRYANT, A.M. 1964. The release of plant cell contents and its relation to bloat. Proc. N.Z. Soc. Anim. Prod. 24, 57

BRYANT, A.M. and LANCASTER, R.J. 1970. The effect of storage time on the voluntary intake of silage by sheep. Proc. N.Z. Soc. Anim. Prod. 30, 77

BRYANT, A.M. and DONNELLY, P.E. 1974. Yield and composition of milk from cows fed pasture herbage supplemented with maize and pasture silages. N.Z. Jl agric. Res., 17, 299

BRYANT, A.M. and NEWTH, R.P. 1975. Evaluation of high moisture forages with ruminants. II. Changes in the chemical composition of herbage after freezing and thawing. N.Z. Jl agric. Res., 18, 375

BRYANT, A.M. 1976. The utilization of whole grain in maize silage by cattle. Proc. N.Z. Soc. Anim. Prod., 36, 76

BRYANT, M.P. and DOETSCH, R.N. 1955. Factors necessary for the growth of Bacteriodes Succinogenes in the volatile acid fraction of rumen fluid. J. Dairy Sci., 38, 340

BURT, A.W.A 1957. The influence of level of feeding during lactation upon the yield and composition of milk. Dairy Sci. Abstr., 19, 435

BUTLER, T.M. 1973. A comparison between concentrates containing barley-soyabean meal and dried molassed beet pulp-groundnut meal for milk production. Ir. J. agric. Res., 12, 329

BUTLER, T.M. 1974. Sources and levels of proteins in cattle feeds. Ir. Grassld. Anim. Prod. Ass. J., p.24

BUTTERY, P.J. 1976. Protein synthesis in the rumen : its implication in the feeding of non protein nitrogen to ruminants. In Principles of Cattle Production, p.145. Ed. H. Swan and W.H. Broster. London; Butterworths

CAMPBELL, I.L. and FLUX, D.S. 1948. The relationship between level of nutrition during the dry period and subsequent performance of dairy cattle. Proc. N.Z. Soc. Anim. Prod., 8, 61

CAMPBELL, I.L., FLUX, D.S. and PATCHELL, M.R. 1955. Factors affecting the non-fatty solids in milk, with particular reference to the New Zealand town milk industry. Proc. N.Z. Soc. Anim. Prod., 15, 132

CAMPBELL, I.L. and DOLBY, R.M. 1959. Recent dairy research work in New Zealand. Part I. Dairy Sci. Abstr., 21, 95

CASTLE, M.E., DRYSALE, A.D. and WAITE, R. 1961. The effect of root feeding on the intake and production of dairy cows. J. Dairy Res., 28, 67

CASTLE, M.E. and WATSON, J.N. 1969. The effect of level of protein in silage on the intake and production of dairy cows. J. Br. Grassld Soc., 24, 187

CASTLE, M.E. and WATSON, J.N. 1970. Silage and milk production, a comparison between wilted and unwilted grass silages made with and without formic acid. J. Br. Grassld Soc., 25, 278

CASTLE, M.E. and WATSON, J.N. 1974. Red clover silage for milk production. J. Br. Grassld Soc., 28, 101

CASTLE, M.E. and WATSON, J.N. 1975. Silage and milk production. A comparison between barley and dried grass as supplements to silage of high digestibility. J. Br. Grassld Soc., 30, 217

CASTLE, M.E. and THOMAS, P.C. 1976. Silage research at the Hannah Institute. A. Rep. Hannah Dairy Res. Inst., (1975), p.61

CHALMERS, M.I. and SYNGE, R.L.M. 1954. Ruminal ammonia formation in casein and herring-meal supplements. J. agric. Sci., Camb., 44, 263

CHALMERS, M.I. 1963. The significance of the digestion of protein within the rumen on the nutrition of the animal. In Progress in Nutrition and Allied Sciences, 91. Ed. D.P. Cuthbertson. Edinburgh; Oliver and Boyd

CHANDLER, P.T. and POLAN, C.E. 1972. Considerations in interpretations of serum amino acids in lactating cows. J. Dairy Sci., 55, 709 (Abstr).

CHANDLER, N.J., ROBINSON, I.B., RIPPER, I.C. and FOWLER, P. 1973. The effect of feeding formaldehyde treated sunflower seed supplement on the yield and composition of milk fat. Aust. J. Dairy Technol., 28, 179

CHRISTENSEN, H.N. 1963. Amino acid transport and nutrition. Fed. Proc., 22, 1110

CHRISTENSEN, H.N. 1964. Free amino acids and peptides in tissues. In Mammalian Protein Metabolism, 1, 105. Ed. H.N. Munro and J.B, Allison. London, Academic Press.

CLANCY, M.J. and WILSON, R.K. 1966. Development and application of a new chemical method for predicting the digestibility and intake of herbage samples. Proc. 10th int. Grassld Congr., p.445

CLARKE, E.M.W., ELLINGER, E.M. and PHILLIPSON, A.T. 1966. The influence of diet on the nitrogenous components passing to the duodenum and through the lower ileum of sheep. Proc. Royal Soc., B 166, 63

CLARK, J.H., SPIRES, H.R. and DERRIG, R.G. 1973. Post-ruminal administration of glucose and Na-caseinate in lactating cows. J. Anim. Sci., 37, 340

CLARK, J.H. 1975. Lactational responses to post-ruminal administration of proteins and amino acids. J. Dairy Sci., 58, 1178

COBBLE, J.W. and HERMAN, H.H. 1951. The influence of environmental temperatures on the composition of milk of the dairy cow. Res. Bull. Mo. agric. Exp. Sta., p.485

COELHO DA SILVA, J.F., SEELEY, R.C., THOMSON, D.J., BEEVER, D.E. and ARMSTRONG, D.G. 1972a. The effect in sheep of physical form and stage of growth on the sites of digestion of a dried grass. Br. J. Nutr. 28, 357

COELHO DA SILVA, J.F., SEELY, R.C., THOMSON, D.J., BEEVER, D.E. and ARMSTRONG, D.G. 1972b. The effect in sheep of physical form on the sites of digestion of a dried lucerne diet. Br. J. Nutr., 28, 43

COLENBRANDER, V.F., BARTLEY, E.E., MORRILL, J.L., DEYOE, C.W. and PFOST, H.B. 1967. Feed Processing. II. Effect of feeding expanded grain and finely ground hay on milk composition, yield and rumen metabolism. J. Dairy Sci., 50, 1966

CONRAD, H.R., HIBBS, J.W., PRATT, A.D. and DAVIS, P.R. 1961. Nitrogen metabolism in dairy cattle. I. The influence of grain and meadow crops harvested as hay, silage, or soilage on efficiency of nitrogen utilization. J. Dairy Sci., 44, 85

CONRAD, H.R. and HIBBS, J.W. 1968. Nitrogen utilization by the ruminant. Appreciation of its nutritive value. J. Dairy Sci., 51, 276

CONWAY, E.J. 1957. In Micro diffusion Analysis and Volumetric Error. 4th edit., London, Crosby Lockwood

COPPOCK, C.E., FLATT, W.P., MOORE, L.A. and STEWART, W.E. 1964a. Effect of hay to grain ratio on utilization of metabolizable energy for milk production by dairy cows. J. Dairy Sci., 71, 1330

COPPOCK, C.E., FLATT, W.P., MOORE, L.A. and STEWART, W.E. 1964b. Relationships between end products of rumen fermentation and utilization of metabolizable energy for milk production. J. Dairy Sci., 47, 1359

CORBIN, E.A. and WHITTIER, E.O. 1965. Composition of milk. In Fundamentals of Dairy Chemistry, p.1. Ed. B.H. Webb and A.H. Johnson. Westport Connecticut; AVI Publishing Co. INC.

CROSS, B.A. 1966. Neuroendocrinology, 1, 217. Ed. L. Martini and W.F. Ganong. London; Academic Press.

DAVEY, A.W.F. 1965. Variations in ruminal pH, volatile fatty acid concentration and proportions of individual acids. Proc. N.Z. Soc. Anim. Prod., 25, 106

DAVIS, C.L., BROWN, R.E. and BEITZ, D.C. 1964. Effect of feeding high grain restricted roughage rations with and without bicarbonates on the fat content of milk produced and proportions of volatile fatty acids in the rumen. J. Dairy Sci., 47, 1217

DAVIS, C.L. 1967. Acetate production in the rumen of cows fed either control or low fibre high grain diets. J. Dairy Sci., 50, 1621

DAVIS, C.L. and BROWN, R.E. 1970. Low-fat milk syndrome. In Physiology of Digestion and Metabolism in the Ruminant, 545. Ed. A.T. Phillipson. Newcastle-upon-Tyne; Oriel Press

DEMARQUILLY, C. 1973. Chemical composition, fermentation characteristics, digestibility and voluntary intake of forage silages : changes compared to the initial green forage. Ann. Zootech, 22, 1

DERRIG, R.G., CLARK, J.H. and DAVIS, C.L. 1974. Effect of abomasal infusion of sodium caseinate on milk yield, nitrogen utilization and amino acid nutrition of the dairy cow. J. Nutr, 104, 151

DIMANT, E., SMITH, V.R. and LARDY, H.A. 1953. Lactose synthesis in the mammary gland perfused with 1-C^{14}-glucose. J. biol. Chem, 201, 85

DODD, F.H. and CLOUGH, P.A. 1962. The effect of various degrees of incomplete milking on milk and butterfat secretion. Proc. 16th int. Dairy Congr., A, 89

DONALDSON, E. and EDWARDS, R.A. 1976. Feeding value of silage : silages made from freshly cut grass, wilted grass and formic acid treated wilted grass. J. Sci. Fd Agric., 27, 536

DOROTYUK, E.N. 1965. Characteristics of milk composition during oestrous in cows. Dairy Sci. Abstr., 27, 183 (Abstr).

DULPHY, J.P. and DEMARQUILLY, C. 1973. Effect of type of forage harvester and chopping fineness on the feeding value of silages. Ann. Zootech, 22, 199

EADIE, J.M., HYLDGAARD-JENSEN, J., MANN, S.O., REID, R.S. and WHITELAW, F.G. 1970. Observations on the microbiology and biochemistry of the rumen in cattle given different quantities of a pelleted barley ration. Br. J. Nutr., 24, 157

EGAN, A.R. and WALKER, D.J. 1973. Resource allocation and ruminant protein production. Proc. III wld. Conf. Anim. Prod., Melbourne, 3, 5(c), 10

ELLIOT, G.M. 1959. The effect on milk yield of the length of milking intervals used in twice a day milking and three times a day milking and incomplete milkings. Dairy Sci., Abstr., 21, 481

el-SHAZLY, K. 1952. Degradation of protein in the rumen of sheep. I. Some volatile fatty acids, including branched-chain isomers found in vivo. Biochem. J., 51, 640

EMERY, R.S., BROWN, L.D. and THOMAS, J.W. 1964. Comparison of corn cobs and hay in ground, restricted-roughage rations affecting milk composition. J. Dairy Sci., 47, 1322

FATIANOFF, N., DURAND, M., ZELTER, S.Z.,and TISSERAND, J.L. 1966. Comparative effects of wilting and of sodium metabisulphate on quality and nutritive value of alfalfa silage. Proc. 10th int. Grassld Congr., p.551

FINKELSTEIN, J.D. 1974. Methionine metabolism in mammals : the biochemical basis for homocystinuria. Metabolism, 23, 387

FISHER, L.J. 1969. Effect of methionine infusion on milk production and plasma-free amino acids of lactating cows. J. Dairy Sci.,52, 943 (Abstr).

FISHER, L.J., LESSARD, J.R. and LODGE, G.A. 1971. Utilization of formic acid treated Sorghum-Sudan silage by dairy cows. Can. J. Anim. Sci., 51, 371

FISHER, L.J. 1972, Response of lactating cows to the intravenous infusion of amino acids. Can. J. Anim. Sci., 52, 377

FISHER, L.J., DONNELLY, P.E., HUTTON, J.B. and DUGANZICH, D.M. 1975. Relationships between levels of feeding and cetain blood metabolites in dairy cows in mid lactation. J. agric. Sci., Camb., 84, 29

FLATT, W.P., MOE, P.W., MOORE, L.A., HOOVEN, N.W., LEHMANN, R.P., ØRSKOV, E.R. and HEMKIN, R.W. 1969. Energy utilization by high producing cows. In Energy Metabolism of Farm Animals, p.221. Ed. K.L. Blaxter, G. Thorbek and J. Kielanowski. Newcastle-upon-Tyne; Oriel Press

FLATT, W.P., MOORE, L.W., HOOVEN, N.W. and PLOWMAN, R.D. 1965. Energy metabolism studies with high producing lactating dairy cows. J. Dairy Sci., 48, 487

FLUX, D.S. 1950. The effect of under nutrition before calving on the quantity and composition of milk produced by two-year-old heifers. J. agric. Sci., Camb., 40, 177

FLUX, D.S. and PATCHELL, M.R. 1950. The comparison of three rations for dairy cattle. Proc. N.Z. Soc. Anim. Prod., 10, 26

FLUX, D.S. and PATCHELL, M.R. 1954. The effect of undernutrition after calving on the quantity and composition of the milk production by dairy cattle. J. agric. Sci., Camb. 45, 246

FLUX, D.S., FOLLEY, S.J. and ROWLAND, S.J. 1954. The effect of adrenocorticotrophic hormone on the yield and composition of the milk of the cow. J. Endocrin., 10, 333

FLUX, D.S. and PATCHELL, M.R. 1957. The effect of short periods of under nutrition after calving on the establishment of lactation in dairy cows. N.Z. Jl Sci. Technol., 38A, 649

FOOT, A.S. 1964. Recent experiments concerning the SNF content of milk. J. Soc. Dairy Tech., 17, 34

GIRARD-GLOBA, A., ROBIN, A.P. and FORESTER, M. 1972. Long term adaption of weanling rats to high dietary levels of methionine and serine. J. Nutr., 102, 209

GOERING, H.K. and WALDO, D.R. 1974. Processing effects on protein utilization by ruminants. Proc. Cornell Nutr. Conf. Feed Mnfr., p.25

GORDON, C.H., DERBYSHIRE, J.C., WISEMAN, H.G., KANE, E.A. and MELIN, C.G. 1961. Preservation and feeding value of alfalfa stored as hay, haylage and direct cut silage. J. Dairy Sci., 44, 1299

GORDON, C.H., DERBYSHIRE, J.C., JACOBSEN, W.C. and HUMPHREY, J.L. 1965. Effect of dry matter in low moisture silage on preservation, acceptability, and feeding value for dairy cows. J. Dairy Sci., 48, 106

GRIFFITHS, R.W. 1969. Some nutritional effects of the addition of rolled barley to an all silage basal diet. Anim. Prod., 11, 286

GRIFFITHS, T.W. and SPILLANE, T.A. 1970. Nutritional effects of cereal and protein supplementation of an all silage basal diet. Anim. Prod., 12, 359

GRIFFITHS, T.W. and SPILLANE, T.A. 1971. Protein and energy inter-relationships in silage-based diets for growing cattle. Anim. Prod., 13, 286

GRIFFITHS, T.W., SPILLANE, T.A. and BATH, I.H. 1971. Studies on the nutritive value of silage with particular reference to the effects of energy and nitrogen supplementation in growing heifers. J. agric. Sci., Camb., 80, 75

GRIFFITHS, T.W. and BATH, I.H. 1973. Effects of energy and nitrogen supplementation of silage diets on rumen fermentation in fistulated heifers. J. agric. Sci., Camb., 80, 89

GRIFFITHS, T.W. and CROWLEY, J.P. 1973. Effects of variations in energy and protein intake during early lactation on nutrient balance and performance of heifers in a beef production system. Ir. J. agric Res. 12, 127

GUJARATI, D. 1970. Use of dummy variables in testing for equality between sets of coefficients in linear regressions : A generalization. The American Statistician 24,(5), 18

HAEGMEISTER, H., KAUFMANN, W. and PFEFFER, E. 1976. Factors influencing the supply of nitrogen and amino acids to the intestine of dairy cows. In Protein Metabolism and Nutrition, EAAP, publ. 16, 441. Ed. D. Cole, K. Boorman, P. Buttery, D. Lewis, R. Neale, and H. Swan. London; Butterworths

HALE, G.D., JACOBSEN, D.R. and HEMKEN, R.W. 1972. Continuous abomasal infusion of casein in lactating Holsteins fed urea supplemented diets. J. Dairy Sci., 55, 689 (Abstr.).

HALFPENNY, A.F., ROOK, J.A.F. and SMITH G.H. 1969. Variations with energy nutrition in the concentrations of amino acids of the blood plasma in the dairy cow. Br. J. Nutr., 23, 547

HANCOCK, J. 1958. The conversion of pasture to milk : The effect of stocking rate and concentrate feeding. J. agric. Sci., Camb., 50, 284

HARRIS, C.E. and RAYMOND, W.F. 1963. The effect of ensiling on crop digestibility. J. Br. Grassld Soc., 18, 204

HARRISON, D.G., BEEVER, D.E., THOMSON, D.J. and OSBOURNE, D.F. 1973. The influence of diet upon the quantity and types of amino acids entering and leaving the small intestine in sheep. J. agric. Sci., Camb. 81, 391

HARRISON, D.G., BEEVER, D.E., THOMSON, D.J. and OSBOURNE, D.F. 1976. Manipulation of fermentation in the rumen. J. Sci. Fd Agric., 27, 617

HEMSLEY, J.A. and MOIR, R.J. 1963. The influence of higher volatile fatty acids on the intake of urea - supplemented low quality cereal hay by sheep. Aust. J. agric. Res., 14, 509

HINDERS, R.G. and OWEN, H.G. 1963. Relationships between efficiency of milk production and ruminal volatile fatty acids of cows fed isocaloric (E.N.E.) rations of varied concentrate levels. J. Dairy Sci., 46, 1246

HINKS, C.E., EDWARDS, I.E. and HENDERSON, A.R. 1976. Beef production from formic acid - treated and wilted silages. Anim. Prod., 22, 217

HOGAN, J.P. 1965a. The digestion of food by the grazing sheep. III. The quantity of protein reaching the small intestine. Aust. J. agric. Res., 16, 347

HOGAN, J.P. 1965b. The digestion of food by the grazing sheep. IV. The effect of mastication on the release of soluble plant nitrogen. Aust. J. agric. Res., 16, 855

HOGAN, J.P. and WESTON, R.H. 1968. Digestion of protein in the intestines of sheep. Proc. Aust. Soc. Anim. Prod., 7, 364

HOGAN, J.P. and WESTON, R.H. 1969. The digestion of pasture plants by sheep. III. The digestion of forage oats varying in maturity and in the content of protein and soluble carbohydrate. Aust. J. agric. Res., 20, 347.

HOGAN, J.P., WESTON, R.H. and LINDSAY, J.R. 1969. The digestion of pasture plants by sheep. IV. The digestion of Phalaris tuberosa at different stages of maturity. Aust. J. agric., Res., 20, 925

HOGAN, J.P. and WESTON, R.H. 1970. Quantitative aspects of mircobial protein synthesis. In Physiology of Digestion and Metabolism in the Ruminant, p.474. Ed. A.T. Phillipson. Newcastle-upon-Tyne; Oriel Press

HOGAN, J.P. 1975. Quantitative aspects of nitrogen utilization in ruminants. J. Dairy Sci., 58, 1164

HOLMES, W., ARNOLD, G.W. and PROVAN, A.L. 1960. Bulk feeds for milk production. I. The influence of level of concentrate feeding in addition to silage and hay on milk yield and milk composition. J. Dairy Res., 27, 191

HOTCHKISS, D.K., JACOBSEN, N.L. and COX, C.P. 1960. Effect of various hay : concentrate ratios and levels of feeding on production and composition of milk of dairy cows. J. Dairy Sci., 43, 872

HUBER. J.T. and BOWMAN, R.L. 1966a. Nutritional factors affecting the solids-not-fat content of milk. J. Dairy Sci., 49, 816

HUBER, J.T. and BOWMAN, R.L. 1966b. Effect of grain level and protein content of the grain for grazing cows on milk composition and yield, and certain blood and rumen constituents. J. Dairy Sci., 49, 395

HUGHES, J.W. 1963. Equipment for the separate and total collection of faeces and urine from dairy cattle. N.Z. Jl agric. Res., 6, 127

HUME, I.D. 1970. Synthesis of microbial protein in the rumen. II. A response to higher volatile fatty acids. Aust. J. agric. Res., 21, 297

HUME, I.D. 1974. The proportion of dietary protein escaping degradation in the rumen of sheep fed on various protein concentrates. Aust. J. agric. Res., 25, 155

HUME, I.D. and PURSER, D.B. 1974. Duodenal flow of dietary and microbial nitrogen in sheep fed subteranean clover harvested at four stages of maturity. Proc. Aust. Soc. Anim. Prod., 10, 399

HUNGATE, R.E. 1965. Quantitative aspects of the rumen fermentation. In Physiology of Digestion in the Ruminant, p.311. Ed. R.W. Dougherty. London; Butterworths

HUNGATE, R.E. 1966. The rumen and its microbes. New York, London; Academic Press

HUTTON, J.B. 1958. Oestrogen function in established lactation in the cow. J. Endocrin., 17, 121

HUTTON, J.B. 1962. Studies of the nutritive value of New Zealand dairy pastures. II. Herbage intake and digestibility studies with dry cattle. N.Z. Jl agric. Res., 5, 409

HUTTON, J.B. and PARKER, O.F. 1966. The effect of grazing time and supplementary feeding of hay on production per cow in early lactation. Proc. N.Z. Soc. Anim. Prod. 26, 71

HUTTON, J.B. and PARKER, O.F. 1967. Meal feeding for milk production. Proc. Rua. Frms. Conf., p.194

HUTTON, J.B., JURY, K.E., HUGHES, J.W., PARKER, O.F. and LANCASTER, R.J. 1971. High moisture silage from mixed pasture herbage as a feed for lactating dairy cows. N.Z. Jl agric. Res., 14, 393

HUTTON, J.B. 1971. Nutrient requirements of N.Z. dairy cattle. Proc. N.Z. Soc. Anim. Prod., 31, 211

HUTTON, J.B. and PARKER, O.F. 1972. The significance of differences in levels of feeding, before and after calving, on milk yield under intensive grazing. N.Z. Jl agric. Res., 16, 95

HUTTON, J.B. 1973. The effect on pasture intake and milk production of variations in the percentage utilization of total available dry matter. Ann. Rep. Res. Div., 1972-73, p.51 : Ministry of Agriculture and Fisheries, Wellington. N.Z.

HUTTON, J.B. 1974. Effects of irrigation and high stocking rates on seasonal dairy production from grasslands. Ann. Rep. Res. Div., 1973-74, p.54 : Ministry of Agriculture and Fisheries, Wellington. N.Z.

HUTTON, J.B. 1975. Growing and using maize on the dairy farm. Proc. Rua. Frms. Conf., p.76

HUTTON, J.B., HUGHES, J.W., BRYANT, A.M., PLUCK, L.J. and TAYLOR. R.E.C. 1975. Evaluation of high moisture forages with ruminants. I. Equipment and techniques for harvesting, processing, and storing high moisture forages. N.Z. Jl agric. Res., 18, 37

HVELPLEND, T. and MOLLER, P.D. 1976. Nitrogen metabolism in the gastro-intestinal tract of cows fed silage. Z. Tierphysiol., Tierernahrg. v. Futtermittelkde, 37, 183

JACKSON, N. and ANDERSON, B.K. 1968. Conservation of fresh and wilted grass in air-tight metal containers. J. Sci. Fd Agric., 19, 1

JACKSON, N. and FORBES, T.J. 1970. The voluntary intake by cattle of four silages differing in dry matter content. Anim. Prod., 12, 591.

JACKSON, N. 1971. Effect of Arachis oil on the conservation of heavily wilted herbage ensiled in plastic containers. I. Effect on silage fermentation quality, nutritive value, nutrient losses and intake by sheep. J. Sci. Fd. Agric., 22, 419

JENNESS, R and PATTON, S. 1959. Principles of Dairy Chemistry. New York; J. Wiley.

JOHANSSON, I. 1952. Studies on udder evacuation in dairy cows. II. The amount and composition of the residual milk after normal milkings. Acta. Agric. Scand., 2, 82

JOHANSSON, I. and CLAESSON, O. 1957. Factors affecting composition in milk. In Progress in the Physiology of Farm Animals, 3, 1005 Ed. J. Hammond. London; Butterworths

KAUFMANN, W. and ROHR, K. 1966. Ergebnisse gaschromatographischer Bestimmung der fluchtigen Fettsauren im Pansen bei unterschiedlicher Futterung. Z. Tierphysiol. Tierernahr Futtermittelk, 22, 1

KAY, M. 1969. Roughage to concentrate balance in foods for the dairy cow. In Proc. 3rd Nutr. Conf. Feed Mfrs. Univ. Nott., p.43 Ed. H. Swan and D. Lewis, London; Churchill

KIRCHGESSNER, M., FRIESEKE, H. and KOCH, G. 1967. Nutrition and Composition of Milk. London; Crosby Lockwood

KLEIBER, M., BLACK, A.L., BROWN, M.A., BLAXTER, C.F., LUICK, J.R. and STADTMAN, F.H. 1955. Glucose as a precursor of milk constituents in the intact cow. Biochem. Biophys. Acta., 17, 252

KORMOS, J. 1967. A study of ensiling wilted and unwilted grass at two stages of maturity. 2. Effect on milk production. Rec. agric. Res., Belf., 16, 57

KRONFELD, D.S. 1976. The potential importance of the proportions of glucogenic, lipogenic and aminogenic nutrients in regard to the health and productivity of dairy cows. Fortschritte in der Tierphysiologie und Tierernahrung, (7), p.6

LABEN, R.C. 1963. Factors responsible for variation in milk composition. J. Dairy Sci., 46, 1293

LANCASTER, R.J., HUTTON, J.B., HUGHES, J.W. and MARSHALL, R.J. 1974. Effect on intake and production of dairy cows of feeding three high-moisture silages having different fermentation characteristics. N.Z. Jl Exptl. Agric., 2, 389

LANCASTER, R.J. 1976. Effect of formaldehyde-treated casein and methionine on the intake of silage by sheep. Proc. N.Z. Soc. Anim. Prod., 36, 127

LAMPO, P.H., WILLEMS, A. and VANSCHOUBROEK, F. 1966. Effect of pregnancy and milk recording district on milk yield and milk composition. Neth. Milk and Dairy Journal, 20, 256

LAWLOR, M.J., GIESECKE, D. and WALSER-KARST, D. 1966. Comparative studies on the digestive physiology of sheep feed on semi-purified or roughage-concentrate diets. Br. J. Nutr., 20, 373

LEES, F.T., McMEEKAN, C.P. and WALLACE, L.R. 1948. The relationship between level of nutrition during dry period and subsequent production of dairy cattle. Proc. N.Z. Soc., Anim. Prod., 8, 60

LEGATES, J.E. 1960. Genetic and environmental factors affecting the solids-not-fat composition of milk. J. Dairy Sci., 43, 1527

LENG, R.A., STEEL, J.W. and LUICK, J.R. 1967. Contribution of propionate to glucose synthesis in sheep. Biochem. J., 103, 785

LENG, R.A. 1970a. Glucose synthesis in ruminants. Adv. vet. Sci., 14, 209

LENG, R.A. 1970b. Formation and production of volatile fatty acids in the rumen. In Physiology of Digestion and Metabolism in the Ruminant, p.406. Ed. A.T. Phillipson. Newcastle-upon-Tyne, Oriel Press

LENKEIT, W. 1972. cited by van ES, A.J.H. and BOEKHOLT, H.A. 1976. Protein requirements in relation to the lactation cycle. In Protein Metabolism and Nutrition, p.441. Ed. D. Cole, K. Boorman, P. Buttery, D. Lewis, R. Neale and H. Swan. London; Butterworths

LEWIS, D. and BUTTERY, P.J. 1973. Ammonia toxicity in ruminants. In Production Disease in Farm Animals, p.201. Ed. J.M. Payne, K.C. Hibbit and B.F. Sansom. London; Balliere Tindall

LING, E.R., KON, S.K. and PORTER, J.W.G. 1961. The composition of milk and the nutritive value of its components. In Milk: The mammary gland and its secretion. 2,195. Ed. S.K. Kon and A.T. Cowie. London; Academic Press.

MacLEOD, G.K. and WOOD, A.S. 1972. Influence of amount and degree of saturation of dietary fat on yield and quality of milk.. J. Dairy Sci., 55, 446

MacRAE, J.C. 1970. Changes in chemical composition of freeze stored herbage. N.Z. Jl agric. Res., 13, 45

MacRAE, J.C. ULYATT, M.J., PEARCE, P.D. and HENDTLASS, J. 1972. Quantitative intestinal digestion of nitrogen in sheep given formaldehyde treated and untreated casein supplements. Br. J. Nutr., 27, 39

MacRAE, J.C. and ULYATT, M.J. 1974. Quantitative digestion of fresh herbage by sheep. II. Sites of digestion of some nitrogenous constituents. J. agric. Sci., Camb., 82, 300

MARTIN, A.K. and BLAXTER, K.L. 1964. The energy cost of urea synthesis in sheep. Proc. 3rd Sym. on Energy Metab, Troon, Scotland, EAAP, publ. 11, p.83

McCARTHY, R.D., PORTER, G.A. and GRIEL, L.C. 1968. Bovine ketosis and depressed fat test in milk; A problem in methionine metabolism and serum lipoprotein abberation. J. Dairy Sci., 51, 459

McCARTHY, R.D., PATTON, R.A. and GRIEL, L.C. 1970. Amino acid nutrition of lactating ruminants. Fed. Proc., 29, 41

McCLYMONT, G.L. and PAXTON, R.P. 1947. The effect of grazing oats on butter-fat content of milk. Need for coarse roughage. Agric. Gaz. N.S.W., 58, 551.

McCLYMONT, G.L. and VALLENCE, W.S. 1962. Depression of blood glycerides and milk fat synthesis by glucose infusion. Proc. Nutr. Soc., 21, xli

McCULLOUGH, M.E. 1966. Relationships between rumen fluid, volatile fatty acids and milk fat percentage and feed intake. J. Dairy Sci., 49, 896

McDONALD, I.W. 1952. The role of ammonia in ruminal digestion of protein. Biochem. J. 51, 86

McDONALD, P. and WHITTENBURY, R. 1967. Losses during ensilage. In Fodder conservation, Proc. Occ. Symp. No.3, Br. Grassld Soc., p.70 Ed. R.J. Wilkins

McDONALD, P., HENDERSON, A.R. and McGREGOR, A.W. 1968. Chemical changes and losses during the ensilage of wilted grass. J. Sci. Fd Agric., 19, 125

McDONALD, P. and EDWARDS, R.A. 1976. The influence of conservation methods on digestion and utilization of forages by ruminants. Proc. Nutr. Soc., 35, 201

McDOWALL, F.H. 1936. The cheese yielding capacity of milk, and its relation to the method of payment for milk for cheese making. N.Z.Jl Sci. Tech., 19A, 137

McMEEKAN, C.P. and WALSHE, M.S. 1963. The inter-relationships of grazing method and stocking rate in the efficiency of pasture utilization by dairy cattle. J. agric. Sci., Camb., 61, 147

MILLER, E.L. 1973. Evaluation of foods as sources of nitrogen and amino acids. Proc. Nutr. Soc., 32, 79

MILNE, J.A. and CAMPLING, R.C. 1972. Intake and digestibility by sheep of artificially dried forages in several physical forms. J. agric. Sci., Camb., 78, 79

MINSON, D.J., RAYMOND, W.F. and HARRIS, C.E. 1960. Studies in the digestibility of herbage. VIII. The digestibility of S37 cocksfoot, S23 ryegrass and S24 ryegrass. J. Br. Grassld Soc., 15, 174

MINSON, D.J. and LANCASTER, R.J. 1963. The effect of oven temperature on the error in estimating the dry matter content of silage. N.Z. Jl agric. Res., 6, 140

MINSON, D.J., HARRIS, C.E., RAYMOND, W.F. and MILFORD, R. 1964. The digestibility and voluntary intake of S22 and H1 ryegrass, S 170 tall fescue, S 48 timothy, S 215 meadow fescue and germinal cocksfoot. J. Br. Grassld Soc., 19, 298

MINSON, D.J. 1966. The apparent retention of food in the reticulo-rumen at two levels of feeding by means of an hourly feeding technique. Br. J. Nutr., 20, 765

MOE, P.W. and TYRRELL, H.F. 1971. Net energy value for lactation of high and low protein diets containing corn silage. J. Dairy Sci., 55, 318

MUNRO, H.N. 1976. Eukaryote protein synthesis and its control. In Protein Metabolism and Nutrition, EAAP, publ. 16, p.3. Ed. D. Cole, K. Boorman, P. Buttery, D. Lewis, R. Neale and H. Swan. London; Butterworths

MURDOCH, J.C. 1960. Effect of pre-wilting herbage on the composition of silage and its intake by cows. J. Br. Grassld Soc., 15, 70

MURDOCH, J.C. 1962. The effect of type of concentrate on milk production when silage is the only roughage offered to cows. J. Br. Grassld Soc., 17, 268

NATIONAL RESEARCH COUNCIL (N.R.C.) 1971. Nutrient requirements of domestic animals, No.3. Nutrient requirements of dairy cattle. Fourth revised ed. Nat. Acad. Sci., Washington, D.C.

NIMRICK, K., HATFIELD, E.E., KAMINSKI, J. and OWENS, F.W. 1970. Quantitative assessment of supplemental amino acid needs for growing lambs fed urea as a sole nitrogen source. J. Nutr., 100, 1301

NOLAN, J.V. and LENG, R.A. 1972. Dynamic aspects of ammonia and urea metabolism in sheep. Br. J. Nutr., 27, 177

NOLAN, J.V., NORTON, B.W. and LENG, R.A. 1973. Nitrogen cycling in sheep. Proc. Nutr., Soc., 32, 93

NORDFELDT, S. 1966. Utilization of hay and/or silage by dairy cows. Proc. 10th int. Grassld Congr., p.588

O'DONOVAN, J., DODD, F.H. and NEAVE, F.K. 1960. The effect of udder infections on the lactation yield of milk and milk solids. J. Dairy Res., 27, 115

OLTJEN, R.R., SLYTER, L.L., WILLIAM, E.E. and KERN, D.L. 1971. Influence of branched chain volatile fatty acids and phenylacetate on rumen micro-organisms and nitrogen utilization by steers fed urea or isolated soy protein. J. Nutr., 101, 101

ORMISTON, E.E., SPAHR, S.L., TOUCHBERRY, R.W. and ALBRIDGE, J.L. 1967. Effects of milking at unequal intervals for a complete lactation on milk yield and composition. J. Dairy Sci., 50, 1597

ØRSKOV, E.R., MAYES, R.W. and PENN, A. 1971. The capacity for the removal of glucose from the small intestine by mature sheep. Proc. Nutr. Soc., 30, 43A

ØRSKOV, E.R. and FRASER, C. 1974. Effect of changes in the glucogenic energy on milk and milk fat in lactating goats. Proc. Nutr., Soc., 33, 9A

ØRSKOV, E.R. and GRUBB, D.A. 1977. The effect of abomasal glucose or casein infusion on milk yield and milk composition in cows in early lactation and negative energy balance. Proc. Nutr. Soc. 36, 58A

OVERMANN, O.R., GARRET, O.F., WRIGHT, K.E. and SANMANN, F.P. 1939 Composition of milk of Brown Swiss cows, with summary of data on the composition of milk from cows of other dairy breeds. Bull. Ill. agric. Exp. Sta., p.457

PAQUAY, R., DE BAERE, R. and LOUSSE, A. 1972. The capacity of the mature cow to lose and recover nitrogen and the significance of protein reserves. Br. J. Nutr., 27, 27

PATCHELL, M.R. 1957. The influence of under nutrition of dairy cows on the yield and composition of milk. N.Z. Jl Sci.,Tech., 38A, 682

PATTON, R.A., McCARTHY, R.D. and GRIEL, L.C. 1968. Lipid synthesis by rumen micro-organisms. I. Stimulation by methionine in vitro. J. Dairy Sci., 51, 1310

PATTON, R.A,. McCARTHY, R.D. and GRIEL, L.C. 1970. Lipid synthesis by rumen micro-organisms. II. Further characterization of the effects of methionine. J. Dairy Sci., 53, 460

PHILPOT. W.N. 1976. Mastitis control yields increased profits. Proc. Rua. Frms. Conf., p.80

POWELL, E.B. 1935. One cause of fat variation in milk. Proc. Amer. Soc. anim. Prod., 31, 40

POWELL, E.B. 9141. Progress report on the relation of the ration to the composition of milk. J. Dairy Sci. 24, 504

PROUD, C.J. 1973. cited by ARMSTRONG, D.G. 1974. Factors affecting the protein value of fresh and conserved feeds. Proc. 5th Gen. Mtg. Eur. Grassld. Fedn., Uppsala, p.114

PURSER, D.B. 1970. Nitrogen metabolism in the rumen : micro-organisms as a source of protein for the ruminant animal. J. Anim. Sci., 30, 988

RATTRAY, P.V. and JOYCE, J.P. 1969. The utilization of perennial ryegrass and white clover by young sheep. Proc. N.Z. Soc. Anim. Prod., 29, 102

REISS, O.K. and BARRY, J.M. 1953. The synthesis of lactose from glucose in the mammary gland. Biochem. J., 55, 783

REIS, P.J. and TUNKS D.A. 1971. Plasma amino acid patterns in sheep receiving abomasal infusion of methionine. Search, 2, 108

REIS, P.J., TUNKS, D.A. and SHARRY, L.F. 1973. Plasma amino acid patterns in sheep receiving abomasal infusions of methionine and cysteine. Aust. J. biol. Sci., 26, 635

RIDDET, I., CAMPBELL, I.L., McDOWALL, F.H. and COX, G.A. 1941. The relation of plane of nutrition to milk production and milk composition in New Zealand. N.Z. Jl Sci. Tech., 23A, 80

ROBERTSON, A., WAITE, R. and WHITE, J.C.D. 1956. Variations in the chemical composition of milk with particular reference to the solids-not-fat. II. The effect of heredity. J. Dairy Res. 23, 82

ROLLERI, G.D., LARSON, B.L. and TOUCHBERRY, R.W. 1956. Protein production in the bovine. Breed and individual variations in the specific protein constituents of milk. J. Dairy Sci., 39, 1683

ROOK, J.A.F. and WOOD, M. 1959. Potassium and lactose in milk in relation to the physiology of milk secretion. Nature, Lond., 184, 647

ROOK, J.A.F. 1961. Variations in the chemical composition of the milk of the cow. Part I. Dairy Sci. Abstr., 23, 251

ROOK J.A.F. and BALCH, C.C. 1961. The effects of intra-ruminal infusions of acetic, propionic and butyric acids on the yield and composition of the milk of the cow. Br. J. Nutr., 15, 361

ROOK, J.A.F. and LINE, C. 1961. The effect of plane of energy nutrition of the cow on the secretion in milk of the constituents of the solids-not-fat fraction and on the concentrations of certain blood plasma constituents. Br. J. Nutr., 51, 109

ROOK, J.A.F., BALCH, C.C. and JOHNSON, V.W. 1965. Further observations on the effects of intra-ruminal infusions of volatile fatty acids and of lactic acid on the yield and composition of the milk of the cow. Br. J. Nutr., 19, 93

ROOK. J.A.F. 1976. Nutritional influences on milk quality. In Principles of Cattle Production, p.221. Ed. H. Swan and W.H. Broster, London; Butterworths

ROWLAND, S.J. and ROOK. J.A.F 1949. Composition of milk of various breeds of cow. Rep. nat. Res. Dairy, Reading, p.42

ROWLAND, S.J., NEAVE, F.K., DODD, F.H. and OLIVER, J. 1959. The effect of Staphylococcus pyogenes infections on milk production. Proc. 15th int. Dairy Congr. 1, 121

ROSEN, H. 1957. A modified ninhydrin colorimetric analysis for amino acids. Arch. Biochem. Biophys., 67,10

SATTER, L.D. and SLYTER, L.L. 1974. Effect of ammonia concentration on rumen microbial protein production in vitro. Br. J. Nutr., 32,199

SATTER, L.D. and ROFFLER, R.E. 1975. Nitrogen requirements and utilization in dairy cattle. J. Dairy Sci., 58, 1219

SCHINGOETHE, D.J., STAKE, P.E. and OWENS, M.J. 1973. Whey components in restricted roughage rations, milk composition, and rumen volatile fatty acids. J. Dairy Sci., 56, 909

SCHMIDT, G.H. and TRIMBERGER, G.W. 1963. Effect of unequal milking intervals on lactation milk, milk fat, and total solids production of cows. J. Dairy Sci., 46, 19

SCOTT, T.W. and COOK, L.J. 1975. Effect of dietary fat on lipid metabolism in ruminants. In Digestion and Metabolism in the Ruminant, p.510. Ed. I.W. McDonald and A.C.I. Warner. University of New England. Publ. unit.

SCHWAB, C.G. and SATTER, L.D. 1973. Response of lactating dairy cows to the abomasal infusion of amino acids. J. Dairy Sci., 56, 664 (Abstr)

SCHWAB, C.G. and SATTER, L.D. 1974. Effect of abomasal infusion of amino acids on lactating dairy cows. J. Dairy Sci., 57, 632 (Abstr)

SCHWAB, C.G., SATTER, L.D. and CLAY, A.B. 1976. Response of lactating dairy cows to abomasal infusion of amino acids. J. Dairy Sci., 59, 1254

SMITH, V.R. 1959. Physiology of Lactation. 5th ed. Ames; Iowa State University Press

SMITH, R.H. 1969. Reviews of the progress of Dairy Science. Section G. Nitrogen metabolism and the rumen. J. Dairy Res., 36, 313

SMITH, G.H. 1971. Glucose metabolism in the ruminant. Proc. Nutr., Soc., 30, 265

SMITH, G.H., McCARTHY, S. and ROOK, J.A.F. 1974. Synthesis of milk from B-hydroxy butyrate and acetate in lactating goats. J. Dairy Res., 41, 175

SMITH, J.S., WAINMAN, F.W. and DEWEY, P.J.S. 1975. The energy value to sheep of a mixed grass silage. Proc. Nutr. Soc., 34, 101A

SNEDECOR, G.W. and COCHRANE, W.G. 1967. Statistical methods. Ames; Iowa State University Press

SNIFFEN, C.J. 1973. Nitrogen utilization as related to solubility of NPN and protein in feeds. Proc. Cornell Nutr. Conf. Feed Mnfr. p.12

SPECHTER, H.H. 1972. Post-ruminal casein infusion of urea fed lactating cows. Ph.D. dissertation, Univ, Guelph, Guelph, Ontario, Canada

SPIRES, H.R., CLARK, J.H. and DERRIG, R.G. 1973. Post-ruminal administration of sodium caseinate in lactating cows. J. Dairy Sci., 56, 664

SPIRES, H.R., CLARK, J.H., DERRIG, R.G. and DAVIS, C.L. 1975. Milk production and nitrogen utilization in response to post-ruminal infusion of sodium caseinate in lactating cows. J. Nutr., 105, 1111

STORRY, J.E. and ROOK, J.A.F. 1962. Effects of large intra-ruminal additions of volatile fatty acids on the secretion of milk constituents. Proc. 16th int. Dairy Congr., A, 64

STORRY, J.E. and ROOK, J.A.F. 1965. Effects of intravenous infusions of acetate, B-hydroxybutyrate, triglyceride and other metabolites on the composition of the milk fat and blood of cows. Biochem. J. 97, 879

STORRY, J.E. and ROOK, J.A.F. 1966. The relationship in the cow between milk-fat secretion and ruminal volatile fatty acids. Br. J. Nutr., 20, 217

STORRY, J.E. 1970. Reviews of the progress of Dairy Science. Section A. Physiology. Ruminant metabolism in relation to the synthesis and secretion of milk fat. J. Dairy Res., 37, 139

STORRY, J.E. 1972. Milk fat; its synthesis and composition in relation to the nutrition of the dairy cow. J. Soc. Dairy Tech., 25, 40

STRUSS, H.E., PETERSON, W.E. and MIX, L.S. 1964. Survey of the utilization of uniform labelled C^{14} glucose in the perfused bovine mammary gland. J. Dairy Sci., 47, 695

SUTHERLAND, T.M. 1976. The overall metabolism of nitrogen in the rumen. In Reviews in Rural Science II : From Plant to Animal Protein, p.65. Ed. T.M. Sutherland, J.R. McWilliam and R.A. Leng. Univ. New England Printing unit

SUTTON, J.D. and JOHNSON, V.W. 1969. Fermentation in the rumen of cows given rations containing hay and flaked maize or rolled barley in widely different proportions. J. agric. Sci., Camb., 73, 459

TAMMINGA, S. 1973. The influence of the protein source on the protein digestion in the ruminant. Z. Tierphysiol. Tierernahrg. v. Futtermittelkde, 32, 185

TAPARIA, A.L. and DAVEY, A.W.F. 1970. The effect on food intake and milk production of adding concentrates to pasture fed cows. N.Z. Jl agric. Res., 13, 616

TEICHMAN, R.E., CARUOLO, E.V. and MOCHRIE, R.D. 1969. Milk production and composition responses to intravenous infusion of L-methionine. J. Dairy Sci., 52, 942 (Abstr.)

TERNOUTH, J.H. 1967. Post-prandial ionic and water exchange in the rumen. Res. Vet. Sci., 8, 283

THOMAS, P.C. 1975. Diet and milk secretion in the ruminant. World Rev. Anim. Prod., 11, 33

THOMAS, P.C. and KELLY, M.E. 1976. The effect of frequency of feeding on milk secretion on the Ayrshire cow. J. Dairy Res., 43, 1

THOMSON, D.J. 1968. The digestibility and utilization of fresh grass, hay and silage by sheep. Anim. Prod., 10, 240

TOUCHBERRY, R.W. 1974. Environmental and genetic factors in the development and maintenance of lactation. In Laction, 3, 349. Ed. B.L. Larson and V.R. Smith. New York; Academic Press

TYRRELL, H.F., MOE, P.W. and FLATT, W.P. 1970. Influence of excess protein intake on energy metabolism of the dairy cow. Proc. 5th Symp. Energy Metab. EAAP., p.69

TYRRELL, H.F., BOLT, D.J., MOE, P.W. and SWAN, H. 1972. Abomasal infusions of water, casein, or glucose in Holstein cows. J. Anim. Sci., 35, 277

TYRRELL, H.F. and MOE, P.W. 1972. Net energy value for lactation of a high and low concentrate ration containing corn silage. J. Dairy Sci., 55, 1106

TYRRELL, H.F. and MOE, P.W. 1975. Symposium. Production efficiency in the high producing cow. Effect of intake on digestive efficiency. J. Dairy Sci., 58, 115

ULYATT, M.J. 1969. Progress in defining the differences in nutritive value to sheep of perennial ryegrass, short rotation ryegrass and white clover. Proc. N.Z. Soc. Anim. Prod., 29, 114

ULYATT, M.J. and MacRAE, J.C. 1971. The sites of digestion of fresh herbage species in the gastro-intestinal tract of sheep. Proc. N.Z. Soc. Anim. Prod., 31, 74

ULYATT, M.J., DELLOW, D.W., EGAN, A.R. and WALKER, D.J. 1973. Quantitative aspects of fermentation in the gastro-intestinal tract of sheep fed fresh herbage. Proc. N.Z. Soc. Anim. Prod., 33, 139

ULYATT, M.J. and MacRAE, J.C. 1974. Quantitative digestion of fresh herbage by sheep. I. The sites of digestion of organic matter, readily fermentable carbohydrate, structural carbohydrate and lipid. J. agric. Sci., Camb., 82, 295

ULYATT, M.J., MacRAE, J.C., CLARKE, R.T. and PEARCE, P.D. 1975. Quantitative digestion of fresh herbage by sheep. iv. Protein synthesis in the stomach. J. agric. Sci., Camb., 84, 453

VALENTINE, S.C. and BROWN, D.C. 1973. Formaldehyde as a silage additive. II. The chemical composition and nutritive value of lucerne hay, lucerne silage, and formaldehyde and formic-acid treated lucerne silages. Aust. J. agric. Res., 24, 939

VALENTINE, S.C. and RADCLIFFE, J.C. 1975. The nutritive value for dairy cows of silage made from formaldehyde-treated herbage. Aust. J. agric. Res., 26, 769

VALLENCE, W.S. and McCLYMONT, G.L. 1959. Depression in percentage milk fat by parenteral glucose infusion and glycerol feeding. Nature, Lond., 183, 466

van ES, A.J.H. and BOEKHOLT, H.A. 1976. Protein requirements in relation to the lactation cycle. In Protein Metabolism and Nutrition, EAAP publ. 16, p.441. Ed. D. Cole, K. Boorman, P. Buttery, R. Neale and H. Swan. London; Butterworth

van't KLOOSTER, A. Th. and BOEKHOLT, H.A. 1972. Protein digestion in the stomachs and intestines of the cow. Neth. J. agric. Sci., 20,272

van SOEST, P. 1973. cited by SNIFFEN, C.J. 1973. Nitrogen utilization as related to solubility of NPN and protein in feeds. Proc. Cornell Nutr. Feed Mnfr. Conf., p.12

VIK-MO, L. and HUBER, J.T. 1971. Abomasal infusion of casein and glucose in lactating cows. J. Dairy Sci., 54, 790

VIK-MO, L. EMERY, R.S. and HUBER, J.T. 1974. Milk protein production in cows abomasally infused with casein or glucose. J. Dairy Sci., 57, 869

VIRTANEN, A.I. 1966. Milk production of cows on protein-free feed. Science, N.Y., 153, 1603

WAITE, R., WHITE, J.C.D. and ROBERTSON, A. 1956. Variations in the chemical composition of milk with particular reference to the solids-not-fat. I. The effect of stage of lactation, season of year and age of cow. J. Dairy Res., 23, 65

WALDO, D.R., KEYS, J.F. and GORDON, C.H. 1973. Preservation efficiency and dairy heifer response from unwilted formic and wilted treated silages. J. Dairy Sci., 56, 129

WALKER, D.J. 1970. Energetics of protein and carbohydrate metabolism in the rumen. Proc. 10th int. Grassld Congr., p.713

WALLACE, L.R. 1958. Wintering cows on autumn saved pasture. Proc. Rua. Frms. Conf., p.216

WALLACE, L.R. 1961. Nutritional requirements of dairy cattle. Proc. N.Z. Soc. Anim. Prod., 21, 64

WALLACE, L.R. and PARKER, O.F. 1966. Comparison of three systems of feeding silage after calving. Proc. Rua. Frms. Conf., p.148

WAYMAN, O., JOHNSON, H.D., MERILAN, C.P. and BERRY, I.L. 1962. Effect of ad libitum or force-feeding of two rations on lactating dairy cows subject to temperature stress. J.Dairy Sci., 45, 1472

WESTON, R.H. and HOGAN, J.P. 1971. The digestion of pasture plants by sheep. V. Studies with subterranean and Berseem clovers. Aust. J. agric. Res., 22, 139

WHEELOCK, J.V., ROOK, J.A.F. and DODD, F.H. 1965. The effect of incomplete milking or of an extended milking interval on the yield and composition of cow's milk. J. Dairy Res., 32, 237

WHEELOCK, J.V., ROOK, J.A.F., DODD, F.H. and GRIFFEN, T.K. 1966. The effect of varying the interval between milkings on milk secretion. J. Dairy Res., 33, 161

WILCOX, C.J., PFAU, K.O., MATHER, R.E. and BARTLETT, J.W. 1959. Genetic and environment influences upon solids-not-fat content of cow's milk. J. Dairy Sci., 42, 1132

WILCOX, C.J., GAUNT, S.N. and FARTHING, B.B. 1971. cited by TOUCHBERRY, R.W. 1974. Environmental and genetic factors in the development and maintenance of lactation. In Lactation, 3, 349. Ed. B.L. Larson and V.R. Smith. New York; Academic Press.

WILLIAMS, L.R., MARTZ, F.A. and HILDERBRAND, E.S. 1970. Feeding encapsulated methionine supplement to lactating cows. J. Dairy Sci., 53, 1709

WILKINS, R.J., WILSON, R.F. and WOOLFORD, M.K. 1974. The effects of formaldehyde on the silage fermentation. Proc. 5th Gen. Mtg. Eur. Grassld. Fed., Uppsala, p.197

WILKINSON, J.M., WILSON, R.F. and BARRY, T.N. 1976. Factors affecting the nutritive value of silage. Outl. Agric., 9, 3

WILSON, G.F. and McDOWALL, F.H. 1966. Ryegrass varieties in relation to dairy cattle performance. I. The influence of ryegrass varieties on milk yield and composition. N.Z. Jl agric. Res., 9,1042

WILSON, G.F. and DOLBY, R.M. 1967. Ryegrass varieties in relation to dairy cattle performance. II. A comparison of the milk yield and composition from a tetraploid and two diploid ryegrass varieties. N.Z. Jl agric. Res., 10, 415

WILSON, G.F., DAVEY, A.W.F. and DOLBY, R.M. 1967. Milk composition as affected by intra-ruminal infusion of volatile fatty acids to cows on a restricted ration. N.Z. Jl agric. Res., 10, 215

WILSON, G.F. and DOLBY, R.M. 1969. Ryegrass varieties in relation to dairy cattle performance. IV. Milk production from cows grazing ryegrass pastures at two stages of growth and receiving two levels of nitrogen fertilizer. N.Z. Jl agric. Res., 12, 489

WILSON, G.F. 1970. The influence of protein supplements on milk yield and composition. Proc. N.Z. Soc., Anim. Prod., 30, 123

WOLFF, J.E. and BERGMAN, E.N. 1972. Gluconeogenesis from amino acids in fed sheep. Am. J. Physiol., 223, 438

WRIGHT, D.E. and HUNGATE, R.E. 1967. Amino acid concentrations in rumen fluid. Appl. Microbiol., 15, 148

WRIGHT, J.A., ROOK, J.A.F. and WOOD, P.D.P. 1974. The responses in milk solids-not-fat and protein contents to improved feeding of dairy cows receiving winter-stall diets and underfed for varying periods. J. Dairy Res., 41, 155

YOUSEF, I.M., HUBER, J.T. and EMERGY, R.S. 1970. Milk protein synthesis as affected by high grain, low fibre rations. J. Dairy Sci., 53, 734

.6 APPENDICES

APPENDIX I. Residual sums of squares for models (i), (ii), (iii), and (iv) relating milk variates to DEI/kg $LW^{0.75}$. (Experiment 3.1)

Residual SS	MODEL (i) (df=19)	(ii) (df=20)	(iii) (df=20)	(iv) (df=21)
Milk (kg)	17.3855	17.6487	18.2958	29.5586
Protein (g)	14222.5938	14574.0277	15622.8647	35257.3608
Fat (g)	48357.4737	48365.1935	49745.2521	143977.6938
Lactose (g)	4185.4172	3983.7451	4071.2270	6548.5445
Fat %	1.0860	1.0988	1.0931	3.8503
Protein %	0.4133	0.4135	0.4190	0.6459
Lactose %	2.7488	2.7627	2.7490	3.3570

APPENDIX 2. Relationship between Δ LW (kg/d), (y), and DEI/kg $LW^{0.75}$/d, (x), in Period III. (Experiment 3.1).

Diet	bx ± SE	c	r	Significance
Silage	-0.55 ± 1.15	1.08	-0.15	
Pasture	-0.67 * ± 0.32	2.12	-0.54	
Silage	-0.66 ± 0.33	1.22	-0.40	***
Pasture		2.10		

APPENDIX 3. Effect of diet and level of feeding on nitrogen partition. (Experiment 3.1).

Item	Diet	Mean	bx ± SE	C	RSD	r
N, % of intake						
Faeces	S	31.7	-0.32 ± 1.2	32.5	2.4	-0.07
	P	34.9		35.8		
Urine	S	42.4	-5.9 * ± 2.2	59.0	4.5	-0.59
	P	48.7		66.2		
Milk	S	17.8	-0.13 ± 1.67	18.1	3.3	-0.02
	P	24.1		24.4		
NR	S	8.1	6.4 *** ± 1.5	-9.7	3.1	0.75
	P	-7.7		-26.4		
N, % of digested						
Urine	S	61	-14.9 * ± 5.3	90	7.2	-0.61
	P	75		103		
Milk	S	26	0.14 ± 3.4	26	4.6	0.01
	P	37		37		
NR	S	12	14.5 *** ± 3.2	-16	4.3	0.79
	P	-7		-40		

x denotes DNI/kg $LW^{0.75}$

APPENDIX 4. Summary of regressions of DEI (MJ/day) on FCM (kg/d), Δ LW (kg/d), and $LW^{0.75}$ (kg); and of (milk energy + ΔLW energy)/$LW^{0.75}$ on MEI/$LW^{0.75}$ in Period III. (Experiment 3.1).

	MEAN VALUES				COEFFICIENTS ± SE			
Diet	DEI	FCM	ΔLW	$LW^{0.75}$	bFCM	bALW	$bLW^{0.75}$	C
Silage	105.8	8.86	-0.52	79.6	2.24 ± 1.79	1.87 ± 4.81	1.27^{**} ± 0.27	-9.08
Pasture	127.4	11.83	-1.59	80.9	3.19^{*} ± 1.29	33.16^{**} ± 8.9	2.27^{**} ± 0.51	-41.80

	Milk Energy + $\Delta LW/LW^{0.75}$	$MEI/LW^{0.75}$	$bMEI/LW^{0.75}$	C
Silage	0.251	1.078	0.459 ± 0.375	-0.1297
Pasture	0.182	1.295	0.459^{**} ± 0.124	-0.4033
Silage			0.450 ± 0.142	-0.182
Pasture				-0.438

APPENDIX 5. Summary of DMI (kg/kg $LW^{0.75}$/d), DEI (MJ/kg $LW^{0.75}$/d), and DNI (g/kg $LW^{0.75}$/d) for Period II. (Experiment 3.2).

Diet	Feeding Level	DEI/LW $^{.75}$	DMI/LW $^{.75}$	DNI/LW $^{.75}$
C.S.	100	1.54	0.140	2.03
	75	1.27	0.114	1.68
	50	0.92	0.085	1.21
F.S.	100	1.41	0.152	1.70
	75	1.00	0.107	1.20
	55	0.72	0.079	0.85
W.S.	100	1.55	0.140	1.99
	75	1.13	0.103	1.46
	50	0.84	0.079	1.08
Diet Means C.S.		1.24	0.113	1.64
F.S.		1.05	0.113	1.25
W.S.		1.17	0.107	1.51
Level Means	100	1.50	0.144	1.91
	75	1.14	0.108	1.45
	50	0.83	0.081	1.19
S.D.		0.099	0.010	0.129
Significance				
Diets		**	N.S.	***
Levels		***	***	***
Diets x Levels		NS	N.S.	N.S.

APPENDIX 6. Relationships between milk variates and DEI/$LW^{0.75}$ (MJ/kg $LW^{0.75}$/d) in Period II (Model (i); Experiment 3.2).

Item	Mean	S.D.	$B\mu \pm SE$	Sign.	Ration	$bx \pm SE$	Sign.	C	R.S.D.	R
Milk Yield (kg/d)	6.08	1.89	0.81 ± 0.06	***	C.S.	3.35 ± 0.09	***	-4.24	0.60	0.96
					F.S.	2.47 ± 0.68	**	-3.56		
					W.S.	2.78 ± 0.66	***	-3.72		
Milk Fat (g/d)	299	83.9	0.82 ± 0.07	***	C.S.	136.4 ± 45.91	**	-163.73	31.7	0.94
					F.S.	93.5 ± 36.18	*	-109.47		
					W.S.	105.8 ± 35.87	**	-121.20		
Milk Protein (g/d)	189	55.3	0.73 ± 0.06	***	C.S.	93.2 ± 25.24	**	-104.17	79.4	0.96
					F.S.	66.7 ± 20.25	**	-76.61		
					W.S.	95.6 ± 19.75	***	-101.98		
Milk Lactose (g/d)	286	92.3	0.81 ± 0.07	***	C.S.	161.4 ± 43.23	**	-203.62	30.6	0.96
					F.S.	116.7 ± 34.51	**	-172.31		
					W.S.	141.4 ± 33.84	***	-192.84		
Milk Fat (%)	4.99	0.58	0.99 ± 0.08	***	C.S.	-0.53 ± 0.33	NS	0.97	0.24	0.93
					F.S.	-0.34 ± 0.27	NS	1.09		
					W.S.	-0.43 ± 0.28	NS	1.11		
Milk Protein (%)	3.15	0.26	0.97 ± 0.13	***	C.S.	-0.29 ± 0.20	NS	0.23	0.14	0.88
					F.S.	-0.10 ± 0.17	NS	0.14		
					W.S.	0.21 ± 0.15	NS	-0.20		
Milk Lactose (%)	4.70	0.18	0.91 ± 0.09	***	C.S.	0.09 ± 0.11	NS	0.48	0.08	0.92
					F.S.	0.09 ± 0.09	NS	0.36		
					W.S.	0.08 ± 0.09	NS	0.36		

(i) μ denotes production in Period I
(ii) x denotes DEI/kg $LW^{0.75}$ in Period II
(iii) mean DEI/$LW^{0.75}$ = 1.19 ± 0.33

APPENDIX 7. Relationship between LW change (kg/d), (y), and DEI/$LW^{0.75}$ (MJ/kg $LW^{0.75}$/d), (x), in Period II. (Experiment 3.2).

Diet	C	b ± SE	Significance	RSD	R	Sign. of Difference b's	Sign. of Difference c's
C.S.	-1.466	0.925 ± 0.8173	NS	0.724	0.393		
F.S.	-1.070	1.239 ± 0.5542	+	0.494	0.645	NS	**
W.S.	-1.494	1.699 ± 0.5269	*	0.470	0.773		
C.S.	-1.968						
F.S.	-1.123	1.289 ± 0.3621	**	0.558	0.596		
W.S.	-1.021						

APPENDIX 8. Summary of DMI (kg/kg $LW^{0.75}$/d), DEI (MJ/kg $LW^{0.75}$/d), DNI (g/kg $LW^{0.75}$/d) for Period II (Experiment 3.3.1).

Diet	Feeding Level	DEI/LW $^{.75}$	DMI/LW $^{.75}$	DNI/LW $^{.75}$
S	100	1.379	0.100	1.82
	75	1.058	0.077	1.40
	50	0.719	0.052	0.95
S + P	100	1.655	0.121	2.73
	75	1.207	0.088	1.99
	50	0.875	0.063	1.44
S + MS				
	100	1.248	0.100	1.09
	75	0.930	0.075	0.81
	50	0.682	0.055	0.60
Diet Means S		1.052	0.076	1.39
S + P		1.246	0.091	2.06
S + MS		0.953	0.077	0.83
Level Means	100	1.427	0.107	1.88
	75	1.065	0.080	1.40
	50	0.759	0.057	1.00
S.D.		0.102	0.012	0.147
Significance				
Diets		***	***	***
Levels		***	***	***
Diets x Levels		NS	NS	**

APPENDIX 9. Relationships between the milk variates and DEI/$LW^{0.75}$ (MJ/kg $LW^{0.75}$/d) in Period II (Model (i). (Experiment 3.3.1).

Item	Mean	S.D.	Bμ ± SE	Sign.	Ration	bx ± SE	Sign.	C	R.S.D.	R
Milk Yield (kg/d)	9.44	2.48	0.60 ± 0.04	***	S	4.04 ± 0.69	***	-2.81	0.70	0.97
					S+P	3.93 ± 0.62	***	-2.02		
					S+MS	3.41 ± 0.97	**	-1.98		
Fat Yield (g/d)	417	95.9	0.54 ± 0.07	***	S	92.5 ± 40.20	*	10.14	40.82	0.93
					S+P	88.3 ± 37.98	*	79.97		
					S+MS	142.0± 57.74	*	-29.09		
Protein Yield(g/d)	257	66.1	0.52 ± 0.05	***	S	131.7 ± 17.13	***	-88.95	17.42	0.97
					S+P	132.3 ± 15.96	***	-51.51		
					S+MS	127.6 ± 24.52	***	-72.83		
Lactose Yield (g/d)	460	119.3	0.65 ± 0.05	***	S	213.5 ± 37.36	***	-151.17	37.90	0.96
					S+P	191.0 ± 33.72	***	-96.09		
					S+MS	184.2 ± 53.27	***	-109.83		
Milk Fat (%)	4.49	0.75	1.08 ± 0.16	***	S	-1.22 ± 0.48	*	0.64	0.46	0.84
					S+P	-1.33 ± 0.43	**	1.24		
					S+MS	0.02 ± 0.66	NS	-0.46		
Milk Prot. (%)	2.76	0.32	0.83 ± 0.06	***	S	0.21 ± 0.10	+	-0.20	0.10	0.96
					S+P	0.13 ± 0.09	NS	0.11		
					S+MS	0.44 ± 0.14	**	-0.35		
Milk Lact. (%)	4.87	0.20	0.94 ± 0.12	***	S	0.21 ± 0.10	*	-0.08	0.10	0.90
					S+P	0.11 ± 0.09	NS	0.19		
					S+MS	0.16 ± 0.14	NS	0.16		

(i) μ denotes production in Period I
(ii) x denotes DEI/kg $LW^{0.75}$ in Period II
(iii) mean DEI/$LW^{0.75}$ = 1.08 ± 0.32

APPENDIX 10. Residual sums of squares for Models (i), (ii), (iv) relating milk variates to DEI/$LW^{0.75}$. (Experiment 3.3.1).

Residual SS	Model (i) (df=20)	(ii) (df=22)	(iii) (df=22)	(iv) (df=24)
Milk (kg)	9.7333	10.1854	9.8664	11.9061
Fat (g)	33326.5174	38374.4013	34412.6888	52355.8723
Protein (g)	6070.1359	6953.5134	6078.3588	12071.7214
Lactose (g)	28731.9646	30722.6323	29255.5163	33452.5138
Fat %	4.3177	5.2672,	5.0056	5.7797
Protein %	0.1985	0.2951	0.2352	0.4269
Lactose %	0.2102	0.2185	0.2183	0.2200

APPENDIX 11. Relationship between LW change (kg/d), (y), and DEI/$LW^{0.75}$ (MJ/kg $LW^{0.75}$/d)($\bar{x}$), in Period II. (Experiment 3.3.1).

Diet	C	b ± SE	Significance	RSD	R	Sign. of difference b's	Sign. of difference c's
S	-3.790	2.371 ± 0.5855	**	0.487	0.837		
S + P	-2.045	1.128 ± 0.4800	*	0.489	0.664	NS	NS
S + MS	-2.852	1.910 ± 0.9885	+	0.710	0.590		
S	-3.074						
S + P	-2.746	1.690 ± 0.3820	***	0.572	0.678	-	NS
S + MS	-2.642						
Total	-2.809	1.680 ± 0.3575	***	0.581	0.685	-	-

APPENDIX 12. Adjusted mean concentrations and molar proportions of rumen VFA immediately before feeding (0h), 4h and 12h later (Experiment 3.3.1).

Diet/Level	Time	Concentration (meq/100 ml) of VFA							Molar % of Total VFA					
		HAc	HPr	IHBu	nHBu	IHVa	nHVa	Total	HAc	HPr	IHBu	nHBu	IHVa	nHVa
S	0h	3.89	0.87	0.08	0.61	0.08	0.10	5.53	70.3	15.6	1.4	9.4	1.4	1.8
S + P		3.42	0.51	0.08	0.53	0.11	0.11	5.02	68.3	15.4	1.6	10.6	2.2	2.1
S + MS		3.42	0.75	0.05	0.49	0.08	0.08	4.86	69.8	15.5	1.1	10.0	1.9	1.8
SED		0.905	0.196	0.019	0.147	0.018	0.027	1.288	1.00	0.98	0.22	0.49	0.32	0.23
Sign.		NS	NS	NS	NS	NS	NS	NS	NS	NS	+	+	NS	NS
Level 100 %		3.53	0.71	0.06	0.58	0.06	0.08	5.07	69.2	16.1	1.2	10.2	1.5	1.7
50 %		3.62	0.70	0.08	0.50	0.11	0.11	5.20	69.7	14.9	1.5	9.7	2.1	2.0
SED		0.739	0.160	0.016	0.120	0.015	0.022	1.052	0.81	0.80	0.18	0.40	0.26	0.18
Sign.		NS	NS	NS	NS	*	NS	NS	NS	NS	+	NS	*	NS
S	4h	5.38	1.66	0.11	0.85	0.17	0.17	8.30	64.2	20.0	1.3	10.4	2.0	2.1
S + P		4.95	1.52	0.08	0.95	0.13	0.15	7.79	63.3	19.6	1.0	12.4	1.8	2.0
S + MS		4.65	1.35	0.09	0.80	0.13	0.15	7.17	64.6	19.0	1.3	11.2	1.8	2.1
SED		0.751	0.207	0.018	0.097	0.029	0.018	1.043	1.63	1.03	0.17	1.20	0.25	0.24
Sign.		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Level 100 %		5.06	1.42	0.08	0.84	0.13	0.14	7.65	65.5	18.6	1.1	11.3	1.7	1.9
50 %		4.93	1.60	0.11	0.84	0.13	0.17	7.85	62.5	20.4	1.3	11.5	2.0	2.2
SED		0.613	0.169	0.014	0.080	0.023	0.015	0.852	1.33	0.84	0.14	0.98	0.20	0.20
Sign.		NS	NS	NS	NS	NS	+	NS	*	+	+	NS	NS	NS

Cont.

APPENDIX 12 (Continued).

Diet/Level	Time	Concentration (meq/100 ml) of VFA							Molar % of Total VFA					
		HAc	HPr	IHBu	nHBu	IHVa	nHVa	Total	HAc	HPr	IHBu	nHBu	IHVa	nHVa
S	12h	4.68	1.30	0.08	0.66	0.10	0.12	6.92	67.4	18.7	1.2	9.7	1.4	1.7
S + P		4.61	1.23	0.07	0.84	0.08	0.11	6.93	66.5	17.8	0.9	12.0	1.2	1.5
S + MS		5.01	1.55	0.06	0.74	0.11	0.10	7.57	66.3	20.1	0.8	9.9	1.6	1.3
SED		0.738	0.275	0.013	0.108	0.022	0.020	1.098	0.69	1.46	0.14	0.91	0.28	0.16
Sign.		NS	NS	NS	NS	NS	NS	NS	NS	NS	+	+	NS	NS
Level 100 %		5.14	1.52	0.07	0.78	0.10	0.12	7.73	6.65	19.4	0.9	10.3	1.3	1.6
50 %		4.39	1.20	0.07	0.71	0.09	0.09	6.55	67.0	18.3	1.0	10.8	1.4	1.4
SED		0.602	0.225	0.011	0.088	0.018	0.016	0.896	0.56	1.19	0.11	0.75	0.22	0.08
Sign.		NS	NS	NS	NS	NS	+	NS	NS	NS	NS	NS	NS	NS

APPENDIX 13. Summary of DMI (kg/kg $LW^{0.75}$/d), DEI (MJ/kg $LW^{0.75}$/d) and DNI (g/kg $LW^{0.75}$/d) in Period II (Experiment 3.3.2).

Diet	Feeding Level	DEI/$LW^{0.75}$	DMI/$LW^{0.75}$	DNI/$LW^{0.75}$
S	100	1.28	0.10	2.45
	75	1.13	0.09	2.16
	50	0.73	0.06	1.41
S + PC_1	100	1.46	0.11	3.24
	75	1.22	0.09	2.66
	50	0.78	0.06	1.69
S + PC_2	100	1.71	0.13	4.13
	75	1.26	0.09	3.04
	50	0.81	0.06	1.94
Diet Means	S	1.05	0.08	2.01
	S + PC_1	1.15	0.09	2.53
	S + PC_2	1.26	0.09	3.04
Level Means	100	1.48	0.11	3.27
	75	1.20	0.09	2.62
	50	0.77	0.06	1.68
S.D.		0.140	0.011	0.281
Significance				
Diets		*	+	***
Levels		***	***	***
Diets x Levels		NS	NS	*

APPENDIX 14. Relationships between the milk variates and DEI/LW$^{0.75}$ (MJ/kg LW$^{0.75}$/d) in Period II (Model (i). (Experiment 3.3.2).

Item	Mean	S.D.	Bμ ± SE	Sign.	Ration	bX ± SE	Sign.	C	R.S.D.	R
Milk Yield (kg/d)	10.11	2.22	0.70 ± 0.09	***	S	2.57 ± 1.03	*	-1.98	0.92	0.93
					S + PC_1	3.70 ± 1.08	**	-2.45		
					S + PC_2	3.52 ± 0.82	***	-2.24		
Milk Fat (g/d)	443	87.1	0.65 ± 0.07	***	S	-4.27 ± 42.43	NS	36.64	38.1	0.92
					S + PC_1	108.69 ± 42.95	*	-26.68		
					S++ PC_2	133.73 ± 33.28	***	-42.39		
Milk Protein (g/d)	278	66.6	0.67 ± 0.11	***	S	84.26 ± 28.23	**	-77.03	25.1	0.94
					S + PC_1	143.14 ± 29.08	***	-133.34		
					S + PC_2	136.30 ± 22.29	***	-103.31		
Milk Lactose (g/d)	489	109.1	0.65 ± 0.09	***	S	144.52 ± 50.22	**	-99.75	45.1	0.93
					S + PC_1	199.69 ± 53.39	**	-125.09		
					S + PC_2	183.5 ± 40.01	***	-113.05		
Milk Fat %	4.44	0.71	0.84 ± 0.09	***	S	-1.36 ± 0.385	**	1.89	0.35	0.90
					S + PC_1	-0.61 ± 0.376	NS	1.36		
					S + PC_2	-0.35 ± 0.302	NS	1.22		
Milk Protein %	2.75	0.24	0.93 ± 0.09	***	S	0.16 ± 0.123	NS	-0.12	0.11	0.92
					S + PC_1	0.44 ± 0.119	**	-0.38		
					S + PC_2	0.33 ± 0.094	**	-0.22		
Milk Lactose %	4.84	0.17	0.75 ± 0.10	***	S	0.17 ± 0.115	NS	0.87	0.10	0.88
					S + PC_1	0.14 ± 0.112	NS	0.88		
					S + PC_2	0.18 ± 0.089	NS	0.84		

(i) μ denotes production in Period I
(ii) x denotes DEI/kg LW$^{0.75}$ in Period II
(iii) overall mean DEI/LW$^{0.75}$ = 1.17 ± 0.33

APPENDIX 15. Residual sums of squares for Models (i), (ii), (iii), and (iv) relating milk variates to DEI/$LW^{0.75}$. (Experiment 3.3.2).

Residual SS	Model			
	(i) (df=20)	(ii) (df=22)	(iii) (df=22)	(iv) (df=24)
Milk (kg)	19.6583	19.7208	20.2589	24.9118
Protein (g)	14502.4534	14928.0064	16272.0326	23046.1341
Fat (g)	33438.4116	35670.4596	43364.5590	75598.4130
Lactose (g)	46787.7961	46963.2276	48017.5487	56338.9853
Fat %	2.7556	2.9205	3.2778	4.2881
Protein %	0.2682	0.2878	0.2986	0.3435
Lactose %	0.2338	0.2346	0.2354	0.2907

APPENDIX 16. Relationship between LW change (kg/d), (y), and DEI/$LW^{0.75}$ (MJ/kg $LW^{0.75}$), (x), in Period II. (Experiment 3.3.2).

Diet		C	b ± SE	Significance	RSD	R	Sign. of difference b's	Sign. of difference c's
S		-3.026	2.110 ± 0.5643	**	0.507	0.764	NS	NS
S + PC_1	8.5 %	-2.209	1.551 ± 0.4235	**	0.390	0.811		
S + PC_2	18 %	-2.514	1.854 ± 0.3346	***	0.384	0.902		
Pooled		-2.723 -2.539 -2.493	1.837 ± 0.2492	***	0.430	0.822		NS
Total		-2.664	1.893 ± 0.2428	***	0.427	0.827		

APPENDIX 17. Adjusted mean concentrations and molar proprotions of rumen VFA immediately before feeding (0h) and 4h later. (Experiment 3.3.2).

Diet/Level	Time	Concentration (meg/100 ml) of VFA							Molar % of Total VFA					
		HAc	HPr	IHBu	nHBu	IHVa	nHVa	Total	HAc	HPr	IHBu	nHBu	IHVa	nHVa
S	0h	3.68	0.88	0.06	0.54	0.07	0.04	5.26	69.9	16.7	1.1	10.4	1.3	0.8
S + PC_1		4.30	1.04	0.06	0.63	0.09	0.03	6.14	70.0	16.9	1.1	10.2	1.5	0.5
S + PC_2		5.47	1.38	0.08	0.79	0.10	0.06	7.89	69.3	17.5	1.1	10.1	1.3	0.8
SED		0.585	0.162	0.010	0.090	0.010	0.013	0.827	0.76	1.02	0.21	0.43	0.16	0.18
Sign.		*	*	NS	*	*	+	*	NS	NS	NS	NS	NS	NS
Level 100 %		5.06	1.25	0.06	0.74	0.08	0.04	7.23	70.0	17.1	1.0	10.2	1.2	0.6
50 %		3.91	0.95	0.07	0.56	0.08	0.04	5.63	69.5	16.9	1.3	10.1	1.5	0.8
SED		0.478	0.132	0.009	0.073	0.008	0.011	0.676	0.62	0.83	0.17	0.35	0.13	0.15
Sign.		*	*	NS	*	NS	NS	*	NS	NS	+	NS	+	NS
S	4h	6.33	1.81	0.13	0.90	0.21	0.20	9.58	65.8	19.05	1.4	9.5	2.2	2.2
S + PC_1		7.31	2.13	0.16	0.99	0.22	0.22	11.04	66.2	19.38	1.5	8.9	2.0	2.0
S + PC_2		8.10	2.02	0.16	1.10	0.24	0.25	11.87	68.2	17.10	1.4	9.3	2.0	2.0
SED		0.569	0.176	0.016	0.097	0.019	0.021	0.742	1.90	1.45	0.12	0.54	0.12	0.20
Sign.		*	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS
Level 100 %		7.59	1.97	0.16	1.01	0.23	0.23	11.19	67.8	17.7	1.4	8.9	2.1	2.1
50 %		6.91	2.00	0.14	0.99	0.21	0.21	10.47	65.7	19.3	1.4	9.5	2.0	2.0
SED		0.465	0.143	0.013	0.079	0.016	0.017	0.606	1.55	1.19	0.10	0.44	0.10	0.17
Sign.		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

APPENDIX 18. PCV %, plasma urea (mg/100 ml) and ∝ amino N (mg/l) immediately before feeding and 4 h later from cows fed silage and receiving abomasal infusions of sodium caseinate, sodium caseinate plus L-methionine, or saline (Mean differences ± SED).

Experiment 3.4.1	0 h	4 h
HC-HS		
% PCV	0.53 ± 0.65	-0.73 ± 0.28
Plasma Urea	0.30 ± 0.72	0.50 ± 0.91
Plasma Amino N	-2.30 ± 2.03	-0.58 ± 3.42
LC-LS		
% PCV	-0.23 ± 0.32	-0.28 ± 0.64
Plasma Urea	0.92 ± 0.68	-0.43 ± 1.05
Plasma Amino N	-6.75 ± 2.99	2.23 ± 4.94
HC-LC		
% PCV	0.75 ± 0.92	-0.45 ± 0.60
Plasma Urea	-0.63 ± 1.24	0.93 ± 1.30
Plasma Amino N	4.45 ± 3.55	-2.80 ± 8.13
Experiment 3.4.2		
CM-S		
% PCV	-0.28 ± 0.56	1.55 ± 0.92
Plasma Urea	-1.48 ± 1.80	-0.70 ± 1.05
Plasma Amino N	-2.09 ± 3.59	23.86 ± 2.04**
C-S		
% PCV	-0.85 ± 0.52	-0.13 ± 0.77
Plasma Urea	-0.35 ± 1.39	1.28 ± 0.86
Plasma Amino N	6.05 ± 3.97	9.92 ± 4.02
CM-C		
% PCV	0.58 ± 0.51	1.68 ± 1.15
Plasma Urea	-1.13 ± 3.17	-1.98 ± 1.83
Plasma Amino N	-8.14 ± 2.83	13.94 ± 5.41+

HC, C = 300 g Na caseinate in 10 l
LC = 150 g Na caseinate in 5 l
HS, S = 10 l saline
LS = 5 l saline
CM = 300 g Na caseinate plus 12 g methionine in 10 l.

APPENDIX 19. Effect of abomasal infusions of 300 g/d sodium caseinate (C) or (C) plus g/d L-methionine (CM) on the concentrations of amino acids (u moles/l) of cows on silage rations immedately before feeding (Oh) and 4h later. (Experiment 3.4.2).

Amino Acid	Time	Mean		CM-S	C-S	(CM-S)-(C-S)
		C	CM	Diff ± SED	Diff ± SED	Diff ± SED
Valine	Oh	210	160	17.2 ± 9.60	57.4 ± 7.0 **	-40.3 ± 11.2 *
	4h	290	300	177.4 ± 23.7 **	155.8 ± 32.0 *	21.5 ± 38.1
	4h - Oh			160.1 ± 30.5 *	98.5 ± 35.9 +	61.6 ± 48.9
Cysteine	Oh	16	42	16.1 ± 8.2	2.2 ± 4.5	13.9 ± 12.4
	4h	18	37	26.5 ± 11.7	8.4 ± 3.9	18.1 ± 11.2
	4h - Oh			10.4 ± 19.2	6.1 ± 5.2	4.2 ± 23.5
Methionine	Oh	9	10	1.6 ± 2.7	-0.6 ± 1.3	2.2 ± 3.2
	4h	26	73	66.0 ± 18.1 *	18.6 ± 9.9	47.4 ± 23.2
	4h - Oh			64.4 ± 19.8 *	19.2 ± 8.7	45.1 ± 23.8
Isoleucine	Oh	109	87	-1.1 ± 2.1	17.0 ± 5.9 +	-18.1 ± 5.4 *
	4h	137	185	111.9 ± 40.1 +	61.7 ± 16.7 *.	50.1 ± 49.4
	4h - Oh			113.0 ± 41.9 +	44.7 ± 17.7 +	68.2 ± 51.9
Leucine	Oh	116	87	-7.4 ± 4.5	17.1 ± 4.8 *	-24.5 ± 2.5 **
	4h	170	245	176.9 ± 58.6 +	98.6 ± 27.4 *	78.2 ± 76.2
	4h - Oh			184.2 ± 63.1 +	81.5 ± 26.4 +	102.7 ± 77.0
Tyrosine	Oh	44	36	5.2 ± 3.8	7.4 ± 5.7	-2.1 ± 8.6
	4h	92	138	108.9 ± 21.4 *	55.2 ± 16.5 *	48.6 ± 34.1
	4h - Oh			98.6 ± 21.6 *	47.9 ± 13.3 *	50.7 ± 31.9
Phenylalanine	Oh	48	45	-0.75 ± 5.7	-3.4 ± 4.0	2.6 ± 5.6
	4h	57	84	41.5 ± 17.9	15.7 ± 8.2	25.8 ± 20.2
	4h - Oh			42.2 ± 20.4	19.1 ± 11.6	23.1 ± 21.3

Cont.

APPENDIX 19. (Cont.)

Amino Acid	Time	Mean C	Mean CM	CM-S Diff ± SED	C-S Diff ± SED	(CM-S)-(C-S) Diff ± SED
∝ Amino-n-butyric	0h	26	27	10.3 ± 4.2 +	7.8 ± 3.4	2.6 ± 2.7
	4h	26	30	12.4 ± 2.9 *	7.5 ± 3.3	4.9 ± 0.9*
	4h - 0h			2.0 ± 1.7	-0.2 ± 3.7	2.2 ± 3.2
Plasma NH_3	0h	609	638	-15.2 ± 47.9	-81.0 ± 15.4*	65.7 ± 60.5
	4h	578	686	-18.7 ± 60.1	-224.9 ± 68.3*	216.1 ± 115.7
	4h - 0h			6.5 ± 48.5	-143.9 ± 74.8	150.4 ± 108.4

APPENDIX 20. The effect of abomasal infusions of glucose (290 g/d), L-methionine (12 g/d), sodium caseinate (300 g/d) or saline (10 l/d) on gross energy partition of cows fed silage. (Experiment 3.4.3).

Variable	Infusate				SED	Sign.
	Casein	Glucose	Methionine	Saline		
Gross Energy Intake (MJ/d)						
Silage	106.9a	109.3a	109.3a	109.6a	± 1.06	NS
Infused	6.8	4.4	-	-		
TOTAL	113.7a	113.7a	109.3b	109.6b	± 1.06	**
Apparent Digestibility %	69.7a	70.0a	68.5a	68.9a	± 0.58	NS
Energy Content (MJ/kg DM) of:						
Milk	24.6a	24.8a	24.7a	24.7a	± 0.28	NS
Faeces	17.6a	17.3a	17.7a	17.5a	± 0.20	NS
Urine	10.2a	8.9b	8.6b	8.4b	± 0.37	*
Energy Excreted (MJ/d)						
Milk	17.3a	17.1a	17.3a	15.7a	± 1.07	NS
Faeces	34.3a	34.0a	34.4a	34.0a	± 0.95	NS
Urine	7.9a	6.8a	7.3a	7.0a	± 0.40	NS
Energy, % of GEI						
Faeces	30.0a	30.0a	31.4a	31.2a	± 0.67	NS
Urine	7.0a	6.1a	6.7a	6.5a	± 0.38	NS
Methane(est)	7.8c	8.0b	8.4a	8.4a	± 0.04	***
Milk	15.2a	15.1a	15.8a	14.4a	± 1.06	NS
Residual(est)	40.0a	40.8a	37.7a	39.5a	± 1.32	NS
Milk + Residual	55.2a	55.9a	53.5a	53.9a	± 0.95	NS
Energy, % of DEI						
Urine	9.8a	8,8a	9.8a	9.5a	± 0.55	NS
Milk	21.8a	21.7a	23.0a	21.0a	± 1.60	NS
Residual	57.3a	58.1a	55.1a	57.4a	± 1.63	NS
Milk + Residual (ME)	78.9a	79.8a	78.1a	78.1a	± 0.73	NS

Means not containing common letters differ significantly (P<0.05)

APPENDIX 21. The effects of abomasal infusions of glucose (290 g/d), L-methionine (12 g/d), sodium caseinate (300 g/d), and saline (10 l/d) on PCV %, plasma urea and ∝ amino N concentrations immediately before feeding (0h) and 4h later. (Experiment 3.4.3).

Variable	Infusate					
	Casein	Glucose	Methionine	Saline	SED	Sign.
PCV %						
0h	31.7a	30.8a	31.1a	30.8a	± 0.84	NS
4h	31.5a	30.1a	30.6a	31.1a	± 0.62	NS
4h - 0h	-0.25a	-0.65a	-0.48a	0.33a	± 0.648	NS
Plasma Urea (mg/100 ml)						
0h	8.25a	8.60a	6.60a	7.90a	± 0.89	NS
4h	15.0a	13.3ab	11.7b	13.9ab	± 1.12	NS
4h - 0h	6.7a	4.8b	5.1ab	6.0ab	± 0.48	*
Plasma Amino N (mg/l)						
0h	41.4a	48.6a	48.8a	41.1a	± 8.44	NS
4h	51.3a	37.6b	37.9b	37.4b	± 4.61	+
4h - 0h	9.9a	-11.0a	-11.0a	-3.7a	±10.84	NS

Means not containing common letters differ significantly ($P<0.05$)

APPENDIX 22. Mean PCV %, and concentrations of plasma urea and plasma ∝ amino N immediately before feeding (0h) and 4h later of cows fed silage rations and receiving abomasal infusions of sodium caseinate (C, 300 g/d), glucose (G, 290 g/d), glucose plus sodium caseinate (G+C), or saline (S, 10 l/d). (Experiment 3.4.4).

Variable	Infusate					Significances		
	C	G	G+C	S	SED	C	G	CxG
PCV %								
0h	29.2	28.2	27.4	28.3	± 0.89	NS	NS	NS
4h	30.0	29.9	29.2	30.4	± 0.45	NS	+	NS
4h - 0h	0.80	1.78	1.80	2.10	± 1.00	NS	NS	NS
Plasma Urea (mg/100 ml)								
0h	8.8	8.3	8.2	8.4	± 0.42	NS	NS	NS
4h	14.4	12.8	12.4	12.8	± 0.86	NS	NS	NS
4h - 0h	5.60	4.43	4.15	4.40	± 0.99	NS	NS	NS
Plasma Amino N (mg/l)								
0h	39.4	43.6	41.6	42.6	± 2.52	NS	NS	NS
4h	53.7	30.0	46.3	35.2	± 4.65	***	NS	NS
4h - 0h	14.23	-13.65	4.70	-7.38	± 5.34	**	+	NS

APPENDIX 23. Mean PCV % and concentrations of ammonia (mg/100 ml), urea (mg/100 ml) and ∝ amino N (mg/l) in plasma, and mean pH and ammonia concentration in rumen fluid (mg/100 ml) immediately before feeding (0h) and 4h later of cows fed pasture (P) or silage (S) rations and receiving abomasal infusions of 300 g/d sodium caseinate (c) or 10 l/d saline (s). (Experiment 3.4.5).

Item	Time	Treatments		Pc-Ps		Sc-Ss		(Pc-Ps)-(Sc-Ss)		Ps-Ss	
		Pc	Sc	Diff ± SED	Sign.	Diff ± SED	Sign.	Diff ± SED	Sign.	Diff ± SED	Sign.
PCV %	0h	31.1	31.0	-2.3 ± 1.51	NS	-1.4 ± 0.55	+	0.9 ± 2.32	NS	0.9 ± 1.00	NS
	4h	31.4	32.3	-0.02 ± 0.93	NS	0.5 ± 1.00	NS	0.5 ± 1.62	NS	-0.4 ± 0.57	NS
	4h - 0h			2.31 ± 1.21	NS	1.9 ± 0.91	NS	-0.4 ± 2.42	NS		
Plasma Urea	0h	10.3	8.2	1.1 ± 1.74	NS	-0.3 ± 0.57	NS	-1.4 ± 2.23	NS	0.7 ± 0.33	NS
	4h	8.7	12.9	1.0 ± 1.20	NS	0.8 ± 1.27	NS	-0.2 ± 2.80	NS	-2.3 ± 0.55	*
	4h - 0h			-0.1 ± 1.32	NS	1.1 ± 1.09	NS	1.2 ± 1.66	NS		
Plasma Amino N	0h	42.3	42.8	5.4 ± 4.27	NS	1.6 ± 3.73	NS	-3.9 ± 10.06	NS	-4.3 ± 2.61	NS
	4h	54.5	56.8	19.3 ± 6.68	*	16.4 ± 4.60	*	-2.9 ± 14.14	NS	-5.2 ± 3.14	NS
	4h - 0h			13.87 ± 5.01	+	14.8 ± 2.64	*	0.9 ± 10.44	NS		
Plasma NH_3	0h	0.83	0.71	0.2 ± 0.07	+	-0.007 ± 0.09	NS	-0.2 ± 0.22	NS	-0.08 ± 0.06	NS
	4h	0.71	0.49	0.02 ± 0.03	NS	-0.21 ± 0.08	+	-0.2 ± 0.13	NS	-0.007 ± 0.02	NS
	4h - 0h			-0.2 ± 0.08	+	-0.20 ± 0.10	NS	-0.02 ± 0.20	NS		
Rumen NH_3	0h	9.4	5.1	1.2 ± 1.68	NS	0.7 ± 0.83	NS	-0.5 ± 2.25	NS	3.8 ± 1.17	*
	4h	18.2	19.4	3.3 ± 5.79	NS	0.3 ± 2.29	NS	-2.9 ± 7.68	NS	4.0 ± 3.16	NS
	4h - 0h			2.1 ± 5.91	NS	-0.4 ± 2.06	NS	-2.4 ± 9.06	NS		
Rumen pH	0h	7.3	7.25	0.2 ± 0.10	+	0.20 ± 0.09	NS	-0.05 ± 0.21	NS	-0.04 ± 0.09	NS
	4h	6.8	7.26	-0.1 ± 0.16	NS	0.15 ± 0.10	NS	0.2 ± 0.26	NS	-0.17 ± 0.05	*
	4h - 0h			-0.3 ± 0.11	*	-0.02 ± 0.17	NS	0.3 ± 0.25	NS		