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USE OF OILSEEDS TO INCREASE FIBER TO STARCH RATIOS OF HIGH ENERGY DIETS OR TO INCREASE DIETARY ENERGY DENSITY FOR EARLY LACTATION COWS

BY

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MEMORANDUM

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TO: H. Agar, Graduate Studies, University Centre

FROM: Dr. J. R. Ingalls, Animal Science

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A thesis submitted to the Faculty of Graduate Studies of the University of Manitoba in partial fulfillment of the requirements of the degree of

MASTER OF SCIENCE

ABSTRACT

Two experiments were conducted to determine the effects of dietary supplementation with fat from oilseeds on feed intake, milk production, milk composition, rumen environment and blood plasma cholesterol levels in dairy cows during early lactation.

Experiment 1 involved thirty Holstein cows, two weeks post-partum, in a split-plot design to determine the effect of substitution of fat and fiber for cereal grain starch on milk production, feed intake, milk composition, rumen volatile fatty acids (VFA) and plasma cholesterol. The fat sources were whole sunflower seed (WSS) and tallow. The three diets were (1) control, (2) 9% WSS and (3) 3.4% tallow with sunflower hulls and meal added in amounts to equal fat, fibre and protein supplemented by WSS in the WSS diet. Fat supplement and forage replaced concentrate resulting in 55% concentrate in the WSS and tallow diets and 67% concentrate in the control diet. Cows were fed an isonitrogenous and isocaloric total mixed ration on an ad libitum basis for 12 weeks. Milk yield, feed intake and body weight changes were similar ($P>0.05$) for all diets. Milk protein percent was lower ($P<0.05$) for cows receiving the WSS diet than for those receiving the control diet. Primiparous cows fed the tallow diet had lower ($P<0.05$) milk fat percentage than those receiving the control diet. This response to diet was not observed for multiparous animals. Fatty acid composition in milk from cows fed WSS and tallow supplemented diets were similar and differed ($P<0.05$) from those of cows fed the control diet. Fat supplemented diets produced milk with lower levels of C8:0-C14:0 fatty acids and

higher levels of C18:0 and C18:1 fatty acids than cows fed the control diet. Rumen fluid VFA proportions were affected by diet resulting in lower ($P<0.05$) acetate:propionate ratios in rumen fluid for cows fed the control diet as would be expected with the lower fiber levels. Blood plasma cholesterol levels were also lower ($P<0.05$) for cows fed the control diet than for those fed the WSS or tallow diets.

Experiment 2 involved eight Holstein cows, 36-61 days post-partum, in a double 4x4 latin square design with 28 day periods to determine the effect of extruded canola seed (ECS) as influenced by niacin or calcium chloride addition on milk production, feed intake, milk composition, rumen environment and plasma cholesterol. The four experimental diets were control; 7.6% ECS; 7.6% ECS plus niacin; and 7.5% ECS plus calcium chloride. Cows were fed ad libitum, a total mixed ration of oat silage, concentrate and long hay in a ratio of 31:60:9 respectively. Milk production, feed intake and milk fat, protein and lactose levels were similar ($P>0.05$) for all diets. Molar percentages of C6:0-C16:0 and C20:0 fatty acids were higher ($P<0.05$) and molar percentages of C18:0 and C18:1 fatty acids were lower ($P<0.05$) in milk from cows fed the control diet when compared to milk from cows fed the ECS diet. Molar percentages of ruminal acetic acid were lower ($P<0.05$) for cows fed the ECS diet while the molar percentages of n-butyric acid were higher ($P<0.05$). Acetate:propionate ratios, ammonia and pH levels in rumen fluid were not affected ($P>0.05$) by diet. Blood plasma cholesterol levels were lower ($P<0.05$) for cows fed the control diet.

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DEDICATED TO MY MOM AND DAD

FOR THEIR LOVE AND

ENCOURAGEMENT

TABLE OF CONTENTS

LIST OF TABLES

Table | Page

LIST OF FIGURES

INTRODUCTION

Energy is considered the number one limiting nutrient in diets of early lactation, high producing dairy cows. The modern dairy cow has the genetic capability to produce milk beyond her ability to consume feed (Patton 1987). Therefore, high yielding cows on conventional diets during early lactation cannot supply sufficient dietary energy to meet requirements for milk production and begin to mobilize body reserves which are replenished later in lactation (Bines et al. 1978). Storage and release of energy from body reserves in this way is less efficient than meeting the cow's need directly from the diet (Van Es 1976). Traditionally cereal grains have supplied most of the energy as starch. But increasing the grain portion of the ration to levels above 55-60% results in problems such as fluctuations in dry matter intake, acidosis and low milk fat problems (Linn 1987). Fat, with a caloric density 2.25 times that of starch, is an attractive means of overcoming limitations in energy supplies during early lactation. Benefits of supplementing dairy rations with fat can fall below the theoretical benefits because fatty acids, particularly unsaturated fatty acids can reduce digestibility of fiber in the rumen (Patton 1987). Reduced fiber digestibility may lower milk fat content by reducing the rumen fluid acetic to propionic acid ratio and, also, lower digestible energy for milk yield (Jenkins and Palmquist 1984). Therefore results of fat supplementation on production traits have been variable as a result of varying experimental factors such as fatty acid composition and physical form of the dietary fat, basal diet composition, productivity and stage of lactation of cows and level of

supplementation.

Current research and practice demonstrate that fat may be added to lactation diets to increase energy intake of high producing cows and/or to reduce starch feeding thereby increasing the ratio of forage to concentrate.

The intent of this study was (1) to determine the effects of replacing cereal grain starch with fiber and fat from whole sunflower seeds or tallow in diets for dairy cows in early lactation; (2) to determine the effects of adding niacin or calcium chloride to extruded canola seed diets expected to decrease milk fat and milk protein percentage.

LITERATURE REVIEW

Lipid Metabolism in the Rumen

The rumen in the ruminant animal is the site of initial metabolization of dietary lipids. Microbial activity in the rumen is responsible for the hydrolysis of esterified dietary lipids and for the subsequent hydrogenation of the liberated unsaturated fatty acids (Harfoot 1981). Furthermore the microorganisms of the rumen are capable of de novo lipid synthesis, using the short-chain (C2-C5) fatty acids produced as end-products of microbial carbohydrate and amino acid metabolism as substrates (Harfoot 1981). These processes of hydrolysis, hydrogenation, and de novo synthesis of microbial lipid within the rumen, have an important bearing on the metabolism of lipid further down the digestive tract and ultimately on the lipid composition of ruminant tissues and milk (Christie 1981).

Hydrolysis

The presence of lipolytic activity in rumen contents has been known for many years. Garton and his co-workers (1958) observed lipolysis of fatty acids from linseed oil and olive oil during incubation with sheep rumen contents but not with boiled rumen contents or saliva. Subsequent investigations have shown that lipolytic ability extends to a wide range of esterified substrates.

The source of hydrolytic enzymes and their distribution among the various rumen constituents, together with their possible mode of action

are less well understood. The hydrolytic enzymes involved appear to be extracellular (Harfoot 1981). There also appears to be preferential activity towards diglycerides (versus triglycerides) (Noble et al. 1974). Faraque et al. (1974) provided evidence to support a contention that the lipases of plant origin were primarily responsible for the hydrolysis of esterified fatty acids in the rumen and that lipases of microbial origin were of secondary importance. Using tissue extracts from pasture grasses they showed that lipases of plant origin remained highly active for periods of at least five hours in the presence of metabolically active rumen microorganisms. Autoclaving of plant extracts resulted in a marked reduction in lipolytic activity compared with that in untreated extracts; the remaining low residual level of lipolytic activity was attributed to the lipolytic activity of microorganisms. These conclusions were not supported by work reported by Dawson and Hemington (1974). They contended that microorganisms were mainly responsible for the hydrolysis of esterified fatty acids in the rumen. Their work showed that the degradation of galactolipids in vitro proceeded at a rate sufficient to account for the rate of disappearance of galactolipids in the rumen. The divergence of theories regarding the source of hydrolytic enzymes indicates the need for further investigation of this area of lipid digestion.

Under normal circumstances the hydrolytic processes in the rumen are both rapid and complete (Hawke and Silcock 1969). Dietary lipids which occur mainly as triglycerides, galactolipids and phospholipids are hydrolyzed efficiently releasing glycerol, galactose and unesterified

fatty acids into the rumen. The glycerol released from esterified lipids is fermented to yield volatile fatty acids (Harfoot 1981).

Biohydrogenation

Analysis of the fatty acid composition of the contents from various parts of the ruminants' digestive tract displayed the extensive ability of the rumen contents to hydrogenate unsaturated dietary components (Noble 1981). Dawson et al. (1974) demonstrated that hydrolysis is indeed a prerequisite for hydrogenation. Liberated C18 unsaturated acids which are the major fatty acid constituents of typical dairy rations are hydrogenated to stearic acid or a number of geometrical (trans) and positional isomers of mono-unsaturated acids (Storry 1981).

The mechanism of biohydrogenation is apparently complex and in spite of extensive investigations, the details still remain largely unresolved. Generally biohydrogenation in the rumen is due largely to bacterial action with protozoan action being of secondary importance (Viviani 1970).

Harfoot et al. (1973) incubated strained rumen contents in vitro with linoleic acid and subsequently fractionated large samples of the incubation mixture into fine food particles, protozoa, bacteria, and cell-free supernatant fluid. These workers showed that 80% of the biohydrogenation of the linoleic acid substrate occurred in association with the fine food particles, negligible changes took place in association with the cell-free supernatant fluid, and only small changes occurred in association with the bacteria and protozoa. In this instance, an increase in the amount of stearic acid in the bacterial fraction was noted. Harfoot et al. (1973) attributed these findings to

the absorption of the linoleic acid substrate to fine food particles and the subsequent hydrogenation of this by extracellular enzymes produced by the bacterial component of the rumen contents. Uptake of stearic acid by the rumen bacteria following hydrogenation was postulated as the reason for the increased stearic acid associated with the bacteria (Harfoot et al. 1973).

The significance of biohydrogenation to the limited range of organisms that carry out this process is unknown. Unsaturated fatty acids are toxic to many microorganisms (Nieman 1954) and it could be postulated that biohydrogenation is a detoxification mechanism (Harfoot 1981). On the other hand, much of the unsaturated fatty acids in the rumen are either esterified or adsorbed onto food particles, thus greatly reducing their concentration in the immediate vicinity of the bacterial cell (Harfoot 1981). The fatty acids of bacterial lipids are largely saturated, and it could be postulated that biohydrogenation serves to convert forage fatty acids into a form suitable for incorporation into the bacterial cell (Harfoot 1981).

The dietary lipid changes that occur within the rumen result in unesterified fatty acids becoming the predominant lipid fraction of the digesta. Stearic acid constitutes the major fatty acid present in digesta and the dietary linoleic and linolenic acids are reduced to minor proportions. The predominant component of the C18 monoenoic acids present is the trans-11 isomer. Following these changes, the unesterified fatty acids are largely adsorbed onto the surface of the particulate matter in the rumen (Lennox et al. 1965). However, although the long chain

unesterified fatty acids constitute by far the major dietary lipid class within the rumen digesta, only 20% of the total lipid is found in the protozoal and bacterial populations (Kenney 1970).

Lipid Biosynthesis in the Rumen

The fatty acids of both the bacterial and protozoal lipids are characterized by high percentages of C13, C14, C15, C16 and C17 branched chain fatty acids, together with several straight chain fatty acids containing an odd number of carbon atoms in addition to palmitic (C16), stearic (C18) and C18 monoenoic fatty acids (Harfoot 1981). The biosynthetic precursors are now known to be the volatile branched and straight chain fatty acids produced in the rumen (Church 1969).

Although certain anaerobic bacteria are capable of de novo synthesis of long chain monounsaturated fatty acids, the synthesis of long chain fatty acids containing more than one double bond is restricted to aerobic organisms (Harfoot 1981). The introduction of these double bonds takes place by desaturation and all desaturase enzyme systems described to date have an obligate requirement for oxygen (Harfoot 1981). However it has been known for some time that unlike most bacteria, mixed populations of bacteria from the rumen contain small amounts of polyunsaturated fatty acids (Viviani 1970). Because of the highly anaerobic conditions which are obtained in the rumen it is assumed that the presence of polyunsaturated fatty acids in rumen bacteria is a result of uptake of preformed unsaturated fatty acids from the rumen (Harfoot 1981).

Studies by Emmanuel (1974) using suspensions of mixed rumen protozoa have shown that considerable synthesis of monounsaturated fatty acids takes place by these organisms. Of the di- and triunsaturated acids present in protozoal lipid 99% and 95% respectively consisted of fatty acids with the double bond nearest the carboxyl group as is the case for polyunsaturated fatty acids in plant tissue. Therefore Emmanuel (1974) suggests that the 18:2 and 18:3 fatty acids of the rumen protozoa were of dietary origin. Emmanuel (1974) resolved that short chain fatty acids served as primer molecules for fatty acid synthesis and that malonate was used as the extender of the carbon chain length.

Bacteria and protozoa are capable of adsorption, uptake and incorporation into their own cells of long chain fatty acids of plant origin (Harfoot 1981). Broad and Dawson (1975) demonstrated a preferential order of uptake by a protozoan suspension of 1-^{14}C labelled long chain fatty acids. The relative uptake of these fatty acids into the protozoal cells were linoleate>oleate>palmitate>stearate. Hawke (1971) incubated mixed rumen bacteria with (1-^{14}C)-linolenic acid. Results indicate extensive biohydrogenation of the linolenic acid substrate and incorporation of this fatty acid into cellular lipids of rumen bacteria.

The adsorption of long chain fatty acids and their corresponding triglycerides onto rumen bacterial cells in the presence and absence of food particles from the rumen has been investigated by Harfoot et al. (1974). Adsorption of fatty acids to the bacteria was greatest with

saturated fatty acids and decreased with increasing unsaturation. The same fatty acids in triglyceride form became adsorbed to a much lower extent and the binding was less tight. Harfoot et al. (1974) observed that the presence of food particles greatly reduced the extent to which fatty acids became adsorbed to the bacteria but had little effect on the extent to which triglycerides were adsorbed to the bacteria. The significance of these adsorption differences between lipids has not been determined.

Lipid Composition of Ruminant Feedstuffs

Conventionally dairy diets contain less than 3.5% ether extract. Up to 50% of forage and 20% of grain ether extractable materials may be non-fatty acid in nature (Palmquist and Jenkins 1980).

Lipids in forage crops comprise 6-8% of the dry weight of leaf tissue and are characterized by their high content of glycolipid (70-80%) and phospholipid (20-30%) (Harfoot 1981). The fatty acid composition of leaf lipids is dominated by the presence of a high proportion of unsaturated fatty acids, especially linoleic acid (C18:2) and linolenic acid (C18:3) along with smaller amounts of oleic acid (C18:1) (Harfoot 1981).

The portion of the dairy diet commonly referred to as concentrates, consists largely of crushed cereal grains and oil seed meals. Adding concentrates to the dairy diet increases the dietary intake of unesterified fatty acids and triglycerides (Harfoot 1981). Linoleic acid (C18:2) predominates in most seed lipids (Palmquist and Jenkins 1980).

The compositions of concentrates vary markedly, but the percentage composition of the total fatty acids present in commercial concentrates is usually within the ranges: C16:0, 15-20%; C18:0, 1-5%; C18:1, 25-35% and C18:2, 30-60% (Harfoot 1981).

Lipid Composition of Rumen Microbes

Viviani et al. (1968) analyzed the lipid composition of mixed rumen bacteria and found that the lipid consisted of 30% phospholipid and 70% non-phospholipid on a weight basis. Also, more than 40% of the non-phospholipid was accounted for by unesterified fatty acids while negligible amounts of glycerol esters were present. The lipid composition of rumen bacteria is characterized by the high proportion of saturated acids present, including branched fatty acids of which C15:0 branched fatty acids were the most abundant. The fatty acid composition of mixed rumen bacteria isolated from sheep (Viviani et al. 1968) and cattle (Tweedie et al. 1966, Williams and Dinusson 1973) as indicated by Harfoot (1981) are shown in Table 1.

The protozoa of the rumen contain a very much wider range of phosphorus containing lipids and a higher proportion of unsaturated fatty acids than do the rumen bacteria (Harfoot 1981). Harfoot (1981) showed that the percentage composition of lipid classes present in mixed rumen protozoa from sheep was as follows: unesterified fatty acids, 10.1; monoglycerides 1.4; diglycerides, 1.0; triglycerides, 1.0; phospholipids, 85.5; sterol esters, waxes 0.7 and unidentified components, 0.3. The total fatty acid composition of mixed protozoa from the rumen

Table 1. Fatty acid composition (percentage by weight) of lipids of mixed rumen bacteria

Fatty acid	Percentage composition of Total lipid[a,1]	Total lipid[a,2]	Total lipid[b,3]	Unesterified fatty acid[b,3]
11:0	0.1	n.d.	n.d.	n.d.
12:0	0.4	4.6	1.2	2.3
12:0 br	0.7	n.d.	n.d.	n.d.
13:0	0.3	n.d.	0.8	0.8
13:0 br	0.7	n.d.	0.6	0.5
14:0	2.3	3.7	3.9	4.0
14:0 br	2.4	n.d.	1.2	1.1
15:0	4.4	n.d.	8.0	7.1
15:0 br	10.1	n.d.	12.7	9.8
16:0	35.2	25.4	31.0	29.7
16:0 br	1.0	n.d.	1.2	0.2
16:1	-	tr	4.0	2.2
17:0	1.8	n.d.	1.6	1.2
17:0 br	1.7	n.d.	n.d.	n.d.
18:0	32.0	20.8	15.0	22.8
18:0 br	n.d.	n.d.	0.1	0.1
18:1	3.9	19.7	6.0	6.4
18:2	3.5	5.6	2.7	2.0
18:3		tr	1.0	1.1
20:0	n.d.	n.d.	0.1	-
Other	-	20.2	8.8	8.8

[a]Bacteria from rumen of cattle.

[b]Bacteria from rumen of sheep.

br - branched

(Harfoot 1981)

[1]Tweedie et al. 1966.

[2]Williams and Dinusson 1973.

[3]Viviani et al. 1968.

of cattle and sheep as reported by Harfoot (1981) is shown in Table 2. Fatty acids C16:0 and C18:0 together accounted for about half by weight of the total fatty acids, while C18:2 and C18:3 made up 11% (Viviani et al. 1968) and 20% (Emmanuel 1974) of the total.

Lipid Digestion, Absorption and Transport

Following extensive metabolization within the rumen, the lipids of the digesta pass through the omasum and abomasum virtually unaltered (Bath and Hill 1967). However the acid environment of the abomasum does give rise to the release of the microbial lipid due to disintegration of the bacteria and protozoa (Noble 1981). Also absorption of long chain fatty acids prior to entrance into the small intestine is possible but quantitatively is of negligible physiological significance (Wood et al. 1963).

Like the rumen contents, the lipid passing into the duodenum under normal dietary circumstances is comprised mainly of unesterified fatty acids (70-80%) together with significant but much smaller amounts of phospholipids (Noble 1981, Palmquist and Jenkins 1980). The unesterified fatty acids are almost wholly associated with the particulate matter such as fragments of the dietary plant material, debris from lysed microbes and desquamated epithelial cells (Noble 1981). Esterified fatty acids are predominantly in phospholipid (Bath and Hill 1967).

In a number of experiments the amount of lipid entering the duodenum markedly exceeded the amount ingested in the diet (Sutton et al. 1970, Bickerstaffe et al. 1972, Outen et al. 1974, 1975). Outon

Table 2. Fatty acid composition (percentage of total fatty acids present) of mixed rumen protozoa

Fatty acid	Source of data: Emmanuel 1974 (from rumen of cow)	Harfoot[a]	Viviani et al. 1968 (from rumen of sheep)
12:0	<0.1	•	0.2
13:0	<0.1	•	0.5
13:0 br	<0.1	•	0.2
14:0	1.2	1.4	1.5
14:0 br	<0.1	1.5	0.5
15:0	2.5	3.4	4.3
15:0 br	3.0	1.5	4.3
16:0	41.0	43.1	37.8
16:0 br	1.5	-	2.3
16:1	-	1.1	6.8
17:0	1.2	-	1.4
17:0 br	3.2	3.7	-
18:0	10.5	9.3	13.5
18.0 br	<0.1	-	0.2
18:1	15.4	18.4	11.5
18:2	17.0	16.1	6.3
18:3	1.9	0.5	4.7
20:0	<0.1	-	0.1
20:0 br	-	-	-
Others	1.5	-	4.2

[a]Harfoot (unpublished observations): from rumen of sheep after 17 hr fast.

• = not determined.

- = not detected.

(Harfoot 1981)

et al. (1974) found increases in fatty acid content of between 75 and 98% when dried grass diets were fed to sheep. Feeding dried clover diets to sheep led to increases of between 12% and 41%. Possible endogenous sources of this increased amount of lipid in the duodenum include contributions from salivary and gastric secretions, from diffusion of tissue fatty acids across epithelial linings and from desquamated epithelial cells (Noble 1981). However, the major source of increased lipid flow is undoubtedly that synthesized by bacterial and protozoal populations in the rumen and subsequently released in the abomasum (Harfoot 1981). White (1985) reported low level fat supplementation of diets fed to Holstein steers resulted in more fat being synthesized in the rumen.

In the duodenum, the digesta is infiltrated by bile, duodenal and pancreatic secretions. However the pH of the proximal one-half of the ruminant intestine remains relatively acid as the neutralizing capacities of the duodenal secretions are insufficient to deal with the large volume of digesta leaving the abomasum (Noble 1981).

The lipid composition of the digesta changes significantly in the distal duodenum and upper jejunum (Noble 1981). Unesterified fatty acids, which made up the major proportion of total lipid in the rumen, abomasum and proximal duodenum decrease with an accompanying increase in the proportion of esterified fatty acids (Noble 1981). Also increased quantities of C18 unsaturated fatty acids are evident due to the secretion of bile lipids into the duodenum (Leat and Harrison 1969).

Scott and Lough (1971) incubated sheep bile with duodenal contents and showed there was a transfer of unesterified fatty acids from particulate matter into micellar solution. This effect increased with pH increases and was later shown to be due to the combined action of the bile salt and phospholipid components (Smith and Lough 1976). Johnson et al. (1974) investigated the region of the small intestine responsible for the digestion and absorption of lipid in ruminants using sheep fitted with pairs of re-entrant cannulae in different parts of the intestine. Limited hydrolysis of esterified fatty acids occurred in the upper jejunum and unesterified fatty acids accounted for the 20% of total fatty acids absorbed here. A further 60% of the total fatty acids in the digesta was absorbed in the middle to lower jejunum and this included fatty acids derived from hydrolysis of both neutral lipids and phospholipids. Once the digesta reached the ileum, absorption of unesterified fatty acids together with hydrolysis and uptake of ester bound fatty acids was all but complete.

Biliary and pancreatic secretions are necessary for the optimal assimilation of fatty acids by ruminant animals. In sheep deprived of bile, the absorption of fatty acids from the small intestine fell to about one-sixth of that of the normal animal (Heath and Hill 1969). In the absence of pancreatic juice the concentrations of lipids in lymph were reduced considerably but not to the levels obtained during bile deprivation and a longer period of deprivation was necessary for the effect to occur (Heath and Morris 1963).

Ruminant bile is characterized by a uniquely high ratio of taurine to glycine bile acid conjugates (Moore and Christie 1984). At low pH (2.5), taurine conjugated bile acids are soluble and partly ionized whereas glycine conjugated bile acids are insoluble at pH 4.5 (Hofmann and Small 1967). Thus in the acidic digesta in the proximal jejunum the taurine conjugated bile acids remain in a partially ionized condition, therefore the micellar phase can affect solubilization. Under similar conditions, glycine conjugates are preferentially adsorbed onto particulate matter (Noble 1981). Harrison and Leat (1972) reported that in the proximal jejunum of sheep, about 60% of the bile salts were partitioned in the soluble micellar phase and 40% in the particulate phase. The pH rises as the digesta moves down the intestinal tract, resulting in conditions more favorable for solubilization and increased transfer of unesterified fatty acids from the particulate matter to the micellar phase and then to the intestinal mucosal cells.

In the upper jejunum, the fatty acids would be taken up by the mucosal cells from a mixed micellar solution consisting mainly of unesterified fatty acids, taurine conjugated bile acids and phosphatidylcholine (Moore and Christie 1984). In the middle and lower jejunum the fatty acids would be taken up by the mucosal cells from mixed micellar solutions consisting of unesterified fatty acids, taurine conjugated bile acids and decreasing proportions of phosphatidylcholine and increasing proportions of lysophosphatidylcholine (Moore and Christie 1984).

Ruminant digestion of lipid is characterized by highly efficient absorption. Andrews and Lewis (1970) determined absorption coefficient for C16 and C18 fatty acids of 83-92%. White (1985) reported the true digestibility of sunflower oil was 83.4%. Furthermore the efficiency of absorption was not lowered by greatly increasing the dietary intake of fatty acids (Heath and Hill 1969).

Although the fatty acid composition of the lipids of lymph are similar to the jejunal digesta unesterified fatty acid fraction, selective absorption and incorporation into lipids may occur (Noble 1981). Overall, the rate of fatty acid uptake and incorporation into the lymph lipids is in the order oleic (C18:1)>palmitic (C16:0)>stearic (C18:0) acid (Noble 1981). Also the efficiency of the absorption of long chain fatty acids (C14-C18) increases with the introduction of a double bond or with a reduction in chain length (Noble 1981).

When ruminant animals are given diets containing supplements of protected fats or oils,large amounts of triacylglycerols enter the duodenum. In these cases 2-monoacylglycerols have an important role in the micellar solubilization of fatty acids as is observed in the digestion of dietary triacylglycerols in the non-ruminant small intestine (Thomson and Dietschy 1981).

Quantitatively the major lipids that enter the ruminant enterocyte from the intestinal lumen consist of unesterified fatty acids, lysophospholipids and under some circumstances 2-monoacylglycerols (Moore and Christie 1984). The mechanism of absorption of these lipids in ruminants is unknown but is it thought that transport from the inner

surface of the microvillus membrane to the smooth endoplasmic reticulum, the site of triacylglycerol synthesis is mediated by a specific fatty acid-binding protein (Ocknor and Manning 1974, 1976).

The synthesis of triacylglycerols in the ruminant enterocyte can occur by the α-glycerophosphate pathway or by the monoacylglycerol pathway but under normal dietary conditions the latter pathway is limited by lack of appropriate substrate (little 2 monoglyceride is absorbed), (Moore and Christie 1984). Thus most fatty acids are incorporated into triglyceride by the α-glycerophosphate pathway with glucose serving as the glycerol precursor (Palmquist and Jenkins 1980). The synthesis of phospholipids in the mucosal cells of the small intestine in ruminant animals probably occurs mainly by the acylation of 1-lysophospholipids absorbed from the intestinal lumen (Moore and Christie 1984). Cholesterol is actively synthesized in the mucosal cells of the ruminant small intestine (Scott and Cook 1975). The mechanism of cholesterol esterification in the ruminant enterocyte has not been reported.

There is selectivity in the incorporation of different fatty acids into the various lipid classes synthesized in the mucosal cells of the ruminant small intestine. Christie and Hunter (1978) report that in the intestinal lymph of sheep given a normal diet, the triacylglycerols contained only 7% of polyunsaturated fatty acids, compared with 27% in the phosphatidylcholine and 24.5% in the cholesterol esters. The greatest proportion of saturated fatty acids was observed in the triacylglycerols and the greatest proportion of mono-unsaturated fatty acids in cholesterol esters.

The lipid components (triacylglycerols, phospholipids, cholesterol and cholesterol esters) and a number of apoproteins synthesized in the bovine enterocyte are assembled into lipoprotein particles in the enterocytes. The lipoproteins are then passed into the lacteals, and from there into the blood system via the intestinal and thoracic lymph ducts (Sterzing et al. 1971).

The two major lipoproteins that are synthesized in the mammalian enterocyte are chylomicrons and very low density lipoproteins (VLDL). These two classes of lipoproteins are concerned with the transport of triacylglycerols. Chylomicron-like particles from bovine plasma are composed of triacylglycerols, 87%; phospholipids, 4%; cholesterol, 4%; cholesterol esters, 2%; and protein, 3%. Bovine VLDL are composed of triacylglycerols, 74%; phospholipids, 7%; cholesterol, 7%; cholesterol esters, 5%; and protein, 8% (Ferreri and Elbein 1982). Chylomicron and VLDL phospholipids consist mainly of phosphatidylcholine (Christie and Hunter 1978; Ferreri and Elbein 1982).

Under normal conditions, three quarters of the lymph lipid is associated with the very low density lipoproteins (VLDL) and one quarter with chylomicrons (Harrison et al. 1974). On entering the plasma, chylomicrons and VLDL acquire apo-E and apo-C by transfer from plasma HDL synthesized in the liver (Imaizumi et al. 1978). Acquisition of apo-C ensures that the catabolism of chylomicrons and VLDL is diverted away from the liver towards extrahepatic tissues (Moore and Christie 1984). Lipoprotein lipase which is bound to the endothelial surfaces of the capillaries that permeate extrahepatic tissues, hydrolyzes core

triacylglycerols of plasma chylomicrons and VLDL particles to fatty acids and partial acylglycerols (Mendelson et al. 1976). These hydrolysis products are then taken up at the site of hydrolysis and utilized for energy production, the synthesis of adipose tissue triacylglycerols or the synthesis of milk fat (Moore and Christie 1981).

Milk Fat Synthesis

During peak lactation, bovine mammary tissue secretes approximately 50 grams of milk fat per kilogram of tissue per day (Clegg 1981). About one-half of milk fatty acids are synthesized de novo in the gland using acetate and β-hydroxybutyrate as carbon sources (Palmquist and Mattos 1978). Acetate is used to a larger extent as only one-sixth of the fatty acids are derived from β-hydroxybutyrate (Palmquist and Mattos 1978). The remaining 50% arises by esterification within the gland of fatty acids absorbed from plasma lipids (Clegg 1981).

The short chain, (C4-C10) and medium chain, (C12-C16) even numbered fatty acids are synthesized in the mammary gland from acetate and β-hydroxybutyrate (Thomas and Chamberlain 1984). This synthesis accounts entirely for the secretion of the short chain acids in milk, but a proportion of the medium chain acids may be taken up preformed from the blood lipids, mainly from the lipoprotein triglycerides (Storry 1981).

Wakil and Gibson (1960) showed that the principal mechanism of fatty acid synthesis in the mammary gland involved two basic reactions. The first reaction concerns the carboxylation of acetyl CoA to form malonyl CoA and is catalyzed by acetyl-CoA carboxylase. Subsequently

under the action of a complex of enzymes which collectively forms fatty acid synthetase, malonyl-CoA is successively condensed with a primer fatty acyl-CoA molecule which is extended by increments of two carbon units. Thomas and Chamberlain (1984) report that although the initial primer molecule in most tissues is acetyl-CoA, in the mammary gland butyrl-CoA formed from 3-hydroxybutyrate or synthesized from acetate can also act as a primer. This major pathway for fatty acid synthesis in the mammary gland is known as the malonyl pathway (Thomas and Chamberlain 1984). A minor pathway through which butyrate is formed by a reversal of B oxidation has been shown in mammary tissue (Moore and Christie 1981). Also direct incorporation of β-hydroxybutyrate as a C4 molecule which may subsequently be elongated by further additions of acetyl-CoA occurs in the mammary gland (Storry 1981).

The long chain, (C18), fatty acids of milk fat are derived entirely from the blood lipoproteins as are some of the medium chain (C12-C16) fatty acids (Storry 1981). The blood lipoproteins are present as VLDL and as chylomicrons. The VLDL may be of intestinal origin or may originate in the liver (Thomas and Chamberlain 1984). Most dietary fatty acids (C12-C18) are absorbed into the lymphatic system from the small intestine and thus bypass the liver entering the arterial circulation via the thoracic duct (Thomas and Chamberlain 1984). Absorption into the portal blood is restricted to fatty acids containing 10-12 carbon atoms or less and therefore is of little quantitative importance (Thomas and Chamberlain 1984). However the liver does contribute VLDL to the blood and derives its fatty acids from either dietary absorption

or from lipolysis in adipose tissue depots (Clegg 1981). Also fatty acids may arise within the liver itself by conversion from lipogenic precursors or lastly by hydrolysis in the liver of triglyceride scavenged from lipoprotein remnants (Clegg 1981).

Incorporation of plasma triglyceride fatty acids into milk fat involves their complete or partial hydrolysis by lipoprotein lipase (LPL), located in the capillary endothelium of the mammary gland (Clegg 1981).

The fatty acids released by the action of LPL are in part incorporated into milk fat without modification, but the mammary gland has a desaturase system with a high affinity towards stearic acid and there is a substantial conversion of C18:0 to C18:1 (Thomas and Chamberlain 1984). The same desaturase system appears to attack medium chain length fatty acids but with much reduced activity (Thomas and Chamberlain 1984).

Mendelson et al. (1976) suggest that hormones responsible for milk secretion might direct dietary fatty acids to the mammary gland by stimulating LPL activity in mammary tissue while suppressing activity in adipose tissue. This inverse relationship of LPL activity in adipose and mammary tissues is co-ordinated during pregnancy and lactation as shown in Figure 1 (Hamosh et al. 1970).

It is difficult to quantify transfer rates for individual dietary acids to milk. The factors influencing the transfer of dietary fatty acids to milk are outlined in Figure 2 (Storry 1981). The transfer rate depends on the amounts of fatty acid provided in the diet, on the level of milk fat production and on the metabolic equilibrium of adipose

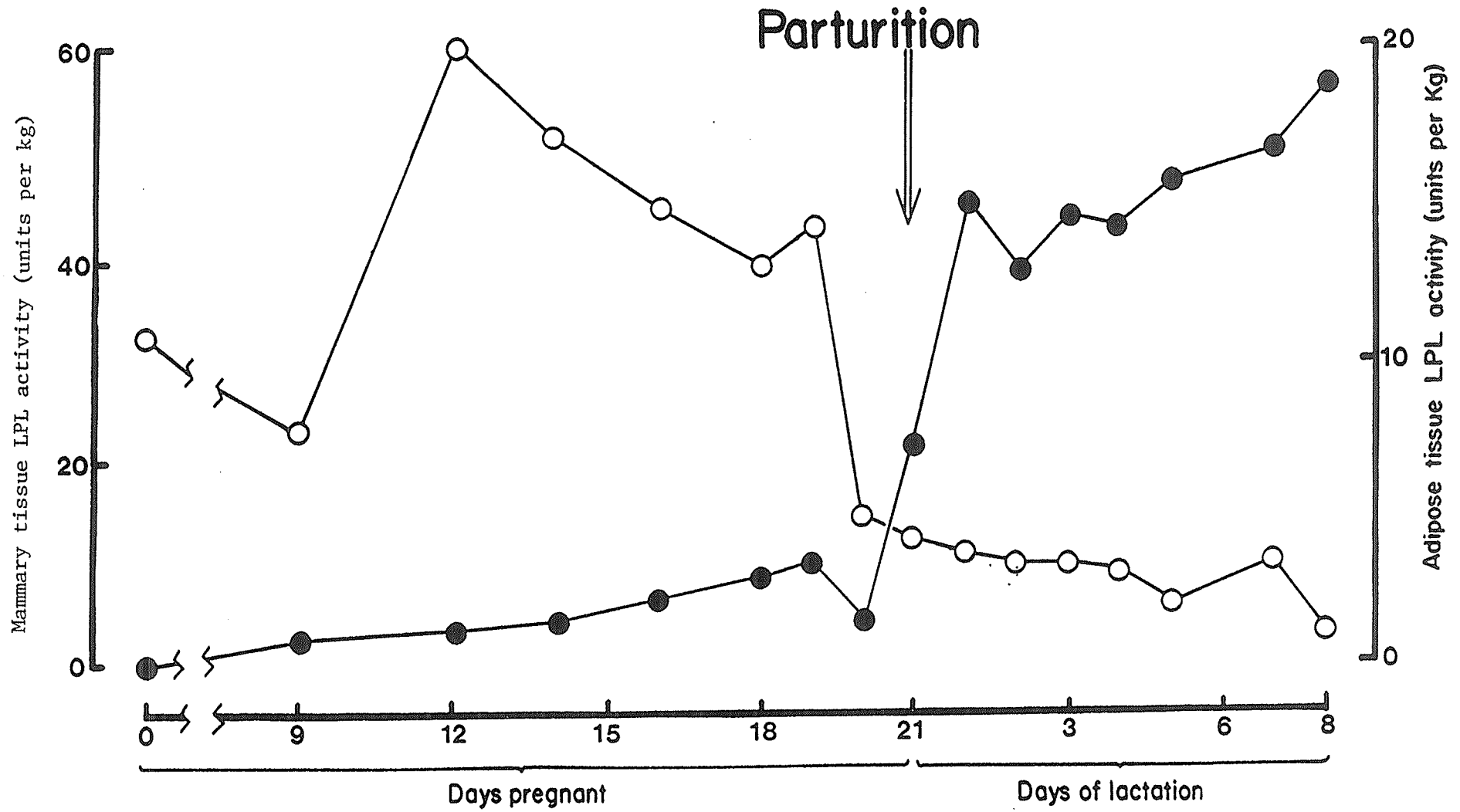

Fig. 1. Reciprocal regulation of lipoprotein lipase (LPL) in adipose and mammary tissues from pregnant and lactating rat.
o - Parmetrial adipose tissue
● - Abdominal/ inguinal mammary glands
1 unit = 1 mmol triacylglycerol hydrolzed per hour
Hamosh et al. (1970) and R.A. Clegg (unpublished results). (Clegg 1981).

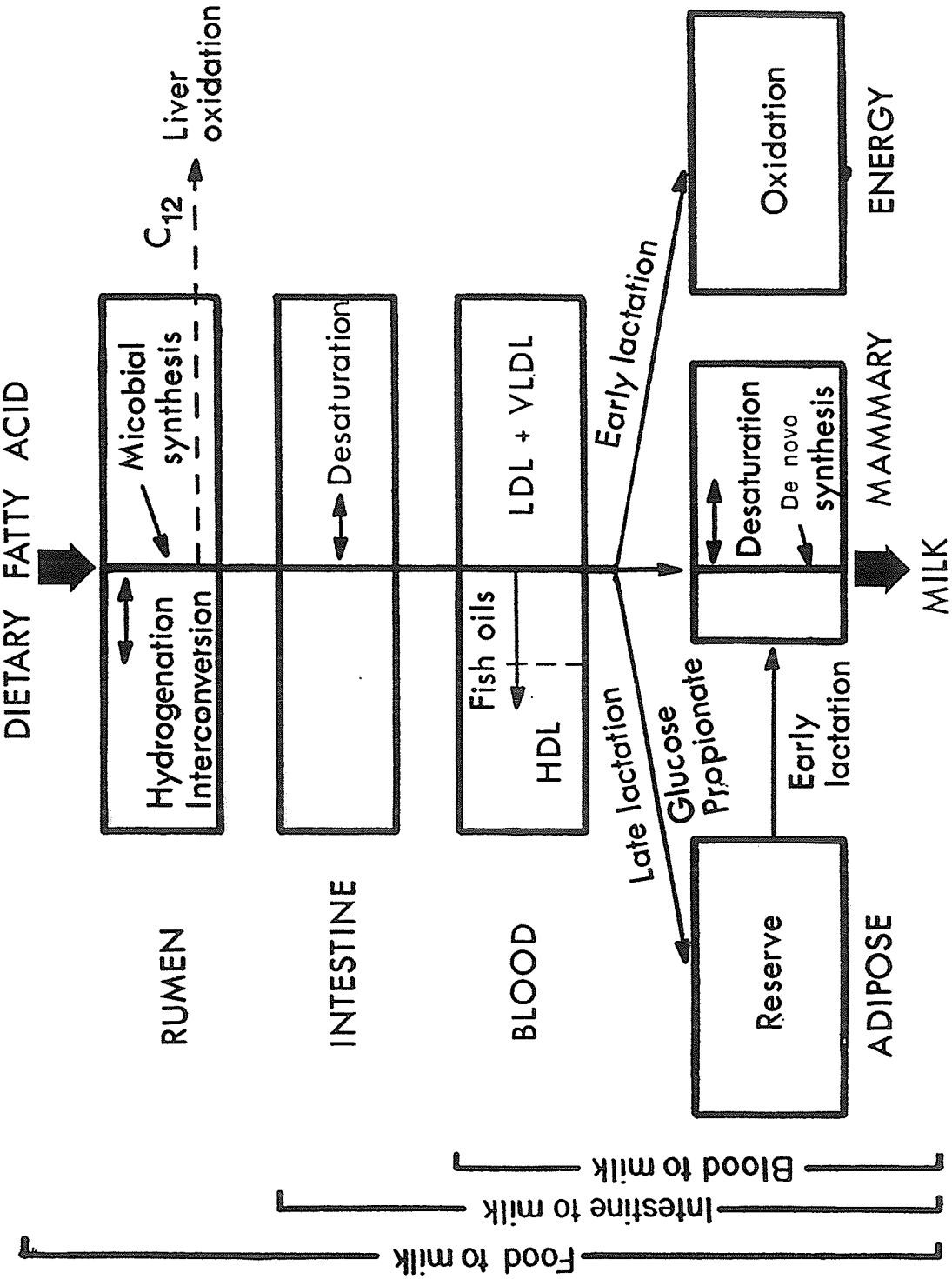

Fig. 2. Factors influencing the transfer of dietary fatty acids to milk. Storry (1981).

tissue as affected by stage of lactation and input of glucogenic nutrients (Storry 1981). Fatty acids of less than 12 carbon atoms show low rates of transfer to milk because of their absorption from the rumen and rapid metabolism by mammary, hepatic and other tissues (Dimenna and Emery 1980). Reported values for net transfers of other fatty acids from diet to milk vary greatly. Measurements of the net transfer of increasing palmitic acid levels from diet to milk range from 29% to 90% compared with values of 19-65% for total C18 fatty acids (Bitman et al. 1973; Banks et al. 1976; Smith et al. 1978). Smith et al. (1978) reported transfers of protected dietary fatty acids to milk from medium and high fat diets of 35% and 25% respectively. Reduced transfer of fatty acids to milk occurs on high grain diets reflecting an increased partition of acids into adipose tissue (Kikeeva 1980). Glascock et al. (1979, 1980) found a decline in transfer of labelled triglycerides to milk from 30% in early lactation to 5% in late lactation. The effect of stage of lactation and proportions of dietary concentrates on the partition of dietary lipids among milk, adipose tissue and oxidation are probably mediated by reciprocal changes in the secretion of growth hormone and insulin (Hart et al. 1978, 1979; Bines et al. 1979) and possibly also of prolactin (Bauman and Currie 1980).

Therefore the fatty acid components of milk fat triglycerides are either synthesized in the tissue or they are derived from the lipids circulating in the plasma. These fatty acids are then esterified into triglycerides mainly by the phosphatidic and diglyceride pathway and finally incorporated into the milk fat globule (Storry 1981). Positional

distributors of fatty acids in the milk triglyceride molecule show definite patterns and probably reflect specificity of the acyltransferases required for the esterification at each position (Moore and Christie 1981).

Fat Supplementation

The use of fats as an energy source in dairy rations has received substantial attention in recent years. The high caloric value of fat (2.25 times that of starch) has heightened this interest in light of the increasing energy requirements to sustain higher milk production on limited feed intakes. Investigators have used saturated fats and oils, unsaturated fats and oils, protected fats and oils, and whole oil seeds as sources of fat for supplementation of rations. Their results have been as variable as the fat sources due to differences in the basal diets supplemented, the fat source used, the frequency of feeding, the level of supplementation and the productivity and stage of lactation of the cows fed the test rations. Milk production, rumen fermentation, milk composition and blood cholesterol levels are influenced by fat supplementation.

Effect of Fat Supplementation on the Rumen

Reports throughout the literature of changes in rumen microbial populations, in ammonia, methane and volatile fatty acid production and of reduced cellulose and fibre digestion in response to dietary lipid supplements clearly show that the basic process of rumen fermentation is affected (Storry 1981). However the mechanism by which fat affects rumen fermentation is not clearly established.

Devendra and Lewis (1974) suggested four theories to explain negative effects of lipid supplementation on fibre digestibility: 1) physical coating of the fibre with fat preventing microbial attack; 2) modified rumen microbial population due to toxic effects of fatty acids on microbes; 3) inhibition of microbial activity from surface active effects of fatty acids on cell membranes; 4) reduced cation availability from formation of insoluble complexes with long chain fatty acids thus causing mineral deficiencies or affecting rumen pH. Orskov et al. (1978) showed that physical coating of dried grass with tallow did not inhibit digestion of fibre in nylon bags suspended in the rumen of sheep fed normal diets but digestion was inhibited in sheep fed tallow. The theory of inhibitory effects of lipids on microbial population and activity is also supported in work done by Maczulak et al. (1981), Henderson (1973), El Hag and Miller (1972). Reports on changes in the rumen associated with high fat diets are conflicting in regard to fibre digestibility, acetate to propionate ratios and feed intake.

Heinrichs et al. (1982) noted a reduction in the quantity and length of initial meals of diets supplemented with fat with a subsequent increase in the number and size of spontaneous meals so that daily consumption was not different. de Visser et al. (1982) also noted that the rate of intake decreased and fat supplemented concentrates were eaten in several small quantities. However overall intake was not depressed in this study.

Steele (1984) noted depressed dry matter intake (DMI) of silage supplemented with unprotected fat however Palmquist and Conrad (1980) did not find a similar decrease with addition of fat to a grain mix. Bines et al. (1978) describes reduced intakes of forage and concentrate as lipid supplementation increased using a protected lipid. MacLeod et al. (1977) found a similar intake depression.

Bernard and Amos (1985) and Smith et al. (1978) reported no significant differences in DMI with whole cottonseed as a lipid source. White (1985) reported that total DMI was not different in animals fed whole sunflower seed diets or free sunflower oil diets. Grumpelt et al. (1984) found significant increases in DMI with higher levels of whole canola seed. Kennelly (1983) found decreases in DMI when rolled or crushed canola seed was used as a fat supplement. Similarly, Sharma et al. (1986) noted depressed DMI with calves fed rations containing pelleted canola seed. Whole sunflower seed incorporation in calf rations did not affect DMI (Sharma et al. 1986). However, Anderson et al. (1984) showed decreased feed intake of diets containing whole sunflower seed.

Since responses in feed intake appear to be variable efforts must be directed ad adjusting for decreases in intake with increased nutrient density and/or in establishing dietary fat levels that do not cause a negative response in feed intake. Feeding systems may also require adjustment if eating behavior is changed by fat addition. Clearly there is much work to be done in this area.

Fibre digestibility was not decreased by supplementation with a

90% tallow and 10% palm kernel oil mixture at 4.9% to 8.4% of total DMI (van der Honing et al. 1983). Tamminga et al. (1983) reporting from the same experiment noted the number of protozoa were severely reduced when fat was fed at 8.4% of DMI. At this high level of supplementation a decrease in total ruminal volatile fatty acid (VFA) was noted and a shift in VFA proportions to more propionate and less butyrate occurred. Palmquist and Conrad (1978) found a lower acetate to propionate ratio when whole raw soybeans were fed but not when relatively saturated hydrolyzed fat was fed. Fibre digestibility was not affected by fat supplementation in this experiment. Sharma et al. (1978) reported slight decreases in the proportion of propionic acid and slight increases in the proportion of acetic acid when cows were fed protected tallow in place of barley and soybean meal. Bines et al. (1978) reported reduced fibre digestibility when a protected lipid was fed. Also concentration of total VFA decreased progressi-ely as the intake of fatty acid increased and ratios of acetate to propionate were reduced on the lipid supplemented diets. Astrup et al. (1976) fed protected vegetable oils and saw no effect on molar percentages of acetic, propionic and butyric acids in rumen fluid.

Ruegsegger and Schultz (1985) showed a significantly lower acetate to propionate ratio of rumen acids when feeding whole soybeans. Volatile fatty acid levels were lower in cows fed canola seed however proportions of VFA were not influenced (Kennelly 1983). Smith et al. (1981) reported that whole cottonseed had no effect on digestibility or availability of dietary fibre. Although there was a tendency for cows receiving whole

cottonseed to have a higher acetate:propionate ratio, the effect was not significant (Moody 1978, Anderson et al. 1979). Murphy et al. (1984) noted decreased rumen digestibility of cellulose with addition of full fat crushed rapeseed to rations. Concentration of total VFA in the rumen decreased with increasing amounts of rapeseed and propionate proportions in the rumen increased slightly with fat supplementation in this study. Sharma et al. (1986) reported marked reductions in apparent digestibility of acid detergent fibre (ADF) in calves fed extruded canola seed and increased apparent digestibility of ADF in calves fed whole sunflower seeds. Rafalowski and Park (1982) noted increased acetate concentration in the rumen with inclusion of sunflower seeds at 20% to 30% of the diet. However ruminal acetate to propionate ratios were unaffected by treatment. McGuffey and Schingoethe (1982) reported no effect on ruminal concentrations of VFA when sunflower seeds were added as a rolled or extruded product to the ration.

Kowalcyzk et al. (1977) observed that successive increments of tallow decreased fibre digestibility and ammonia (NH_3) concentrations in the rumen of sheep and suggested decreased fibre digestibilities may be mediated by low NH_3. Tamminga et al. (1983) also recorded reductions in ruminal NH_3 concentrations after supplementing cow diets with tallow. Contrarily Palmquist and Conrad (1978) have observed increased NH_3 levels in the rumen on high fat diets. Orskov et al. (1978), working with sheep, did not witness lowered NH_3 levels with fat supplementation. Sharma et al. (1978) reported no change in NH_3 levels when protected tallow was fed.

It appears that effects on ruminal metabolism with supplemental fat are rather inconsistent. However it has been established that fatty acids particularly polyunsaturates, are inhibitory to microbial growth (Henderson 1973, Galbraith et al. 1971). Decreased fibre digestibility and subsequent fluctuations in total VFA and VFA proportions have been reduced by addition of metal cations particularly calcium (Palmquist and Jenkins 1980).

Miller et al. (1970) showed that digestibility of malt distillers grains was increased by removing lipids or by the addition of calcium salts. Galbraith et al. (1971) showed bacteriacidal effects of long chain fatty acids and how they could be greatly diminished by the presence of Ca^{2+} and Mg^{2+} ions. El Hag and Miller (1972) observed reversal of depression in fibre digestibility by fatty acids in the artificial rumen by addition of Ca^{2+} ions but not with other cations.

Palmquist and Jenkins (1980) postulated that calcium improves digestibility of high fat rations by forming insoluble soaps which remove the fatty acids from solutions so they are no longer available to bind to the rumen microbiota. Jenkins and Palmquist (1982) in an in vitro study determined that the more water soluble forms of calcium (i.e. calcium chloride) increased insoluble soap formation and digestibility more rapidly and extensively. This study also revealed that fatty acids differ in the extent to which they react with cations to form insoluble soaps. Increasing saturation and chain length both increased the extent of insoluble soap formation and is in agreement with earlier work (Demarne et al. 1970). Jenkins and Palmquist (1982) pointed out that

soap formation in the rumen rquires time and high fat diets with calcium may improve digestibility less if solids turnover is faster.

In a later study, Jenkins and Palmquist (1984) tried to overcome the influences which limit formation of soap in the rumen when fat and calcium were fed separately by adding fat to rations as preformed soaps. However fatty acid digestibility was reduced as was digestibility of energy. These decreases in digestibility were attributed to incomplete dissociation of soap in the abomasum or reformation of calcium soaps in the intestines and subsequent excretion in the feces, due to excess calcium (Jenkins and Palmquist 1984).

Most recently Palmquist et al. (1986) determined the effect of dietary fat and calcium source on insoluble soap formation in the rumen. They reported that addition of fats to diets tended to lower the ratio of acetic to propionic acids (A:P) in the rumen and that dicalcium phosphate and limestone partially corrected the A:P ratio and that soluble calcium chloride completely restored the A:P ratio to that of a control diet with no added fat. However Palmquist (1986) concluded that beneficial effects of adding calcium to high fat diets were not caused by increased calcium soap formation in the rumen.

Chalupa et al. (1984) suggested that hard fats (saturated long chain fatty acids) and calcium salts of long chain fatty acids did not interfere with fermentation by rumen microbes sustained in batch cultures. Later, Chalupa et al. (1986) using an in vivo study, reported only mild responses in the rumen to calcium salts and hard fats confirming the effectiveness of these fat forms in protecting microbes from adverse effects of fat. However Chalupa et al. (1986) note that dietary buffers

should be used with calcium salts to maintain ruminal pH so that dissociation of salt does not occur.

Storry (1981) suggests that effects of dietary lipids on rumen fermentation and fibre digestibility can be reduced by feeding them as natural unextracted seeds or in various protected forms. Palmquist and Jenkins (1980) did not observe reduced fibre digestibility and proposed minimal effects of fat on fibre digestibility in practical diets due to relatively high food intakes, high fibre rations and low retention times.

Fat Supplementation and Milk Production

Clapperton and Steele (1985) observed that dietary supplementation with tallow at 3% of total dry matter intake (DMI) increased milk yield of cows fed this ration whereas soybean oil at the same level had no effect on milk production. Murphy and Morgan (1983) found no change in milk yield when rations were supplemented at 5.2% of DMI with tallow. de Visser et al. (1982) fed a mixture of 11% tallow and 1% palm kernel oil at 7.6% of DMI and reported increases in milk production. Mattias et al. (1982) fed concentrates containing 5% added tallow to first and second lactation Holstein cows in early lactation. Both first- and second- lactation cows fed fat produced more fat-corrected milk, but heifers only, responded with a higher milk-fat percentage, while older cows had an equal milk fat percentage but produced more milk. Palmquist and Conrad (1980) experimented with tallow and blended vegetable fat at both 3.3% and 5% of DMI. Milk yield was decreased with tallow at 5% of DMI and was unaffected by the other treatments. Wrenn et al. (1978) observed increases of 8-10% in milk production with addition of unprotected

tallow at 2.5% of DMI. Goering et al. (1977) fed soybean oil or cottonseed oil to supply 5.8% fat in the ration compared to 3.3% lipid for the unsupplemented control. Yields of milk were not affected by treatment.

Recent work using protected fats and oils resulted in similar variation in response. Vicente et al. (1984) fed 5.6% of the DMI as protected tallow and witnessed significant increases in yields of milk. This finding was in agreement with similar work done by Murphy and Morgan (1983). Smith et al. (1978) fed protected tallow to give fat percentages in the rations of 2.9% (no fat supplement) 8.4% and 14.3%. Treatments did not affect milk yields. A parallel response was reported by Sharma et al. (1978). Wrenn et al. (1978) reported increases in milk yield of between 8% and 10% when protected tallow was fed at 2.5% (actual tallow) of DMI. Goering et al. (1977) fed protected soybean oil and protected cottonseed oil to supply 5.8% fat in the ration (control was 3.3% fat). Yields of milk were not affected by treatment. Astrup et al. (1976) using protected coconut oil, protected hydrogenated soybean oil and protected soybean oil as lipid supplements reported increases in milk yield. Goering et al. (1976) fed a protected safflower oil supplement at 3.4% (actual safflower oil) of DMI and observed no effect on milk production.

The use of whole seeds as lipid sources has increased due to availability, handling ease and the natural protection offered by the seed coat. Bernard and Amos (1985) fed whole cottonseed or pelleted whole cottonseed at 12.5% of DMI. There was no treatment effect on milk yield. Ruegsegger and Schultz (1985) fed heat treated whole soybeans to increase fat percentage in the ration by 2% over the control. There was some

stimulation of milk production with supplementation. Grumpelt et al. (1984) fed whole canola seed at 0, .5, 1.0, 1.5 and 2.0 kg/day and found no effect on milk production. Murphy et al. (1984) reported milk production was not influenced by supplementation with full fat crushed rapeseed at up to 12% of DMI. Handy and Kennelly (1983) fed 3.6% of DMI as whole canola seed and did not influence milk yield. Smith et al. (1981) fed whole cottonseed at 0 to 25% of a complete ration. There was no effect on milk yield. Moody (1978) also found no effect on milk production using cottonseeds. Frank (1981) investigated the effects of whole rapeseed on straw and hay based rations at 2% of the ration dry matter. Significantly higher yields of milk were observed with the straw but not with the hay-based diet.

Finn et al. (1985) fed cows in early lactation rolled sunflower seeds at 10% DMI. There was no improvement in milk yield with lipid supplementation. White et al. (1985) noted significant increases in milk yield with inclusion of whole sunflower seed versus sunflower oil. Anderson et al. (1984) fed 10% of DMI as whole cottonseed, 5% of DMI as extruded soybeans or 12% of DMI as whole sunflower seed. The whole sunflower seed diet decreased milk production. However Rafalowski and Park (1982) noted significant increases in milk yield when cows were fed 10% of DMI as whole sunflower seed. McGuffey and Schingoethe (1982) fed sunflower seeds and noted no improvement in milk yield.

Although results do vary, there generally seems to be either a lack of response in milk yield with dietary lipid supplementation or an improvement. Palmquist (1986) reviewed a summary of many milk production

trials completed by Østergaard et al. (1981) that demonstrated a curvilinear response in milk yield which was greater for high-yielding cows (Figure 3).

Østergaard et al. (1981) also noted a linear increase in milk yield with protected fats but a curvilinear response to unprotected fats (Figure 4).

There appears to be a need for further investigation of the effect of dietary fat supplementation on milk production so that responses are more predictable.

Effect of Fat Supplementation on Milk Composition

The yield of total fat in milk depends upon the combined contributions of fatty acids derived from intramammary synthesis and from lipoprotein triglycerides or unesterified fatty acids of blood plasma (Storry 1981). Increases in the proportions of dietary fatty acids and their respective mono-unsaturated counterparts together with decreased proportions of all other acids have been characteristic of milk fat when fat is added to the ration (Christie 1981). The effects of fat supplements on milk fat secretion reflect the interaction of the positive influences of the fats in increasing the supply of preformed fatty acids to the mammary gland, and the negative influences that accrue through the inhibition of mammary acetyl-CoA carboxylase activity and through the impact of the fats on fermentation in the rumen (Thomas and Chamberlain 1984).

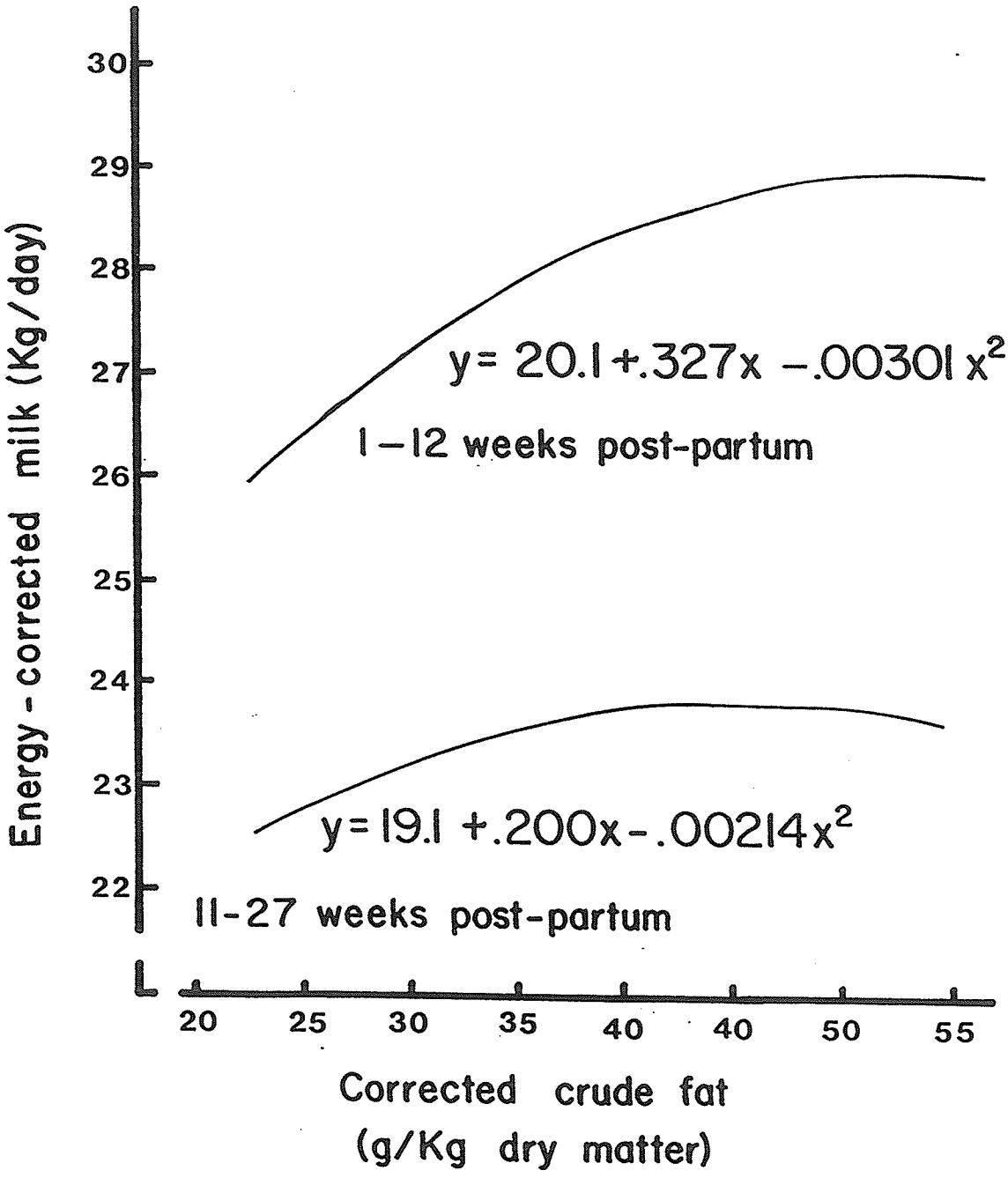

Fig. 3. Quantitative relationships between the content of corrected crude fat in the ration and milk yield. (After Østergaard et al., 1981) Palmquist (1986).

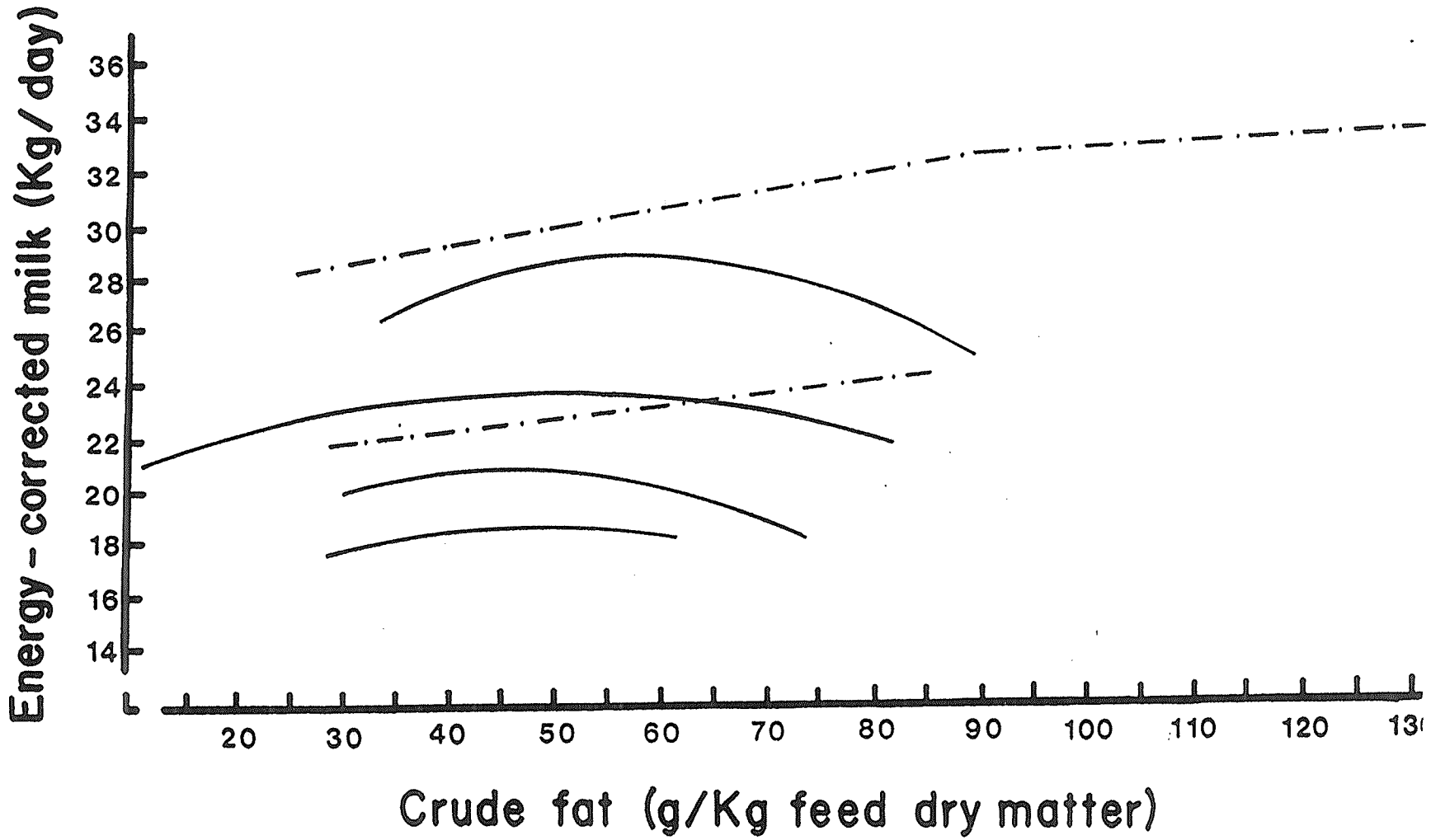

Fig. 4. Effects of increasing amounts of animal fat (protected -.- and unprotected —) on milk yield. Individual curves are composited from different experiments and represent cows of equal production potential. Østergaard et al. (1981).

Current literature suggests that milk fat yield is generally decreased when unprotected, unsaturated fats were fed (Clapperton and Steele 1985, Steele 1984, de Visser et al. 1982, Clapperton 1982, Banks et al. 1980, Goering et al. 1976). Feeding unprotected saturated fat increased milk fat yield (Steele 1984, Clapperton and Steele 1983, Palmquist and Conrad 1980) or had no effect on milk fat yield (Murphy and Morgan 1983, Palmquist and Conrad 1978, Wrenn et al. 1978). Thomas and Chamberlain (1984) reviewed a number of experiments using a variety of fat sources at moderate levels of inclusion and concluded that milk fat yield is increased with saturated fat supplements but is not changed or is reduced with unsaturated fat supplements.

The "vegetable" oils used as fat supplements in animal diets contain high proportions of unsaturated long chain fatty acids. Total unsaturated fatty acid contents as percents of the fat are 87.5% for canola oil, 69.5% for cottonseed oil, 80.8% for soybean oil and 85.2% for sunflower seed oil (Table 3) (National Research Council 1984). The most commonly used comparatively saturated fat source is tallow with an unsaturated fatty acid content of 43.9%. There are also various animal-vegetable fat blends used with varying saturated and unsaturated fatty acid compositions. Hydrogenated vegetable oil refers to a relatively saturated product.

The negative effects on milk fat percentage noted previously when referring to unprotected unsaturated fat supplementation are therefore characteristic of supplementation with most free vegetable oils. Christie (1981) reviewed several feeding trials in which vegetable oil supplements were added to the diets and found there was either no

Table 3. Fatty acid composition of various fat sources

Feed name	Fatty acids, % of the fat									
	C8:0	C10:0	C12:0	C14:0	C16:0	C16:1	C18:0	C18:1	C18:2	C18:3
Animal tallow	-	-	0.9	3.7	24.9	4.2	18.9	36.0	3.1	0.6
Canola oil	-	-	-	-	4.8	0.5	1.6	53.8	22.1	11.1
Cottonseed oil	-	-	-	0.8	22.7	0.8	2.3	17.0	51.5	0.2
Soybean oil	-	-	-	0.1	10.5	0.2	3.8	22.8	51.0	6.8
Sunflower oil	-	-	-	-	5.9	-	4.5	19.5	65.7	-

(National Research Council 1984)

significant effect on milk fat yield or it was depressed relative to that of the non-fat supplemented control animals. Banks et al. (1982) reported that soybean oil fed as free oil decreased milk fat content whereas the oil present in crushed whole soybeans maintained or increased the proportion of milk fat relative to a low-fat control diet. The crushed, unextracted seed protects the rumen from the deleterious effects of the free oil and causes a slower, more controlled release of fat into the rumen (Banks et al. 1982).

Incorporation of protected fats, both unsaturated and saturated into the diet resulted in increases in milk fat yield (Vicente et al. 1984, Murphy and Morgan 1983, Palmquist and Moser 1981, Kronfeld et al. 1980, Smith et al. 1978, Wrenn et al. 1978 and Sharma et al. 1978). Feeding of oilseeds resulted in increases in milk fat (White et al. 1985, Smith et al. 1981, Frank 1981, Banks et al. 1980, Moody 1978 and Christenson et al. 1978), or more commonly no change in milk fat yield (Ruegsegger and Schultz 1985, Finn et al. 1985, Grumpelt et al. 1984, Murphy et al. 1984, Rafalowski and Park 1982, Thomke 1981 and Anderson et al. 1979).

The decreases in milk fat reported with addition of fat are due to lowered outputs of fatty acids (C4-C16) synthesized de novo in the mammary gland (Banks et al. 1982). This depressed intramammary synthesis of fatty acids has been attributed to: 1) an altered production of VFA in the rumen, decreasing supplies of acetate or β-hydroxybutyrate to the mammary glans; and/or 2) to a direct inhibition of mammary acetyl CoA carboxylase activity through increased mammary uptake of long chain

fatty acids from plasma triglyceride (Banks et al. 1982, Moore and Christie 1981, Storry 1981). Evidence for the first mechanism is based largely on observed changes in ratios of the major rumen VFA in response to dietary lipid supplements and the absence of this effect is often interpreted as evidence for the alternative inhibition of mammary lipogenic enzyme activities (Christie 1981).

Fatty acid composition is affected by dietary fat supplementation regardless of whether the effect on milk yield is positive or negative. Positive responses in milk fat occur because of increased uptake of plasma fatty acids with smaller proportional decreases in fatty acids synthesized de novo in the mammary gland (Yang et al. 1978). Decreased de novo synthesis of short chain fatty acids in the mammary gland with increased uptake of long chain fatty acids is a clearly established phenomenon (Palmquist and Jenkins 1980).

Dietary fats can alter the fatty acid composition of milk fat in a number of ways. Fatty acids may be absorbed and transported to the mammary gland where they are esterified unchanged; altered in the rumen by hydrogenation and esterified in the hydrogenated form in the mammary gland; or fatty acids may be modified by desaturation in the mammary gland before esterification (Christie 1981). Also the decrease in de novo synthesis in the mammary gland of C4 to C16 fatty acids mediated by dietary fat supplementation at the ruminal and/or mammary gland significantly affects the proportions of the different fatty acids found in milk fat.

Christie (1981), in reviewing several experiments in which high

levels of tallow were fed to lactating cows, reports that yields and relative percentages of stearic (C18:0) and oleic (C18:1) fatty acids were increased and those of medium chain (C14:0-C16:0) fatty acids tended to decrease.

Clapperton and Steele (1985) reported the proportion of short chain and of 16-carbon fatty acids, (palmitic, palmitoleic), in milk were reduced and that of 18-carbon fatty acids, (stearic, oleic, linoleic, linolenic), increased with addition of soybean oil or tallow to the diet. Steele (1984) noted a decreased yield of C4 to C14 fatty acids in milk fat with both tallow and groundnut oil additions and decreased palmitic (C16:0) acid with the groundnut oil. Banks et al. (1980) fed soya oil and reported decreased proportions of short and intermediate chain fatty acids and increased proportions of C18 acids. Palmquist and Conrad (1978) report similar findings with additions of ground raw soybeans or hydrolyzed fat to the diet.

Feeding protected fat resulted in a 40% to 50% depression of fatty acid synthesis in the mammary gland and increased transfers of dietary fatty acids to milk fat (Smith et al. 1978). Wrenn et al. (1978) found lowered concentrations of lauric (C12:0) and myristic (C14:0) acid and 40% increases in oleic acid (C18:1) concentration by feeding protected tallow. Sharma et al. (1978) reported increased concentrations of stearic (C18:0) and oleic (C18:1) acids in milk while other fatty acids, (C6-C16), were decreased with higher dietary concentrations of protected fat. Similarly, Dunkley et al. (1977) found feeding protected tallow caused significant decreases in weight percentage of all fatty acids

except acetic (C4:0), palmitoleic (C16:1), stearic (C18:0) and oleic (C18:1).

The feeding of oilseeds also caused depressions in short and medium chain fatty acids synthesized de novo in the mammary gland and increased proportions of dietary long chain fatty acids. However White et al. (1985) reported increased molar proportions of milk fat short chain fatty acids with diets supplemented with whole sunflower seeds versus diets supplemented with free sunflower oil. Handy and Kennelly (1983) noted significantly higher levels of stearic (C18:0), oleic (C18:1), linoleic (C18:2) and linolenic (C18:3) fatty acids in milk fat and reduced levels of lauric (C12:0), myristic (C14:0) and palmitic (C16:0) acids when feeding ground whole canola seed. The fatty acid profile obtained in feeding cracked soya beans showed decreased shorter chain fatty acids and increased C18:0 fatty acid (Banks et al. 1980). Thomke (1981) reported increases in C18 acids from 43% to 51% when whole rapeseed was fed. Feeding sunflower seeds caused lower proportions of C14 and C16 fatty acids in milk fat and higher levels of C18 fatty acids (Finn et al. 1985, Rafalowski and Park 1982 and McGuffey and Schingoethe 1982).

Milk protein content is often reduced by dietary fat supplementation but milk protein yield is not always adversely affected (Thomas and Chamberlain 1984). Decreases in protein content can most often be seen when protected fats are fed (Murphy and Morgan 1983, Palmquist and Moser 1981, Sharma et al. 1978, Smith et al. 1978 and Dunkley et al. 1977). However some workers have recorded no decrease in milk protein (Kronfeld et al. 1980, Goering et al. 1976, Wrenn et al. 1976), while Astrup et al. (1976) actually noted increases in milk protein percentage when

diets were supplemented with protected fat. Unprotected fats have either negatively affected protein contents (Murphy and Morgan 1983, de Visser et al. 1982) or had no effect (Clapperton and Steele 1983, Palmquist and Conrad 1978, Goering et al. 1977). Additions of oilseeds as the fat supplement have usually not affected milk protein levels (Bernard and Amos 1985, Ruegsegger and Schultz 1985, Murphy et al. 1984, Handy and Kennelly 1983, Rafalowski and Park 1982), however in some cases milk protein content was decreased by oilseed addition (Anderson et al. 1984, Smith et al. 1981). In terms of individual milk proteins, reductions have been observed in both casein and lactalbumen (Smith et al. 1978, Dunkley et al. 1977).

Emery (1978) reviewed means of increasing milk protein and suggested that additions of lipids to rations might limit blood glucose levels due to lack of dietary carbohydrates. Since glucose is the substrate for lactose synthesis and it is also a major source of energy (in addition to acetate), for milk protein synthesis, the decrease in milk protein might be due to restricted availability of glucose (Emery 1978). Smith et al. (1978) suggest the decrease also may be due to a direct effect of the fat supplement on synthesis of these milk components. Dunkley et al. (1977) postulated excessive protected lipid supplement in the ration might decrease carbohydrate available to rumen microbes enough to depress production of microbial protein. Horner et al. (1986) noted depressed microbial protein synthesis when 15% whole cottonseed was fed. Palmquist and Moser (1981) concluded that dietary fat may impair amino acid transport into the mammary gland and milk protein synthesis by

inducing insulin resistance (inability of insulin to stimulate tissue glucose utilization). There appears to be no clear understanding of the mechanism by which fat supplements reduce milk protein content.

The problem of milk protein depression associated with fat supplementation has been removed in a limited number of experiments using niacin. Horner et al. (1985) fed 6 grams of niacin with whole cottonseed and alleviated a milk protein depression noted when whole cottonseed was fed alone. In a later study Horner et al. (1986) added .07% niacin to a diet supplemented with 15% whole cottonseed. Results indicate niacin increased ruminal protozoa numbers and microbial protein synthesis which may affect milk protein levels.

Effect of Fat Supplementation on Blood Cholesterol

Increased blood cholesterol levels have occurred when feeding protected tallow (Smith et al. 1978, Sharma et al. 1978, MacLeod et al. 1977), protected polyunsaturates (Yang et al. 1978, Wrenn et al. 1978, 1975, Goering et al. 1976) and whole oilseeds (Finn et al. 1985, Park and Rafalowski 1983, Rafalowski and Park 1982, Christensen et al. 1978). Bitman et al. (1973) proposed that hypercholesterolemia in cows fed high fat diets could be caused by an obligatory intestinal synthesis of cholesterol required for absorption and transport of elevated amounts of circulating total lipids and fatty acids. Nestel et al. (1978) reported that the fat induced hypercholesterolemia was primarily due to an increased intestinal biosynthesis of cholesterol and that increased reabsorption of endogenous cholesterol, facilitated by the simultaneous absorption of fat, might have contributed to the hypercholesterolemia.

In contrast to response of blood, milk cholesterol level was not elevated by high fat diets (Rafalowski and Park 1982, Sharma et al. 1978, MacLeod et al. 1977, Wrenn et al. 1978). Therefore cholesterol does not transfer directly from blood to milk thereby suggesting the existence of separate metabolic pools for cholesterol of blood and milk (Wrenn et al. 1976).

Although there has been extensive work completed regarding the effects to be expected when various dietary fats are included in the rations of dairy cows, much remains to be done. Overall, inclusion of modest amounts of dietary fat appear to result in favorable responses, however the benefits are not always observed. The effect of chosen fats on the fatty acid composition of milk fat are quite predictable. However changes in milk fat, milk protein and milk yield are far less predictable and require further research.

EXPERIMENT 1

Effect of Whole Sunflower Seed or Tallow on Production of Dairy Cows in Early Lactation

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ABSTRACT

Thirty Holstein cows were used to determine the effect of supplementary fat in the form of whole sunflower seeds (WSS) or tallow on feed intake, milk production, milk composition, rumen environment and blood cholesterol. The thirty cows at two weeks post partum were divided into three groups, each consisting of four heifers and six cows assigned to one of three experimental diets in a split-plot design. The experimental period was 12 weeks in duration. The three diets were control, 9% WSS, and 3.4% tallow diet supplemented with sunflower meal and hulls in amounts proportional to that of the WSS diet. Fat supplement and forage replaced concentrate in the fat diets resulting in 55% concentrate in sunflower seed and tallow diets and 67% in the control diet. Cows were fed ad libitum, an isonitrogenous, isocaloric total mixed ration of corn silage, haylage, and concentrate with hay fed separately at 10% of dry matter intake. Acid detergent fibre levels were 14.0, 18.5 and 18.3% for the control, sunflower seed and tallow diets respectively and neutral detergent fibre ranged from 30.8 to 31.9%. Milk yield and feed intake were similar ($P>0.05$) for all treatments. Milk fat percentage displayed a significant diet by parity interaction ($P<0.05$) with primiparous cows fed the tallow diet having a lower ($P<0.05$) milk fat percentage than primiparous cows fed the sunflower seed diet. Milk protein percent was lower ($P<0.05$) for cows receiving the sunflower seed diet than for those receiving the control diet. In general, the fatty acid composition of milk fat from cows fed the sunflower seed diet was similar to that of cows fed the tallow diet. Milk

fat from cows fed the control diet had higher ($P<0.05$) levels of C8:0, C10:0, C12:0, C14:0, C18:3 and lower ($P<0.05$) levels of C18:0 and C18:1 fatty acids than those fed the sunflower seed or tallow diets. Cows fed the sunflower seed diet had lower ($P<0.05$) levels of C14:1 and C16:0 in milk fat than cows fed the control or tallow diets and higher ($P<0.05$) levels of C20:0 fatty acids in milk fat from multiparous cows. Rumen fluid from cows fed the control diet was higher ($P<0.05$) in propionic acid at both two and six hours post-feeding and lower ($P<0.05$) in iso-butyric and acetic acid at six hours and n-butyric at two hours post-feeding than for cows fed sunflower seed or tallow diets. Cows fed the sunflower seed diet had higher ($P<0.05$) rumen fluid levels of n-butyric acid at six hours post-feeding and lower ($P<0.05$) levels of n-valeric acid at two hours post-feeding than cows fed the control diet. Acetate: propionate ratios in rumen fluid were lower ($P<0.05$) for cows fed the control diet while rumen fluid pH taken two hours post-feeding was lower ($P<0.05$) for cows fed the control diet than for those fed the tallow diet. Blood plasma cholesterol levels were lower for cows fed the control diet than for those fed the fat-supplemented diets.

INTRODUCTION

Milk production in early lactation may be limited by energy intake. Increasing grain amounts in diets can increase energy intakes but grain proportions above 55 to 60% of the ration dry matter can cause a chain reaction of counterproductive events (Linn 1987). The starch of the grain or lack of fibre that the grain allows causes a shift to reduced ruminal acetate:propionate ratios which induces a glucogenic response. The glucogenic response results in adipose tissue 1) competing with the mammary gland for lipogenic substrate, effectively reducing acetate availability to mammary gland; 2) taking up and esterifying increased amounts of long chain fatty acid; and 3) decreasing mobilization of adipose long chain fatty acid for milk synthesis thus causing the low milk fat syndrome (Palmquist and Jenkins 1980). Therefore fat with its increased energy density (2.25 times that of starch) offers the potential to maintain high energy density while allowing for increased fibre content to prevent depression of milk fat.

The use of whole seeds as lipid sources has increased due to availability, handling ease and the natural protection offered by the seed coat. Unsaturated fats are toxic to certain cellulolytic and methanogenic bacteria of the rumen (Henderson 1973) resulting in decreased fibre digestibility. Feeding whole oilseeds as a means of supplying fat in a protected form has increased due to the advantages of partial rumen bypass of oil in the whole seed and/or to slow release of fat in the rumen allowing complete biohydrogenation thereby minimizing deleterious effects on fibre digestibility (Palmquist and Jenkins 1980)

and/or effects at the mammary level.

Manitoba produced 85% of the sunflower seed produced in Canada in the 1987 crop year with 28,300 hectares seeded. Sunflower seeds contain 17 to 20% crude protein and 35 to 40% oil and therefore may be considered an attractive feed supplement.

White (1985) fed whole sunflower seeds to lactating cows and increased milk fat percentage when compared to cows consuming diets containing free sunflower oil. Rafalowski and Park (1982) determined that 10% inclusion of whole sunflower seeds increased milk yield while maintaining milk fat percent in dairy cows during early lactation.

This study was designed to determine the effect of feeding isocaloric diets when replacing cereal grain starch with fibre and fat from whole sunflower seeds or tallow in diets of dairy cows in early lactation.

MATERIALS AND METHODS

Thirty Holstein cows, twelve primiparous and eighteen multiparous, were placed on one of the three experimental diets. Multiparous cows were grouped into three similar production groups based on previous lactation performance, and assigned to an experimental diet. Primiparous cows were to be randomly and evenly divided between the three test groups. However, the actual assignment resulted in one experimental group with three primiparous cows and the other with five primiparous cows. Therefore each experimental group consisted of six multiparous cows and three, four or five primiparous cows. The cows were placed on test for twelve weeks starting fourteen days post-partum.

The three experimental diets were: 9% whole sunflower seed (WSS); no fat supplement (control) and 3.4% tallow with 2.75% sunflower hulls and 2.75% sunflower meal (Table 4). The sunflower hull, sunflower meal and tallow diet was designed to imitate the whole sunflower seed diet with fat source and form being the primary difference. The three diets were fed once daily on an ad libitum basis as total mixed rations with corn silage and grass silage. Alfalfa hay was fed separately. The WSS and tallow diets contained 55.25% concentrate, 19.5% corn silage, 15.0% grass silage and 10.25% long hay on a dry matter basis. The control ration was 66.75% concentrate, 13.5% corn silage, 9.75% grass silage, and 10% long hay on a dry matter basis. All rations were formulated to be isonitrogenous and isocaloric (Table 5). The fatty acid composition of each ration is listed in Table 6.

Table 4. Ingredient composition of the total mixed rations, % dry matter

Ingredient	Total mixed ration		
	Sunflower seed	Control	Tallow
Barley	31.5	50.6	31.5
Distiller's dried grains	4.0	3.9	4.0
Canola meal	3.0	4.2	3.1
Soybean meal	5.2	5.3	5.2
Whole sunflower seed	9.0	-	-
Sunflower hulls	-	-	2.75
Sunflower meal	-	-	2.75
Tallow	-	-	3.4
Dicalcium phosphate	.4	.5	.4
Limestone	.5	.7	.5
Cobalt iodized salt	.4	.3	.4
Urea	.25	.25	.25
Molasses	.7	.7	.7
Vitamin-mineral premix†	.3	.3	.3
Corn silage	19.5	13.5	19.5
Alfalfa silage	15.0	9.75	15.0
Long hay	10.25	10.0	10.25
	100.0	100.0	100.0

†Provided per kilogram of diet: 5250 IU vitamin A, 450 IU vitamin D_3, 6 IU vitamin E, 6.6 mg copper, 24.1 mg zinc, 20.4 mg manganese, 0.5 mg selenium, and 327.6 mg magnesium.

Table 5. Nutrient composition of the total mixed rations, % dry matter

Nutrient	Total mixed ration		
	Sunflower seed	Control	Tallow
Crude protein, %	17.2	16.8	17.0
Ether extract, %	6.7	3.1	6.9
Acid detergent fibre, %	18.5	14.0	18.3
Neutral detergent fibre, %	31.9	31.2	30.8
Calcium, %	.8	.7	.75
Phosphorus, %	.5	.5	.5
Energy (kcal/g)	4.7	4.5	4.8

Table 6. Fatty acid content of the total mixed rations, molar percentages

Fatty acid	Total mixed ration		
	Sunflower seed	Control	Tallow
C8:0	.1	-	.1
C10:0	-	-	-
C12:0	.2	.2	.3
C14:0	.6	1.0	1.8
C14:1	.5	.3	.8
C16:0	29.4	35.7	29.7
C16:1	1.2	1.2	2.1
C18:0	8.5	6.1	10.7
C18:1	37.0	34.6	32.5
C18:2	10.0	13.9	12.2
C18:3	10.6	7.0	9.9
C20	.5	-	-

Feed intake and milk production were recorded daily throughout the experimental period. Milk samples were taken for three consecutive 24 hour periods during the first week on test, (three weeks post-partum), and alternate weeks thereafter (5, 7, 9, 11, 13 weeks post-partum). The milk samples were composited weekly for each cow and a sample taken to the Manitoba Department of Agriculture Dairy Laboratory for milk fat, protein and lactose analyses. A second milk sample was retained (-20°C) for milk fatty acid analysis. Dry matter content of the three concentrates, corn silage and grass silage was tested weekly and samples were composited monthly for chemical analysis. Hay samples were taken monthly, tested for dry matter content and kept for chemical analyses.

Rumen liquor samples were taken via an esophageal tube (Ingalls et al. 1980) during the 11th week and again in the 12th week on test. These samples were taken at two hours post-feeding and at six hours post-feeding on alternate weeks. The pH of the rumen liquor was recorded at the time of sampling using a Model 598S-50 pH meter from Cole-Parmer Instrument Company. Heparinized tail blood samples were taken at the same time as rumen samples were taken. Rumen and blood samples were centrifuged and the supernatants extracted and stored (-20°C) for later analysis.

Feed dry matter was determined by drying samples for three days at 60°C. Feed samples were analyzed for nitrogen, phosphorus, ether extract, and acid detergent fibre according to the Association of Official Analytical Chemist (AOAC 1984). Calcium levels were determined

according to the AOAC (1975). Gross energy was determined using a Parr Adiabatic Oxygen Bomb Calorimeter. Neutral detergent fibre was determined by the method developed by Goering and Van Soest (1970).

Fatty acid composition of the feed was determined by extracting fat using a method adopted from Bligh and Dyer (1959). The molar ratios of the fatty acids were determined by methylating the feed fat samples according to Metcalf et al. (1966) and separating the fatty acids by gas liquid chromatography with a Varian Vista 6000 (Varian Canada Inc., Georgetown, Ontario) using a 2.43 meter by .64 centimeter column packed with GP 3% SP 2310/2% SP 2300 on 100/120 Chromosorb WAW (Supelco Canada Ltd. Oakville, Ontario). Peak areas for each fatty acid were analyzed using a Varian 402 Data System (Varian Canada Inc., Georgetown, Ontario) and expressed as a percentage of total fatty acids detected.

Milk fat, protein and lactose were measured by infrared spectroscopy using a Foss MS300 Infra-red Analyzer (Milk-O-Scan 203B Type 17920). Fat was extracted from milk samples for fatty acid analysis according to Lambert (1964). The molar ratios of milk fatty acids were determined by methylation of the milk samples according to Metcalf et al. (1966) and separation by gas liquid chromatography (GLC) with a Varian Vista 6000 (Varian Canada Inc., Georgetown, Ontario) using a 2.43 meter by 2.43 centimeter column packed with GP 3% SP 2310/2% SP 2300 on 100/120 Chromosorb WAW (Supelco Canada Ltd., Oakville, Ontario). Peak areas for each fatty acid were analyzed using a Varian 402 Data System (Varian Canada Inc., Georgetown, Ontario) and expressed as a percentage of total fatty acids detected. Identification of individual

fatty acids was accomplished by the use of a GLC reference standard (Nu Chek Prep Inc., Elysian, Minnesota) which established retention times for each fatty acid.

Volatile fatty acid levels in rumen fluid were determined by gas liquid chromatography according to Erwin et al. (1961). Blood plasma samples taken 6 hours post-feeding were analyzed for cholesterol by the Veterinary Services Laboratory of the Manitoba Department of Agriculture and blood plasma cholesterol levels in samples taken 2 hours post-feeding were determined using an enzymatic colorimetric method of Sigma Chemical Company (Procedure No. 351), St. Louis, Missouri.

This experiment was a split plot design with repeated measures for milk production, feed intake and milk composition. The analyses of the data for milk production, feed intake and 4% fat-corrected milk included the following factors in the model: calving season as a block; diet; parity of the cow; cow; week; and two-way interactions of diet with parity, week with parity, and diet with season. Data were collected weekly over the twelve week experimental period for milk production and feed intake. There were two calving seasons, one of which included cows calving between November 4, 1984 and January 17, 1985 and the other including cows calving between February 3, 1985 and April 19, 1985.

Milk composition data were collected biweekly and the analyses of these data (milk protein, lactose, fat and fatty acid levels) included the same factors in the model as were present for milk production with the exception of calving season which was excluded as a block.

The data for volatile fatty acid (VFA) levels in the rumen fluid, blood cholesterol, and rumen fluid pH were collected during the final

two weeks of the experimental period for each cow. Sampling times were two hours post-feeding one week and six hours post-feeding on the alternate week and therefore samples were analyzed separately. The data for blood cholesterol, rumen pH and VFA were analyzed as a completely randomized design. The analyses of these data included the following factors in the model: diet, parity of cow, and the two-way interaction of diet with parity. Since there were duplicate samples of rumen fluid used during the analysis of VFA there were two observations per cow for this parameter while there was a single observation per cow for rumen fluid pH and blood cholesterol.

All analyses were performed using the General Linear Model (GLM) Procedure (SAS Institute Inc. 1985). Multiple comparison analysis of treatment least square means was completed using the Bonferroni option under GLM (SAS Institute Inc. 1985).

RESULTS AND DISCUSSION

Dry matter intake (DMI) was not affected ($P>0.05$) by diet (Table 7, Appendix 1-3). This is in agreement with results reported by White (1985) and Rafalowski and Park (1982) using similar levels (9 and 10% respectively) of whole sunflower seed and Heinrichs et al. (1982), and Palmquist and Conrad (1980) using similar levels of tallow (5 and 3.3% respectively). Mertens (1985) noted that neutral detergent fibre (NDF) has been shown to be the chemical characteristic most consistently related to the intake potential of feeds. The NDF levels determined for the three experimental diets (Table 5) are within 1.1 percentage points of each other and thus this factor would support the similar consumption of all diets. Parity did affect ($P<0.05$) DMI, primiparous cows consuming less feed per day than multiparous cows (Table 7, Appendix 1-3).

Milk production was not significantly affected ($P>0.05$) by diet however primiparous cows produced less milk ($P<0.05$) per day than multiparous cows (Table 7, Appendix 4-6). The milk yields achieved during this trial were quite high and according to Østergaard et al. (1981) a response in milk yield might be expected since high yielding cows in early lactation tend to benefit to a greater extent from fat supplementation. Many researchers have reported increases in milk yield in response to dietary supplementation with tallow (Clapperton and Steele 1985; de Visser et al. 1982; Mattias et al. 1982). However others (Murphy and Morgan 1983; Palmquist and Conrad 1980) reported no milk yield response to tallow additions. White (1985) noted significant

Table 7. Effect of supplemental dietary fat for dairy cows in early lactation on least square means for daily dry matter intake, milk production and body weight change

Parameter	Diet					
	Sunflower seed	S.E.	Control	S.E.	Tallow	S.E.
Dry matter intake:						
Primiparous cows (kg) x	18.14	.65	18.43	.52	17.36	.69
Multiparous cows (kg) y	22.64	1.08	21.80	1.08	22.11	1.37
Combined (kg)	20.39	.75	20.12	.65	19.73	.85
Milk production:						
Primiparous cows (kg) x	34.99	1.92	33.83	1.52	31.05	2.04
Multiparous cows (kg) y	43.08	3.13	38.93	3.13	39.89	3.96
Combined (kg)	39.03	2.17	36.38	1.89	35.47	2.47
4% Fat-corrected milk:†						
Primiparous cows (kg) x	21.05a	.83	20.33ab	.66	17.15b	.88
Multiparous cows (kg) y	22.83	1.07	21.59	1.07	22.70	1.35
Combined (kg)	21.94	.77	20.96	.67	19.92	.88
Body weight change: (Day 84-Day 1)						
Primiparous cows (kg)	15.75	9.95	26.42	8.90	23.00	11.49
Multiparous cows (kg)	11.90	12.89	-1.3	14.89	5.33	18.23
Combined (kg)	13.83	9.58	12.56	9.48	14.16	11.95

a,b - Row means with different letters are statistically different ($P<0.05$).

x,y - Designates difference between primiparous cows and multiparous cows for measured parameters.

† - Designates a treatment by parity interaction ($P<0.05$).

S.E.- Standard error.

increases in milk yield with whole sunflower seed versus sunflower oil as did Rafalowski and Park (1982) feeding whole sunflower seed. However, Finn et al. (1985) and McGuffey and Schongoethe (1982) feeding 9.7 and 9.9% whole sunflower seeds respectively noted no improvement in milk yield with sunflower seed added to cow diets as a fat source.

Cow performance expressed as 4% fat-corrected milk (FCM) was affected ($P<0.05$) by parity with dietary treatment affecting ($P<0.05$) primiparous cows but not multiparous cows (Table 7). The primiparous cows on the tallow diet produced less ($P<0.05$) 4% FCM than those on the sunflower seed diet (Table 7). There were fewer primiparous cows fed the tallow diet due to a treatment assignment error and therefore results may be less indicative of a true response. A parity effect with fat supplemented diets has not been well documented although Mattias et al. (1982) reported first lactation cows fed 5% added tallow responded with higher milk fat percentage while second lactation cows produced more milk to provide an overall significant postive effect on fat-corrected milk when compared to a control.

The body weight changes over the trial period were not different ($P>0.05$) for cows fed the three diets (Table 7). The diets were formulated to be isocaloric and without changes in DMI or milk production, body weight changes would not be expected to differ.

Milk fat percentage was low for all diets (Table 8). The minimum

Table 8. Effect of supplemental dietary fat for dairy cows in early lactation on least square means for daily milk fat, milk protein, and milk lactose content

Parameter	Diet					
	Sunflower seed	S.E.	Control	S.E.	Tallow	S.E.
Milk fat:†						
Primiparous cows (%)	3.43a	.19	3.13ab	.17	2.36b	.22
Multiparous cows (%)	2.67	.22	2.94	.22	3.33	.22
Combined (%)	3.05	.16	3.03	.15	2.79	.17
Milk protein (%)	2.87b	.07	3.11a	.06	2.96ab	.08
Milk lactose (%)	5.04	.08	5.17	.07	4.96	.09

a,b - Row means with different letters are statistically different ($P<0.05$).

† - Designates a treatment by parity interaction ($P<0.05$).

S.E. - Standard error.

recommended level of acid detergent fibre in order to maintain milk fat percentage is 21% (National Research Council 1978). The calculated dietary levels (Table 5) are up to seven percentage points lower than this and explain the low milk fat yields. The control diet purposely was formulated to cause a milk fat depression however the 4% increase in ADF allowed by substituting fat and forage for cereal grain starch in the fat supplemented diets did not cause the expected increase in milk fat percent but did result in increased acetate proportions (Table 10). There was a parity effect on milk fat percentage which shows lower milk fat percentages for primiparous cows fed the tallow supplemented diet when compared to primiparous cows fed the whole sunflower seed diet. Again the lesser number of primiparous cows assigned to the tallow diet may be influencing the results since usually supplementing feed with an unprotected saturated fat such as tallow, increases milk fat percentage (Steele 1984; Thomas and Chamberlain 1984) or does not affect it (Murphy and Morgan 1983; Palmquist and Conrad 1978). Other researchers report that feeding of sunflower seeds resulted in increases in milk fat (White et al. 1985) or more commonly no change in milk fat percent (Finn et al. 1985; McGuffey and Schingoethe 1982; Rafalowski and Park 1982).

Milk protein percent was lower ($P<0.05$) for cows fed the sunflower seed diet when compared to cows fed the control diet (Table 8, Appendix 7). Milk protein levels also tended to be lower for the tallow supplemented diet however the difference was not significant (Table 8). Assuming the hull of the sunflower seed is actually protecting the lipid, Smith et al. (1978) postulated that when protected fats are fed sig-

nificant quantities of energy are unavailable for fermentation and microbial growth thereby reducing microbial protein and glucose precursors to the animal. Emery (1978) suggested that additions of lipids to rations might limit blood glucose levels and since glucose along with acetate are the major sources of energy for milk protein synthesis, the decrease in milk protein might be due to restricted availability of glucose. Palmquist and Moser (1981) concluded that dietary fat may impair amino acid transport into the mammary gland and milk protein synthesis by inducing insulin resistance. Since microbial protein or plasma glucose were not measured in this experiment it is difficult to support or deny these theories. Additions of oilseeds as fat supplements have often not affected milk protein levels (Bernard and Amos 1985; White 1985; Rafalowski and Park 1982) however milk protein drops have been reported (Anderson et al. 1984; Smith et al. 1981).

Milk lactose percentage was not affected ($P>0.05$) by diet or parity (Table 8, Appendix 7).

Molar percentages of C8:0 to C14:0 and C18:3 fatty acids in milk were higher ($P<0.05$) for cows fed the control diet than for those fed the tallow or sunflower seed diets (Table 9). Decreased de novo synthesis of short chain fatty acids in the mammary gland along with increased uptake of dietary long chain fatty acids is an established effect of lipid addition to the diet (Palmquist and Jenkins 1980). Molar percentage of C14:1 fatty acid in milk was higher ($P<0.05$) for cows fed the control vs the WSS diet and molar percentage of C16:0 fatty acid in milk was lower ($P<0.05$) for cows fed the sunflower seed diet

(Table 9). Cows fed the sunflower seed diet had the highest ($P<0.05$) molar percentage of C18:0 in the milk fat followed by cows fed the tallow diet followed by cows fed the control diet (Table 9). Molar percentages of milk fat C18:1 fatty acid were lower ($P<0.05$) for cows fed the control diet than for those fed the other diets (Table 9). Other researchers feeding sunflower seeds noted lower proportions of C14:0 and C16 fatty acids in milk fat and higher levels of C18 fatty acids (Finn et al. 1985; Rafalowski and Park 1982; McGuffey and Schingoethe 1982). Christie (1981) reviewed several experiments in which high levels of tallow were fed and reported higher percentages of C18:0 and C18:1 and lower levels of C14 and C16 fatty acids which corresponds with the results of this experiment (Table 9). Molar percentages of milk fat C20:0 fatty acid were different ($P<0.05$) for the two parity groups (Table 9). Multiparous cows fed the sunflower seed diet had higher ($P<0.05$) molar percentages of C20:0 in milk fat than those fed the control or tallow diets (Table 9). The differences in C20:0 fatty acid levels in milk for primiparous cows were not significant ($P>0.05$) (Table 9). This diet effect with the multiparous cows is in agreement with the theory of increased long chain fatty acid proportions of fat supplemented diets but the parity effect is not easily explained.

Molar percentages of acetic acid and isobutyric acid in rumen fluid samples taken six hours post-feeding were lower ($P<0.05$) for cows fed the control diet than for those fed the sunflower seed and tallow diets (Table 10). Molar percentages of propionic acid in

Table 9. Effect of supplemental dietary fat for dairy cows in early lactation on least square means of fatty acids (molar %) in milk fat

Fatty acid	Diet					
	Sunflower seed	S.E.	Control	S.E.	Tallow	S.E.
C4:0	1.26	.07	1.18	.06	1.24	.07
C6:0	1.82	.11	2.04	.10	1.83	.12
C8:0	1.46a	.09	1.85b	.08	1.45a	.10
C10:0	3.15a	.21	4.64b	.19	3.15a	.23
C12:0	3.47a	.22	5.46b	.20	3.58a	.24
C14:0	11.08a	.32	14.02b	.30	11.31a	.35
C14:1	1.16a	.11	1.75b	.10	1.49ab	.12
C16:0	22.55a	.39	27.65b	.36	27.03b	.43
C16:1	1.64	.13	2.03	.12	1.89	.14
C18:0	17.04c	.59	10.69a	.55	13.79b	.65
C18:1	30.41b	1.10	24.52a	1.01	29.05b	1.20
C18:2	3.21	.15	3.05	.14	2.98	.17
C18:3	.27a	.02	.36b	.01	.29a	.02
C20:0†	1.37c	.08	.66a	.07	.98b	.09
C20:0 Primiparous	1.19	.13	.71	.11	1.15	.15
C20:0 Multiparous	1.56b	.10	.60a	.10	.82c	.10

a,b,c - Row means with different letters are statistically different ($P<0.05$).

† - Designates a treatment by parity interaction ($P<0.05$).

S.E. - Standard error.

Table 10. Effect of supplemental dietary fat for dairy cows in early lactation on least square means of volatile fatty acid levels (molar %) in rumen fluid 2 and 6 hours post-feeding

Volatile fatty acids*		Diet					
		Sunflower seed	S.E.	Control	S.E.	Tallow	S.E.
Acetic acid	2 hours	59.83	.96	57.45	.90	58.84	1.05
	6 hours	60.07a	.85	56.50b	.80	60.88a	.93
Propionic acid	2 hours	20.13b	1.13	26.10a	1.06	20.78b	1.23
	6 hours	20.82b	1.13	28.94a	1.06	21.72b	1.24
Isobutyric acid	2 hours	1.15	.09	1.06	.08	1.36	.10
	6 hours	1.09a	.07	.80b	.07	1.18a	.08
n-Butyric acid	2 hours	15.01a	.64	11.20b	.60	14.63a	.70
	6 hours	14.45a	.77	10.02b	.72	12.51ab	.84
Isovaleric acid	2 hours	2.15	.16	1.87	.15	2.45	.18
	6 hours	1.75	.15	1.47	.14	1.78	.16
n-Valeric acid	2 hours	1.73b	.13	2.32a	.12	1.93ab	.14
	6 hours	1.82	.14	2.27	.14	1.93	.16

*Sampling time (2 versus 6 hours post-feeding) was significant except for acetic and n-valeric acid

a,b - Row means with different letters are statistically different ($P<0.05$).

S.E. = standard error.

rumen fluid samples taken at both two and six hours post-feeding were higher ($P<0.05$) for cows fed the control diet than cows fed the fat-supplemented diets (Table 10). Molar percentages of n-butyric acid in rumen fluid taken at both two and six hours post-feeding were lower ($P<0.05$) for cows fed the control diet than cows fed the sunflower seed diet and at two hours post-feeding also lower than cows fed the tallow diet. Molar percentages of n-valeric acid in rumen fluid taken at two hours post-feeding were higher ($P<0.05$) for cows fed the control diet than for cows fed the sunflower seed diet (Table 10). The acetate: propionate ratio (A:P) in rumen fluid was lower ($P<0.05$) six hours after feeding for cows fed the control diet versus the fat supplemented diets and tended to be lower at two hours post-feeding although the difference for the tallow diet was not significant (Table 11). The changes in volatile fatty acid proportions appear to reflect the lower fibre level of the control diet rather than any adverse effect of fat supplementation on fibre digestibility. Kloppenstein and Owen (1981) note that A:P ratios of ruminal fatty acids of 2.6 to 3.2 are normal. The A:P ratio of the fat supplemented diets were within this range (Table 11) however the A:P ratios of the control diet were lower ($P<0.05$). Reports on changes in the rumen associated with high fat diets are conflicting in regard to fibre digestibility and A:P ratios. The improvement in A:P ratio with fat supplementation in this experiment supports the concept of substituting fat for starch in the ration to maintain a high energy intake while increasing fibre content to prevent milk fat depression. The lack of response in milk fat percentage

Table 11. Effect of supplemental dietary fat for dairy cows in early lactation on least square means for the rumen fluid acetate:propionate ratio (A:P) and pH taken 2 and 6 hours post-feeding

Parameter		Diet					
		Sunflower seed	S.E.	Control	S.E.	Tallow	S.E.
A:P	2 hours	3.07a	.17	2.26b	.16	2.88ab	.19
	6 hours	2.94a	.15	2.01b	.14	2.87a	.17
pH	2 hours	6.87ab	.10	6.50a	.10	6.92b	.11
	6 hours	6.71	.09	6.41	.09	6.53	.10

a,b - Row means with different letters are statistically different ($P<0.05$).

S.E. - Standard error.

(Table 8), even though the rumen environment has been positively influenced is confounding. The fibre levels were higher in the sunflower seed and tallow diets, however they are lower than the 21% ADF recommended for maintaining normal fat test (National Research Council 1978).

Lower rumen fluid pH for cows fed the control diet was noted although it was only significantly lower ($P<0.05$) than pH for cows fed the tallow diet at two hours post-feeding (Table 11). The higher concentrate level of the control diet and increased propionic acid in the rumen fluid justify this trend. Rumen fluid pH levels ranged from 6.41 to 6.92.

Blood plasma cholesterol levels were different for the two parity groups at two hours post-feeding with lower ($P<0.05$) blood plasma cholesterol levels observed in multiparous cows fed the control diet (Table 12). Blood plasma cholesterol levels in primiparous cows at two hours post-feeding followed a similar pattern, however the increase in levels for cows fed fat-supplemented diets over cows fed the control diet was statistically significant ($P<0.05$) for only the cows fed the tallow diet. Blood plasma cholesterol levels in samples taken six hours post-feeding were lower ($P<0.05$) for cows fed the control diet than for cows fed the sunflower seed or tallow diets (Table 12). Several researchers, (Finn et al. 1985; Rafalowski and Park 1982; Thomke 1981; Smith et al. 1978) have reported this effect. Bitman et al. (1973) and Nestel et al. (1978) suggest increased intestinal biosynthesis of cholesterol is required for absorption and transport of increased circulating lipids and fatty acids.

Table 12. Effect of supplemental dietary fat for dairy cows in early lactation on least square means for blood plasma cholesterol (Mmol/litre) taken 2 and 6 hours post-feeding

Parameter	Diet					
	Sunflower seed	S.E.	Control	S.E.	Tallow	S.E.
Cholesterol (2 hours)†						
Primiparous	5.04ab	.38	4.51b	.34	6.46a	.44
Multiparous	6.42a	.35	3.76b	.35	7.22a	.39
Combined	5.73b	.27	4.13c	.25	6.84a	.30
Cholesterol (6 hours)						
Combined	6.92a	.38	4.62b	.36	8.28a	.42

a,b - Row means with different letters are statistically different (P 0.05).

† - Designates a treatment by parity interaction (P<0.05).

S.E. - Standard error.

EXPERIMENT 2

Effect of Extruded Canola Seed as Influenced by Calcium Chloride and Niacin on Dairy Cows in Early Lactation

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ABSTRACT

Eight multiparous Holstein cows were used to evaluate the effects of extruded whole canola seed (ECS) as influenced by calcium chloride (CaCl) and niacin on feed intake, the rumen environment, milk production, milk composition and blood cholesterol. The cows were assigned to a double 4x4 latin square design with 28 day periods. The four experimental diets were control, 7.6% ECS, 7.5% ECS plus CaCl and 7.6% ECS plus niacin. The three diets containing the ECS were, on a dry matter basis, 60% concentrate, 31% oat silage and 9% long hay. The control diet consisted of 61% concentrate, 30% oat silage and 9% long hay on a dry matter basis. Cows were fed ad libitum a total mixed ration of oat silage and concentrate with hay fed separately. Acid detergent fibre levels were 20.2, 20.8, 20.7 and 20.9% for the control, ECS, ECS plus CaCl and ECS plus niacin diets respectively. Neutral detergent fibre levels ranged from 30.7 to 32.3%. Feed intake, milk production and milk protein, lactose and fat levels were similar ($P>0.05$) for all diets. Molar percentages of milk fatty acids C6:0 to C16:0 and C20:0 were higher ($P<0.05$) for cows fed the control diet than for cows fed the ECS diet or the ECS plus niacin diet. Molar percentages of C18:0 and C18:1 milk fatty acids were lower ($P<0.05$) for cows fed the control diet than for cows fed the ECS diet. Rumen fluid taken two hours post-feeding contained higher ($P<0.05$) molar percentages of acetic acid and lower ($P<0.05$) molar percentages of n-butyric acid for cows fed the control diet than for cows fed the ECS diet. Molar percentages of acetic acid taken six hours post-feeding were higher ($P<0.05$) for cows

fed the ECS plus CaCl diet than for cows fed the ECS diet. Rumen fluid acetate:propionate ratios, ammonia, and pH levels were similar ($P>0.05$) for all diets. Blood plasma cholesterol levels taken at two and six hours post-feeding were lower for cows fed the control diet than for cows fed the three ECS diets but the difference was significant ($P<0.05$) at two hours post-feeding between the control and ECS diets only and at six hours post-feeding between the control and ECS plus niacin diets only.

INTRODUCTION

Fats and oils are useful dietary ingredients for increasing energy intake during early lactation because of their high energy content. In practice, however, concentrate diets for cows contain only limited amounts of supplementary fats and oils because at high concentrations, they impair digestion and metabolism in the rumen (Bines et al. 1978). This is due to an inhibitory effect of fatty acids, particularly poly-unsaturates on microbial growth (Palmquist and Jenkins 1980).

A decrease in milk fat percentage is generally experienced when unprotected, unsaturated fats are fed (Clapperton and Steele 1985, Clapperton 1982, Banks 1980). This decrease is due to lowered outputs of fatty acids synthesized de novo in the mammary gland attributed to altered production of VFA in the rumen and/or direct inhibition of mammary acetyl CoA carboxylase activity. The addition of calcium particularly CaCl has improved fibre digestibility and acetate: propionate ratios when fat supplemented diets are fed (Palmquist et al. 1986, Jenkins and Palmquist 1982).

Milk protein percentage has also been reduced by feeding high fat diets (de Visser et al. 1982, Smith et al. 1978, Christenson et al. 1978). The mechanism by which fat supplements reduce milk protein content are not clear although theories include restricted availability of glucose due to lack of dietary carbohydrates (Emery 1978), depression of microbial protein synthesis due to decreased carbohydrate availability to rumen microbes (Dunkley et al. 1977), or impaired amino acid transport into the mammary gland and milk protein synthesis by inducing

insulin resistance (Palmquist and Moser 1981). Horner et al. (1985) fed 6 grams of niacin with whole cottonseed and alleviated a milk protein depression noted when whole cottonseed was fed alone.

Canola is Canada's most important oilseed crop with 4.2 million tonnes produced in the 1988 crop year. Approximately 15% of the canola produced in Canada is grown in Manitoba. Canola oil is highly unsaturated with 87.5% of the fatty acids containing one or more double bonds (National Research Council 1984). Feeding extruded whole canola seed to dairy cows allows this unsaturated oil access to the rumen environment where it exerts a detrimental effect upon the microbial population (Grumpelt 1987) and resulting fat test.

This study was undertaken to determine the response of cows in early lactation to extruded canola seed as influenced by calcium chloride or niacin.

MATERIALS AND METHODS

Eight multiparous cows ranging from 36 to 61 days post-partum were used in a double 4x4 latin square design with 28 day periods.

The four experimental diets were control; 7.6% extruded whole canola seed (ECS); 7.5% ECS plus CaCl; and 7.6% ECS plus niacin (Table 13). The whole canola seed was extruded at 104-110°C using a Brady power take off extruder. The rations were fed ad libitum once a day as total mixed rations with oat silage as a roughage. Long hay was fed separately. The three rations containing the ECS were, on a dry matter basis, 60% concentrate, 31% oat silage, and 9% long hay. The control ration consisted of 61% concentrate, 30% oat silage and 9% long hay on a dry matter basis. The nutrient composition of each experimental ration is listed in Table 14 and the fatty acid content is listed in Table 15.

Feed intake and milk production were recorded daily throughout the experimental period. Milk samples were taken for three consecutive 24 hour periods each week of the trial. Milk sample composites were sent to the Manitoba Department of Agriculture Dairy laboratory for analysis of milk fat, protein and lactose while a sample was retained (-20°C) for milk fatty acid analysis. Dry matter content of the four concentrates and oat silage was tested weekly and samples were composited monthly for chemical analysis. Hay samples were taken during each period, tested for dry matter content and kept for chemical analysis.

Dry matter percentages of the feed were determined by drying samples for three days at 60°C. Feed samples were analyzed for nitrogen

Table 13. Ingredient composition of the total mixed rations, % dry matter

Ingredient	Total mixed ration			
	Control	ECS	ECS + CaCl	ECS + niacin
Barley	36.25	27.30	27.00	27.30
Distiller's dried grains	7.30	7.55	7.50	7.55
Canola meal	16.00	15.10	15.00	15.10
Limestone	.95	.95	.90	.95
Dicalcium phosphate	.40	.50	.50	.50
Salt	.35	.35	.35	.35
Extruded canola seed	-	7.60	7.50	7.60
Dairy herd premix†	.25	.25	.25	-
Niacin premix‡	-	-	-	.25
Calcium chloride	-	-	.60	-
Oat silage	29.90	31.40	31.40	31.40
Hay	8.60	9.00	9.00	9.00
	100.00	100.00	100.00	100.00

† Provided per kilogram of diet: 4375 IU vitamin A, 375 IU vitamin D_3, 5 IU vitamin E, 5.5 mg copper, 20.1 mg zinc, 17 mg manganese, .04 mg selenium and 273 mg magnesium.

‡ Nutrient levels are the same as listed for dairy herd premix plus 274.5 mg niacin per kilogram of diet.

Table 14. Nutrient composition of the total mixed rations, % dry matter

	Total mixed ration			
Nutrient	Control	ECS	ECS + CaCl	ECS + niacin
Crude protein, %	19.3	19.8	19.8	19.7
Ether extract, %	3.4	6.8	6.9	7.0
Acid detergent fibre, %	20.2	20.8	20.7	20.9
Neutral detergent fibre, %	32.3	31.8	30.7	32.1
Calcium, %	.9	.9	1.0	.9
Phosphorus, %	.6	.6	.6	.6
Energy (kcal/g)	4.3	4.5	4.4	4.5

Table 15. Fatty acid content of the total mixed rations, molar percentages

Fatty acid	Total mixed ration			
	Control	ECS	ECS + CaCl	ECS + niacin
C12:0	.2	.2	.2	.2
C14:0	1.4	1.3	1.3	1.3
C16:0	21.6	18.1	18.1	17.9
C16:1	.6	.6	.7	.7
C18:0	3.0	3.3	3.2	3.3
C18:1	23.6	36.4	36.0	35.6
C18:2	32.4	20.9	22.1	22.3
C18:3	17.2	19.2	18.0	18.3
C20:0	-	-	.4	.4
	100.0	100.0	100.0	100.0

phosphorus, calcium, ether extract, acid detergent fibre, gross energy and fatty acid content as in previous manuscript (MacInnis Mabon and Ingalls 1989).

Milk fat, protein and lactose were measured by infrared spectroscopy as in previous manuscript (MacInnis, Mabon and Ingalls 1989). Milk fatty acid levels, and volatile fatty acids in rumen fluid were determined using the same methods as in previous manuscript (MacInnis Mabon and Ingalls 1989). Ammonia levels in the rumen fluid were measured by the method of known addition using an Ammonia Electrode - Model 95-10, Orion Research, Cambridge, Massachusetts.

Blood plasma cholesterol levels were determined using an enzymatic colorimetric method of Sigma Chemical Company (Procedure No. 351), St. Louis, Missouri.

This experiment was a 4x4 replicated latin square. The data were collected during the final two weeks of each four week experimental period for the following parameters: milk production, feed intake, milk protein, milk fat, milk lactose, milk fatty acids, volatile fatty acid (VFA) levels in rumen fluid, rumen fluid ammonia levels, rumen fluid pH, and blood plasma cholesterol levels. All data were analyzed as standard latin squares with treatment, period, square and cow effects in the model.

Milk protein and milk lactose data contained hour of sampling (morning or afternoon) as an additional block in the analysis. The data for blood plasma cholesterol levels and rumen fluid ammonia, pH, and VFA levels were collected two hours post-feeding one week and six hours post-feeding on the alternate week. Therefore samples (two hours or six hours) were analyzed separately.

The VFA levels in rumen fluid, cholesterol levels in blood plasma and fatty acid levels in milk fat were analyzed in duplicate.

Milk fat was analyzed as a standard latin square with treatments, periods, square and cow effects in the model. However adjustments had to be made to the data to compensate for a sampling error that resulted in milk samples being analyzed that were either from the morning milking or the afternoon milking and not a proper daily blend. During periods 1 and 2 milk fat data represented morning milk levels while in period 3 afternoon milk was analyzed. The error was realized so that in period 4 both morning and afternoon milk samples were analyzed. In order to determine daily milk fat percentages, the mean difference between overall morning milk fat levels and afternoon milk fat levels was calculated. Morning milk fat data from periods 1 and 2 and afternoon milk fat data from periods 3 and 4 were compared and were different by 2.67 percentage points. The average proportion of daily milk production obtained in the morning milking was calculated to be 62.15% and that obtained in the afternoon, 37.85% of total daily production. These percentages were determined by averaging individual daily cow production data from morning milkings and then afternoon milkings. Daily milk fat data for each cow were calculated using the following equations:

Morning fat % = afternoon fat % - 2.67

Afternoon fat % = morning fat % + 2.67

Daily milk fat % = (morning fat % x 62.15%) + (afternoon fat % x 37.85%)

All analyses were performed using the General Linear Models (GLM) Procedure (SAS Institute Inc. 1985). Multiple comparison analysis of treatment least square means was completed using the Bonferroni option under GLM (SAS Institute Inc. 1985).

RESULTS AND DISCUSSION

Feed dry matter intake (DMI) was not influenced ($P>0.05$) by the addition of extruded canola seed (ECS) although DMI tended to be higher for ECS diets (Table 16, Appendix 8). Grumpelt (1987), using slightly higher levels of ECS (7.9%) reported increased intake of diets supplemented with either ECS or whole canola seed (WCS) when compared to a control. Sharma et al. (1978) observed lower ($P<0.05$) DMI by calves of a diet supplemented with 18% pelleted canola seed and equal ($P>0.05$) DMI of a diet supplemented with 18% extruded canola seed when compared to the control diet. Murphy et al. (1984) noted similar DMI for cows fed 0, 1 or 2 kilograms of crushed rapeseed. Kennelly (1983) reported decreased ($P<0.05$) DMI of diets supplemented with 7.2% ground WCS when compared to a control. The neutral detergent fibre levels determined for the four experimental diets (Table 14) were within 1.6 percentage points of each other and since NDF has been shown to be the chemical characteristic most consistently related to the intake potential of feeds (Mertens 1985), this factor would support similar DMI of all diets.

Milk production was not affected ($P>0.05$) by the addition of extruded canola seed (Table 16, Appendix 8). This is in agreement with results reported by Murphy et al. (1984), Kennelly (1983), Handy and Kennelly (1983) and Thomke (1981) using ground canola seed. Grumpelt (1987) observed increased milk production in response to dietary supplementation with extruded canola seed. Smith et al. (1980) fed extruded soybeans at 7% of the diet and noted higher milk yields when compared to a control or whole cottonseed diets. Østergaard et al.

Table 16. Effect of dietary inclusion of extruded canola seed (ECS), ECS plus calcium chloride (CaCl), or ECS plus niacin on least square means for daily dry matter intake, milk production, milk protein percentage and milk lactose percentage of dairy cows

	Diet				
Parameter	Control	ECS	ECS + CaCl	ECS + niacin	S.E.
Dry matter intake (kg)	21.74	22.18	22.26	22.83	.43
Milk production (kg)	37.89	37.44	37.19	37.47	1.03
Milk protein (%)	2.88	2.96	3.00	2.97	.04
Milk lactose (%)	5.04	4.92	4.92	4.89	.04

S.E. - Standard error.

(1981) demonstrated a curvilinear response in milk yield which was greater for high yielding cows in early lactation. The production levels achieved in this experiment (Table 16) are high and cows were in early lactation so a milk yield response might be expected.

There was no response ($P>0.05$) in milk protein or milk lactose (Table 16, Appendix 9) percentages to added ECS. Grumpelt (1987), Murphy et al. (1984), Kennelly (1983), Thomke (1981) and Frank (1981) reported similar findings using extruded, rolled or ground canola seed. Smith et al. (1980), using 7% extruded soybeans recorded no effect on milk protein percent. Decreases in milk protein content are quite often observed when protected fats are fed (Palmquise 1980), while unprotected fats have either negatively affected protein percentage (Murphy and Morgan 1983, de Visser et al. 1982) or had no effect (Clapperton and Steele 1983, Palmquist and Conrad 1978). Addition of oilseed other than canola as the fat supplement have not affected milk protein (Bernard and Amos 1985, Ruegsegger and Schultz 1985, Rafalowski and Park 1982) or decreased milk protein percentage (Anderson et al. 1984, Smith et al. 1981). The problem of milk protein depression associated with fat supplementation was offset by addition of niacin in an experiment by Horner et al. (1985) using whole cottonseeds. Hutjens (1987) suggests that the possible mode of action of niacin may be one or more of the following 1) to increase coenzyme levels causing improved nutrient use by the rumen microbes and/or; 2) to increase microbial protein synthesis providing more protein for milk production and tissue use; and/or 3) to increase rumen volatile fatty acid (VFA)

production with a more desirable VFA pattern; 4) to normalize the energy status of high producing cows by optimizing weight loss and minimizing ketosis. The diet containing ECS plus niacin was designed to offset a possible milk protein drop caused by the fat supplementation. However there was no decrease in milk protein percent with ECS addition nor was there a positive effect on milk protein caused by the supplementary niacin (Table 16).

Milk fat percentages were low for all diets but not different ($P>0.05$) (Table 17, Appendix 9). These values had to be calculated (as outlined previously) due to a milk sample collection error. The acid detergent fibre levels of the four diets (Table 14) should support normal milk fat levels (National Research Council 1978), however the rumen fluid acetate:propionate ratios (Table 21) were lower than the 2.6-3.2 noted by Kloppenstein and Owen (1981) as supporting a normal fat test. A depression in milk fat percentage was expected in cows fed extruded canola seed since Grumpelt (1987) observed a significant milk fat depression with cows fed 7.9% ECS. The drop in milk fat percent with ECS diets would appear to be due to the more rapid and/or increased release of the oil in the rumen because of the extrusion process. Current literature suggests that milk fat percentage is generally decreased when unprotected, unsaturated fats are fed (Clapperton and Steele 1985, Steele 1984, de Visser et al. 1982, Clapperton 1982, Banks 1980). The decreases in milk fat reported with addition of fat are due to lowered outputs of fatty acids synthesized de novo in the mammary gland (Banks et al. 1982) attributed to an

altered production of VFA in the rumen decreasing supplies of acetate or B-hydroxybuterate to the mammary gland and/or to a direct inhibition of mammary acetyl CoA carboxylase activity through increased mammary uptake of long chain fatty acids from plasma triglyceride (Moore and Christie 1981).

The extrusion process appears to destroy the protective structure of the seed which affords a slower more controlled release of fat into the rumen when oilseeds are fed whole (Grumpelt 1987, White 1985, Smith et al. 1981) or ground (Murphy et al. 1984, Kennelly 1983, Banks et al. 1982). Steele (1985) observed that the concentration of extracellular oil and not the total amount of oil caused a decrease in milk fat percentage. Although the calculated data shows no milk fat response to dietary fat supplementation in this experiment other reseachers have improved fibre digestibility with calcium (El Hag and Miller 1972, Miller et al. 1970) and particularly calcium chloride (Palmquist et al. 1986, Jenkins and Palmquist 1982). Inclusion of calcium chloride to the ECS supplemented diet in this experiment did not affect milk fat percentage (Table 17), nor did it improve the A:P ratio in rumen fluid as reported by Palmquist et al. (1986). The low milk fat percentages observed in cows fed the control diet are unexpected. The particle length of the oat silage as a roughage source may have affected milk fat yields as Pallauf (1974) concluded that a decrease in the particle length of the roughage has more effect on milk fat percentages than a decrease in the crude fibre content of the ration. The possibility of a carryover effect from one experimental period to the next was raised

Table 17. Effect of dietary inclusion of extruded canola seed (ECS), ECS plus calcium chloride (CaCl), or ECS plus niacin on least square means of calculated daily percentages and of calculated morning and afternoon fat percentages of dairy cows

	Diet							
Parameter	Control	S.E.	ECS	S.E.	ECS + CaCl	S.E.	ECS + niacin	S.E.
Milk fat % (calculated)†	2.76	.12	2.80	.12	2.78	.12	2.59	.11
Milk fat % (6:30 a.m.)	1.75	.12	1.79	.12	1.77	.12	1.58	.11
Milk fat % (2:30 p.m.)	4.42	.12	4.45	.12	4.43	.12	4.24	.11

S.E. - Standard error.

† - Daily milk fat % = (morning fat % x 62.15%) + (afternoon fat % x 37.85%).

by Kennelly (1983) to explain depressed milk production and milk fat percentages in changeover experiments using graded levels (3.6-10.8%) of crushed whole canola seed or canola oil. Storry and Sutton (1969) suggested that in regards to ration adaptation one week is required after a change in ration to stabilize ruminal fermentation whereas 3 weeks seem to be necessary for stabilization of milk fat production. Samples for milk fat determination in this experiment were from weeks 3 and 4 of the 4 week experimental periods. Although roughage particle size or experimental adaptation period may be involved in causing the unusually low milk fat percentage in cows fed the control and ECS diets, the unfortunate need to use calculated fat values based on averages may warrant caution when interpreting these results. Diet did not affect ($P>0.05$) 4% fat corrected yields (FCM) in this experiment (Table 18). This is in agreement with Grumpelt (1987), Murphy et al. (1984), Thomke (1981) and Frank (1981) in feeding extruded, whole or crushed canola seed. Kennelly (1983) reported decreased 4% FCM when feeding finely ground canola seed at 3.6, 7.2 and 10.8% DMI.

Fatty acid composition of milk fat was significantly affected ($P<0.05$) by supplementation with extruded canola seed with and without CaCl or niacin (Table 19). Molar percentages of C6:0 to C16:0 fatty acids in milk were higher ($P<0.05$) for cows fed the control diet than cows fed the ECS diet and the ECS plus niacin diets and higher but not significantly ($P>0.05$) than those of cows fed the ECS plus CaCl diet (Table 19). Decreased de novo synthesis of short chain fatty acids in the mammary gland along with increased uptake of dietary long chain

Table 18. Effect of dietary inclusion of extruded canola seed (ECS), ECS plus calcium chloride (CaCl), or ECS plus niacin on least square means for 4% fat-corrected milk (FCM) of dairy cows

	Diet							
Parameter	Control	S.E.	ECS	S.E.	ECS + CaCl	S.E.	ECS + niacin	S.E.
4% FCM† (kg)	21.08	.49	20.98	.50	20.85	.49	20.54	.48

S.E. - Standard error.

† - Based on calculated milk fat % values.

Table 19. Effect of dietary inclusion of extruded canola seed (ECS), ECS plus calcium chloride, or ECS plus niacin on least square means of fatty acids (molar %) in milk fat of dairy cows

	Diet				
Fatty acids	Control	ECS	ECS + CaCl	ECS + niacin	S.E.
C4:0	1.01	.92	1.00	.94	.05
C6:0	1.71a	1.35b	1.54ab	1.42b	.07
C8:0	1.46a	1.03b	1.22ab	1.10b	.07
C10:0	3.50a	2.28b	2.79ab	2.52b	.21
C12:0	4.04a	2.67b	3.28ab	2.99b	.24
C14:0	11.98a	9.04b	10.26ab	9.57b	.50
C14:1	1.60a	1.18b	1.36ab	1.18b	.09
C16:0	24.44a	18.55b	20.38ab	18.76b	1.02
C16:1	.78	.64	.65	.62	.05
C18:0	14.32b	18.61a	16.58ab	18.68a	.71
C18:1	30.94b	39.17a	36.31ab	37.45ab	1.57
C18:2	2.78	2.70	2.76	2.79	.10
C18:3	.28	.28	.27	.28	.01
C20:0	1.08b	1.48a	1.55a	1.42a	.08

a,b - Row means with different letters are statistically different ($P<0.05$).

S.E. - Standard error.

fatty acids is an established effect of dietary fat supplementation (Storry 1981). It is interesting to note that the ECS plus CaCl diet caused less of a decline in the molar percentages of C6:0-C16:0 fatty acids in milk and less of an increase in C18:0 and C18:1 fatty acids than the other ECS diets (Table 19). Depressed intramammary synthesis of fatty acids with fat supplementation has been attributed to an altered production of VFA in the rumen and/or inhibition of mammary acetyl-CoA carboxylase activity (Thomas and Chamberlain 1984). The ECS plus CaCl diet allowed only a non-significant decrease ($P>0.05$) in acetate levels in the rumen fluid taken 2 hours post-feeding when compared to the control whereas the ECS diet caused significantly lower ($P<0.05$) acetate levels than the control (Table 20). The acetate levels of the rumen fluid at 6 hours post-feeding was lower ($P<0.05$) for the ECS diet compared to the ECS plus CaCl diet (Table 20). It appears that calcium chloride reduced the depression in intramammary synthesis of fatty acids by increasing supplies of acetate. However a significant increase in the A:P ratio was not achieved with calcium chloride although the ratio was higher for cows fed the ECS plus CaCl diet than those fed the ECS diet (Table 21).

Molar percentages of C18:0, C18:1 and C20 were higher ($P<0.05$) for cows fed the ECS diet than for cows fed the control diet (Table 19). The fatty acid profiles of the four diets are given in Table 15. The level of C18:1 (oleic) fatty acid is higher in the ECS diets and the levels of C16:0 (palmitic) and C18:2 (linoleic) fatty acids are lower in the ECS diets when compared to the control diet (Table 15). Grumpelt (1987) reported lower ($P<0.05$) molar percentages of C6:0-C16:0

Table 20. Effects of dietary inclusion of extruded canola seed (ECS), ECS plus calcium chloride (CaCl), or ECS plus niacin on least square means of volatile fatty acid levels (molar %) in rumen fluid taken 2 hours and 6 hours post-feeding from dairy cows

Volatile fatty acids*		Diet				
		Control	ECS	ECS + CaCl	ECS + niacin	S.E.
Acetic acid	2 hours	51.47a	47.23b	50.23ab	50.21ab	.79
	6 hours	49.87ab	47.40b	52.72a	49.36ab	1.10
Propionic acid	2 hours	23.85	24.37	23.33	22.99	.69
	6 hours	24.58	23.73	23.21	25.88	1.13
Isobutyric acid	2 hours	1.37	1.30	1.31	1.37	.14
	6 hours	1.28	1.45	1.39	1.15	.08
n-Butyric acid	2 hours	18.46b	21.81a	20.43ab	20.41ab	.79
	6 hours	19.06	21.94	17.65	18.53	.12
Isovaleric acid	2 hours	1.94	2.39	2.10	2.09	.15
	6 hours	1.96	2.05	2.14	1.64	.13
n-Valeric acid	2 hours	2.93	2.91	2.60	2.93	.13
	6 hours	3.25	3.43	2.89	3.45	.21

a,b - Row means with different letters are statistically different ($P<0.05$).

* - Sampling time (2 versus 6 hours post-feeding) was significant except for acetic, propionic, isobutyric and n-butyric acids.

S.E. - Standard error.

Table 21. Effect of dietary inclusion of extruded canola seed (ECS), ECS plus calcium chloride (CaCl), or ECS plus niacin on least square means for rumen fluid ammonia levels, pH and acetate: propionate ratios (A:P) taken at either 2 hours or 6 hours post-feeding from dairy cows

Parameter		Diet				
		Control	ECS	ECS + CaCl	ECS + niacin	S.E.
Ammonia (mg/100 ml)						
	2 hours	15.63	14.23	17.10	17.48	1.21
	6 hours	17.60	17.80	16.66	17.51	2.01
A:P	2 hours	2.21	1.97	2.20	2.23	.08
	6 hours	2.10	2.06	2.30	1.88	.11
pH	2 hours	6.59	6.64	6.44	6.38	.12
	6 hours	6.33	6.49	6.61	6.49	.08

S.E. - Standard error.

fatty acids and higher molar percentages of C18:0-C20.0 fatty acids when a ECS diet was fed compared to a control. Handy and Kennelly (1983) noted increased C18 fatty acids and decreased C12:0, C14:0 and C16:0 fatty acids when ground canola seed was fed. Murphy et al. (1984) observed increases in milk fatty acids with chain lengths longer than C16 and decreases in fatty acids with chain lengths equal to or less than C16 when crushed rapeseed was fed.

Rumen pH and rumen ammonia levels were not influenced ($P>0.05$) by fat supplementation (Table 21). Rumen pH levels ranged from 6.33 to 6.64. Grumpelt (1987) reported no effect on rumen pH or ammonia with 7.9% ECS. Murphy et al. (1984) and Kennelly (1983) noted increases in rumen pH with 11.76% and 7.2% added ground canola seed respectively. Tamminga et al. (1983) and Kowalcyzk et al. (1977) observed decreased ($P<0.05$) levels of NH_3 in the rumen when tallow was added. However Kennelly (1983) and Sharma et al. (1978) using 7.2% crushed canola and 5, 10 and 15% protected tallow respectively, found no change in NH_3 levels.

Blood plasma cholesterol levels in samples taken two hours post-feeding were greater for cows fed the ECS diets but only significantly greater ($P<0.05$) for cows fed the ECS diet when compared to the control (Table 22). Blood plasma cholesterol levels in samples taken six hours post-feeding were also greater for cows fed the ECS diets but only significantly greater ($P<0.05$) for cows fed the ECS plus niacin diet when compared to the control (Table 22). Several researchers (Finn et al. 1985, Park and Rafalowski 1983 and Christensen et al. 1978) have

Table 22. Effect of dietary inclusion of extruded canola seed (ECS), ECS plus calcium chloride (CaCl), or ECS plus niacin on least square means for blood plasma cholesterol levels (Mmol/litre) taken 2 hours and 6 hours post-feeding from dairy cows

		Diet				
Parameter		Control	ECS	ECS + CaCl	ECS + niacin	S.E.
Cholesterol	2 hours	5.29b	6.66a	6.11ab	6.35ab	.32
	6 hours	5.31b	5.99ab	6.13ab	6.44a	.26

S.E. - Standard error.

noted this effect. An increased intestinal biosynthesis of cholesterol required for absorption and transport of increased circulating lipids and fatty acids is the probable cause (Nestel et al. 1978, Bitman et al. 1973).

GENERAL DISCUSSION

Dietary fat supplementation with whole sunflower seeds or tallow in Experiment 1 allowed maintenance of the energy level provided by the control diet while increasing dietary fibre content. Dietary fat supplementation in Experiment 2 increased the energy density of the ECS diets relative to the control diet and utilized CaCl or niacin to offset expected negative effects of fat supplementation. Although both experiments involved the addition of fat to diets of dairy cows in early lactation, the objectives were quite different.

DMI was not affected ($P>0.05$) by fat supplementation in Experiment 1 and 2. A decrease in dry matter intake of diets supplemented with fat to increase energy density (as in Experiment 2) could eliminate the desired effect of increasing energy intake and a drop in DMI of diets supplemented with fat to allow for higher dietary fibre (as in Experiment 1) would reduce overall nutrient intake and probably performance. Addition of tallow, WSS and ECS at the levels tested in Experiments 1 and 2 did not decrease DMI.

Milk production was not influenced ($P>0.05$) by diet in Experiment 1 and 2. A response in milk production was not expected in Experiment 1 since diets were formulated to be isocaloric. But the lack of response to the increased energy density of the ECS diets in Experiment 2 is confounding.

Milk protein percent was decreased ($P<0.05$) when cows were fed whole sunflower seeds in Experiment 1. Milk protein depressions when fat is added may be due to: 1) depressed production of microbial protein

due to decreased carbohydrate availability in the rumen (Dunkley et al. 1977); 2) impairment of amino acid transport into the mammary gland and milk protein synthesis by induced insulin resistance (Palmquist and Moser 1981); 3) or simply a decrease in protein synthesis due to restricted availability of glucose since lipids cannot be converted into lactose in the animal and dietary carbohydrates are reduced in fat supplemented diets. Niacin was added to a ECS diet in Experiment 2 as a result of reported alleviation of a milk protein depression by addition of niacin (Horner et al. 1985). However, a milk protein depression was not produced with ECS as an added fat source in Experiment 2 and the niacin treatment had no effect on milk protein.

Milk fat percentages in Experiments 1 and 2 were low (Table 8, Table 17) and were not greatly influenced by diet. A parity by diet interaction in Experiment 1 was noted with primiparous cows fed the tallow diet producing milk with lower ($P<0.05$) milk fat percent than primiparous cows fed the sunflower seed diet. This is unusual since unprotected saturated fats such as tallow usually increase milk fat percentage or do not affect milk fat yield (Steele 1984; Palmquist and Conrad 1980; Wrenn et al. 1978; Palmquist and Conrad 1978). The limited number of animals assigned to the tallow diet may be influencing the results. An improvement in milk fat percentage of cows fed the fat supplemented diets in Experiment 1 was expected since the forage:starch ratio was considerably increased in these diets when compared to the control diet. The fat supplemented diets in Experiment 1 were higher in ADF than the control diet but all diets were well below the 21% ADF

recommended for normal fat test (Table 5) and all diets contained a similar level of NDF. There was no response in fat test to the increased ADF allowed by the fat supplemented diets but increased ($P<0.05$) acetate proportions in the rumen were noted. The expected improvement in milk fat test for cows fed the higher fibre fat supplemented diets in Experiment 1 were not realized possibly because the ADF values were still too low or the fats supplied from WSS or tallow caused decreased intramammary fatty acid synthesis with proportionally lower increased uptake of plasma fatty acids.

A depression in milk fat percentage was expected in Experiment 2 with cows fed the extruded canola seed as a result of the observations of Grumpelt (1987). But the very low milk fat percentage of cows fed the control diet was unexpected. Particle length of the oat silage, carryover effects or inaccuracies in the calculated fat data are possible reasons for the depressed milk fat. Calcium chloride (CaCl) was added to a ECS diet in Experiment 2 as a result of reports suggesting improved fibre digestibility and acetate:propionate (A:P) ratios (Palmquist et al. 1986; Jenkins and Palmquist 1982). Inclusion of CaCl did not affect A:P ratios or milk fat percentage. But acetate levels in the rumen fluid were higher for cows fed the ECS plus CaCl diet at two and six hours post-feeding than for cows fed the ECS diet however only significantly different at six hours. Also molar percentages of C6:0-C16:0 milk fatty acids declined less and molar percentages of C18:0 and C18:1 increased less with the ECS plus CaCl diet when compared with the ECS diet. Therefore CaCl appears to affect rumen fermentation and warrants further study.

Fatty acid composition of milk fat was affected by fat supplementation in Experiments 1 and 2. Decreased de novo synthesis of short chain fatty acids in the mammary gland along with increased uptake of dietary long chain fatty acids is an established effect of lipid addition to the diet (Palmquist and Jenkins 1980). The results of Experiments 1 and 2 are in agreement with these findings. The intermediate response of milk fatty acids from cows fed the ECS plus CaCl in Experiment 2 are noteworthy and may be a result of the increased proportion of acetate in the rumen.

Molar percentages of VFA were affected by diet in Experiments 1 and 2. Clearly rumen VFA in Experiment 1 were influenced by the lower fibre level of the control diet. The decrease ($P>0.05$) in the acetate:propionate ratios for cows fed the control diet results in an A:P ratio substantially lower than suggested by Kloppenstein and Owens (1981) to allow normal fat test. The increase in the fibre content allowed by the use of WSS or tallow caused decreases ($P>0.05$) in the proportion of propionic acid in rumen fluid and increases ($P<0.05$) in the proportions of acetic, n-butyric and isobutyric acids. Acetate:propionate ratios were increased ($P<0.05$) by fat supplemented diets probably because of the increased fibre content. The effects in Experiment 2 were less dramatic but feeding the ECS diet resulted in lower acetic acid proportions and higher butyric acid proportions when compared to feeding the control although differences were significant at two hours post-feeding only. Acetate to propionate ratio was not affected by diet in Experiment 2 but the low A:P ratios determined for all diets suggests

an upset in rumen environment which may be due to reduced particle length of the primary roughage. The reduction in the A:P ratios is reflected in the low milk fat percentages experienced by cows receiving all four experimental diets. The ECS plus CaCl diet supported higher ($P<0.05$) acetate proportions than the ECS diet and suggests a positive effect of CaCl on the rumen although the response did not cause improved milk fat percentage.

Blood plasma cholesterol levels were predictably increased ($P<0.05$) by fat supplementation the form of WSS, tallow and ECS in Experiments 1 and 2. Increased intestinal biosynthesis of cholesterol is required for absorption and transport of increased lipids and fatty acids (Bitman et al. 1973; Nestel et al. 1978).

SUMMARY

1. Dry matter intake and milk production were not affected by a) replacing cereal grain starch with fibre and fat from whole sunflower seeds or tallow or b) by increasing energy density with extruded canola seed.
2. Increasing dietary fibre level by replacing starch with fat and fibre did not improve milk fat percentage when NDF levels were similar.
3. Acetate:propionate ratios were increased in fat supplemented diets in Experiment 1 as would be expected with the higher fibre levels. Calcium chloride addition to a ECS diet increased acetic acid proportions in rumen fluid.
4. Sunflower seed supplementation decreased milk protein levels.
5. Supplementing with ECS did not decrease milk protein percent and the addition of niacin did not change milk protein percent.
6. Supplementing with ECS did not cause a further reduction in fat test and the addition of CaCl did not improve milk fat test.
7. Generally fat supplementation increased the proportion of longer chain fatty acids and decreased the proportion of shorter chain fatty acids in milk fat.
8. Plasma cholesterol level was increased by fat supplementation.

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A P P E N D I C E S

Appendix 1. Analysis of variance for dry matter intake for multiparous (M) and primiparous (P) cows (Trial 1)

Parameter	Source	Degrees of freedom	Type III mean square	F value	Significance level
Dry matter intake (M)	Treatment (Trt)	2	794.32	0.21	NS
	Season	1	2296.75	0.73	NS
	Cow (Trt*season)	14	3700.59	26.06	.0001
	Week	11	1868.17	13.16	.0001
	Trt*week	22	298.87	2.11	.0047
	Week*season	11	124.02	0.87	NS
	Error	154	141.98		
Dry matter intake (P)	Treatment (Trt)	2	785.77	1.04	NS
	Season	1	240.31	0.32	NS
	Cow (Trt*season)	8	752.84	6.71	.0001
	Week	11	1085.21	9.68	.0001
	Trt*week	22	109.30	0.97	NS
	Week*season	11	36.00	0.32	NS
	Error	88	112.16		

NS - Not significant (P>0.05).

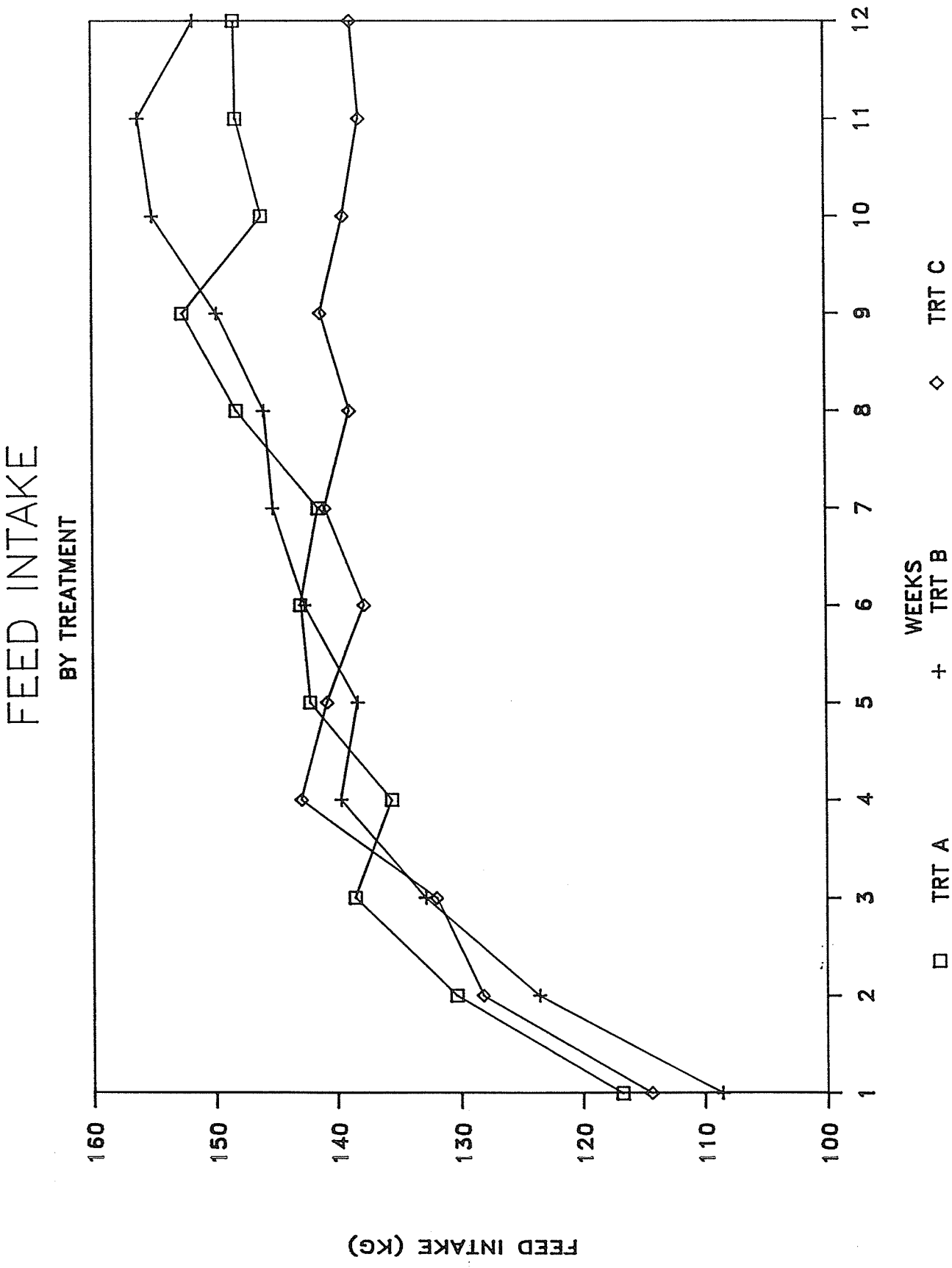
Appendix 2

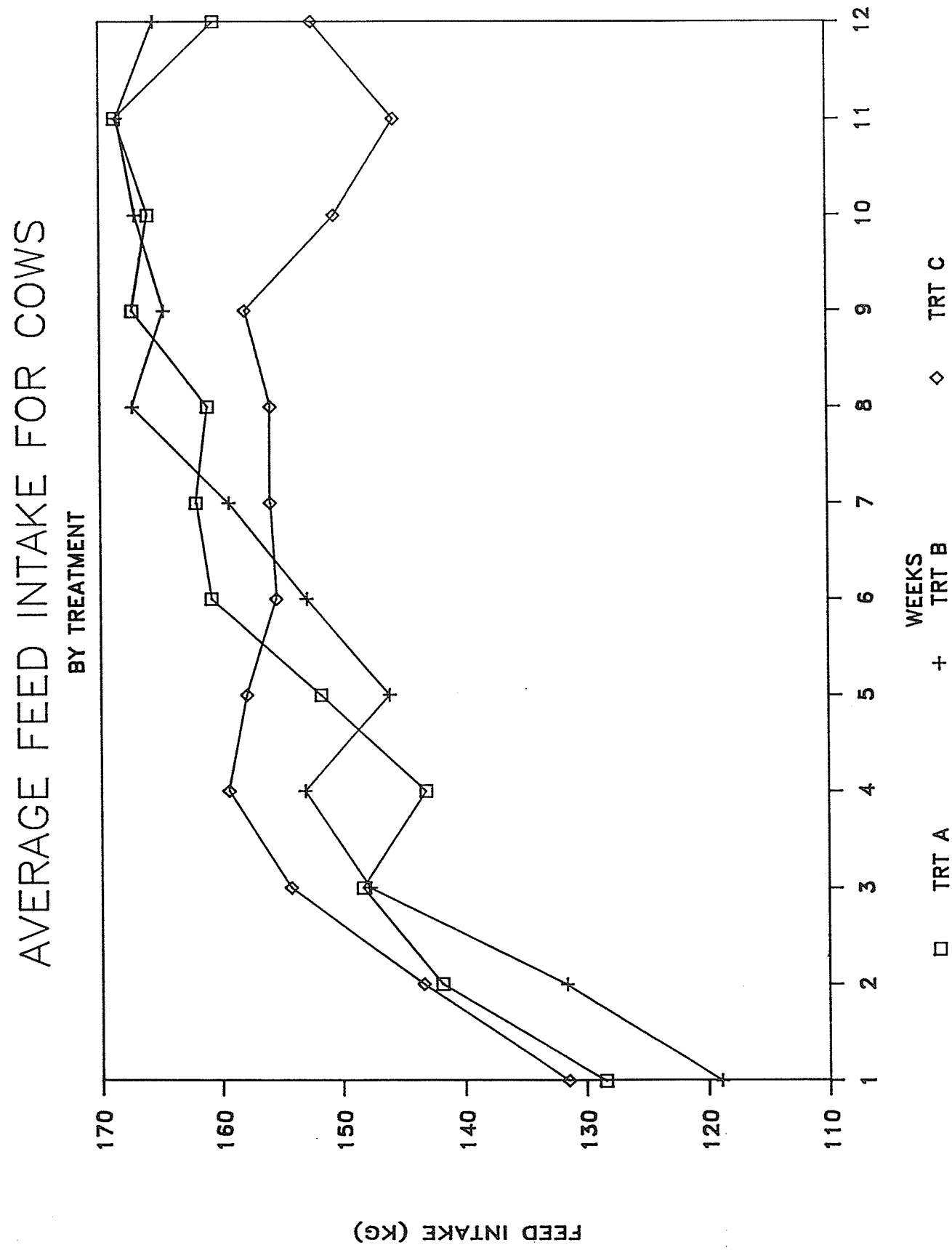
Feed intake graphs for Experiment 1

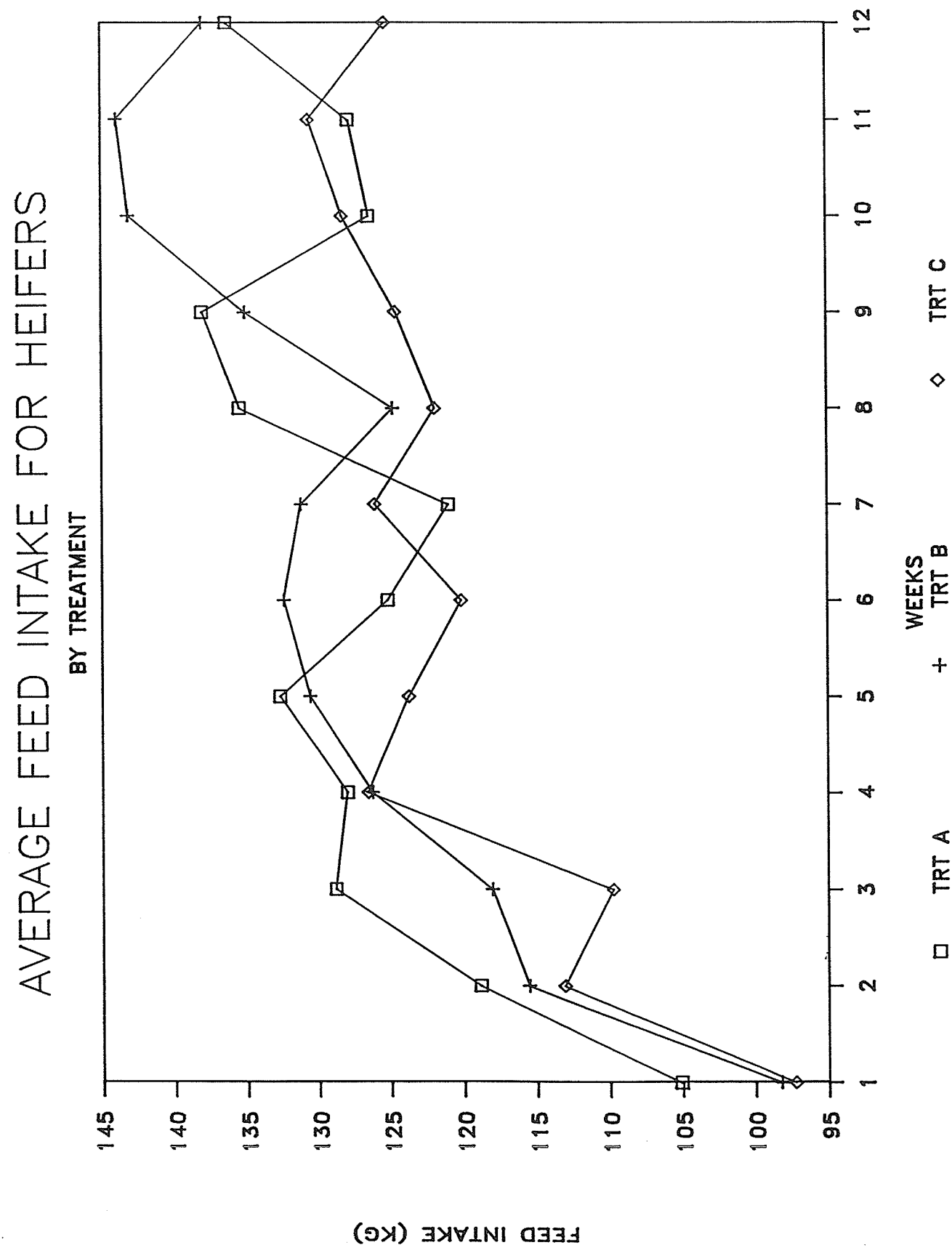
Trt A - Whole sunflower seeds

Trt B - Control

Trt C - 3.4% tallow + sunflower hulls + meal


FEED INTAKE
BY TREATMENT
FEED INTAKE (KG)
160
150
140
130
120
110
100
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
TRT A
TRT B
TRT C


AVERAGE FEED INTAKE FOR COWS
BY TREATMENT
FEED INTAKE (KG)
170
160
150
140
130
120
110
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
TRT A
TRT B
TRT C


AVERAGE FEED INTAKE FOR HEIFERS
BY TREATMENT
FEED INTAKE (KG)
145
140
135
130
125
120
115
110
105
100
95
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
TRT A
TRT B
TRT C

Appendix 3

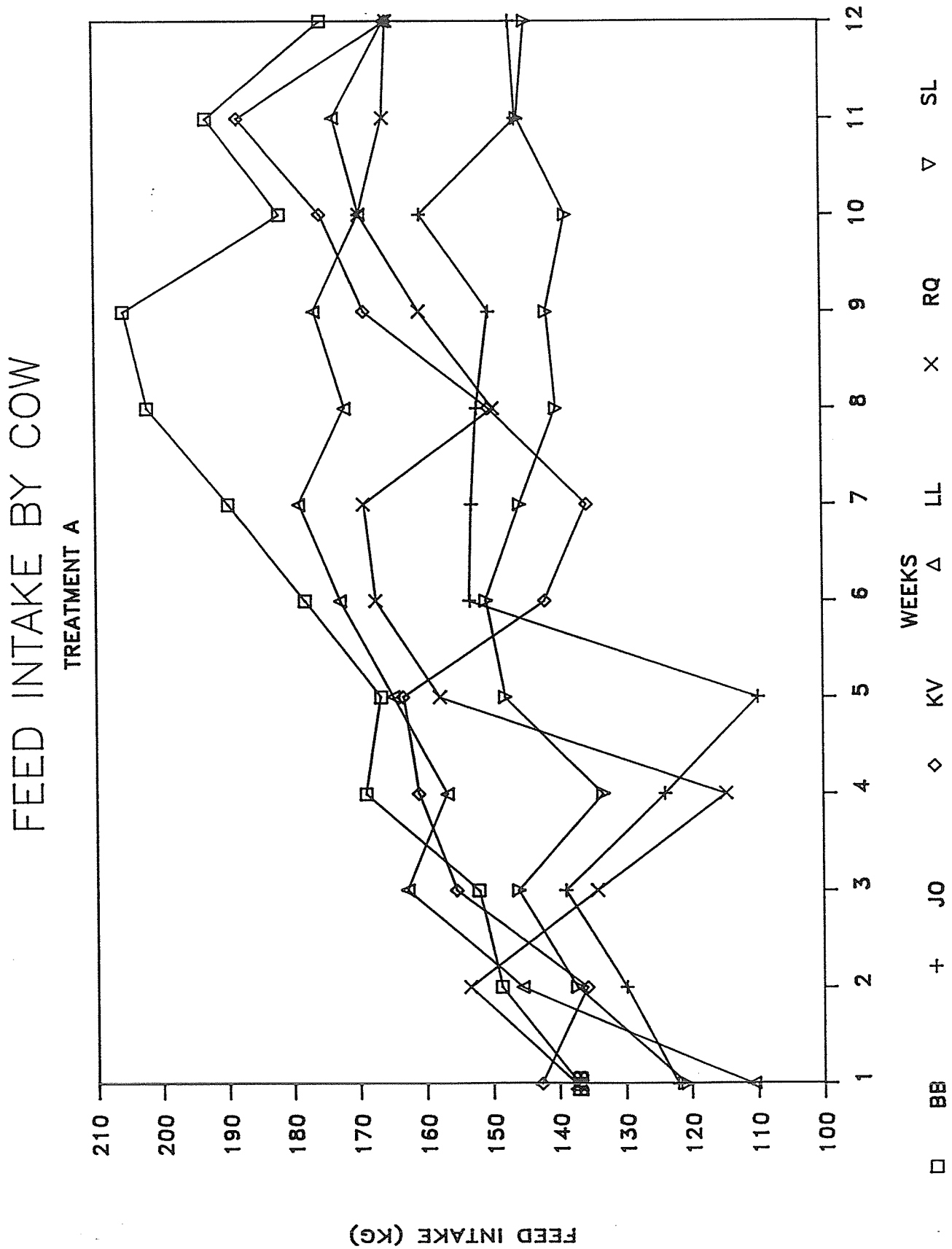
Feed intake graphs for Experiment 1

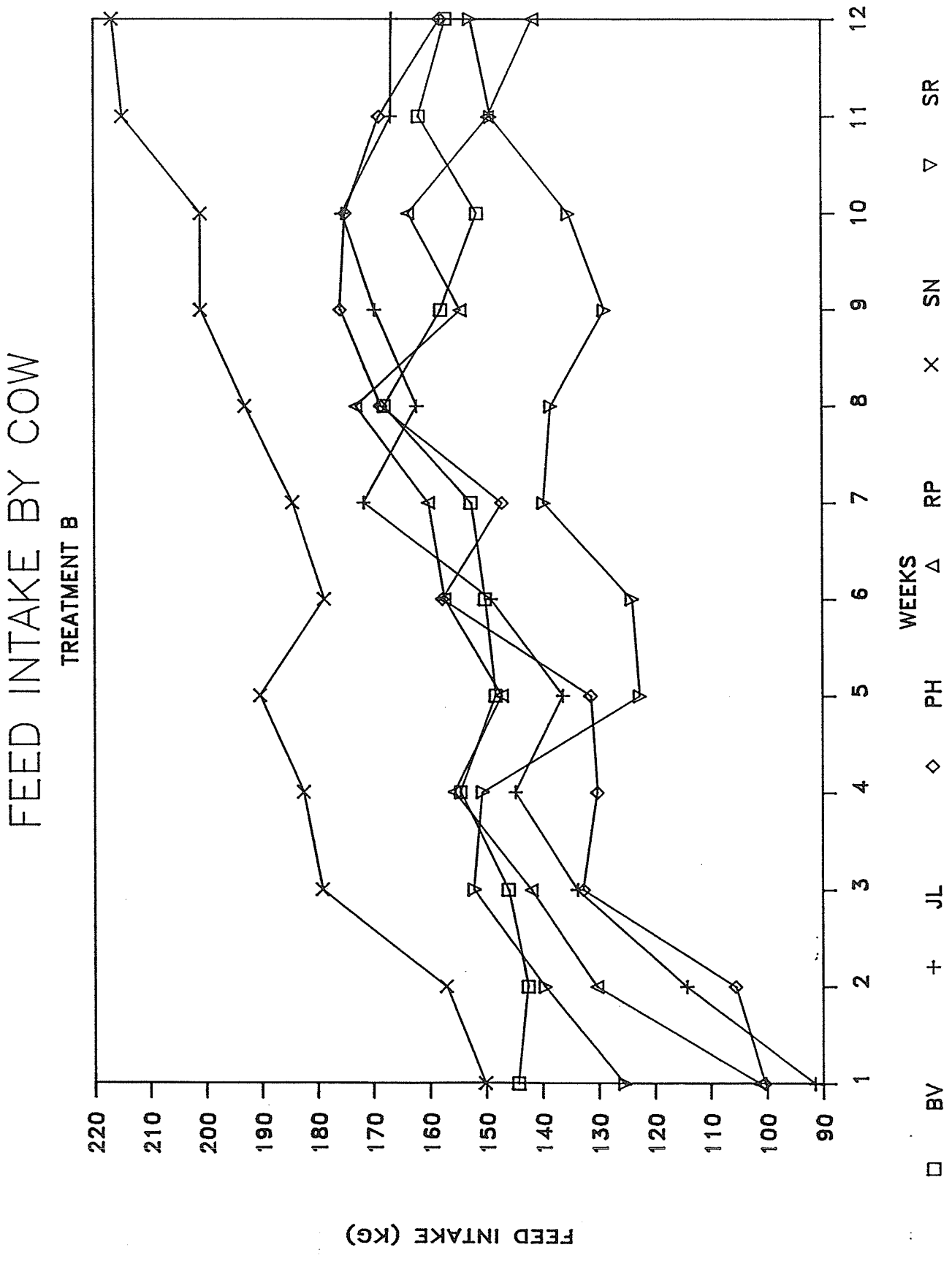
□ + ◇ △ X ▽ = individual cows on treatments

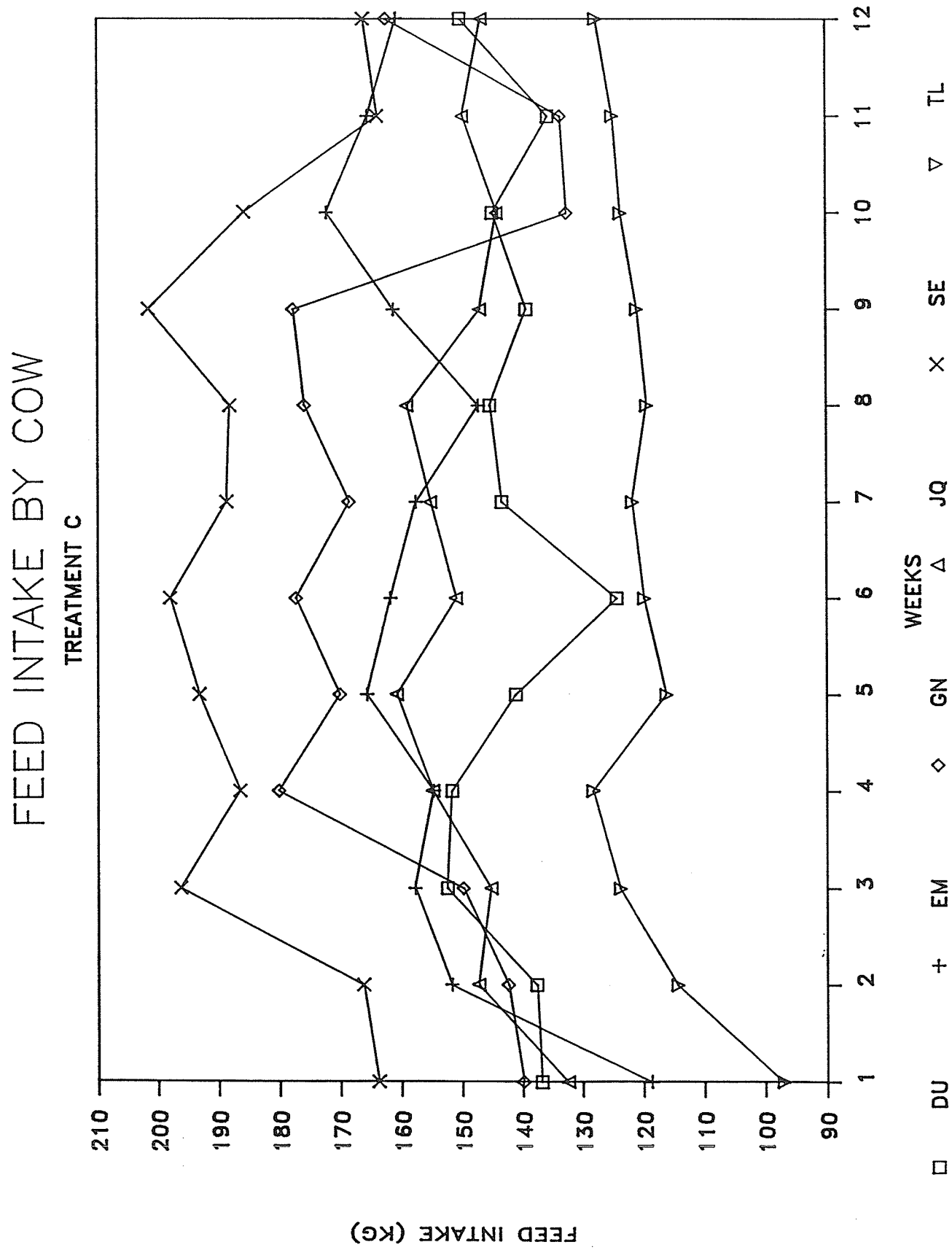
Treatment A - 9% Whole sunflower seed

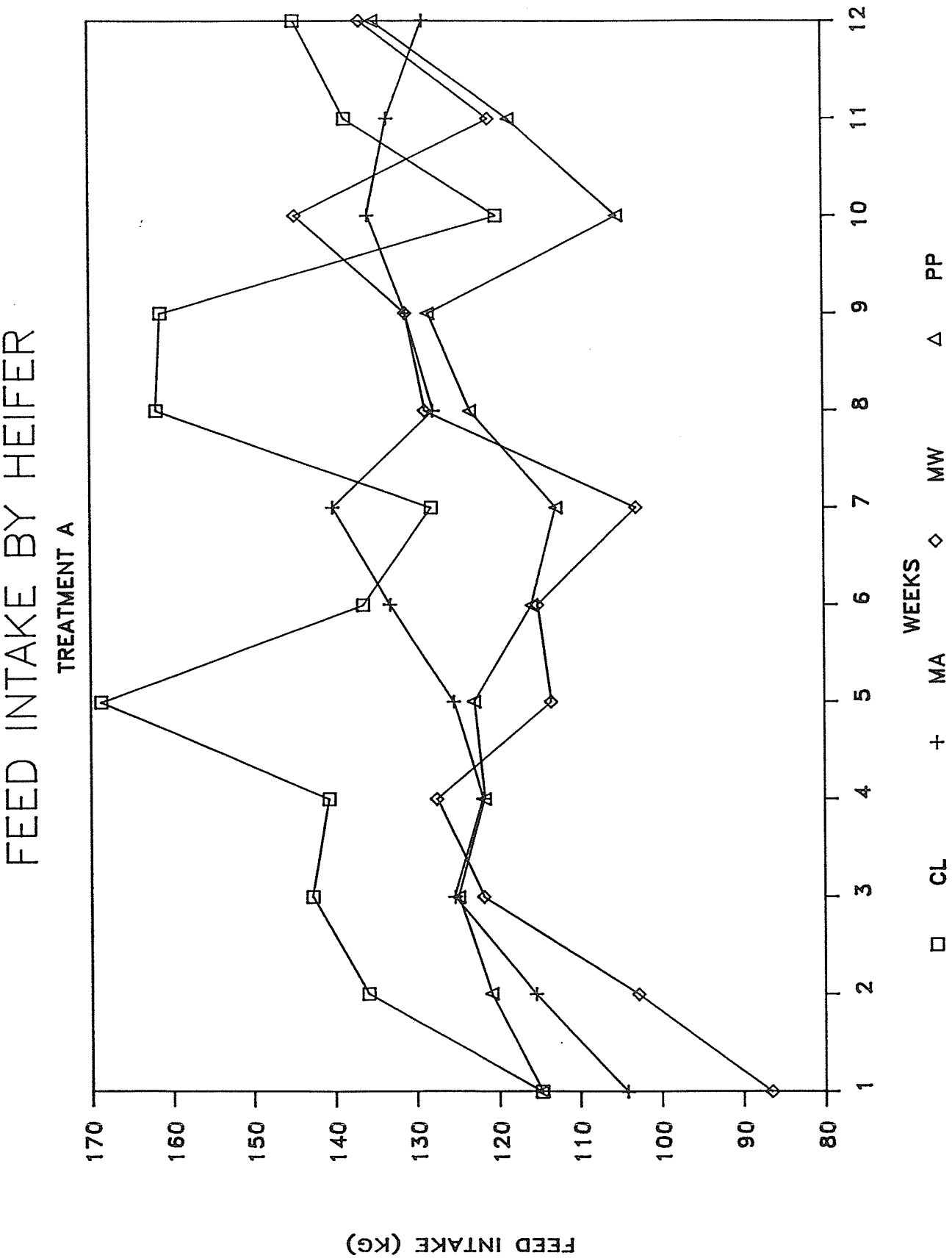
Treatment B - Control

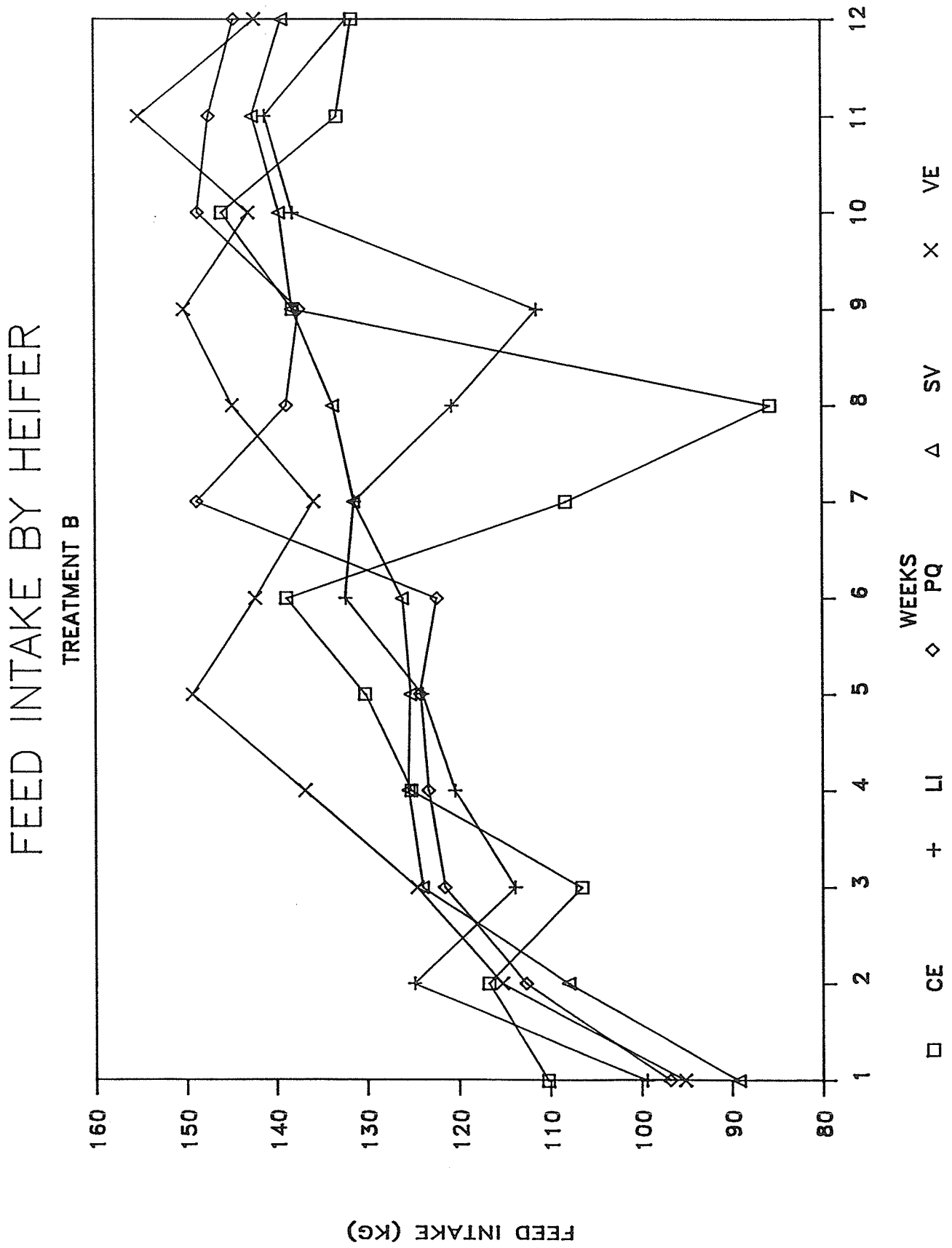
Treatment C - 3.4% Tallow plus sunflower meal and hulls

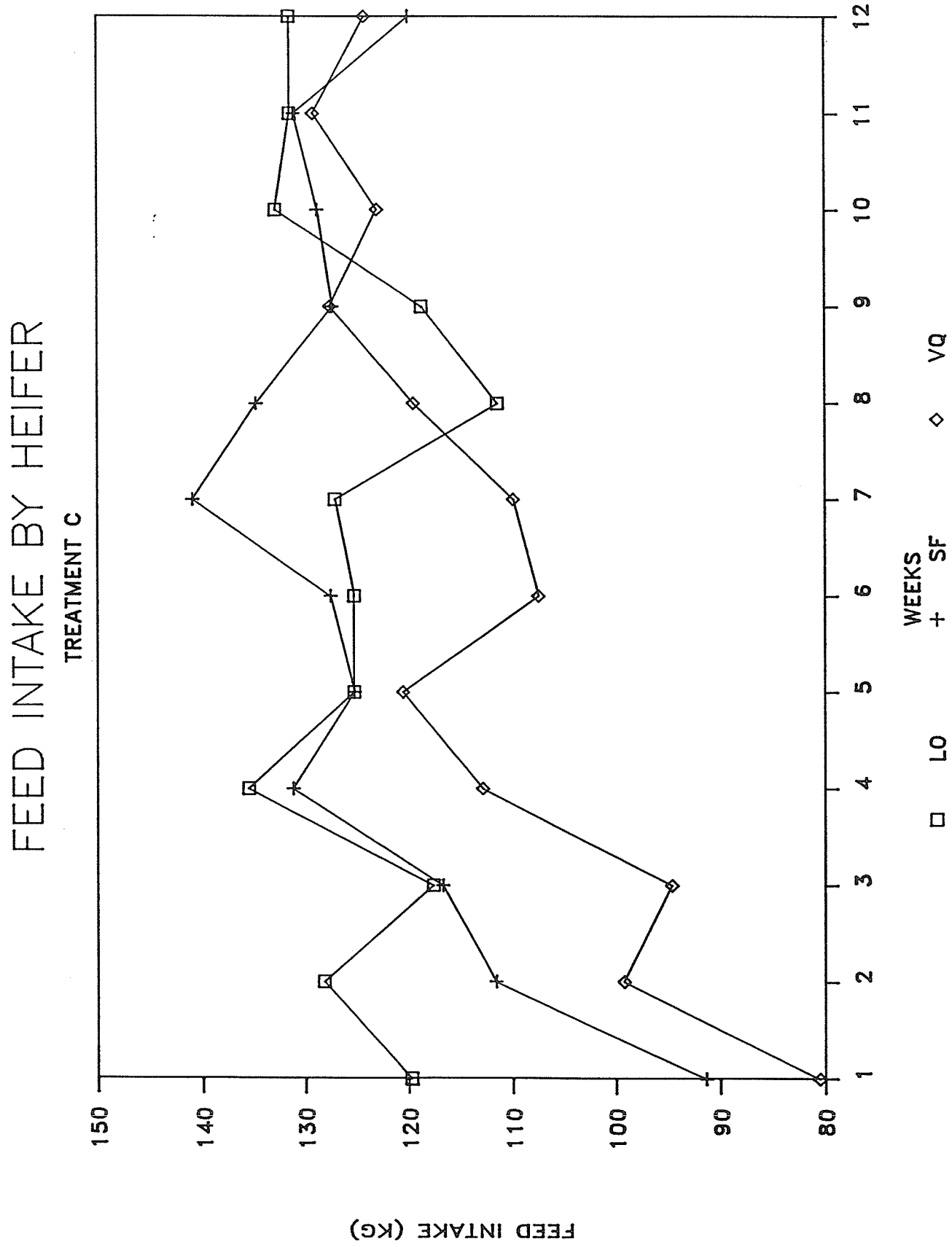

FEED INTAKE BY COW
TREATMENT A
FEED INTAKE (KG)
210
200
190
180
170
160
150
140
130
120
110
100
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
BB
JO
KV
LL
RQ
SL


FEED INTAKE BY COW
TREATMENT B
FEED INTAKE (KG)
220
210
200
190
180
170
160
150
140
130
120
110
100
90
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
□ BV
+ JL
◇ PH
△ RP
× SN
▽ SR


FEED INTAKE BY COW
TREATMENT C
FEED INTAKE (KG)
210
200
190
180
170
160
150
140
130
120
110
100
90
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
□ DU
+ EM
◇ GN
△ JQ
× SE
▽ TL


FEED INTAKE BY HEIFER
TREATMENT A
FEED INTAKE (KG)
170
160
150
140
130
120
110
100
90
80
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
CL
MA
MW
PP


FEED INTAKE BY HEIFER
TREATMENT B
FEED INTAKE (KG)
160
150
140
130
120
110
100
90
80
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
CE
LI
PQ
SV
VE


FEED INTAKE BY HEIFER
TREATMENT C
FEED INTAKE (KG)
150
140
130
120
110
100
90
80
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
LO
SF
VQ

Appendix 4. Analysis of variance for milk production for multiparous (M) and primiparous (P) cows (Trial 1)

Parameter	Source	Degrees of freedom	Type III mean square	F value	Significance level
Milk production (M)	Treatment (Trt)	2	18328.99	0.60	NS
	Season	1	41467.49	1.35	NS
	Cow (Trt*season)	14	30755.56	102.62	.0001
	Week	11	1516.61	5.06	.0001
	Trt*week	22	419.47	1.40	NS
	Week*season	11	281.84	0.94	NS
	Error	154	299.70		
Milk production (P)	Treatment (Trt)	2	4532.80	0.70	
	Season	1	3333.14	0.51	
	Cow (Trt*season)	8	6499.66	40.38	
	Week	11	1111.75	6.91	
	Trt*week	22	276.54	1.72	
	Week*season	11	166.25	1.03	
	Error	88	160.96		

NS - Not significant (P>0.05).

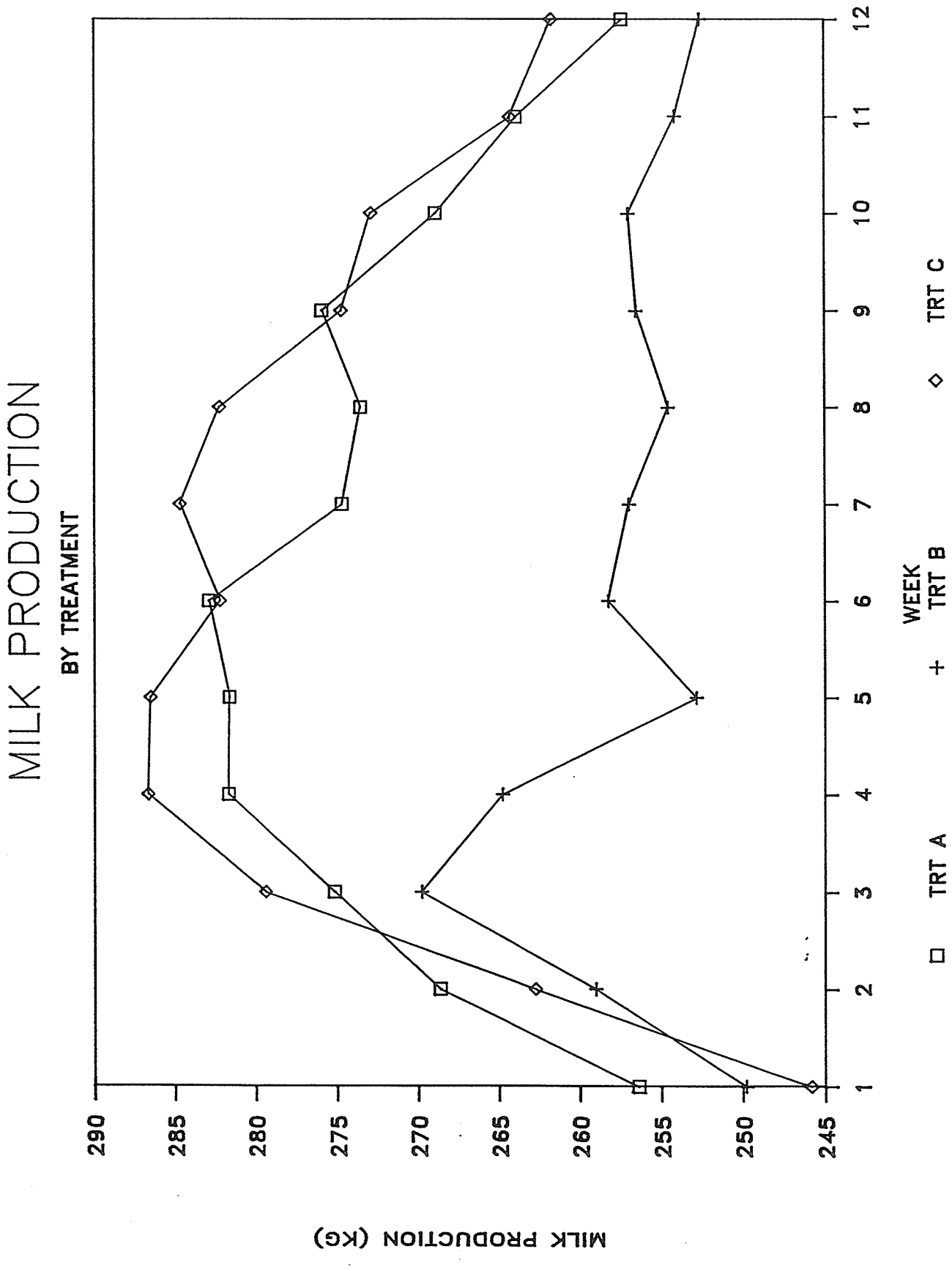
Appendix 5

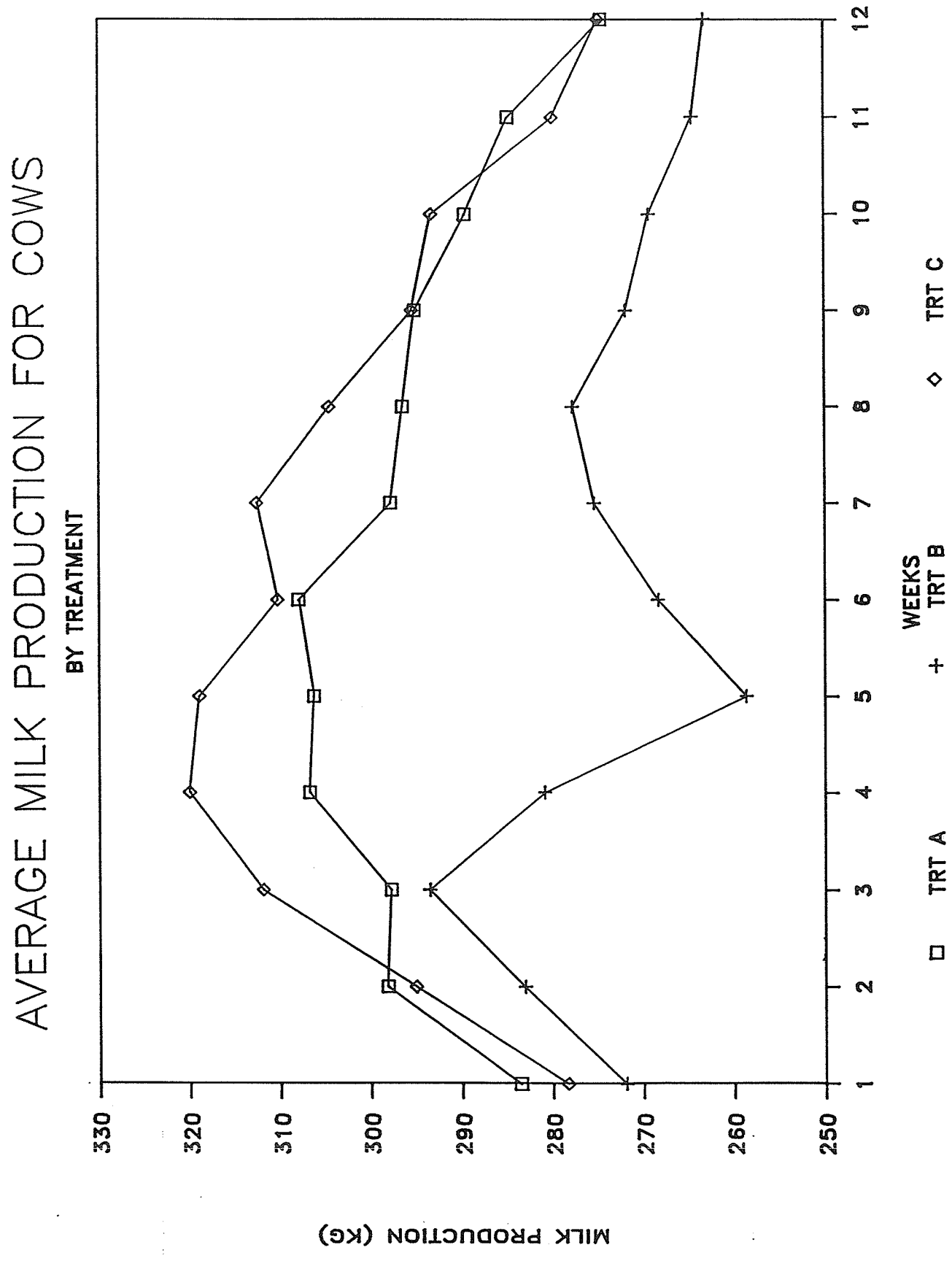
Milk production graphs for Experiment 1

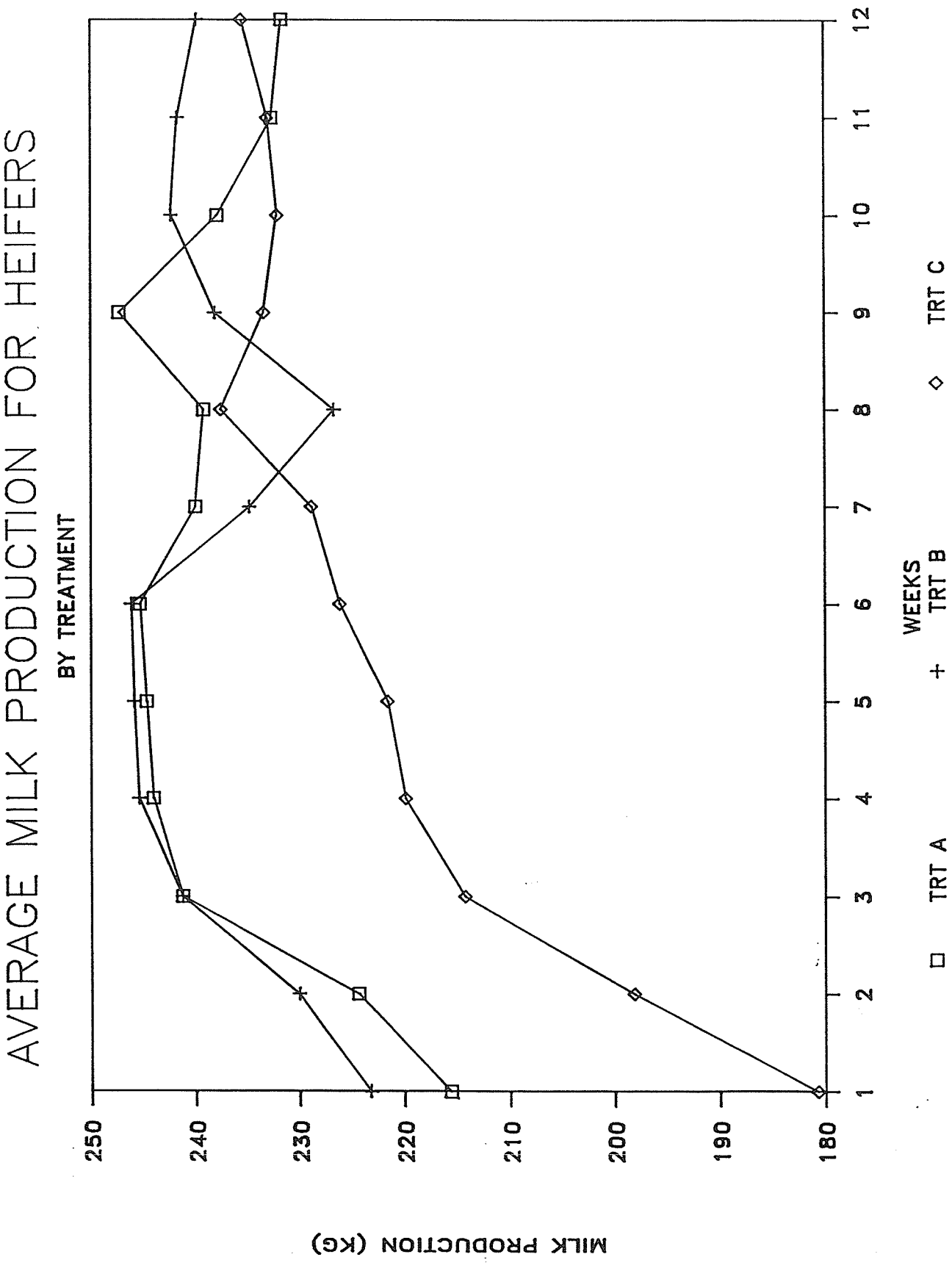
Trt A - 9% Whole sunflower seed

Trt B - Control

Trt C - 3.4% Tallow plus sunflower seeds and hulls


MILK PRODUCTION
BY TREATMENT
MILK PRODUCTION (KG)
290
285
280
275
270
265
260
255
250
245
1
2
3
4
5
6
7
8
9
10
11
12
WEEK
TRT A
TRT B
TRT C


AVERAGE MILK PRODUCTION FOR COWS
BY TREATMENT
MILK PRODUCTION (KG)
330
320
310
300
290
280
270
260
250
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
TRT A
TRT B
TRT C


AVERAGE MILK PRODUCTION FOR HEIFERS
BY TREATMENT
MILK PRODUCTION (KG)
250
240
230
220
210
200
190
180
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
TRT A
TRT B
TRT C

Appendix 6

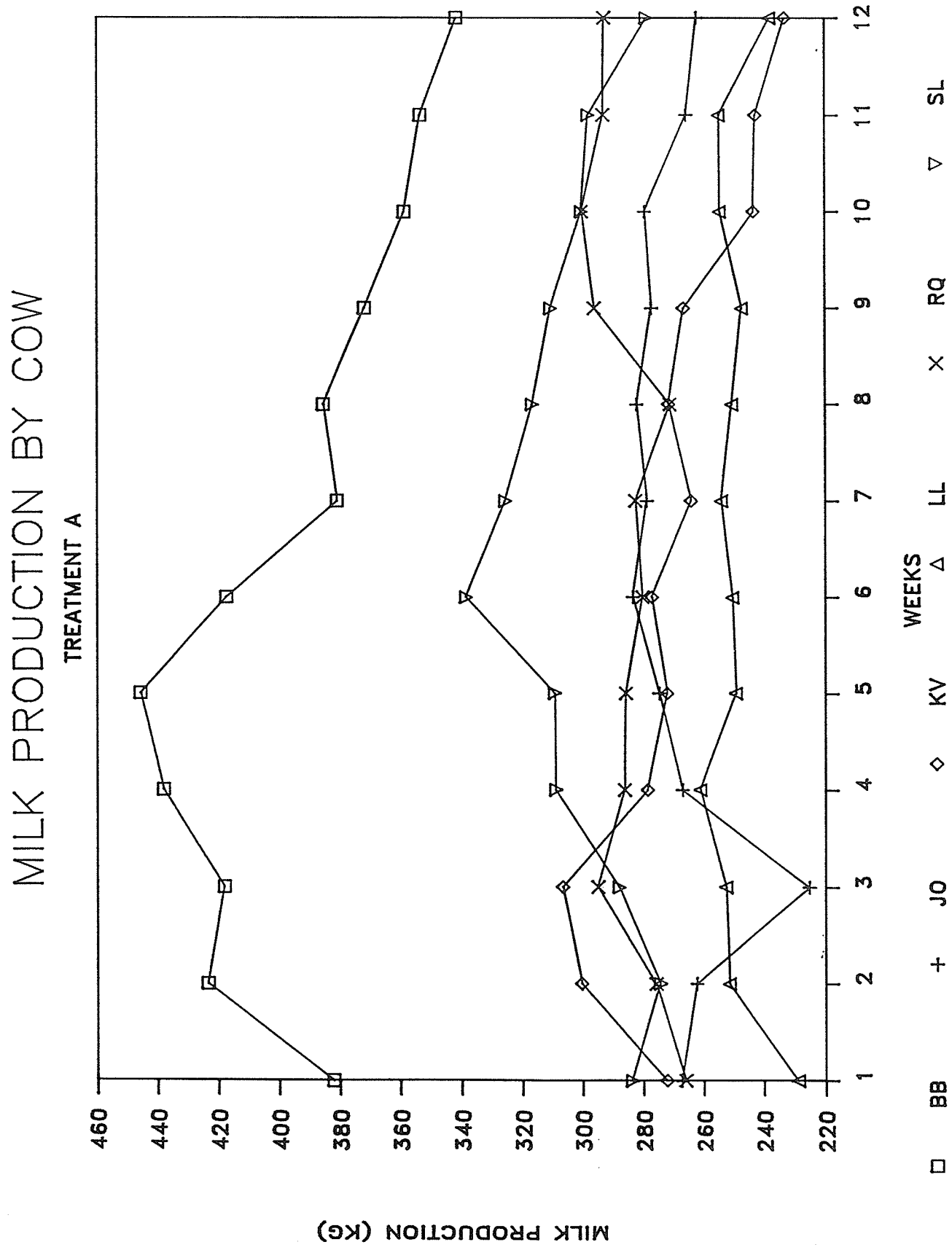
Milk production graphs for Experiment 1

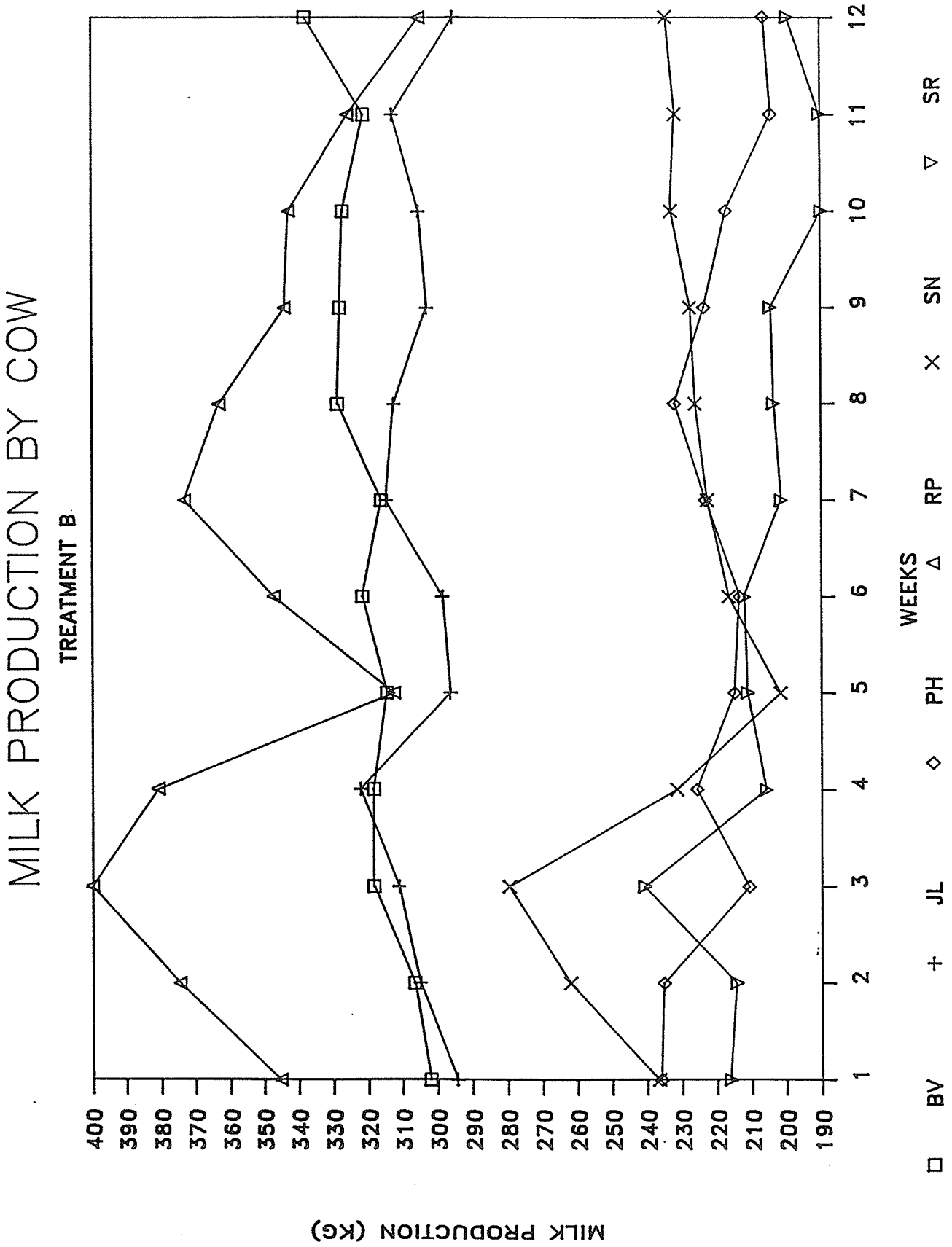
□ + ◇ △ × ▽ = individual cows on treatments

Treatment A - 9% Whole sunflower seed

Treatment B - Control

Treatment C - 3.4% Tallow plus sunflower meal and hulls


MILK PRODUCTION BY COW
TREATMENT A
MILK PRODUCTION (KG)
460
440
420
400
380
360
340
320
300
280
260
240
220
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
BB
JO
KV
LL
RQ
SL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TREATMENT B
MILK PRODUCTION (KG)
400
390
380
370
360
350
340
330
320
310
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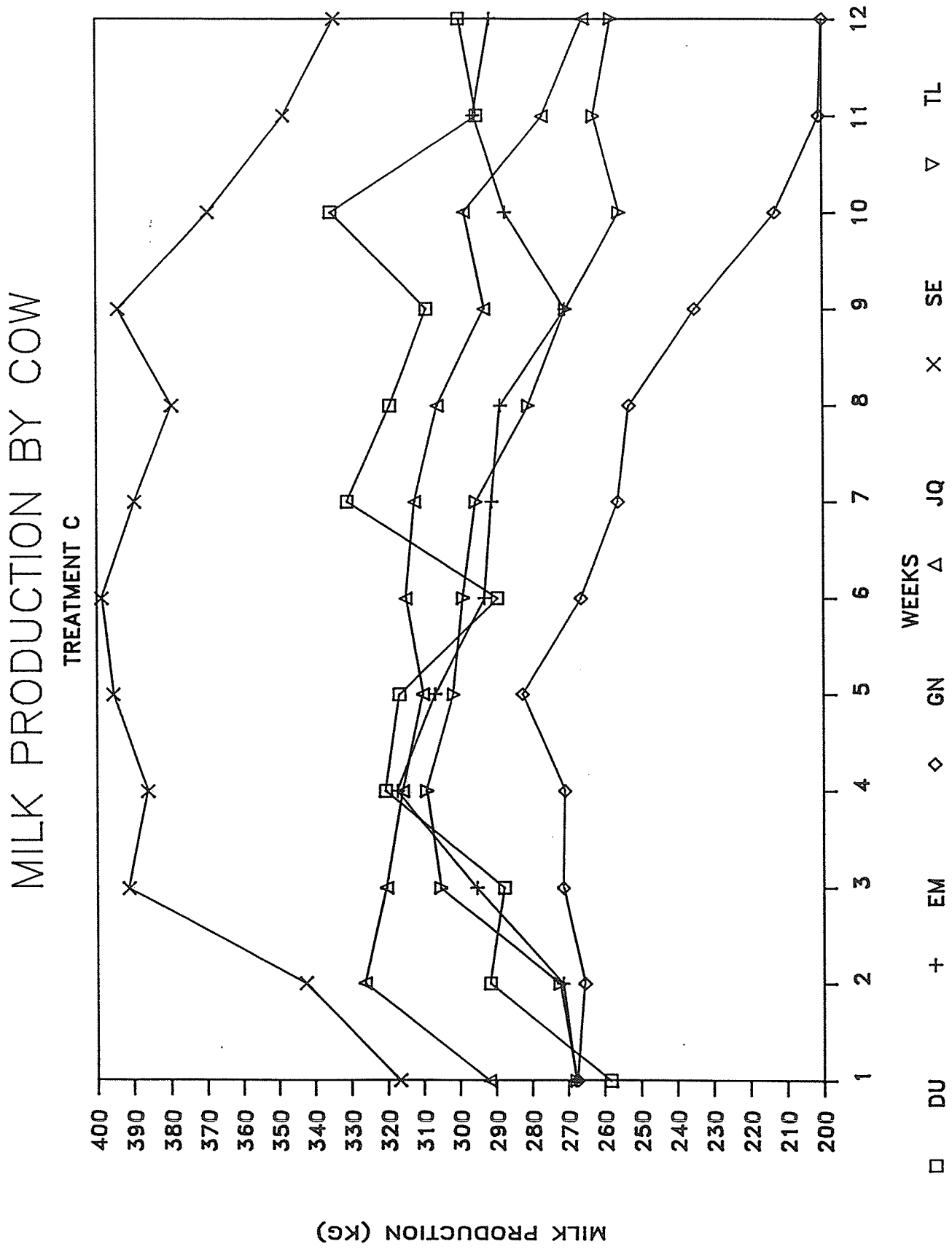
290
280
270
260
250
240
230
220
210
200
190
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
□ BV
+ JL
◇ PH
△ RP
× SN
▽ SR


MILK PRODUCTION BY COW
TREATMENT C
MILK PRODUCTION (KG)
400
390
380
370
360
350
340
330
320
310
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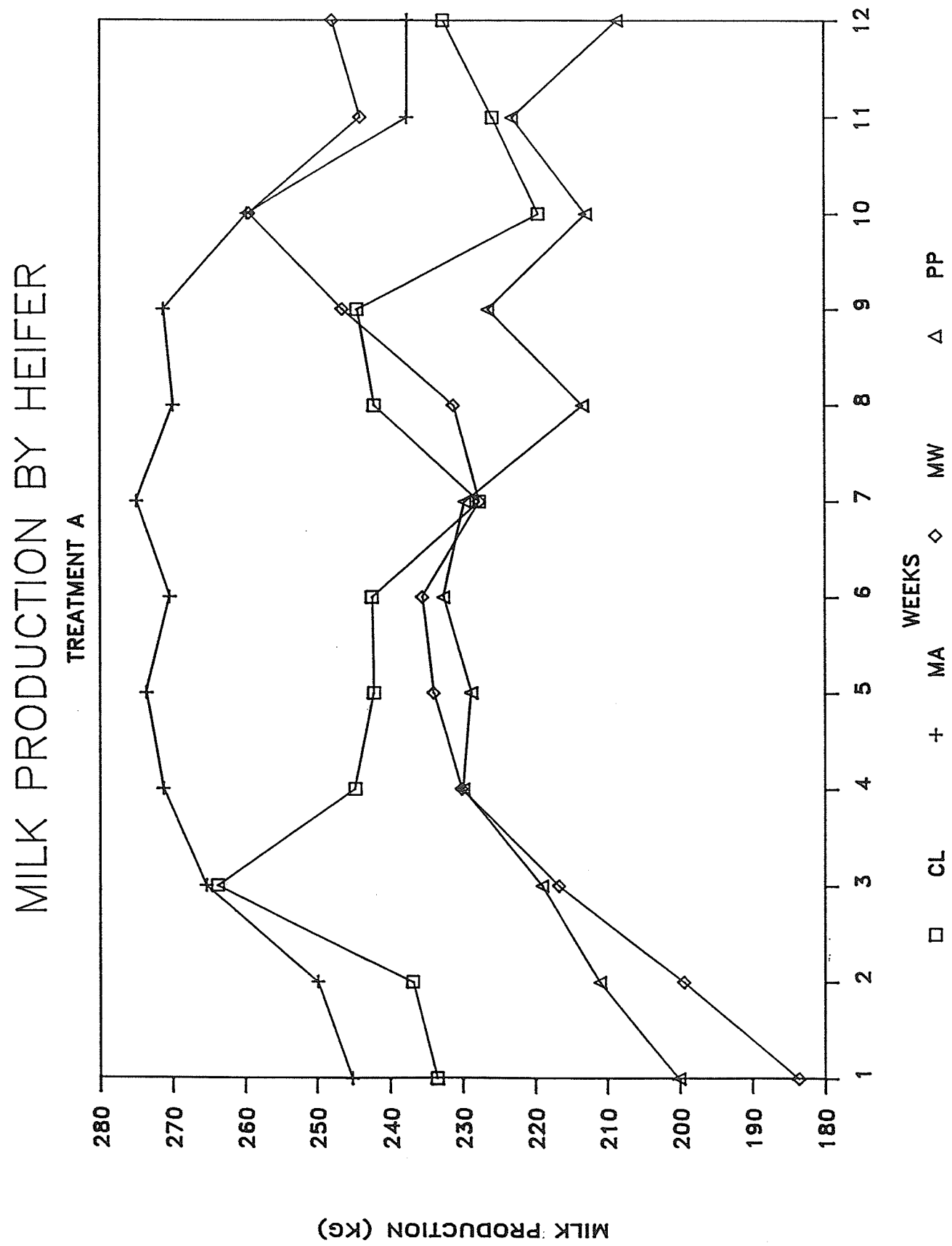
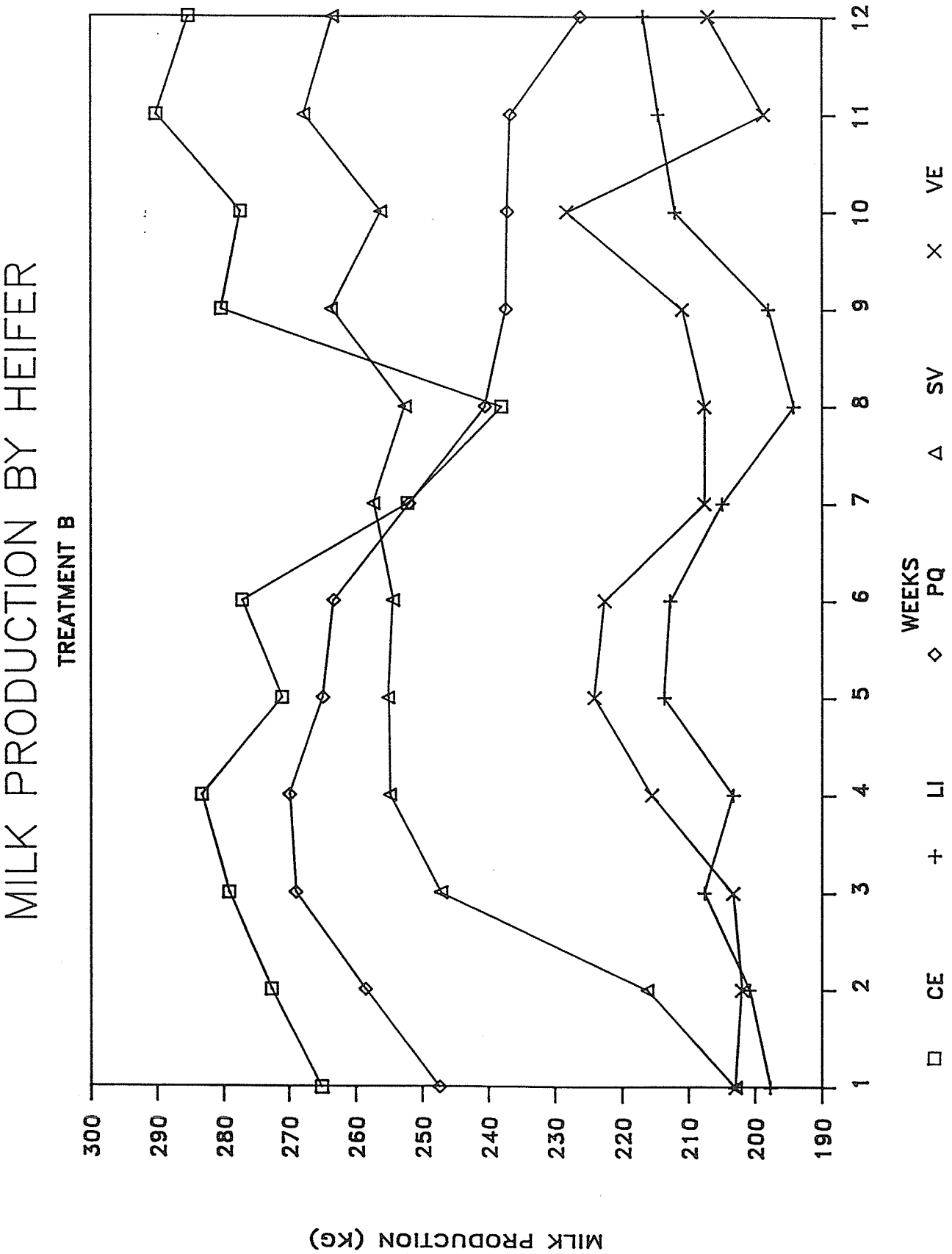
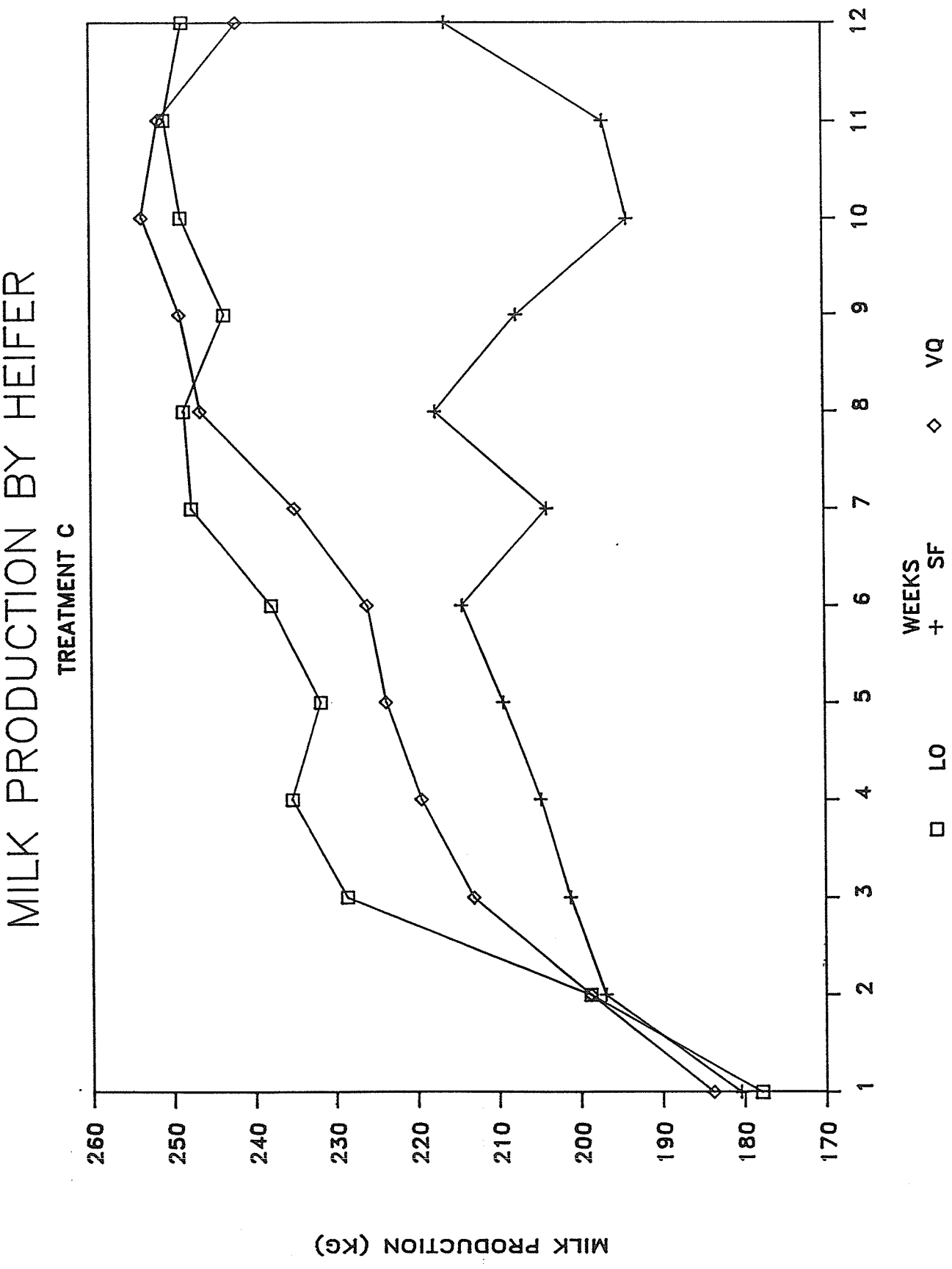
290
280
270
260
250
240
230
220
210
200
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
DU
EM
GN
JQ
SE
TL


MILK PRODUCTION BY HEIFER
TREATMENT A
MILK PRODUCTION (KG)
280
270
260
250
240
230
220
210
200
190
180
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
□ CL
+ MA
◇ MW
△ PP


MILK PRODUCTION BY HEIFER
TREATMENT B
MILK PRODUCTION (KG)
300
290
280
270
260
250
240
230
220
210
200
190
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
□ CE
+ LI
◇ PQ
△ SV
× VE


MILK PRODUCTION BY HEIFER
TREATMENT C
MILK PRODUCTION (KG)
260
250
240
230
220
210
200
190
180
170
1
2
3
4
5
6
7
8
9
10
11
12
WEEKS
□ LO
+ SF
◇ VQ

Appendix 7. Analysis of variance for milk protein and milk lactose levels in Trial 1

Parameter	Source	Degrees of freedom	Type III mean square	F value	Significance level
Milk protein	Treatment (Trt)	2	0.89	4.43	.0231
	Type	1	0.62	3.07	NS
	Trt*type	2	0.03	0.16	NS
	Cow (Trt*type)	24	0.20	13.23	.0001
	Week	5	0.03	1.76	NS
	Trt*week	10	0.01	0.65	NS
	Type*week	5	0.01	0.93	NS
	Error	130	0.02		
Milk lactose	Treatment (Trt)	2	0.65	2.52	NS
	Type	1	0.43	1.67	NS
	Trt*type	2	0.42	1.62	NS
	Cow (Trt*type)	24	0.26	16.95	.0001
	Week	5	0.03	1.83	NS
	Trt*week	10	0.01	0.55	NS
	Type*week	5	0.03	1.85	NS
	Error	130			

NS - Not significant (P>0.05).

Appendix 8. Analysis of variance for dry matter intake and milk production (Trial 2)

Parameter	Source	Degrees of freedom	Type III mean square	F values	Significance level
Dry matter intake	Square	1	1571.63	10.90	.0040
	Treatment (Trt)	3	157.37	1.09	NS
	Period	3	705.78	4.89	.0117
	Cow (Square)	6	771.90	5.35	.0025
	Trt*Period*Cow (Square)	18	144.21	1.74	NS
	Error	32	82.72		
Milk production	Square	1	109.86	0.13	NS
	Treatment (Trt)	3	66.59	0.08	NS
	Period	3	5797.28	6.91	.0027
	Cow (Square)	6	4687.44	5.59	.0020
	Trt*Period*Cow (Square)	18	839.29	6.55	.0001
	Error	32	128.17		

NS - Not significant (P>0.05).

Appendix 9. Analysis of variance for milk protein, milk lactose and milk fat levels (Trial 2)

Parameter	Source	Degrees of freedom	Type III mean square	F values	Significance level
Milk protein	Square	1	0.04	0.41	NS
	Treatment (Trt)	3	0.13	1.36	NS
	Period	3	0.95	10.18	.0004
	Cow (Square)	6	0.39	4.14	.0087
	Hour	1	0.98	64.37	NS
	Trt*Period*Cow (Square)	18	0.09	6.15	.0001
	Error	201	0.02		
Milk fat	Square	1	0.58	0.87	NS
	Treatment (Trt)	3	0.49	0.73	NS
	Period	3	1.70	2.52	NS
	Cow (Square)	6	2.05	3.03	.0314
	Trt*Period*Cow (Square)	18	0.68		.0001
	Error	190	0.23		
Milk lactose	Square	1	0.19	2.08	NS
	Treatment (Trt)	3	0.24	2.64	NS
	Period	3	0.88	9.54	.0005
	Cow (Square)	6	0.19	2.03	NS
	Hour	1	0.25	6.82	NS
	Trt*Period*Cow (Square)	18	0.09	2.48	.0012
	Error	202	0.04		

NS - Not significant