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THE EFFECT OF ENERGY INTAKE LEVEL ON THE DIGESTIBILITY  
OF HIGH ENERGY RATIONS BY CATTLE

by

Wayne Carl Figroid

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A Dissertation Submitted to the Faculty of the  
COMMITTEE ON AGRICULTURAL BIOCHEMISTRY AND NUTRITION

In Partial Fulfillment of the Requirements  
For the Degree of

DOCTOR OF PHILOSOPHY

In the Graduate College

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GRADUATE COLLEGE

I hereby recommend that this dissertation prepared under my  
direction by Wayne Carl Figroid  
entitled The effect of energy intake level on the digestibility  
of high energy rations by cattle  
be accepted as fulfilling the dissertation requirement of the  
degree of Doctor of Philosophy

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SIGNED: Wayne Carl Figliach

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## ABSTRACT

Total collection digestion studies were conducted with steers in which 0, 5, 10, and 15 percent animal fat was added to 60, 75, and 90 percent concentrate rations.

Dry matter consumed as a percent of body weight decreased ( $P < .01$ ) with increasing ration fat and concentrate levels. Increasing fat level decreased ( $P < .05$ ) apparent dry matter, gross energy, and chloroform-methanol-hydrochloric acid lipid digestibilities. Increasing concentrate level increased ( $P < .01$ ) dry matter, gross energy, crude protein, acid detergent fiber, and chloroform-methanol-hydrochloric acid lipid apparent digestibilities.

Lipid ingested per unit of body weight was least at each fat level for 90 percent concentrate rations. These rations resulted in the greatest decreases in digestibilities and performance, implicating a lipid X concentrate interaction as the limiting factor.

Increasing fat and concentrate levels increased ( $P < .01$ ) fecal soap excretion to a maximum of 24 percent of fecal dry matter, or to 31 percent of ingested lipid. Apparent calcium digestibility decreased (excretion increased) as fecal soap excretion increased. Gross energy per unit fecal dry matter increased with both increasing fat and concentrate levels.

When on constant feed increasing fat level reduced ( $P < .01$ ) rumen pH. Increasing concentrate level lowered ( $P < .01$ ) rumen pH on both constant and ad libitum feed. Increasing fat and concentrate

levels depressed ( $P < .05$ ) acetic and increased ( $P < .05$ ) propionic acid concentrations of constant and ad libitum feed rumen samples.

Ten percent added fat slightly decreased digestible energy intake, but 15 percent added fat and increasing concentrate levels drastically reduced digestible energy intake.

A 90 percent concentrate ration was fed to cattle at several levels to evaluate, by multiple regression analysis, the effect of feed intake on apparent digestibilities.

The narrow range of feed intakes attained precluded identification of the feed intake, digestibility relationship from power functions of first through fourth powers, or sums of these, for polynomial, logarithmic, or exponential expressions.

Apparent dry matter, gross energy, and crude protein digestibilities declined 4.5, 5.0, and 8.9 percentage units, respectively, when feed intake was increased from mean levels of 0.97 to 1.81 times calculated maintenance requirements. Calculated from regression equations ( $P < .01$ ), changes in digestibilities for increasing feed intakes from 1 to 2 times maintenance were in percentage units -5.4 to -6.3 (dry matter), -6.0 to -7.1 (gross energy), and -10.3 to -11.6 (crude protein).

Four 90 percent concentrate rations (10 to 16 percent crude protein) were fed to cattle to evaluate the effect of diet protein level on apparent digestibilities, and to calculate metabolic fecal nitrogen by extrapolation to zero nitrogen intake.

Increasing protein increased ( $P < .01$ ) apparent digestibilities of dry matter, gross energy, and crude protein by approximately 5, 5,

and 20 percentage units, respectively, or by 0.76, 0.78, and 2.90 percentage units per percent protein.

Fecal nitrogen was regressed on feed nitrogen, both expressed on the basis of 100 g dry matter of (1) feed, (2) feces and feed, respectively, and (3) feces. The resulting equations were (1)  $y = 0.418 + 0.2099x - 0.0060x^4$  (0.418 g fecal nitrogen per 100 g feed dry matter at zero nitrogen intake), (2)  $y = 2.20 + 0.609x$ , and (3)  $y = 2.63 + 0.0827x$ . Equation (1) was not significant ( $P < .05$ ); equations (2) and (3) were significant ( $P < .05$ ). Correlations were higher ( $P > .05$ ) when nitrogen was expressed on the basis of fecal, rather than feed, dry matter.

Grams fecal nitrogen per 100 g fecal dry matter were regressed on grams digestible nitrogen per 100 g dry matter of (1) feed and (2) feces. Equations best describing these relationships were (1)  $y = 2.75 + 0.54x$  and (2)  $y = 2.91 + 0.084x$ .



## INTRODUCTION

As world population continues to increase and man's knowledge expands, there is an increasing awareness of the critical importance of food production. This concern has extended beyond the quantity of food produced to the more important concept of food quality. A ready means of improving human nutrition is to increase the proportion of animal protein products in the diet, an acceptable solution because people are accustomed to and like to eat meat. It is a reasonable solution, for as human food production increases, there will also be an increased quantity of by-products available for use as animal feeds. However, in general these by-products--straws, rinds, pulps, etc.--are high in cellulose and indigestible materials and are therefore utilized much more effectively by ruminants than nonruminants. Since pressures on the food supply by the human population will continue to increase, it is easy to hypothesize that feedstuffs for animal production will be more and more relegated to the by-products of human food production. These pressures on animal feedstuffs will require not only an increased utilization of by-product feeds by ruminants, but also a reduced and more judicious use of concentrate feeds, such as grains and oil meals, that are necessary human and non-ruminant sources of energy and protein. Animal nutritionists, assessing these facts, have increasingly emphasized the importance of investigating the utilization of energy and protein for meat production by farm animals.

Certainly an important by-product feed ingredient available to livestock producers is animal fat. Fat is unique among by-product feed-stuffs in that it is a concentrated source of energy and, at levels of 4 or 5 percent of the diet, has been routinely added to ruminant rations in the western United States. While the results of considerable research are available with respect to the use of fat at these levels, there is a dearth of information regarding its use at levels of 10 to 15 percent of the ration. Experiment I was undertaken to study the effect of dietary fat and concentrate levels on ration digestibility.

With increasing pressures for the maximum effective utilization of concentrates by animals, it is becoming more important to define energy losses incurred in the digestion and metabolism of these feeds. Because the efficiency of energy utilization by ruminant digestive processes is relatively low, digestion usually represents the single greatest loss of ingested energy to the animal. As one examines the literature, the importance of digestion trials becomes apparent. Yet, it is also apparent that relatively little attention has been given to the level of feed consumption, with respect to a percent of body weight or to maintenance energy requirements, as it related to digestive efficiency. Since the level of feed consumption is known to influence the efficiency of digestive processes, the following question logically arises: Does the feedstuff under study have a single mean digestion coefficient, or is there a family of digestion coefficients that pertain to the various levels of possible feed intake? Experiment II examines

the effect of feed intake level on the digestibility of high concentrate rations.

Protein, as the highest priced and least plentiful component of ruminant rations, is unfortunately rather inefficiently utilized. As with energy, digestive losses represent a substantial inefficiency in its utilization. This digestive cost must be determined with cattle consuming typical high concentrate rations before protein requirements of cattle can be understood. One of the least understood protein digestive losses in ruminants is that of metabolic nitrogen excretion in the feces. This cost has been well assessed with roughage diets fed to cattle, but the limited information available on high concentrate diets is somewhat ambiguous. Experiment III was undertaken to examine the influence of diet protein level on ration component digestibilities and to determine metabolic fecal nitrogen excretion of cattle consuming high concentrate rations.

## REVIEW OF LITERATURE

### Effect of Dietary Fat on Ration Utilization and Rumen Function

The literature pertaining to the addition of fat to cattle rations deals primarily with low levels of fat and relatively high roughage rations. Little information is available regarding the addition of large quantities of fats to typical cattle finishing rations. However, some information is available from sheep experiments, and because of obvious digestive system similarities to cattle, these data have been included. Evidence from mineral metabolism and microbiological studies has also been cited in an effort to explain the often observed depressed performance and apparent microbial toxicity associated with the feeding of higher levels of fats to ruminants.

### Fecal Soaps

Level of particular minerals and fat in the ration are the principal determinants of the quantity of fecal soaps produced, which may be substantial. Because of the formation of ether-insoluble fecal soaps, which must undergo acid hydrolysis prior to extraction, fat digestibility may not be reliably determined by the ether extract method.

Bohman and Lesperance (20) added 0.5 lb fat per head per day to grass hay fed to cattle and noticed that the percentage of fecal soaps was significantly higher when fat was fed. When both 3.0 lb alfalfa and 0.5 lb fat were added to the basal ration, the amount of soaps was statistically different from, and intermediate between, the nonfat ration

and those with fat only. Even with no added dietary fat the feces contained 2.3 percent fecal soaps. This did not appear to be an artifact, as fecal soaps were not measurable in alfalfa hay samples. Digestibility of the added fat was 6 percent lower when fecal fat was corrected for fecal soaps.

Dijkstra (46) observed that the digestibility of ether extract was increased 30 percent or more by fat additions, while the digestibility of lipids extracted by tetrachloromethane improved 10 to 20 percent for the rations to which fat was added. It was postulated that the difference in digestibilities of fats extracted by the different methods was due to the formation of insoluble calcium and magnesium soaps. Substantiating evidence has been offered by Esplin et al. (53). Five percent fat additions to 70 percent concentrate alfalfa hay cattle rations increased ( $P < .05$ ) fecal soap excretion over the control ration. Fecal soaps as a percent of feces were 2.30, 3.63, and 3.94, respectively, for control, tallow, and hydrolyzed vegetable and animal fat rations.

Roberts and McKirdy (147) also suggested that analysis of feces by the conventional ether extract method does not remove all of the fecal fat. This is illustrated by a comparison of the percent fecal ether extract (2.93) with the percent fecal crude fat (8.10), which includes fecal soaps, for a 95 percent concentrate ration containing 5 percent prime animal tallow.

Ward and Reid (176) have also reported that the ether extract of feces does not provide a reliable indication of the digestibility of fat. They used cows fed varying mineral intakes but consuming otherwise

identical diets. Total lipid excretion, expressed as a percentage of intake and measured by acid hydrolysis and chloroform extraction, did not change with calcium intake. The total ether extractives excreted in the feces were, however, higher ( $P < .01$ ) in the group fed the low calcium level. This was attributed to increased excretion of soaps in the feces of cows in the higher calcium groups and to the fact that these soaps were insoluble in ether. Crockett and Deuel (39) have eloquently urged the importance of considering fecal soap excretion when determining fat digestibility. They observed with rats that the digestibility of hydrogenated lard melting at 55 C was 62 percent, while that of lard melting at 61 C was 21 percent. However, if no allowance had been made in the calculation for the soap fractions, the digestibilities would have been 100 and 91 percent, respectively.

#### Rumen pH and Added Fat Rations

Reports of pH measurements on rumen contents of animals fed fats are almost nonexistent. Putnam, Oltjen, and Bond (143) did, however, note that rumen pH was lower ( $P < .01$ ) when 5 percent soybean oil was added to an all-concentrate steer ration but was somewhat higher when oil was added to a 23 percent alfalfa hay steer ration. In contrast, Brethour, Sirny, and Tillman (24) observed no changes in diurnal rumen pH values when 10 percent corn oil was added to a ground milo-cottonseed hull lamb ration.

#### Rumen Volatile Fatty Acids and Added Fat Rations

Knowledge regarding rumen volatile fatty acid concentrations with added fat rations is also very limited. Brooks et al. (26) reported lower rumen total volatile fatty acid concentration (3.2 mg per 100 ml rumen fluid) when 3.2 percent fat was added to 90 percent cottonseed hull sheep diets. However, the volatile fatty acid concentration returned to normal when alfalfa ash was added to the basal ration plus fat. Esplin et al. (53), on the other hand, reported higher ( $P > .05$ ) total volatile fatty acid concentrations for two 4 percent added fat rations than for the control ration. Molar percents of the short chain fatty acids were essentially the same across all groups for the 70 percent concentrate steer rations studied. Using all-concentrate rations, Putnam et al. (143) noted that addition of 5 percent soybean oil to the diet resulted in a greater ( $P < .05$ ) ruminal total volatile fatty acid concentration and an increase in propionate concentration at the expense of acetate. With a 23 percent alfalfa hay mixed ration, 5 percent added oil increased the total ruminal volatile fatty acid concentration. Dietary cod-liver oil decreased the proportion of acetic acid while increasing the proportion of propionic acid in rumen ingesta (137, 152). In addition the total concentration of volatile fatty acids in the rumen fluid increased (152).

#### Influence of Dietary Lipid on Ration Digestibility

The fatty acid composition of lipids commonly added to livestock rations differs (116): Tallow is composed of (as glycerides) in percent,

palmitic 24 to 32, stearic 20 to 25, oleic 37 to 43, and myristic plus linoleic 5 to 9 acids; and corn oil contains palmitic 8 to 12, oleic 19 to 49, and linoleic 34 to 62 percent acids as glycerides.

Cattle. Kellner and Kohler [in Czerkawski, Blaxter, and Wainman (40)], as early as 1896, studied the addition of 700 g of arachis (peanut) oil to a basal ration in cattle calorimetric experiments. Digestibility of the diet and methane production were not depressed when the oil was emulsified, although the same quantity of oil not emulsified depressed both parameters. Czerkawski et al. (40) subsequently unequivocally demonstrated the depressing influence of dietary lipid on rumen methane production.

Animal fat with a melting point of about 42 C and an iodine number of 45.5 was fed to cattle by Bohman and Lesperance (20). Three treatments--0.5 lb fat, 3 lb alfalfa plus 0.5 lb fat, and 3 lb alfalfa--were added to a basal ration of grass hay. Digestibility of ether extract in the added fat rations increased ( $P < .05$ ) from 45.6 to 83.2 percent. However, added fat decreased ( $P < .05$ ) the digestibilities of organic matter, crude fiber, and nitrogen-free extract and had no effect ( $P > .05$ ) on digestibilities of gross energy, dry matter, or crude protein. Total digestible nutrients increased ( $P < .05$ ). Depression of digestibilities, except for that of crude fiber, was corrected by the addition of alfalfa. Fecal calories increased by only 0.05 kcal per g with the addition of dietary fat. It was concluded that the effect of dietary fat on ration digestibility was not the major factor limiting its utilization by cattle.



Albin and Durham (1) added 2 percent beef tallow to a 95 percent concentrate steer ration and noted that feed consumption was unchanged. However, digestion coefficients for dry matter, crude protein ( $P < .05$ ), and gross energy were lower for the tallow than for the basal ration.

Three 70 percent concentrate fattening rations were fed to steers by Esplin et al. (53). Tallow and a combination of hydrolyzed vegetable and animal fat were melted and added to alfalfa hay to give an added fat level of 4 percent of the diet. The former had an iodine number of 49.8 with 3.3 percent free fatty acids, while corresponding values for the latter were 100.4 and 67.6 percent, respectively. On a fatty acid basis, hydrolyzed vegetable and animal fat contained 77 percent  $C_{18}$  acids, compared to 62 percent for tallow. One percent dicalcium phosphate was added to each ration. Fat and tallow additions improved ( $P > .05$ ) the digestibilities of dry matter, crude protein, crude fiber, and gross energy. There were increases in ether extract ( $P < .01$ ) and gross energy ( $P < .05$ ) digestibilities and in total digestible nutrients ( $P < .05$ ) due to lipid additions. Estimates, by difference of the digestibilities of added lipids, were 93.4 percent for tallow and 91.2 percent for hydrolyzed vegetable and animal fat. An 80 percent concentrate ration (barley, oats, beet pulp, and alfalfa), with and without 7 percent added tallow, was fed to Hereford steers at the Washington Station (48). Digestibilities of dry matter and crude fiber were reduced ( $P < .05$ ) when tallow was added, although crude protein digestibility was not affected. Feed consumption of the added tallow ration was depressed by 10 percent.

Steers consumed a basal ration (grain plus 0.5 lb hay) or the basal ration plus 5 percent prime animal tallow in work reported by Roberts and McKirdy (147). Percent ether extract of the rations was 3.14 and 9.50, respectively. Both dry matter and gross energy digestion coefficients were lower ( $P>.01$ ) for the added fat ration. Ether extract digestibility was greater ( $P<.01$ ) when fat was added to the ration, and crude fat digestibility, calculated by difference, for animal tallow was 90.1 percent. This is in agreement with the value of 93.4 percent reported by Esplin et al. (53). In addition, when fat was fed there was an increase ( $P<.01$ ) in fecal gross energy ( $P<.05$ ). This was also observed by Bohman and Lesperance (20).

Page, Erwin, and Roubicek (140) studied the addition of 10 percent tallow to a 65 percent roughage ration, containing dehydrated alfalfa meal and cotton gin trash, fed to normal and vitamin A deficient steers. Digestion coefficients calculated by the chromic oxide ratio technique indicated that tallow increased ether extract and crude protein digestibilities and depressed crude fiber digestibility in the vitamin A deficient group. Nitrogen-free extract digestibility was depressed by tallow in both vitamin status groups.

Lofgreen (107) determined the net energy for production for yellow grease, bleachable tallow, and acidulated cottonseed foots by feeding them at levels of 2.5, 5.0, 7.5, and 10.0 percent of the ration to beef heifers. Net energy for production for each fat at all levels was 2.59 Mcal per kg. There were no differences ( $P>.05$ ) in the performance of animals fed the various fat levels except for a decrease in feed

required per unit of gain as the fat level increased. Animals receiving tallow consumed less feed and gained less than those fed the other two fats.

Ward et al. (177) fed cattle a mixed ration containing corn cobs and reported that digestibilities of all ration components except protein and ether extract were reduced ( $P < .01$ ) by the addition of 400 g corn oil to the basal ration. Alfalfa ash only partially counteracted the detrimental effect of corn oil.

Sheep. Typical of a number of papers dealing with fat additions to sheep rations is one by Bujisse (28), who fed a 70 percent pelleted lucerne meal, 30 percent concentrate ration with and without tallow added at levels of 1.5 to 6.0 percent. All amounts of added tallow, particularly 3.0 percent or more, depressed digestibility of crude fiber. Digestibilities of dry matter, organic matter, crude protein, and nitrogen-free extract were not significantly affected, while that of fat increased from about 48 percent for the basal ration to 78 percent for the highest fat ration.

Higher levels of fat were used by Dijkstra (46) in experiments in which lambs were fed lucerne meal alone or with 2.7, 5.5, 8.2, and 11.0 percent steamed destructor fat. Digestibilities of crude protein were reduced approximately 5 percentage units and crude fiber approximately 10 percentage units for 8.2 and 11.0 percent added fat rations. However, digestibilities of both fractions at 0, 2.7, and 5.5 percent levels of added fat were essentially equivalent.

Brethour, Sirny, and Tillman (24) studied the addition of 15 percent animal fat to a ground milo-cottonseed hull lamb ration. Amounts fed differed to maintain approximately isocaloric and isonitrogenous intakes. Fat addition to the ration resulted in decreases ( $P .05$ ) in the digestibilities of both dry and organic matter.

Cameron and Hogue (32) allotted lambs to mixed hay-grain rations containing 90, 50, or 10 percent alfalfa meal and 0 or 15 percent added corn oil. Lambs consuming 50 and 90 percent alfalfa diets containing corn oil gained as rapidly as those without corn oil. Within the oil group, lambs fed the low fiber diet gained less ( $P .05$ ) and consumed 30 percent less feed. The corn oil diets were also consumed at a rate of 30 percent less than diets without oil.

Swift et al. (161) added 3.7 or 7.1 percent corn oil to a lamb-fattening ration containing 43 percent mixed hay and having an ether extract level of about 2.2 percent. The low level of corn oil increased the digestibilities of dry matter, crude protein, ether extract, crude fiber, nitrogen-free extract, lignin, and gross energy. The high level of corn oil decreased all digestion coefficients, except that of ether extract, to values below those obtained for the basal period. Addition to the diet of the low level of corn oil (34 g) containing 319 kcals gross energy increased digestible energy by 314 kcals. Adding the second 34 g of corn oil increased digestible energy of the ration by only 83 kcals.

Davison and Woods (42) ran digestion trials with sheep to determine the effects of adding 5 percent corn oil or fatty acids to a 46 percent corn cob ration with 1 percent dicalcium phosphate. Both corn oil and the fatty acid mixture (in percent, myristic 2, palmitic 26, stearic 16, oleic 48, and linoleic 8) decreased ( $P < .01$ ) the digestibilities of dry matter, cellulose, and ash and reduced the digestibility of crude protein ( $P < .05$ ). Both lipid sources increased ( $P < .01$ ) the digestibility of ether extract, and while corn oil depressed ( $P < .01$ ) nitrogen retention, the fatty acid mixture did not. It was observed that addition of 5 percent stearic or 5 percent oleic acid to the ration decreased ( $P < .05$ ) the digestibilities of dry matter and organic matter and tended to reduce crude protein digestibility much more than did 1 percent lauric acid. Oleic acid decreased cellulose digestibility more than did lauric or stearic acids, while ash digestibility was lower for stearic and oleic acid rations than for lauric acid or control rations. Ether extract digestibility was lower ( $P < .05$ ) for the stearic acid ration than for oleic and lauric acid rations. It was apparent that the fatty acids depressed ration digestibility as much as did neutral fat.

Sheep and cattle rations without added fat may contain as much total lipid as do rations to which fat has been added, but added fat apparently depresses cellulose utilization more than does lipid, originally present in the feed--especially when alfalfa ash is not present. This might be expected, because Weenink (182) has reported that leaf lipids have a high content of galactolipids. About 60 percent of the acetone-soluble neutral lipid fraction of forages, grasses, and clovers

consists of galactesyl glyceryl esters of fatty acids, largely linolenic acid.

Several generalizations are readily made from the literature regarding lipid additions to ruminant rations. Such additions usually depressed feed consumption compared to the same ration without added lipid (32,34,48,75,76,107,119,143), although this is not to imply that there is no literature in which feed consumption remained unchanged or increased slightly when lipid was added (1,72).

Another important consideration is that live weight gains were often decreased by the addition of lipid to the ration (34,107,119,143). This was, however, attributed to decreased feed consumption, for when feed consumption was only slightly depressed or remained about the same as control rations, live weight gains were usually superior to those of cattle on control rations (72,74,75,76). Similar observations were made on the addition of corn oil to lamb rations. It was noted that high levels of oil markedly reduced feed consumption and live weight gain of animals consuming low fiber rations, while only slightly reducing them for animals fed high fiber levels (24,32).

#### Influence of Minerals on Ration Digestibility

Many workers have observed the relationship between dietary ions and the utilization of dietary fat by ruminants. The addition of minerals to ruminant rations containing added lipid has been studied in an effort to define rumen lipid metabolism and lipid digestion.

Tillman, Sirny, and MacVicar (165) reported that addition of 28 g of alfalfa ash per day to a lamb ration containing 60 percent cottonseed hulls and 2.4 percent corn oil increased ( $P < .01$ ) the digestibilities of dry matter, crude fiber, ether extract, nitrogen-free extract, organic matter, and gross energy. Crude protein digestibility was increased ( $P < .05$ ) and the digestibility of crude fiber showed the greatest response to the addition of alfalfa ash. White et al. (183) showed for a lamb ration containing 5 percent corn oil that 30 g of alfalfa ash, or 4.4 g calcium, or 4.4 g calcium plus 0.86 g phosphorus restored cellulose digestibility to normal. Nitrogen retention was increased over the basal ration level by the three mineral treatments. Calcium or calcium plus phosphorus addition to the basal ration did not alter cellulose digestibility. Phosphorus alone or a trace mineral mixture of copper, molybdenum, manganese, cobalt, iron, zinc, and boron did not restore cellulose digestibility.

Davison and Woods (44) studied the addition of calcium carbonate, calcium chloride, or magnesium carbonate to 46 percent ground corn cobs containing 5 percent corn oil. Calcium as carbonate or chloride (8 and 12 g, respective, per sheep per day) was equally effective in alleviating corn oil depression of digestibilities. Magnesium carbonate at 7 g per day succeeded only in decreasing ( $P < .05$ ) crude protein digestibility. At mineral levels of less than one-half of these amounts, only calcium chloride was effective ( $P < .05$ ) in alleviating the depressing effects of corn oil on organic matter digestibility.

In another study, Davison and Woods (43) reported that, when 5 percent corn oil was added to rations containing approximately 40 percent corn cobs or alfalfa hay, lambs did not efficiently utilize corn oil if the ration contained less than 0.3 percent calcium.

Brethour et al. (24) reported that addition of sodium or potassium bicarbonate did not improve the utilization of 15 percent added fat sheep rations. Grainger et al. (68) reported that iron partially alleviated the depressive action of corn oil on cellulose digestibility, while potassium further decreased the digestibility of cellulose. Magnesium and zinc caused sheep to go off feed.

White and his associates (183) suggested that a possible explanation of the calcium-fat relationship lies in the formation of calcium soaps. Indeed, Grainger and Stroud (69) and Grainger et al. (68) showed that corn oil decreased the apparent digestibility of calcium and increased excretion of fecal soaps by lambs. Tillman and Brethour (164) also reported that corn oil decreased calcium digestibility. However, Davison and Woods (43,44) did not find a significant change in fecal excretion of calcium when corn oil was added to a basal ration. This was also true with rations containing added calcium. However, calcium did increase the digestibility of energy in rations containing corn oil, thus suggesting that the calcium-fat relationship is not due simply to the formation and excretion of soaps.



### Digestion of Lipids

Many scientists studying lipid digestion in ruminants have attempted to describe the sequence of events undergone by lipids as they pass through the digestive tract.

Rumen Hydrolysis. Garton, Hobson, and Lough (65) first reported that rumen microorganisms could predigest fats by hydrolyzing the ester linkages between fatty acids and glycerol. They incubated linseed oil with sheep rumen contents in vitro and observed that a considerable proportion of the esterified fatty acid residues were liberated as free fatty acids, which subsequently underwent hydrogenation. Boiled rumen contents did not exhibit this lipolytic activity, nor did sheep saliva, and it was concluded that rumen microorganisms were responsible for the lipolysis. Fifty to 60 percent of lipids present in the rumen of sheep fed a variety of rations consisted of free higher fatty acids.

Anaerobic in vitro incubations of olive oil and cocoa butter by Garton, Lough, and Vioque (66) resulted in the liberation of 68 and 40 percent, respectively, of the esterified fatty acids. This suggested that hydrolysis of triglycerides by microorganisms may be related to the degree of unsaturation of their component fatty acids, the more saturated glycerides being less readily emulsified in rumen contents than the less saturated.

Despite the extensive hydrolytic activity of the rumen, no appreciable degradation of long-chain fatty acids was observed in incubation experiments with sheep rumen contents in experiments reported by

Lough and Vioque (109). In vivo work by McCarthy (113) substantiates the latter observation.

Rumen Hydrogenation. Perhaps the first unequivocal evidence of microbial hydrogenation was obtained from studies by Reiser and Reddy (146) on goats which were fed for several weeks diets containing 10 percent cottonseed oil or linseed oil. At slaughter, six hours post feeding, samples of total fatty acids in the rumen were obtained following saponification of portions of the rumen contents. Determination of iodine numbers showed that unsaturated fatty acids of the dietary oils had undergone a considerable amount of hydrogenation. This was confirmed by Ulyatt, Czerkowski, and Blaxter (170), who demonstrated that 85 percent of the double bonds of unsaturated fatty acids were hydrogenated in the rumen.

Rumen Soap Formation. Lough and Vioque (109) reported that short-chain, water soluble fatty acids were readily absorbed from the rumen but very little absorption of long-chain acids apparently occurred. A greater proportion of long-chain acids are therefore available for soap formation. Davison and Woods (44) showed that, in the presence of calcium, fatty acids from hydrolyzed neutral fats formed calcium soaps, which are insoluble and precipitate. They were therefore ineffectual in decreasing rumen microbial activity. Conversely, sodium and potassium form soluble soaps in an aqueous medium and they decreased cellulose digestion (24).

Abomasum. Results of experiments by Davison and Woods (43) showed an increase in digestible energy when calcium was added to

rations containing corn oil. For this to occur, fatty acids and calcium must dissociate to permit the absorption of fatty acids. Garton (64) measured the pH of sheep abomasal contents, finding it to be approximately 3, and reported that the solubility of calcium increased as it passed from the rumen into the abomasum. The greater acidity (due to gastric juice) in the abomasal liquor resulted in an immediate liberation of calcium. He determined soluble calcium values in rumen liquor of 0.5 mg percent when sheep consumed a ration of oats, chopped oat straw, ground maize, and blood meal, and of 18 mg percent with a chopped meadow hay diet. Corresponding calcium values for abomasal contents were 8 and 31 mg percent, respectively.

Small Intestine. Davison and Woods (44) have suggested that calcium and fatty acids pass into the small intestine in the dissociated state and that calcium is probably absorbed before the pH becomes neutral. Fatty acids are then absorbed in the presence of bile or perhaps immediately upon passing from the abomasum. Some soaps of course escape in the feces, and there is also a combination of calcium with metabolic fat (fatty acids) to form soaps in the gut (11).

Evidence of a similar nature was reported by Lennox and Garton (106), who noted that, of the lipids entering the small intestine of sheep, unesterified fatty acids, primarily stearic and isomers of  $C_{18}$  unsaturated acids, predominated. When the digesta reached the ileum, the uptake of fatty acids was nearly complete, as was the hydrolytic release of esterified fatty acids.

Theoretical Mechanisms for Lowered Digestibility of Added Fat Rations

Several attempts to explain the depression in digestibility caused by addition of lipids to ruminant diets have centered around the postulation that lipids coat particles of feed, thereby interfering with enzymatic digestion of the feed.

Coating Theory of Lowered Digestibility. Ward et al. (177) studied this theory by adding 2.4 percent (of the ration) corn oil to the 45 percent concentrate portion of the diet, with the result that little oil came into contact with the remainder of the ration, cottonseed hulls. Digestibilities of protein, fiber, and nitrogen-free extract were improved over those for the basal diet, but the differences were not significant ( $P > .05$ ). Oil additions to the entire ration reduced ( $P < .01$ ) the digestibilities of all ration components, except that of protein which was reduced at the 10 percent level of probability. Later Brethour et al. (24) conducted a similar study with sheep fed a 10 percent corn oil ration. Corn oil was mixed with the entire basal ration or with the concentrate portion of the ration only, with roughage and concentrate being fed separately. Both methods of corn oil addition decreased gains and the efficiency of feed utilization. This evidence appears to contradict that of Ward et al. (177) and does not support the concept that the effect of supplemental fat is produced by coating the fibrous portion of the ration.

The data of White et al. (183) also seem to refute this hypothesis. They utilized high roughage rations with 5 percent added corn oil fed to sheep over three successive periods. The ratio of fat to

cellulose in the ration was constant throughout the experiment. A progressive depression of cellulose digestion, rather than an initial depression with no further decrease, was reported. It was suggested that the purely physical effect (coating of feed particles) on cellulose digestion should have been eliminated by passage of the fat-coated cellulose from the rumen. However, recovery of cellulose digestibility after fat was deleted from the ration was not obtained for 10 and 17 days for the two test groups. This suggested that supplemental fat decreased microbial metabolic activity and/or modified the rumen microbial population concerned with cellulose digestion.

It was subsequently shown by Ulyatt and Czerkowski [in Czerkowski and Breckenridge (41)] that long-chain fatty acids were almost wholly adsorbed onto particles of digesta when linseed oil acids were infused into the rumens of sheep.

One might therefore conclude that, while added dietary lipids coat feed particles, this does not seem to be the primary reason for reduced ration digestibility when lipid is added to the diet.

Lipids and Microbial Metabolism. An explanation of the deleterious effects of fat and oil additions to ruminant rations may be found in the results of in vitro studies in which workers have added fat and fatty acids to microbial cultures.

Kodicek and Worden (99) have proposed the following mechanisms to explain the inhibitory action of fatty acids on microbes (p.84):

- (1) a direct chemical action upon the metabolism of the bacteria, or upon the availability of some metabolite present in the medium. . . .

(2) a physiochemical mechanism. If the unsaturated fatty acids were to form a monolayer around the bacteria it might exert its effect in at least three different ways:

- a. by changing the permeability of the adjacent surfaces
- b. by exerting some chemical influence
- c. or by altering the surface tension and so interfering with bacterial division.

The vast majority of the literature has dealt with the physiochemical mechanism hypothesis.

Fatty Acid Inhibition of Microbial Growth. Nieman (138) studied the influence of fatty acids in the normal solubility range of 5 to 50 ppm on the growth of pure strains of various organisms. The effect was either an inhibition or promotion of growth, with the effect a function of the concentration and nature of the fatty acids and of the bacterial species involved. Generally only gram-positive organisms are susceptible to the action of fatty acids in minute amounts. [The maximum proportion of gram-positive bacteria in rumen digesta occurs with animals fed high grain rations, according to Hungate (92). This may explain the greater influence of added dietary lipid on ruminants consuming high concentrate rations.] Growth inhibition was caused by both saturated and unsaturated fatty acids. Antibacterial activity of unsaturated fatty acids increased with the number of double bonds, and natural cis forms were generally more active than trans isomers. Antibacterial activity of saturated fatty acids was optimal for a chain length of 12 carbons. It was concluded that the inhibitory effect of long-chain fatty acids was best explained by the adsorption of the fatty acid to the cell surface, thereby altering the absorption and excretion powers of the cell.

Of the saturated fatty acids investigated by Hassinen, Durbin, and Bernhardt (82), only  $C_8$  and  $C_{10}$  inhibited the growth of both gram-positive and gram-negative bacteria. There was a positive correlation between the increase in bacteriostatic effect and the decrease in water solubility. With each increase of two carbon atoms from  $C_8$ , which was effective through  $C_{12}$ , the bacteriostatic effect increased on an average of 3.5 times. Molar solubility decreased about 3.4 times with each increase of two carbon atoms from  $C_8$  to  $C_{18}$ .

Eighteen carbon unsaturated fatty acids inhibited methane production by methanogenic bacteria in in vitro work reported by Demeyer and Henderickx (45). Cis-unsaturated fatty acids were much more active than either trans isomers or saturated fatty acids, and for cis isomers, toxicity increased with the number of double bonds. Both observations are in agreement with those of Nieman (138). Various esters were inactive, indicating the importance of the free carboxyl group. A physiochemical mechanism of inhibition was assumed.

Oleic, linoleic, and linolenic acids caused complete inhibition of growth and lactic acid production of Lactobacillus helveticus in work reported by Kodicek and Worden (99). Methyl esters of linoleic and linolenic acids did not show this inhibition, which is in agreement with the inactivity of various esters noted by Demeyer and Henderickx (45). Lecithin, cholesterol, calciferol, lumisterol, alpha-tocopherol, alpha-tocopherol acetate, and calcium chloride all reversed this inhibition suggesting that the action of fatty acids was bacteriostatic rather than bactericidal.

In vitro cellulose digestion studies with fatty acids added to the medium at 1.25 mg per ml were reported by Davison and Woods (42). Lauric, oleic, and stearic acids decreased cellulose digestibility, while butyric and valeric acids increased ( $P < .01$ ) it. These effects were also observed by Bentley et al. (9). Acetic and caproic acids were without effect but caprylic, capric lauric, myristic, palmitic, stearic, oleic, and linoleic acids decreased ( $P < .01$ ) cellulose digestion (42).

Evidence of a metabolic nature that substantiates the inhibitory action of long-chain fatty acids on microorganisms has come from numerous experiments. Hassinen, Durbin, and Bernhart (82) postulated that differences in the nutritional value of various fats might be due to effects of the fatty acid components on intestinal microflora. Nath et al. (136) went one step further and analyzed the cecal flora of rats on similar sucrose diets and found the number of coliform organisms decreased in the ceca of most rats fed high levels of corn oil. This group of organisms is generally believed to be, to a great extent, responsible for the intestinal synthesis of vitamins. It therefore appears that high levels of corn oil exert their effect directly on the microflora and indirectly on the animal.

Boutwell et al. (23) arrived at a similar conclusion as the result of their rat studies. They suggested that substitution of corn oil for butterfat resulted in a decreased synthesis of vitamins by intestinal flora, an explanation of the superior nutritive value of butterfat over corn oil in some rations. Mannering, Orsini, and Elvehjem (111)



demonstrated that isocaloric substitution of fat for dextrin decreased the growth of rats by lowering bacterial synthesis of riboflavin.

Bactericidal Agents. Dubos (47) reported that all fatty acids which he tested exerted a bacteriostatic effect on tubercle bacilli in a protein-free medium, with unsaturated fatty acids having the most pronounced effect. The toxicity was abolished either by esterification or by addition of crystalline serum albumin to the medium.

Stanley and Adams (156) have concluded that salts of fatty acids which function as anionic detergents are bactericidal agents primarily because of their surface-tension lowering capacity. They found that aqueous solutions of sodium salts of the most effective acids were very soapy, whereas those of the ineffective acids were not. It thus appeared that a correlation existed between bactericidal action and surface tension. That in vitro bactericidally effective acids occurred only in a form which permitted their surface-tension reducing action to be effective, i.e., as soluble salts, substantiates this hypothesis. Ethyl esters unable to form sodium salts were entirely ineffective in vitro. Although all bactericidally effective aliphatic acids were marked surface-tension depressants, this must not be regarded as the sole criterion of bactericidal effectiveness. The total acids of cod liver oil and some individual acids having 19 or more carbon atoms were found to be good surface-tension depressants but were relatively noneffective bactericides.

It was concluded that, in addition to being a good surface-tension depressant, a bactericidally effective acid must also have a

molecular weight of about 256 and contain approximately 16 carbon atoms. Below this, bactericidal and surface-tension effectiveness decreased while molecular weights above 256 caused a drop in bactericidal effectiveness usually without a drop in surface-tension effect. Some correlation between chemical structure and surface tension was revealed, although it was concluded that the aliphatic acid bactericidal effectiveness was due to a certain combination of physical properties common to all of these acids rather than to any specific chemical structure. Regardless of structure, the surface tension of solutions of sodium salts of the acids decreased with an increase in molecular weight to 19 carbons and then began to increase slightly. Double bonds had no appreciable effect on surface tension. Position of the carboxyl group in the higher fatty acids was important, because a great change in solubility was caused by moving the carboxyl group from the alpha-carbon to any other carbon atom. This may explain the lack of toxicity of various esters of saturated fatty acids observed by Demeyer and Henderickx (45), which they attributed to the lack of a free carboxyl group. Palmitic and stearic acids were without bactericidal properties while their isomers possessed high bactericidal activity. These workers summarized supporting evidence reported by Larson, in which he found that all pellicle-forming organisms ceased to grow at the surface when the surface tension of the medium was below 45 dynes per cm.

Domagk, as reported by Shelton et al. (153), observed much improved germicidal activity when a large aliphatic residue was attached to the quaternary nitrogen atom. These workers prepared simple

aliphatic quaternary ammonium bromide salts with straight-chain alkyl groups of 6 to 18 carbon atoms. Little germicidal activity was obtained when the long-chain alkyl group contained fewer than 8 carbon atoms, but as the alkyl group lengthened, germicidal activity increased substantially, reaching a maximum at 16 carbon atoms. This evidence lends support to the theory that salts of fatty acids behave similarly to the synthetic detergents.

In the same vein, salts of fatty acids may behave in a manner similar to some antibiotics. Anderson et al. (3) reported that subtilin showed surface-tension lowering effects. Subtilin is tensioactive, and amounts required for antibiotic effect are within the range of surface-tension activity.

Bactericidal Mechanisms. Valko in 1946 (172) suggested that the biochemical effects of surface-active agents may be at least partially explained as follows. Surface-active ions possess strong affinity for proteins and therefore readily combine with them. This combination results in a disturbance of the intermolecular structure of proteins by upsetting the balance of electrostatic forces and non-Coulombic cohesion in the molecule. Simultaneously, the interaction of proteins with the solvent molecules may cause significant changes, and as a result, bonds between the compounds of conjugated proteins may be disrupted. Denaturation and unfolding of protein molecules, inactivation of enzymes, and thus bacteria are the end results of these processes.

Hotchkiss (90) proposed the following mechanism to explain the bactericidal action of surface-active agents. The first stage of the surface-active agent-bacteria interaction is pictured as a combination

of surface-active ions with oppositely charged sites on the bacterial surface. This process may be prevented, or perhaps reversed, through competition of ions such as phosphatides, other detergents, hydrogen, and hydroxyl ions. If the surface-active agent's hydrophobic groups have the required affinity for the bacterial surface, adsorption of a small amount will result in irreversible damage to the cell membrane. The bacterial cell is now unable to repair itself and begins to autolyze, with cell constituents undergoing enzymatic degradation and nitrogen and phosphorus compounds being released from the cell in increasing quantities. Low concentrations of surface-active agents apparently kill bacteria only when they initiate this sequence of changes.

Baker, Harrison, and Miller (7) are in agreement with Valko (172) and Hotchkiss (90) with regard to the twofold action of synthetic detergents on bacterial metabolism. First, a disorganization of the cell membrane occurs due to high surface activity of the detergent, and secondly, there is a denaturation of proteins essential to metabolism and growth.

Certain surface-active compounds such as phospholipids are able to markedly modify the activity of detergents (7). Phospholipids (lecithin, cephalin, and spingomyelin) possess a polar-nonpolar structure and presumably have an affinity similar to that of detergents for bacterial cells. Since phospholipids, even at high concentrations, do not inhibit bacterial metabolism, they could protect the cell by altering the membrane structure to prevent penetration by detergents. Baker et al. (7) demonstrated that phospholipids were ineffective unless added before or simultaneously with the detergent.

Another explanation of the negligible activity of the salts of fatty acids in vivo appears to lie in their ready adsorption by protein (10). Addition of very small amounts of blood plasma to the medium increased the activity of bacilli four to eight times. This is in agreement with the observation by Dubos (47) that the microbial toxicity of fatty acids was abolished by addition of crystalline serum albumin to the medium.

#### Absorption of Lipids From the Gut

Certainly less is known about digestion of lipids in the ruminant intestine, or their absorption, than is the case for other farm mammals. For example, Itoh and Kayashima (94) were unable to find gastric lipase in the cow. No lipolytic activity toward olive oil was found in extracts of the upper jejunum, though, according to Uchino and Mori (169), an esterase capable of hydrolyzing triacetin was present. However, Koref and Munoz (100) found pancreatic lipase in increasing amounts in the pancreas of calf embryos, suckled calves, and mature cows.

Fatty Acid-Absorption Studies. Assimilation of lipids from the small intestine was examined by Felinski et al. (54). It was observed with sheep fed 700 g of a mixed diet, that about 25 g of lipid were transported from the intestine via the lymph in 24 hours. As with other mammals, this chylomicron lipid consisted of 70 to 80 percent triglycerides and 15 to 20 percent phospholipids, in addition to small amounts of sterols, free fatty acids, and sterol esters. The daily amount of stearic acid transported ranged from 5.6 to 7.4 g. In another absorption

study, Heath and Hill (84) recently reported no differences ( $P > .01$ ) in the absorption of oleic (92 percent), palmitic (93 percent), and stearic (87 percent) acids from sheep intestine. It therefore appears that stearic acid is readily absorbed by the sheep, although this is not the case for swine. Howard et al. (91) fed pigs diets containing 10 percent beef tallow or 10 percent maize oil and noted the apparent digestibilities of lipid were respectively 44 and 79 percent.

On the other hand there is no reason to suppose that, in general, absorption of long-chain fatty acids from the small intestine of the ruminant follows a pattern different from that in other mammals. Evidence of a more empirical nature, reported by Moore, Noble, and Steele (130), lends credence to this theory. After collecting data from the abomasal infusion of linoleic and linolenic acids with sheep, they suggested that there was no reason to suppose the mechanisms of digestion and absorption of the infused triglycerides were different from those in monogastric animals. Supporting data were also presented by Heath and Morris (85) resulting from their studies with sheep and lambs on the role of bile and pancreatic juice in lipid digestion and absorption. Intestinal lymph duct cannulae and chronic fistulae of the bile and pancreatic ducts were employed. In both sheep and lambs the lipid content of intestinal lymph fell to low values when the animals were deprived of bile or pancreatic juice. It was shown that the return of bile to the intestine of a lamb with normal pancreatic secretion led to  $^{14}\text{C}$ -activity from labeled tripalmitin appearing quickly in the lipids of intestinal lymph.

It has been suggested by Holmes and Deuel (89) that an inverse relationship exists between the coefficient of digestibility and the melting point of fats. The critical temperature above which there is a marked decrease in digestibility in man appears to be about 50 C. Fat digestibility in rats is similarly affected. This was studied by Crockett and Deuel (39) using bland lard and two hydrogenated lards with melting points of 48 C, 55 C, and 61 C, respectively. The coefficient of digestibility for bland lard was 94.3 percent, while those for low and high melting point hydrogenated lards were 63.2 and 21.0 percent. It was concluded that reduced digestibility of the hydrogenated lards was not due to a failure of lipolysis but to the animals' inability to absorb the resulting palmitate and stearate. This hypothesis was supported by the observation that the quantity of fatty acids set free by hydrolysis far exceeded that required to produce a satisfactory emulsion. It was postulated that emulsification was incomplete with high melting fats or that the fat particles may have been too large to be absorbed through the intestinal mucosa. In related work, Steenbock, Irwin, and Weber (158) observed that more hydrogenated fats were less quickly absorbed by the rat than were less saturated fats. Studies by Augur, Rollman, and Deuel (6) indicated that lecithin additions to the diet of rats greatly enhanced the absorption of fats, particularly those with higher melting points.

Micelle Formation. Hofmann and Borgstrom (88) determined the physical-chemical state of lipid in the intestinal lumen during fat digestion to be of a micellar phase, containing principally the products of lipolysis solubilized in a bile salt solution, in continuous

equilibrium with an emulsified oil phase. Interspecies differences were shown to exist in lipid composition of the two phases as a result of the extent of lipolysis characteristic of each species. The micellar phase in the pig was composed primarily of fatty acids and monoglycerides, according to Freeman et al. (61). In sheep, because of the extensive lipolysis (65) and hydrogenation (146) that occur in the rumen, micellar lipid was largely free fatty acids together with lysolecithin (104). Fatty acids were the major components of the micellar phase of both species and, because of the role of micellar lipid in fat absorption, were therefore the principal form in which lipid was absorbed.

In swine, the uptake by the small intestine of fatty acids from a mixed micellar solution was shown to be nonspecific (61), but in the mucosal cell the rate at which fatty acids were incorporated into triglycerides was very specific, according to Freeman (60). This step was not rate-limiting and did not influence the rate of fatty acid removal from the lumen. Transport of triglycerides out of the cell as chylomicrons may limit the rate of fat absorption, but it is unlikely that it is fatty acid specific. Thus, the luminal oil phase-bile salt solution equilibrium, especially the rate and extent of formation of mixed micelles, is important in controlling the absorption of fatty acids. Fatty acids establish an equilibrium between the two phases in a rapid and apparently nonspecific manner, and therefore the extent to which various fatty acids can be solubilized in the micellar phase becomes the key question.



Fatty acids exhibited marked differences in their solubility properties in bile salt solutions. Palmitic and stearic acids behaved as typical nonpolar solutes, and the low saturation ratios of these acids and their high saturation values at various bile salt concentrations were patently two factors which limited their absorption. It was noted that pH was capable of influencing micellar capacity. Also, the concentration of calcium ions in the intestinal contents of both sheep and pigs was considerably above the threshold to restrict distribution of stearic acid in the micellar phase. However, the pH of the sheep duodenum was lower than that for the pig, suggesting that the interaction of calcium ions with stearic acid was less pronounced in the sheep than in the pig. The properties of trans-fatty acids, which are produced as a result of the hydrogenation of unsaturated fatty acids in the rumen, apparently did not present a barrier to their efficient absorption.

Other than solubility, the most important factor influencing the micellar capacity of fatty acids such as palmitic and stearic was the type of amphiphiles in the dispersion. Amphiphiles present in the intestinal lumen of the pig were largely of exogenous origin, derived from monoglycerides and polar fatty acids with lysolecithin playing a minor role in micelle formation (61). In contrast, there was a great dearth of exogenous amphiphile in the sheep, and of the phospholipid passing along the tract, 60 to 85 percent was accounted for by lysolecithin (104). It was apparently derived in situ from the action of phospholipase secreted in pancreatic juice on biliary lecithin (103).

It was suggested that in the ruminant, lysolecithin replaces the function of monoglyceride in micelle formation in the monogastric animal. The efficient absorption of stearic acid by the sheep indicated an adequate supply of this highly effective amphiphile (54). Lysolecithin was effective in increasing the solubilization of long-chain saturated fatty acids and increased the solubilization of stearic acid into the micellar phase.

The foregoing appears to be a reasonable explanation of the superior ability of nonruminating calves (70) and lambs (22) to utilize dietary animal fats, and perhaps of the greater effectiveness of dietary animal fats versus vegetable oils for the young ruminant.

#### Summary

It is possible to make a number of generalizations from the literature concerning the influences of added dietary lipid on ruminant performance, ration digestibility, and rumen mineral and microbial metabolism.

Added dietary lipid, except at very low levels, depresses the digestibility of crude fiber most drastically of the ration components, followed by nitrogen-free extract. Crude protein digestibility is often slightly enhanced, while ether extract digestibility is significantly increased. The digestibility of gross energy and total digestible nutrients is often greater or at least remains unchanged if the level of dietary lipid added is not excessively high.

Experiments with sheep have indicated that the depression of ration component digestibility is alleviated by the addition of a

significant proportion of alfalfa to the diet. Alfalfa ash and calcium are effective as is alfalfa hay in this respect.

The literature indicates sheep are better able to utilize high levels of dietary fat than are cattle. Low levels--4 or 5 percent--of added lipid often enhance daily liveweight gain and reduce the amount of feed required per unit of gain. Lipid additions to ruminant rations usually depress feed consumption compared to the same ration without added fat, and if this is of sufficient magnitude, daily gains will also be reduced. It has been noted that high fat, low crude fiber rations are less well utilized than are high fat, high crude fiber rations.

Added dietary lipid may increase total ruminal volatile fatty acid concentration and shift the concentration in favor of propanoic acid, while slightly reducing the pH of the rumen contents. In addition, dietary lipid reduces the quantity of methane production by ruminants.

It has become apparent that dietary lipid increases the percent fecal soaps. These are not accounted for by the ether extract technique, and the digestibility of crude fat determined by this method is not accurate.

The action of dietary fat with regard to its lowering of digestibility is more than a physical coating of feed particles. More important, added fat influences microbial metabolism. Fats are hydrolyzed and hydrogenated to saturated fatty acids by microorganisms in the rumen. These fatty acids may complex with various ions present in the rumen to form soaps, or they may pass unchanged into the small intestine. Some

ions, for example, calcium, form inactive soaps which are precipitated, while others, such as potassium, form soluble soaps which act as bactericidal agents by functioning as detergents. Unsaturated fatty acids are more active in this respect than are saturated fatty acids.

One of the important mechanisms of this bactericidal activity is that of lowering the surface tension of solutions. In this respect, longer-chain fatty acids are more effective than short-chain fatty acids, which are devoid of bactericidal properties and may even promote bacterial growth. It is believed that surface-active agents complex with protein of the bacterial cell wall, causing disorganization of the cell membrane, followed by denaturation of proteins essential to metabolism and growth. In this regard, it is known that added dietary fats can inhibit the production of B vitamins by microbes, and microbial cells in the presence of active soaps lose cell constituents and die.

A relationship is known to exist between the coefficient of digestibility and the chain length and saturation of fatty acids. This results from the equilibrium existing in the intestinal lumen between the micellar and emulsified oil phases and thus the rate at which various fatty acids can be solubilized in the micellar phase for absorption by the gut mucosa. Another factor influencing the micellar capacity of fatty acids such as palmitic and stearic is the type of amphiphile present in the gut. The ruminant has an endogenous source of the very effective amphiphile lysolecithin, which allows efficient absorption of these two fatty acids. This is the the case for monogastric animals.

Effect of Feed Intake Level  
on Ration Digestibility

One of the many factors that potentially influence the ultimate value of a feedstuff for ruminant animals is the declining nutritive value of the diet as the level of feed intake increases (11). This depression in nutritive value could be the result of either a reduction in true digestibility or an increased loss of absorbed material as heat, or a combination of the two (127). Experimentally it has been observed to be the result of a reduction in digestibility. Reid (144), for example, suggested that the major reason for increased gross energy requirements per unit of milk for high producing cows is a decreased digestibility of the diet when fed in amounts sufficient to support high milk production.

The literature regarding level of feed intake versus digestibility is about equally divided between cattle and sheep. Before this information can be meaningfully united, any differences in the digestive powers of these two species must be noted. To define these differences Cipolloni et al. (35) analyzed the results of 1,912 digestion trials and found that for roughages, apparent organic matter digestion coefficients were 3 percentage units higher for cattle. Sheep tended to digest concentrate feeds to a greater extent than did cattle. Several workers (16,17,178) have reported results similar to these. In general, absolute differences between the two species remained small, and both responded in a similar manner to various levels of feed intake.

### Dry Roughages and Silages

Several workers have reported that increasing levels of roughage intake did not depress digestibility by herbivorous animals (55,67,71, 179). In general, however, these experiments have been marred by less than adequate design and lack sufficient valid data from which satisfactory conclusions can be drawn.

Generally, the digestibility of forages--long, ground, or ground and pelleted--decreases with increasing levels of intake (5,11,12,13,16, 33,141,175). Blaxter and Graham (13) observed that the extent to which digestibility was depressed appeared to be related to fineness of grind of the forage. They fed sheep either dried grass that had been coarsely chopped, or one of the two cube types made from dried grass and ground to pass through 1/4 or 1/16-inch sieves. Feeding was at the rate of 600 and 1500 g per day. Apparent gross energy digestibilities were decreased by 3.5, 5.2, and 9.8 percentage units for the chopped, medium, and fine ground rations, respectively, at the high level of intake. The quantity of food and its physical form had a marked effect ( $P < .05$ ) on the digestibility of all ration components except ether extract. Crude fiber digestibility was depressed by 40 relative percent, while the digestibility of nitrogen-free extractives fell by only 12 relative percent. This was attributed to a more rapid rate of passage of the finely ground feed. Both Armstrong (5) and Blaxter (11) have reported similar results with respect to fineness of grind of roughages.

Several workers have reported on studies in which digestibility, associated with level of intake, appeared to be related to forage

quality. Armstrong (5), Blaxter (11), and Waite, Johnson, and Armstrong (175) noted that the rate of fall of energy digestibility with increasing levels of feed intake was greater for feedstuffs of poorer digestibility (at the maintenance level of nutrition) than it was for forage of high quality. However, Blaxter (11) is of the opinion that this is due to the fact that a unit increase in nutritional level necessitates a greater increase in the dry food intake for those animals consuming the poorer ration.

Experimental evidence regarding the effect of level of intake on silage digestibility is limited. In 1897, Jordan and Jenter (95) reported a decline in digestibilities when the level of a corn silage-hay mixture fed was doubled to ad libitum amounts. Watson et al. (181) observed that when corn silage was fed to steers at levels of 8.0 kg per day to ad libitum, apparent digestibilities of dry matter, organic matter, crude fiber, and nitrogen-free extract progressively decreased ( $P < .01$ ) with increasing levels of intake. Plane of nutrition did not affect ( $P > .05$ ) nitrogen and ether extract digestibilities.

#### Mixed Rations

Results of many detailed experiments have demonstrated that the digestibilities of rations composed of both concentrates and forages are depressed as the level of consumption by ruminants increases. According to Reid (144), "Though the degree of effect varies from ration to ration, increasing inputs of feed result in an ever decreasing nutritive value (e.g., TDN) per unit of feed ingested. . ." (p. 84).

Blaxter (11) reported the results of 21 experiments involving a total of 194 determinations of digestibility and noted that an increase in feeding level resulted in a decrease in digestibility. As early as 1911 Eckles (49) reported that a cow fed a low-concentrate mixed ration at maintenance digested 73.8 percent of the entire ration, while the same cow, receiving a liberal quantity of the ration, digested it only to the extent of 66.3 percent. A second cow consuming 50 percent less feed under similar circumstances digested 72.2 and 67.0 percent of the ration, respectively. Generally, crude protein, crude fiber, nitrogen-free extract, and crude fat were digested to a greater extent at the maintenance level of feeding. The same worker in 1913 (50) reported that cows under essentially the same regimen digested 71.2 percent of the ration at the maintenance level of nutrition and 65.6 percent at higher levels of feed consumption. He concluded that digestion coefficients determined at maintenance levels are not applicable to cows in heavy milk production consuming large quantities of feed.

Data supporting this conclusion were reported from many sources. Schneider and Ellenberger (151) in 1927 indicated that reducing the quantity of low concentrate ration fed because of a decline in milk production apparently increased the digestibility of all nutrients with the exception of ether extract. The greatest increase, 16 percentage units, occurred with crude fiber, while digestibilities of dry matter and nitrogen-free extract increased over 4 percentage units.

Slight decreases in the digestibilities of rations consisting of alfalfa hay wafers and 16 or 37 percent of a corn-soybean meal mixture



occurred when the level of feed intake changed from maintenance to 2 to 3 times maintenance (56). Blaxter and Wainman (15) fed a ration in the proportion of 2 to 1, hay to oats, to sheep and cattle at levels of from 1 to 3 times the maintenance nutritional level. For both species, the percent food energy lost as feces increased as the feeding level increased. For 1, 2, and 3 times maintenance, the absorbed energy for steers and sheep were, respectively, 61.1, 60.9; 59.7, 60.2; 58.3, 59.5 in kcal per 100 kcal ingested. A pelleted mixture of 55 percent hay and 45 percent corn meal was given to sheep at three nutritional levels--maintenance, ad libitum, and intermediate intakes--in work by Paladines et al. (141). Fecal energy loss as a percentage of gross energy intake increased ( $P < .01$ ) as the level of intake increased. Forbes, Braman, and Kriss in 1928 (57) fed steers a ration of equivalent quantities of alfalfa hay and corn meal at five planes of nutrition as multiples of maintenance--0, 0.5, 1.0, 1.5, and 2.0. They reported a decrease in digestible energy at an increasing rate, after an initial increase, as a result of lowered digestion of carbohydrate and protein.

Moe, Tyrrell, and Reid (128) reported total digestible nutrient values for rations composed of concentrates and either early- or late-cut hay. They employed various levels of intake as percentages of the total digestible nutrients determined at the maintenance plane of nutrition. More than 60 observations with cows producing from 0 to 120 pounds of milk per day indicated that, as the level of food consumption increased from 1 to 6 times maintenance, the relative total digestible

nutrient value decreased from 100 to 77 percent. Concentrate to roughage ratios of the rations were not reported.

Truter and Louw (167) observed that 82.4 percent of the dry matter of equal parts lucerne hay and crushed maize was digested by sheep at maintenance levels, but this decreased ( $P < .05$ ) to 79.9 percent at 1.5 times maintenance. Increasing the plane of nutrition to 2.0 times maintenance did not bring about a further decrease ( $P .05$ ) in digestibility. Coefficients of digestibility of cellulose and ether extract also decreased ( $P < .05$ ) as the plane of nutrition increased to 1.5 times maintenance, while crude protein digestibility declined ( $P < .01$ ) 5.2 percentage units over the entire range. Watson et al. (180) also found that crude protein digestibility was depressed to a greater extent than was any other ration constituent.

The energy metabolism of steers was studied at seven planes of nutrition, in multiples of maintenances--0, 0.5, 1.0, 1.5, 2.0, 2.5, and 3.0--using a ration consisting of equal proportions of alfalfa and corn meal (58). As feed consumption increased from 1.0 to 3.0 times maintenance, there was a slight but continuous drop in digestible energy. This decrease, as also reported in a 1928 paper by Forbes et al. (57), was due mainly to the nitrogen-free extract and crude protein fractions. Marston (112) used a cubed ration consisting of wheat, lucerne hay, and cane molasses in the ratio of 5:4:1 by weight fed to sheep at 0.5, 1.0, 1.5, and 2.0 multiples of the maintenance level of nutrition. Dietary digestible energy progressively decreased from 83.6 percent at the lowest level of intake to 79.8 percent at the highest level.

Rations containing alfalfa and corn or concentrates in the proportions of 1:1 and 1:2 were fed to cattle at six levels of nutrition: 0.5, 1.0, 1.5, 2.0, 2.5, and 3.0 times maintenance in work reported by Forbes et al. (59). They reported that cows consuming 2.72 times the maintenance requirement of nutrition digested all constituents of the ration less efficiently than did cows at 1.38 times maintenance. Dry matter digestion coefficients for steers fed the six planes of nutrition, from low to high, were 75.4, 76.1, 74.7, 73.0, 72.1, and 69.3. Organic matter, crude protein, crude fiber, and gross energy digestibilities followed the same trend, while ether extract and nitrogen-free extract digestibilities showed a steady decline. Mitchell and Hamilton (126) used a ration containing 73 percent ground corn and 24 percent alfalfa hay fed to steers at full feed (2.2 percent of body weight),  $4/5$ ,  $3/5$ ,  $2/5$ , and  $1/5$  that amount to study digestibility. The lowest level of feeding was associated with the most complete digestibility of all nutrients. Nitrogen-free extract, ether extract, and dry matter digestibilities responded to increasing levels of feed intake by progressively decreasing. Crude protein and crude fiber tended to give higher digestibilities at lower feed intakes.

In an extensive study by Leaver, Campling, and Holmes (105) sheep were fed rations consisting of 1:1 and 1:4 ratios of hay to concentrates in amounts increasing by 200 g from 600 to 1400 g dry matter per day. For both diets, as the level of feeding increased there was a significant decline ( $P < .001$ ) in organic matter and crude fiber digestibilities, 1.3 and 5.9 percentage units greater, respectively, for the high concentrate

ration. The depression in digestibility of the crude-fiber components of the low and high concentrate diets accounted for only 39 and 29 percent, respectively, of the decline in organic matter digestibility. Thus, part of the decrease in digestibility must have been due to reduced digestibility of nonstructural components of the diet. The mean retention time of food in the alimentary tract at the high level of feeding was 60 percent of that at the low level for both diets. The relationships between level of feeding and organic matter or crude fiber digestibilities were linear ( $P < .01$ ) for the low concentrate diet and curvilinear ( $P < .05$ ) for the high concentrate diet. Greater depressions in digestibility occurred as the proportion of concentrates in the diet increased, the decline in organic matter digestibility per increment of maintenance level being 4.8 and 5.5 percentage units, respectively, for the low and high concentrate rations. This finding is in general agreement with that of Brown (27). Curvilinear relationships have also been shown in the data of Forbes et al. (57) and Blaxter and Wainman (15).

Bloom et al. (19) conducted digestibility studies with lactating cows using four ratios of hay to grain: 75:25, 55:45, 35:65, and 15:85 at high, medium, and low levels of feeding. Average dry matter digestibilities for high, medium, and low feeding levels were 57.6, 57.8, and 58.2 percent, but there were greater differences in dry matter digestibilities between high and low planes of nutrition on the two highest concentrate rations than on the two lowest concentrate rations. Crude protein digestion coefficients tended to be greater at the lower planes of nutrition. In a similar study, Brown (27) fed cattle diets having

ratios of concentrates to hay of 4:1, 2:1, and 1:4. Feeding levels ranged from approximately maintenance to 5 times maintenance for the 4:1 ratio, from maintenance to 3.5 times maintenance for the 2:1 ratio, and from maintenance to 2.8 times maintenance for the 1:4 ratio. The observed mean coefficients of dry matter digestibility were 63.9 and 61.6, respectively, for low and high feeding levels of the 1:4 ratio ration. For the 2:1 grain to hay ratio dry matter digestibilities were 72.5, 68.7, and 67.3 for the low to high planes of nutrition. Calculated dry matter digestibilities for the 4:1 ration decreased from 78.1 percent at the maintenance level to 62.7 percent at 5 times the maintenance level of nutrition. Regression coefficients indicated that dry matter digestibility decreased 3.84 and 2.02 percentage units with each increase in feeding level of one unit of maintenance, respectively, for 4:1 and 2:1 grain to hay rations. Nevertheless, definite conclusions could not be drawn concerning the influence of grain level on the depression in digestibility associated with feeding level. Although both regression coefficients were significantly different from zero, they were not significantly different from each other.

Sheep and cattle were fed mixtures of hay and maize from 100 percent hay and 0 percent maize to 100 percent maize and 0 percent hay in 20 percent increments in work reported by Blaxter and Wainman (16). Feeding all rations to both species at a nutritional level of about 2 times maintenance resulted in depressed gross energy digestibility ( $P < .05$ ) and nitrogen digestibility over the same rations fed at one-half that amount. The decline in digestibility resulting from an

increased plane of nutrition did not appear to be related to the percent concentrate in the ration for either species.

Contrary to the bulk of the literature, a number of reports have indicated little or no depression in digestibilities of mixed rations with increasing levels of intake. Almost without exception the conclusions drawn from these experiments can be seriously questioned on the basis of the small range of levels of nutrition or levels of feed intake studied.

Kleiber, Goss, and Guilbert (97) fed heifers a ration consisting of about equal proportions of alfalfa, molasses meal, dried beet pulp, and corn starch at 0.5, 1.0, and 1.5 times the maintenance level of nutrition. Energy digestion coefficients were, respectively, 80.6, 81.4, and 80.5. In another study, sheep weighing about 55 kg were fed 300, 600, 900, and 1200 g per day of a 5:4:1 mixture of lucerne hay, maize meal, and peanut meal (67). Between the low and high feeding levels, digestibilities declined by the following percentage units: dry matter, 1; gross energy, 2; and crude protein, 1. However, crude fiber and ether extract digestibilities increased 3 percentage units.

Andersen et al. (2) reported a variable effect of level of intake upon the digestibility of mixed rations consisting of 80 percent concentrates and 20 percent hay. In separate experiments, levels of intake ranged from 1.1 to 2.1 and from 0.6 to 3.1 times the maintenance level of nutrition, while in a third trial a ration containing 60 percent of the same concentrate mixture and 40 percent of the same hay was fed at levels of 1.1 to 1.9 times the maintenance requirement. There was

no effect of level of intake on the digestibility of dry matter in the first trial, whereas increasing intakes markedly depressed dry matter digestibility, respectively, from 85.7 to 74.3 percent for the 0.5 and 2.7 times maintenance levels in the second trial. In a third trial, dry matter digestibility was 78 percent at the low level of feed consumption and 69 percent at the highest level. In another experiment, eight rations composed of 0, 25, 50, or 75 percent of a concentrate mixture and 100, 75, 50, or 25 percent, respectively, of each of two hays were fed to steers. Feed consumption of the early-cut hay rations ranged from 1.0 to 2.5 times the maintenance level, while the late-cut hay diets were consumed at levels from 1.0 to 1.9 times that level. The level of intake did not affect ( $P>.05$ ) digestible energy values of rations containing either hay.

Mumford et al. (135) fed rations of clover hay and ground corn in ratios of 1:1 to 1:5 to steers at the following planes of nutrition: maintenance, ad libitum and maintenance plus 1/3 or 2/3 the difference between maintenance and ad libitum feed intakes. Dry matter, carbohydrate, and crude protein digestibilities decreased with increasing feed intake. This was especially pronounced with the 1:1 ration, but was less apparent at higher levels of feed intake with the 1:3 and 1:5 rations. Digestion coefficients of none of the nutrients were affected by plane of nutrition when the ration was composed of hay, ground corn, and linseed meal in the ratio of 1:4:1. Conflicting evidence is offered in the following two experiments by Lassiter, Huffman, and Duncan (101). Three alfalfa hay to grain ratios (80:20, 50:50, and 20:80) were fed to

nonlactating cows at three levels of feed intake (70, 100, and 130 percent) with the highest level equal to the maximum level of feed intake. Dry matter, crude protein, and crude fiber digestibilities increased ( $P < .01$ ) respectively, 4, 18, and 4 percentage units as the level of feed intake increased from the minimum to maximum level. In a second, essentially duplicated experiment (102), the same hay to grain ratios were used, with 70, 100, and 130 percent levels of feed intake in which the medium level was equivalent to 24 pounds of feed per day. As the level of feed intake increased, there was a definite trend toward higher digestion coefficients. Digestibility differences between the 70 and 130 percent levels of feed consumption were about 6 percentage units for dry matter, crude protein, crude fiber, and organic matter. This trend was most pronounced with crude fiber digestion and with the 20:80 hay to grain ratio ration.

#### Submaintenance Digestibilities

With some diets, a parabolic response has been observed in which digestibility of the submaintenance ration is lower than that of the same diet consumed at the maintenance level of nutrition.

This phenomenon was observed by Blaxter and Graham (12) when sheep were fed dried grass at the rate of 2000 or 2900 kcal per day. They responded with an increase in energy digestibility, respectively, from 59.7 to 62.0 percent. Hale, Duncan, and Huffman (71) observed that when cows were fed 10, 20, or 30 lb of alfalfa hay per day, the apparent digestibilities of dry matter, crude protein, and crude fiber were, respectively, about 9, 4, and 9 percentage units lower for the 10 than 20



lb ration. Forbes, Braman, and Kriss (57,58) reported that steers fed equal proportions of alfalfa hay and corn meal responded to an increase in plane of nutrition from 0.5 to 1.0 times maintenance by increasing the percent gross energy digestibility. This was due mainly to the increased digestibility of crude fiber but partly to that of protein also.

Kleiber, Goss, and Guilbert (97) observed that the digestible energy of a ration consisting of about equal proportions of alfalfa, molasses meal, dried beet pulp, and corn starch increased 0.8 absolute percent when the plane of nutrition was raised from 0.5 to 1.0 times the maintenance level.

#### Metabolizable Energy Versus Digestible Energy

Of the three energy losses--urinary, fecal, and methane--considered in the determination of metabolizable energy, fecal energy losses are by far the most important in determining the nutritive value of foodstuffs (11). Thus, digestible energy and metabolizable energy of a foodstuff, which differ by methane and urinary energies, are generally highly correlated. However, it has been observed that the decrease in apparent digestibility of energy at higher levels of intake is compensated for to some extent by a concomitant decrease in energy losses as urine and methane (16,56). If energy losses as urine and methane entirely compensated for the decrease in apparent digestibility, it is apparent that the metabolizable energy content of a ration, when expressed as a percent of the ration's gross energy content, would remain constant

regardless of level of intake. In this case, then, the depression of digestibility caused by high feed intakes is really more of an academic problem than it is a practical one (142). That this does not occur with all rations is shown by the work of Blaxter and Graham (13) who fed sheep 600 and 1500 g of dried grass, coarsely chopped or ground through a medium or fine screen, and noted the compensation amounted to only about 1 to 3 percentage units when the apparent digestibility depression ranged from 4 to 10 percentage units, respectively.

Flatt (56) fed 37 and 16 percent concentrate rations containing corn, soybean meal, and alfalfa at 1, 2, and 3 times the maintenance level of nutrition and found only slight decreases in digestibility. However, decreases in methane and urine energies were completely compensatory, allowing metabolizable energy expressed as a percent of gross energy to remain almost constant regardless of the level of feed intake.

Blaxter and Wainman (16) conducted an extensive study with sheep and cattle fed mixtures of hay and maize from 100 percent hay and 0 percent maize to 100 percent maize and 0 percent hay at levels of 1 and 2 times the maintenance requirement. The decrease in energy digestibility associated with feeding level was approximately 2 to 5 percentage units for the 60 percent and higher maize rations consumed by both cattle and sheep. At these higher proportions of maize, urine and methane energy decreased sufficiently to permit metabolizable energy, when expressed as a percent of gross energy, to remain unaffected by level of feeding. However, the percentage of metabolizable energy decreased with increased

feeding levels with diets containing high levels of hay. This decrease amounted to 4 percentage units on the all-hay rations.

### Summary

The effect of feeding level on apparent digestibility appeared to be somewhat variable from station to station, as well as between trials within stations. However, most data indicated some depression in apparent digestibility as level of intake increased.

With all forage rations, the depression in digestibility with increasing levels of intake was greater with forages of poor quality than good quality and was more pronounced when forages were finely ground and pelleted than when fed in the long or chopped form. Apparently the digestibility of crude fiber was depressed more than was that of the nitrogen-free extractives. This supports the observation that, at higher levels of intake, rate of passage through the digestive tract was faster than at lower feed intakes. A progressive decline in the digestibilities of ration components also occurred with silage rations.

The depression in digestibility of mixed rations associated with increased feeding levels appears to be influenced by factors other than feeding level alone. Generally, crude fiber, of all the ration components, exhibited the greatest decline in digestibility with increased feed intake, but digestibilities of nonstructural components also declined. Greater depressions in digestibility may have occurred as the proportion of concentrates in the diet increased. The relationships between level of feeding and organic matter and crude fiber digestibilities were linear for low concentrate rations and curvilinear for high concentrate

rations. A number of reports indicated little or no depression in digestibility of mixed rations with increasing levels of intake. However, almost invariably conclusions drawn from these experiments can be seriously questioned because of the small range in level of feed intake employed.

With some rations, a parabolic response was observed in which the digestibility of the submaintenance ration was lower than that of the same diet consumed at the maintenance level of nutrition.

Some data indicated that metabolizable energy (as a percent of gross energy) did not decline nearly as much as did digestible energy with increased levels of feed intake.

Metabolic Fecal Nitrogen and Digestibility  
Determinations With Respect to  
Dietary Nitrogen Levels

Nitrogenous compounds excreted in the feces consist in part of undigested or unabsorbed food nitrogen and in part of metabolic fecal nitrogen. The latter includes (a) endogenous catabolism and (b) alimentary-lining nitrogen, derived from the lining of the gut as a result of digestive processes (25). Included in metabolic nitrogen of the feces is nitrogen originating from a variety of sources--epithelial cells, bacteria, mucus, and residues from bile and digestive juices (150). Under conditions of adequate, above maintenance feeding, digestive processes are evidently the dominant sources, according to Mitchell and Bert (124). Actually the term "metabolic products" was introduced as early as 1884 by workers in the laboratory of Carl Voit (in 145) to designate that fraction of the fecal matter not traceable to food. The

existence of fecal metabolic nitrogen, as distinguished from undigested nitrogen, was reported by both Thomas (163) and Mitchell (120) who observed that feces excreted on a nitrogen-free diet always contained nitrogen compounds.

In the large intestine metabolic fecal products, whatever their origin, serve as nutrient media for resident bacteria, which may make up a predominant part of the fecal nitrogen and fecal dry matter on low-residue diets consumed by men (110). Osborne and Mendel (139) reported that bacteria made up about one-third of the weight of rat feces. Metabolic fecal nitrogen is not a biochemical entity that can be chemically assayed (38). Various means have been devised to separate food residue nitrogen from metabolic nitrogen in the feces in attempts to estimate the true digestibility of dietary protein. The several chemical methods used to distinguish nitrogen from these two sources were based upon assumptions concerning the solubility of part of the fecal nitrogen in various reagents, for which no adequate support may be found (150).

However, two in vivo methods are in common use for the determination of metabolic fecal nitrogen. The fecal nitrogen excretion of animals fed a nitrogen-free diet can be determined directly and has been used by some workers, including Mitchell and Carman (125) as a metabolic fecal nitrogen value.

A second method is often attributed to Titus (166). He determined the digestibility of alfalfa protein by varying the protein content of the ration while total food consumption remained constant. His work indicated that, within the range of nitrogen intake investigated

(5.1 to 13.6 percent ration crude protein), there was a linear relationship between fecal nitrogen, feces corrected to an 80 percent water content, and feed nitrogen. However, contrary to popular belief, he refused to accept the extrapolation of this regression line to the point of zero nitrogen intake as the value for metabolic fecal nitrogen, as was subsequently done by Bosshardt and Barnes (21) and Bell et al. (8).

#### Influence of Protein Level on Ration Digestibility

That increased levels of dietary natural protein result in improved ration digestibility by ruminants appears well established. This is substantiated by digestibility studies with sheep and cattle consuming both roughage and mixed diets.

Woods, Gallup, and Tillman (185) formulated semi-purified lamb rations in which various oil meals supplied over 90 percent of the total nitrogen. With but few exceptions, there was a progressive increase in the digestibilities of diet components as the level of protein in the soybean oil meal, sesame oil meal, and cottonseed meal diets was increased from 4 to 6 and then 8 percent. This was especially evident with crude fiber digestibility, which increased from 30 to 73 percent when the soybean oil meal diet protein increased from 4 to 8 percent. Under the same dietary change, crude protein digestibility increased from 27 to 58 percent and organic matter digestibility increased from 60 to 78 percent.

Gallup and Briggs (63) fed cattle 10 lb of several prairie hays and 0 to 3.0 lb of cottonseed meal daily, and found that the digestibilities of all nutrients were greatest in the rations of highest protein

content. However, as protein content of the rations increased from 5.5 to 13.4 percent, only the digestibilities of crude protein and ether extract increased in a somewhat regular manner. Crude protein digestibility increased from 33.3 to 60.1 percent. Changes in digestibilities of other ration components were small and appeared to be related to variations in hay quality and composition rather than to total protein content of the ration.

Klosterman et al. (98) reported digestion coefficients determined by the chromic oxide method for steers fed hay and a full feed of ground ear corn plus 0.75 or 1.50 lb of soybean oil meal per head daily. Dry matter and crude protein digestion coefficients for the low soybean oil meal averaged, respectively, 62.3 and 49.9, while for the higher soybean oil meal ration, corresponding figures were 65.0 and 57.8.

Eleven, 14, and 17 percent protein rations containing 97, 88, and 82 percent barley, respectively, with soybean oil meal added to increase the protein level, were fed to steers (96). Dry matter digestion coefficients were 72, 78, and 81, and were different ( $P < .05$ ) from each other. Apparent nitrogen digestion coefficients of 59, 68, and 75 were also different ( $P < .01$ ) from each other.

French, Glover, and Duthie (62) examined a large quantity of world data for cattle, sheep, and goats and related (y), the digestibility coefficient, to (x), the percentage of crude protein in the dry matter of feed, by the equation  $y = 70 \log x - 15$ . This equation is applicable to rations composed of both herbage and mixed feeds, for as the data suggests, it is the total percentage of crude protein in the

ration which determines its digestibility. In addition, it was noted that the digestibility of crude protein in the feed increased rapidly at low protein levels (from about 2 to 9 percent) and thereafter rose more slowly as the crude protein content increased.

In general, rations exhibit a depression in dry matter digestibility at low protein feeding levels (30). The depression of apparent coefficients of digestibility appears to be most marked for crude protein and less so for dry matter. The depression in protein digestibility in such rations seems more apparent than real, since metabolic fecal nitrogen is not generally considered in such calculations (30,31). The decrease in the digestibilities of nonprotein ration components, on the other hand, may be of real significance in that they may reflect the activities of microorganisms in the digestive processes.

Several workers have conducted microbiological studies in an effort to explain the influence of protein level on ration digestibility. Moir and Williams (129) reported an extremely high correlation ( $r = 0.98$ ) between level of protein intake and concentration of free microorganisms in the rumen. Later, using sheep, Williams et al. (184) found that, at all levels of starch feeding, the addition of protein significantly increased the mean dry matter digestibility from 55 to 61 and 71 percent for low, medium, and high protein diets, respectively. The low and high protein diets contained, respectively, 3.5 and 13.1 percent protein. Adding high levels of starch to the low, medium, and high protein diets yielded, respectively, 21, 44, and 54 million rumen bacteria per cubic millimeter. The bacteria count at the medium protein level



was higher ( $P < .05$ ) than at the low protein level, as was that at the high protein level ( $P < .01$ ).

This was substantiated by the work of Burroughs et al. (29). The dry matter digestion coefficient for a 4:4:1 starch:corn cob:alfalfa hay cattle diet was only 13.2. By adding 1 lb of casein and thereby increasing ration protein to 11.1 percent, roughage dry matter digestibility increased sharply to 46.4 percent. Increasing the ration protein to 17.4 percent by the addition of another pound of casein increased roughage digestion to 53.5 percent. Average rumen bacterial counts in billions per gram for these rations were 24.8, 47.1, and 48.4, respectively.

#### Factors Influencing the Excretion of Metabolic Fecal Nitrogen

Before the importance of metabolic fecal nitrogen can be assayed, factors influencing its excretion need evaluating, as must be its origins. Many factors have been implicated and subsequently studied as potential influences on the rate of metabolic fecal nitrogen excretion: dry matter intake, fecal excretion of dry matter, influence of indigestible dietary substances, crude fiber content of the diet, body weight or surface area, protein content of the diet, proportions of dietary carbohydrate and fat, diethylstilbestrol, and whether metabolic fecal nitrogen was determined on a nitrogen-free diet.

Dry Matter Intake. Mitchell (121) compiled a large quantity of data regarding the excretion of metabolic fecal nitrogen by human subjects and noted that the amount of dry matter consumed seemed to be the dominant factor influencing metabolic fecal nitrogen excretion. Fecal

nitrogen per 100 g of dry matter consumed tended to increase as food consumption increased. Mitchell also compiled similar data on dogs and drew the same conclusion. In a further study with rats to determine the influence of food consumption on metabolic nitrogen excretion in the feces, he plotted dry matter consumed per 100 g of body weight against metabolic fecal nitrogen per 100 g of dry matter consumed. With the influence of body weight removed, a fairly consistent trend toward the excretion of larger amounts of metabolic nitrogen in the feces with increasing feed consumption was noted. Similarly, Schneider (150) studied the relationship between metabolic nitrogen and dry matter intake with rats and determined the average correlation coefficient to be 0.962. The relationship was clearly a linear one. Schneider (149) also compiled data from 1160 rat metabolism determinations and calculated the correlation coefficient between food intake and metabolic nitrogen of the feces to be 0.74. The partial correlation coefficient of food intake and metabolic fecal nitrogen, independent of body weight, was also 0.74.

Data accumulated by Swanson and Herman (160) from experiments with dairy heifers fed roughage rations provide indirect evidence of the relationship between feed intake and metabolic fecal nitrogen. During two low nitrogen feeding periods in which feed consumption increased by 50 percent in the last period, they observed that total fecal nitrogen did not increase proportionally, resulting in a decline in fecal nitrogen per kilogram of feed intake.

With lambs fed an 0.11 percent nitrogen diet consisting of straw, starch, sugar, and oil, Sotola (155) reported that, in general, as the

quantity of dry matter ingested increased, total fecal nitrogen was a good measure of metabolic fecal nitrogen, as the total daily nitrogen intake amounted to only 12 percent of the fecal nitrogen output.

Fecal Excretion of Dry Matter. A nitrogen balance study was conducted by Meyer (117) with rats fed equivalent amounts of the basal ration plus 0, 5, 15, or 30 percent cellulose. He noted that fecal dry matter excretion was more satisfactory than food consumption as a reference base for estimating metabolic nitrogen. The correlation of metabolic fecal nitrogen with food consumption was 0.60, but was 0.78 with fecal dry matter excretion. In addition less variation was present when metabolic fecal nitrogen concentration was related to indigestible dry matter than when related to dietary cellulose levels.

In experiments on himself, Heupke (86) consumed a constant diet to which were added in successive periods varying amounts of nitrogen-poor ballast foods. He observed that the total amount of fecal nitrogen voided per day bore a direct relationship to the daily dry weight of the feces. He concluded that for every 20 g of fecal dry matter, 1 g of fecal nitrogen and 1 to 2 g of ether extract must be regarded as secretory products.

Ellis et al. (51) determined the metabolic fecal nitrogen excretion of lambs fed a purified diet containing 0.004 percent nitrogen. Metabolic fecal nitrogen was expressed in two ways: milligrams per gram of dry matter intake, which averaged 2.39, and milligrams per gram of fecal dry matter excreted, which averaged 7.17. Coefficients of variation for the two systems averaged 9.21 and 6.86, respectively, and

it was felt that a closer relationship existed between metabolic fecal nitrogen and excreted dry matter than between metabolic fecal nitrogen and dry matter intake. Mukherjee (134) and Hironaka, Bailey, and Kozub (87) also found that the relationship between metabolic fecal nitrogen and fecal dry matter excretion was closer for cattle and sheep than was the relationship between metabolic fecal nitrogen and dry matter intake.

The fecal excretion of dry matter and its relationship to metabolic fecal nitrogen excretion, though highly correlated, is confounded by the factors that determine dry matter excretion, namely, body size, level of food consumption, and proportions of crude fiber and indigestible substances in the food.

Influence of Indigestible Dietary Substances. Mitchell (123) reported that the excretion of metabolic nitrogen in the feces is related, for equal intakes of dry matter, to the indigestible matter contained in the diet. In studying this, Mendel and Fine (115) fed dogs a basal diet of meat, sugar, and lard or the basal plus agar and bone ash. Absolute fecal nitrogen increased due to the addition of the indigestible materials to the diet, although the nitrogen intake did not vary. For one dog, fecal nitrogen was increased 60 percent by the addition of 10 g of indigestible materials, and fecal nitrogen was augmented 133, 133, and 192 percent by the addition to the diet of another dog of 6, 7, and 13 g, respectively, of such materials.

Blaxter and Wood (18) fed baby calves a semi-synthetic nitrogen-free liquid diet formulated so that cod-liver oil replaced the dried skim milk of a normal diet. The excretion of metabolic fecal nitrogen

was found to be higher during the semi-synthetic diet periods than during periods of normal feeding. Apparent energy digestibilities were 66 percent for the semi-synthetic diet and 93 percent for the normal diet; however, statistical analysis of these results showed that there was no significant change in fecal nitrogen excretion.

Crude Fiber Content of the Diet. That the amount of roughage in the diet is an important factor in determining the quantity of metabolic nitrogen excreted in the feces was demonstrated by the work of Mitchell (120). Rats subsisting on a starch, lard, butterfat diet, with and without filter paper ad libitum, were compared, and it was observed that the excretion of fecal nitrogen per 100 g of food ingested was 173 and 122 mg, respectively. During periods in which filter paper was consumed, fecal nitrogen increased by an average of 42 relative percent.

Meyer (117) studied nitrogen balance with rats fed a basal diet to which 0, 5, 15, or 30 percent cellulose was added at the expense of the basal. Feeding was conducted to maintain equal intakes of the basal diet. Metabolic nitrogen excreted per gram of food intake was 1.38, 1.44, 1.75, and 1.81 mg, respectively, for the four cellulose levels. Values for the 15 and 30 percent cellulose rations were higher ( $P < .05$ ) than that for the 0 percent ration.

Hutchinson and Morris (93) studied the crude fiber-metabolic fecal nitrogen relationship with the same goats fed a starch, sawdust, oat straw, sugar diet, with and without 15 to 24 percent paper replacing starch. Paper invariably increased fecal nitrogen excretion, as shown by values of 468 and 528 mg per 100 g of dry food intake, respectively,

for rations without and with added paper. In another experiment, goats were fed low and high fiber diets consisting of oats and maize and different proportions of paper and starch. Daily fecal nitrogen excretion increased on the average of from 1.07 g on the low fiber to 1.57 g on the high fiber ration, an increase of 47 relative percent. The ration was somewhat unacceptable to the goats, and food intake was low.

Similar results were obtained by Steenbock, Nelson, and Hart (159), who observed that when 350-lb calves consumed rations consisting of varying proportions of milk, starch, and straw, fecal nitrogen excretion was always greater than ingested nitrogen and decreased as the quantity of straw in the ration was decreased.

Body Weight or Surface Area. Schneider (149), working with rats, investigated factors determining the amount of metabolic nitrogen in the feces and was able to show the existence of two distinct fractions of metabolic fecal nitrogen. One fraction is constant for each animal, but varies among different animals roughly in proportion to body size. The other fraction is variable in proportion to the intake of dry matter and indigestible non-nitrogenous matter. The former is so small that, with amounts of food permitting growth, its effect on the ratio of total metabolic nitrogen to dry matter consumed is inappreciable. Using rats (over 50 g) and pigs he determined the coefficient of variation of the ratio involving the constant fraction of metabolic fecal nitrogen and body weight to be 37.5, while that of the ratio involving the former and body surface was only 24.5. It was concluded that the constant fraction

of metabolic fecal nitrogen varies more closely in proportion to body surface than in proportion to body weight.

Mitchell (121) used rats weighing between 40 and 220 g to evaluate the influence of body weight on the excretion of metabolic fecal nitrogen. He reported a tendency for the smaller rats to excrete less metabolic fecal nitrogen in relation to food consumed than the larger, although the effect of body weight was small, amounting to only 0.031 g per 100 g of dry matter consumed.

Schneider (149) used data obtained from 1160 rat metabolism determinations to calculate the association,  $r$ , between body weight and metabolic fecal nitrogen. This was found to be 0.502. The partial correlation of body weight and metabolic fecal nitrogen, independent of food intake, was 0.499.

In the same vein, Swanson and Herman (160) noticed that yearling dairy heifers fed hay and straw excreted more fecal nitrogen on practically the same quantity of the same diet during their last low-nitrogen period than during their first such period when they were 40 kg lighter in weight.

Protein Content of the Diet. (a) Total fecal nitrogen - Titus (166) in his famous paper of 1927 reported that a linear relationship existed between the total nitrogen content of the feces of a steer (when corrected to uniform water content) and the nitrogen content of its feed. Mitchell and Bert (124), working with rats, also demonstrated that a linear relationship may be expected between the ratio of fecal nitrogen to dry matter consumed and to the level of dietary protein

within a range of from 0 to 20 percent. Blaxter and Mitchell (14), working with ruminants, observed a linear mathematical relationship between fecal nitrogen per 100 g of dry matter ingested and nitrogen content of the feed expressed on a dry basis. However, Harris, Work, and Henke (79) noted little mathematical relationship.

(b) Metabolic fecal nitrogen - Despite the well-established relationship between food protein level and total fecal nitrogen, no such relationship between dietary protein and metabolic fecal nitrogen has been determined. Voit (174) appears to be the only worker to have studied this possible relationship. He showed that, for any type of diet fed to dogs, the greater the dietary nitrogen intake, the greater the excretion of metabolic nitrogen in the feces. The ratios of metabolic fecal nitrogen per square meter of body surface to the energy intake in percent of the requirements were slightly different for protein, starch, and sugar. However, Voit attached no great significance to these differences.

Proportion of Dietary Carbohydrate. Voit (174) was unable to show that dietary carbohydrates influenced metabolic fecal nitrogen appreciably differently than did protein or sugar.

Proportion of Dietary Fat. Voit (174) found a substantial difference in the ratio of metabolic fecal nitrogen per unit of body surface to the energy intake in percent of the requirements for fat as compared to those for protein, starch, and sugar. Although these ratios were energy-dependent, with nutrients on a dry matter basis, the value of the ratio for fat was still less than 40 percent of that for sugar,



the next lowest ratio. He felt that this was of "undoubted significance."

Mitchell (122), however, working with rats several years later, failed to confirm Voit's results. He observed that the substitution of an equivalent weight of fat for starch did not affect the excretion of metabolic fecal nitrogen, even though the total amount of dry fecal material produced was considerably increased.

Diethylstilbestrol. McLaren et al. (114) studied the influence on metabolic fecal nitrogen excretion of 2 mg of diethylstilbestrol per day fed lambs consuming a 0.28 percent nitrogen ration containing 52 percent wheat straw plus molasses and concentrates. Metabolic fecal nitrogen values for diethylstilbestrol treated and control lambs were 0.495 and 0.469 g per 100 g of dry matter ingested, respectively. This difference was not significant ( $P > .05$ ). After correcting for undigested nitrogen, the corresponding values were 0.444 and 0.427, again not different ( $P > .05$ ) from each other.

Nitrogen-free Diet. Bosshardt and Barnes (21) studied the excretion of metabolic fecal nitrogen with mice consuming diets of various protein levels or a protein-free diet. The data, with the exception of that obtained with the protein-free diet approximated a straight line. Extrapolation of this line to the point of zero nitrogen intake gave a value of 322.9 mg of metabolic fecal nitrogen per 100 g of food consumed, as opposed to 300 mg per 100 g of food consumed for the protein-free diet.

Schiftan (148) conducted a similar study with swine and reported that regression lines, representing three protein sources fed up to 15 percent of the diet and extrapolated to 0 percent protein intake, converged closely on the metabolic fecal nitrogen value obtained with a protein-free diet.

Harris and Mitchell (78) fed sheep a low-nitrogen ration containing 36 percent wheat straw and wood pulp, with and without various levels of urea. The mean observed value for the ratio of fecal nitrogen per 100 g of dry matter consumed at the 0.136 percent nitrogen level was 0.55 g. That computed from the regression equation at the same dietary nitrogen level was 0.54 g. The difference between these two values was not significant ( $P > .05$ ).

Working with steers fed alfalfa hay and paper pulp Titus (166) concluded that it was probably not accurate to determine metabolic fecal nitrogen with animals consuming a nitrogen-free ration. However, the validity of his conclusion has been questioned, because he did not feed a nitrogen-free ration in this study.

Hutchinson and Morris (93) used sheep and goats fed essentially nitrogen-free diets to evaluate ruminant levels of metabolic fecal nitrogen. It was necessary to feed nearly all animals by stomach tube, as they would not eat voluntarily. With sheep fed a high level of the diet, apparently the fiber accumulated in the rumen, and the animals became ill and eventually died.

The use of nitrogen-free diets to determine the metabolic fecal nitrogen excretion by ruminants has been questioned on grounds that it

seems impossible to maintain normal rumen function with such a diet. In addition, to be consumed, nitrogen-free ruminant rations usually must contain large quantities of highly indigestible feedstuffs such as straw. Thus, these become confounding factors in the evaluation of metabolic fecal nitrogen excretion.

#### Metabolic Fecal Nitrogen Excretion of Ruminants and Nonruminants

There appear to be significant differences in metabolic fecal nitrogen excretion levels for ruminant and nonruminant animals.

Monogastric Animals. Mitchell (120) in 1924 reported the metabolic fecal excretion of rats to be about 120 mg per 100 g of food ingested. Metabolic fecal nitrogen excretion by swine, in grams per 100 g of food ingested, have ranged from about 0.07 reported by Schneider (150) to 0.10 determined by Mitchell and Bert (124) to a high of 0.18 reported by Bell et al. (8). Differences in swine values are partially a function of dietary crude fiber levels.

Mitchell (121) compiled the results of 22 experiments with humans and calculated the average metabolic fecal nitrogen excretion to be about 0.2 g per 100 g of dry matter ingested. He also reviewed data from 4 dog experiments in which the metabolic fecal nitrogen level was 0.17 g per 100 g of dry matter ingested.

Nonruminating Calves. Blaxter and Wood (18) determined metabolic fecal nitrogen excretion for 30 kg calves fed a semi-synthetic liquid diet. They obtained values of 0.45, 0.42, and 0.41 g per 100 g of dry matter ingested. It was observed that the nitrogen-free diet

used in these determinations had an apparent energy digestibility of 66.5 percent, thus precluding direct comparison with typical monogastric metabolic fecal nitrogen values.

Lofgreen and Kleiber (108) used  $^{32}\text{P}$  labeled casein to measure the proportion of fecal nitrogen which was of metabolic origin. Using 39 kg calves fed purified liquid diets, they obtained metabolic fecal nitrogen excretion values of 0.27 g per 100 g of dry matter intake. This value was considered to be a more valid estimate of metabolic fecal nitrogen excretion than those of the previously mentioned workers (18), who used nitrogen-free rations of low digestibility.

Sheep. Hutchinson and Morris (93), using a semi-purified nitrogen-free diet containing about 50 percent roughage, determined metabolic fecal nitrogen excretion values for lambs weighing between 14 and 24 kg. Using the extrapolation method, they calculated metabolic fecal nitrogen excretion to be 0.50 g per 100 g of dry matter food intake. Miller, Morrison, and Maynard (118) calculated, from 14 experiments, an average value of 0.55 g of metabolic nitrogen per 100 g of dry matter intake. Results similar to those reported in these papers were also obtained by Hamilton and Robinson (77), Turk, Morrison, and Maynard (168), and Harris and Mitchell (78).

A somewhat lower value of 0.435 was determined by McLaren et al. (114), using a 0.28 percent nitrogen ration containing 52 percent wheat straw.

Hironaka et al. (87) used sheep and cattle fed 0 to 100 percent concentrate rations and found that metabolic fecal nitrogen determined

by extrapolation was 0.89 g per 100 g of fecal dry matter. Ellis et al. (51) used lambs weighing between 60 and 80 lb fed a 0.004 percent nitrogen purified ration containing 40 percent Solka Floc. They reported metabolic fecal nitrogen levels of 2.39 and 7.17 mg per g of dry matter ingested and excreted, respectively. Singh and Mahadevein (154) recently determined metabolic fecal nitrogen levels of adult rams fed concentrate mixtures plus wheat bhusa given to appetite. By extrapolation, metabolic fecal nitrogen was found to be 0.214 and 0.473 g per 100 g of dry matter ingested and excreted, respectively. In a second trial, the same animals were changed to an almost nitrogen-free diet and corresponding metabolic fecal nitrogen values were increased to 0.254 and 0.545. Values from these last two studies (51,154) are dramatically lower than those reported by the previously mentioned workers, and they compare favorably with the results of Lofgreen and Kleiber (108) for baby calves receiving a liquid diet.

Cattle. Swanson and Herman (160) fed yearling dairy heifers chopped wheat and oat straws and measured a fecal nitrogen excretion of 0.53 g per 100 g of dry matter consumed. Colburn, Evans, and Ramage (36), by regression equations, measured the true digestibility of protein in forage consumed by ruminants. From this value they estimated metabolic fecal nitrogen as 0.60 g per 100 g of dry matter consumed. Using a lower roughage ration containing about 75 percent wheat straw and 25 percent starch and maize, Hutchinson and Morris (93) determined fecal nitrogen excretion for cows at about 0.45 g per 100 g of dry food intake.

Harris et al. (79) fed steers unusual, relatively high concentrate rations containing 15 percent bagasse, the remainder of the ration being essentially pineapple syrup, sugar cassava meal, and soybean oil meal. Little difference was noticed in the fecal nitrogen excretion of steers fed at the 1.44 or 13.81 percent crude protein levels so these values were averaged to 0.489 g fecal nitrogen per 100 g of dry matter intake.

Much lower metabolic fecal nitrogen values were reported by Morris and Wright (132). They determined a value of 0.33 g per 100 g of food intake from the results of 14 unpublished experiments. Harris and Loosli (80) determined, by the extrapolation method, the metabolic fecal nitrogen excretion of calves weighing 97 kg and consuming mixed rations at several protein levels. They obtained a value of 0.37 g per 100 g of dry matter ingested. Morris and Wright (131), in another experiment, were successful in getting a steer to eat a low nitrogen diet of sago-pith meal, which is low in fiber and contains little nitrogen. At amounts of feed sufficient to cover energy requirements, it was determined that 0.405 g of fecal nitrogen was excreted per 100 g of dry matter intake. The latter three metabolic fecal nitrogen values for cattle (80,131,132) are considerably lower than those reported by other workers; however, they are not nearly as low as those reported by Ellis et al. (51) and Singh and Mahadevein (154) for sheep.

#### Summary

Supplementing rations, especially low protein rations, with protein generally leads to dramatic increases in the apparent

digestibilities of ration components, and in particular that of protein.

It seems abundantly clear that metabolic fecal nitrogen plays an important part in the nitrogen economy of the animals. Metabolic nitrogen of the feces consists of two fractions: nitrogenous secretions from the body into the intestine and waste products of the digestive mechanism. The use of the extrapolation method for determining metabolic fecal nitrogen seems better suited for ruminants than does the nitrogen-free diet method. Body size, digestibility of the diet, dry matter intake, and crude fiber content of the diet all influence the excretion of metabolic fecal nitrogen, although the latter two are most important in this respect. Strangely enough, the most accurate reference base for metabolic fecal nitrogen is the fecal excretion of dry matter.

The excretion of metabolic fecal nitrogen appears to be substantially higher for ruminants than for monogastric animals. This may be attributable to the use of low digestibility, high crude fiber diets in ruminant determinations. The question remains unanswered because of the lack of data for ruminants consuming highly digestible diets, thus precluding direct comparison between the two kinds of animals. The limited data available for ruminants consuming high concentrate diets, however, suggest that there may be a true difference between ruminants and non-ruminants with respect to metabolic fecal nitrogen excretion.

## EXPERIMENTAL PROCEDURE

Young, growing cattle used in three series of digestion trials were individually housed in 8' x 16' concrete-floored pens. Each pen was equipped with an automatic watering cup and an aluminum shade which covered the feed bunk and one-half of each pen.

Cattle were individually weighed at the initiation and termination of each collection period. They were fed ad libitum twice daily until 5 days prior to the 5-day collection periods, at which time they were assigned to a constant level of feed intake.

Sufficient feed was mixed in one batch to last through the constant feed and collection periods of each experimental period. Feed ingredients between periods were not necessarily from the same source. The major ingredient of each ration was steam processed-flaked milo (sorghum grain) prepared by the method of Hale et al. (73).

The total collection method was employed, and the feces were recovered from pen floors with a dust pan and putty knife. Collections were made at dawn and dusk, in addition to several times during the day. Pen floors were washed at the termination of each 24-hour period. Feces collected during each 24-hour period were well mixed and an approximate 500-g aliquot, plus a representative sample of feed, were saved for laboratory determinations. Both feed and fecal samples were initially dried to constant weight in a forced-air oven at 45°C for Experiment I and at 90°C for Experiments II and III.



Dry matter, ether extract, and crude protein were determined according to A.O.A.C. (4) methods. Acid detergent fiber was determined by the method of Van Soest (173). Calcium was analyzed from a perchloric acid digest by flame photometry according to Coleman Bulletin D-248B (37), and phosphorus was determined by the method of Hawk, Oser, and Summerson (83). Gross energy was measured in an adiabatic bomb calorimeter. The pH of rumen fluid samples was measured with a Beckman Model N pH meter.

The fecal soap determination outlined by Bohman and Lesperance (20) was found to be inadequate for the extraction of high levels of soaps from feces. The 0.5 N hydrochloric acid solution used by these workers was not as effective as a 1.0 N solution applied directly to the sample or refluxing with a 1 percent hydrochloric acid-chloroform solution. The latter two methods were less consistent and did not remove as much soap from the samples as the method subsequently used and described below. Chloroform-methanol-hydrochloric acid (60:40:1 Vol.) lipid extractions were performed according to a modification of the procedure of the Heath and Hill (84), in which refluxing was replaced by blending at high speed for 14 minutes with a Sorvall Omni-mixer. Chloroform-methanol lipid extractions were performed identically to the previous extraction, with the exception that hydrochloric acid was deleted from the solvent solution. Fecal soaps were defined as chloroform-methanol-hydrochloric acid lipid extract minus chloroform-methanol lipid extract.

Rumen volatile fatty acids were prepared for analysis according to Erwin, Marco, and Emery (52). Analysis of 0.20 or 0.28 microliter

samples was by gas-liquid chromatography utilizing a Beckman GC-5 chromatograph with a hydrogen flame detector. The 0.3 by 182.9 column was packed with a partition liquid composed of 5 percent neo-pentylglyco adipate and 2 percent phosphoric acid and an inert support compound, Chromosorb W (60-80 mesh). Column and detector temperatures were 115° and 170°C, respectively, and the hydrogen flow rate was 17 ml per minute. The quantity of each volatile fatty acid present was determined by dividing the area under its curve by 1, 2, 3, and 4 for acetate, propionate, butyrate, and isobutyrate, and valerate and isovalerate, respectively, according to Theurer and Sawyer (162). Corrected area, when placed on a percentage basis, gave the molar percent (moles per 100 moles) of each fatty acid.

Data for the first experiment were statistically evaluated by least squares analysis of variance techniques according to Harvey (81). Data for the last two experiments and part of Experiment I were analyzed by Generalized Stepwise Multiple Regression Analysis for first through fourth power expressions (Charles K. Huszar, Computer Center, University of Arizona). In addition, for Experiment III, data were subjected to analysis of variance methods according to Steel and Torrie (157). Duncan's New Multiple Range Test (157) was employed to test differences between treatment means at the 5 percent level of significance.

Experiment I. Effect of Dietary Fat  
and Concentrate Levels on  
Ration Digestibility

Digestion studies were conducted with 12 half-sibling steers weight  $615 \pm 59$  lb (sd) at the initiation of the first collection.

were allotted to a randomized complete-block experimental design utilizing three blocks and four rations: 0, 5, 10, and 15 percent added animal fat. This permitted three observations per treatment per period. The experiment was conducted over three periods, each successively involving a higher concentrate ration, increasing from 60 to 75 to 90 percent (Tables 1 through 3). Each steer remained on the same level of fat throughout the experiment. Forty-day ad libitum feeding periods were utilized to "adjust" the animals to new rations prior to constant feed (90 percent ad libitum) periods. At the termination of each ad libitum and constant feed period, rumen samples were taken for volatile fatty acid and pH determinations.

Steer 17 (5 percent fat treatment) died after collection 1 and was not replaced. A volatile fatty acid sample for steer 20, collection 3, could not be obtained.

Experiment II. Effect of Feed Intake  
Level on the Digestibility  
of High Energy Rations

Eight heifer calves weighing  $455 \pm 38$  lb (sd) were randomly allotted to two 4 x 4 Latin-square experimental designs. Each Latin-square consisted of four animals, four periods, and four treatments. Treatments consisted of a 90 percent concentrate ration (Table 4) fed at approximately 1.0, 1.3, 1.5, and 1.8 times the maintenance level of energy consumption according to the California Net Energy Standards (171). The experimental design permitted two observations at approximately the same feed intake, per period.

Table 1. Sixty percent concentrate experimental rations. Experiment I, a, b

Item	Percent Added Fat			
	0	5	10	15
Ground alfalfa hay	20.00	20.00	20.00	20.00
Cottonseed hulls	20.00	20.00	20.00	20.00
Steam-processed milo	48.80	42.65	36.20	29.70
Cottonseed pellets	4.50	5.50	7.00	8.50
Molasses	5.00	5.00	5.00	5.00
Animal fat	-	5.00	10.00	15.00
Dicalcium phosphate	0.60	0.65	0.60	0.60
Urea	0.60	0.70	0.70	0.70
Salt	0.50	0.50	0.50	0.50
	100.00	100.00	100.00	100.00
Gross energy/kg, kcal	4,260	4,637	4,862	5,117
Crude protein %	13.95	14.45	14.36	13.80
Acid detergent fiber %	27.36	27.14	28.80	25.86
Lipid % <sup>c</sup>	4.02	9.44	13.93	19.57
Lipid % <sup>d</sup>	3.36	8.74	14.10	19.58
Ether extract %	1.51	7.16	13.26	18.00
Calcium %	0.64	0.61	0.70	0.75
Phosphorus %	0.37	0.36	0.38	0.46

a. Vitamin A added at the rate of 1500 I.U./lb of ration.

b. Analysis on dry matter basis.

c. Chloroform-methanol-hydrochloric acid extracted lipid.

d. Chloroform-methanol extracted lipid.

Table 2. Seventy-five percent concentrate experimental rations.  
Experiment I,a,b

Item	Percent Added Fat			
	0	5	10	15
Ground alfalfa hay	15.00	15.00	15.00	15.00
Cottonseed hulls	10.00	10.00	10.00	10.00
Steam-processed milo	64.70	58.00	51.50	45.00
Cottonseed pellets	3.50	5.20	6.70	8.20
Molasses	5.00	5.00	5.00	5.00
Animal fat	-	5.00	10.00	15.00
Dicalcium phosphate	0.50	0.50	0.50	0.50
Urea	0.50	0.50	0.50	0.50
Salt	0.50	0.50	0.50	0.50
Ground limestone	0.30	0.30	0.30	0.30
	100.00	100.00	100.00	100.00
<hr/>				
Gross energy/kg, kcal	4,366	4,723	4,982	5,272
Crude protein %	13.83	14.05	13.19	14.17
Acid detergent fiber %	17.96	20.19	20.30	20.08
Lipid % <sup>c</sup>	4.86	8.15	14.97	21.45
Lipid % <sup>d</sup>	3.78	8.66	13.59	19.90
Ether extract %	3.05	7.90	13.48	18.60
Calcium %	0.66	0.62	0.61	0.63
Phosphorus %	0.43	0.41	0.35	0.39

a. Vitamin A added at the rate of 1500 I.U./lb of ration.

b. Analysis on dry matter basis.

c. Chloroform-methanol-hydrochloric acid extracted lipid.

d. Chloroform-methanol extracted lipid.

Table 3. Ninety percent concentrate experimental rations.  
Experiment I.<sup>a, b</sup>

Item	Percent Added Fat			
	0	5	10	15
Ground alfalfa hay	5.0	5.0	5.0	5.0
Cottonseed hulls	5.0	5.0	5.0	5.0
Steam-processed milo	79.3	73.1	66.5	59.8
Cottonseed pellets	3.5	4.7	6.3	8.0
Molasses	5.0	5.0	5.0	5.0
Animal fat	-	5.0	10.0	15.0
Dicalcium phosphate	0.5	0.5	0.6	0.5
Urea	0.5	0.5	0.5	0.5
Salt	0.5	0.5	0.5	0.5
Ground limestone	0.7	0.7	0.6	0.7
	100.0	100.0	100.0	100.0
<hr/>				
Gross energy/kg, kcal	4,392	4,683	5,111	5,254
Crude protein %	14.02	13.22	13.57	14.44
Acid detergent fiber %	11.44	13.98	11.06	11.34
Lipid % <sup>c</sup>	4.58	8.17	13.94	19.09
Lipid % <sup>d</sup>	3.20	7.52	15.06	19.34
Ether extract %	3.02	8.35	14.96	19.70
Calcium %	0.56	0.65	0.54	0.68
Phosphorus %	0.43	0.49	0.40	0.46

a. Vitamin A added at the rate of 1500 I.U./lb of ration.

b. Analysis on dry matter basis.

c. Chloroform-methanol-hydrochloric acid extracted lipid.

d. Chloroform-methanol extracted lipid.

Table 4. Experimental ration. Experiment II.<sup>a, b</sup>

Item				
Ground alfalfa hay				10.0
Steam-processed milo				81.5
Cottonseed pellets				1.5
Molasses				5.0
Dicalcium phosphate				0.5
Urea				0.5
Salt				0.5
Ground limestone				0.5
				<u>100.0</u>
Periods				
	1 <sup>c</sup>	2 <sup>c</sup>	3 <sup>c</sup>	4
Gross energy/kg, kcal	4,335	4,532	4,346	4,305
Crude protein %	13.00	12.33	13.37	13.23

a. Vitamin A added at the rate of 1500 I.U./lb of ration.

b. Analysis on dry matter basis.

c. Feed was refused by the following animals in periods 1, 2, and 3, respectively: heifers 7; 7 and 8; 8.

Each period of the digestion experiment consisted of at least a 10-day ad libitum feeding interval plus the constant feed and collection periods. Two animals refusing feed during the first three periods were reassigned to the same treatments for a later make-up collection.

Regression equations were developed to determine the effect of level of energy intake on the digestibility of a high concentrate ration.

Experiment III. Metabolic Fecal Nitrogen and  
Digestibility Determinations With Cattle  
Consuming High Concentrate Rations

Eight steers weighing  $532 \pm 30$  lb (sd) were randomly allotted to a completely random experimental design. Two steers were assigned to each of four essentially isocaloric 90 percent concentrate ration treatments (Table 5). Crude protein levels were approximately 10, 12, 14, and 16 percent. After completion of a 21-day ad libitum feeding interval, a constant feed period began, followed by a 5-day collection phase. Constant feed intake per day was equal to 10 percent of metabolic body weight ( $\text{weight}^{0.75}$ ) determined just prior to the constant feed period. At the end of the first collection phase, steers were re-allotted to the four treatments, with each steer assigned to a different treatment than in period 1. After the same length time intervals, a second collection was made. The two animals refusing feed during the constant feed or collection periods were reassigned to the same treatments for a later make-up collection.

Regression equations were developed from these data and the data of Experiment II in an effort to determine a metabolic fecal nitrogen



Table 5. Experimental rations. Experiment III.<sup>a,b</sup>

Item	Percent Protein			
	10	12	14	16
Cottonseed hulls	10.00	10.00	10.00	10.00
Steam-processed milo	83.15	77.20	71.27	65.85
Soybean oil meal	-	6.00	12.00	17.50
Molasses	5.00	5.00	5.00	5.00
Dicalcium phosphate	0.55	0.50	0.37	0.25
Salt	0.50	0.50	0.50	0.50
Ground limestone	0.80	0.80	0.85	0.90
	100.00	100.00	100.00	100.00
<u>Period 1</u>				
Gross energy/kg, kcal	4,382 <sup>c</sup>	4,345	4,342	4,349 <sup>c</sup>
Crude protein %	9.65 <sup>c</sup>	11.82	15.12	14.77 <sup>c</sup>
<u>Period 2</u>				
Gross energy/kg, kcal	4,286	4,374	4,343	4,391
Crude protein %	10.30	13.10	14.91	17.53

a. Vitamin A added at the rate of 1500 I.U./lb of ration.

b. Analysis in dry matter basis.

c. Feed was refused by the following animals in period 1:  
steers 23 and 24.

value by extrapolation to zero nitrogen intake. Relationships between fecal nitrogen and dry matter ingestion and excretion were also examined, as was the influence of diet protein level on ration component digestibilities.

## RESULTS AND DISCUSSION

### Experiment I

#### Performance Data and General Considerations

Performance data were collected as an incidental part of this experiment, and validity of these data may correctly be questioned because of the manner in which the experiment was conducted. Parameters were measured during restricted feed periods associated with collections, as well as during ad libitum feeding periods, and the cattle were rumen-pumped, bled, and muscle-biopsied six times during the course of the experiment. In addition, relatively short time periods at each concentrate level were employed (56 days for the lower concentrate levels and 28 days for the highest concentrate level), and only three animals per fat level were utilized. However, because of the equal treatment of all animals on the several concentrate and fat levels, perhaps differences between treatments are valid approximations of actual treatment differences. It is important to realize that lipid levels of the rations were not those indicated in various table headings. To those amounts should be added the lipid content of the control ration, giving total ration lipid levels of approximately 4, 9, 14, and 19 percent (Tables 1 through 3) based on chloroform-methanol-hydrochloric acid extraction.

From the data in Table 6, it is apparent that as the level of dietary fat increased, average daily gain continued to decrease, with but one exception. As expected, feed per unit of gain (feed conversion)

Table 6. Performance data by ration (period) and treatment.<sup>a</sup>  
Experiment I.

Item	% Added Fat				Means
	0	5	10	15	
<u>60% Concentrate ration<sup>b</sup></u>					
Mean initial wt, lb	469	491	506	475	485
Feed consumption as % of body weight	2.85	2.77	2.40	2.00	2.50
Feed consumption/day, lb	15.60	15.56	13.58	10.32	13.76
Feed/gain	5.70	5.85	5.73	6.75	6.01
Avg daily gain, lb	2.74	2.66	2.37	1.53	2.32
<u>75% Concentrate ration<sup>b</sup></u>					
Mean initial wt, lb	622	640	639	561	615
Feed consumption as % of body weight	2.70	2.46	2.23	1.98	2.34
Feed consumption/day, lb	19.17	16.72	15.39	12.11	15.85
Feed/gain	6.36	5.90	8.01	12.39	8.16
Avg daily gain, lb	3.19	2.90	1.96	1.48	2.38
<u>90% Concentrate ration<sup>c</sup></u>					
Mean initial wt, lb	801	766	748	644	740
Feed consumption as % of body weight	1.83	2.10	1.85	1.22	1.75
Feed consumption/day, lb	14.96	16.68	14.38	7.99	13.50
Feed/gain	16.54	6.87	6.10	-	9.84
Avg daily gain, lb	0.90	2.43	2.36	0.05	1.44
<u>Means</u>					
Feed consumption as % of body weight	2.58	2.51	2.16	1.84	
Feed consumption/day, lb	16.90	16.32	14.45	10.53	
Feed/gain	6.62	5.97	6.57	8.81	
Avg daily gain, lb	2.55	2.71	2.20	1.20	

a. Data not subjected to statistical analysis, except feed consumption as % body weight: see Tables 7 and 8.

b. Fed for 56 days.

c. Fed for 28 days.

figures were in general higher as the level of dietary fat increased. However, comparing across concentrate levels, 5 percent added fat rations resulted in the lowest feed requirements per unit of gain and the highest daily gain of the four fat levels. Closer inspection of Table 6 reveals that the 0 percent fat level permitted the largest daily gains, with the lowest and next to lowest feed conversions for 60 and 75 percent concentrate rations, respectively. Two of the three cattle consuming the 0 percent fat, 90 percent concentrate ration went "off feed," greatly reducing average daily gain and increasing the feed conversion for steers consuming the ration. Thus, when summing across all concentrate levels, one is left with the perhaps mistaken impression that the 5 percent fat level resulted in the most favorable gain and feed conversion.

Average daily gain and feed per unit of gain were found to be intrinsically related to feed consumption as a percent of body weight, and appeared to vary largely as a function of feed consumption (Table 6). Feed consumption as a percent of body weight decreased with both increasing fat and concentrate levels (Tables 7, 8). The one exception to this was the previously noted 0 percent fat, 90 percent concentrate ration treatment. The use of a nested analysis imposed the limitation that the difference between two means cannot be declared significant by Duncan's New Multiple Range Test unless the larger subset in which both means are contained is a significant range (157). Thus, from data in Tables 7 and 8, as well as in analyses of variance Tables 27 and 28 (see p. 108 and 109), one can see that both concentrate and fat levels

Table 7. Dry matter consumed as a percent of body weight by added fat levels within concentrate level. Experiment I.

% Concentrate	% Added Fat			
	0	5	10	15
60	2.85 <sup>a</sup>	2.77 <sup>a</sup>	2.40 <sup>a,b</sup>	2.00 <sup>b</sup>
75	2.70	2.46	2.23	1.98
90	1.83 <sup>a</sup>	2.10 <sup>a</sup>	1.85 <sup>a</sup>	1.22 <sup>b</sup>
Means	2.58 <sup>a</sup>	2.51 <sup>a</sup>	2.16 <sup>b</sup>	1.84 <sup>c</sup>

a,b,c. Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 8. Dry matter consumed as a percent of body weight by concentrate levels within added fat level. Experiment I.

% Added fat	% Concentrate		
	60	75	90
0	2.85 <sup>a</sup>	2.70 <sup>a</sup>	1.83 <sup>b</sup>
5	2.77	2.46	2.10
10	2.40	2.23	1.85
15	2.00 <sup>a</sup>	1.98 <sup>a</sup>	1.22 <sup>b</sup>
Means	2.50 <sup>a</sup>	2.34 <sup>a</sup>	1.75 <sup>b</sup>

a,b. Means with different superscripts within row are significantly different ( $P < .05$ ).

influenced ( $P < .05$ ) feed consumption, although depressions ( $P < .05$ ) were limited to 60 and 90 percent concentrate and 0 and 15 percent fat levels, respectively.

When comparing across concentrate levels, consumption of the 0 percent fat rations was greatest and different ( $P < .05$ ) from that of 10 and 15 percent fat rations (Table 7). The aforementioned influence of two cattle going "off feed" must be considered when evaluating the magnitude of these differences. Comparing across fat levels, less ( $P < .05$ ) of the 90 percent concentrate rations were consumed (Table 8). These observations regarding the influence of fat and concentrate levels on feed consumption agree favorably with the literature and the three pertinent papers (32,48,107) cited in this work.

It is imperative to realize that the terms lipid and fat as used in this experiment refer not to conventional ether extract values but to lipid determined by chloroform-methanol-hydrochloric acid extraction. In Table 9 are compared lipid values of feed samples that underwent ether, chloroform-methanol, and chloroform-methanol-hydrochloric acid lipid extractions. These data indicate substantial differences in the quantity of lipid determined by the different methods, especially for the lower concentrate rations containing 0 and 5 percent added fat. Ether extract values at these fat levels were considerably lower than corresponding chloroform-methanol and chloroform-methanol-hydrochloric acid extract values. Therefore, comparison of ration lipid values determined by the chloroform-methanol-hydrochloric acid technique with

Table 9. Percent ether extract, chloroform-methanol extract, chloroform-methanol-hydrochloric acid extract, and "fecal soaps" of feed. Experiment I.

Item	Ether Extract	Chloroform- methanol Extract	Chloroform- methanol- hydrochloric Acid Extract	"Fecal Soaps" <sup>a</sup>
<u>60% Concentrate Ration</u>				
0% Added Fat	1.51	3.36	4.02	0.66
5% Added Fat	7.16	8.74	9.44	0.70
10% Added Fat	13.26	14.10	13.93	-0.17
15% Added Fat	18.00	19.58	19.57	-0.01
<u>75% Concentrate Ration</u>				
0% Added Fat	3.05	3.78	4.86	1.08
5% Added Fat	7.90	8.66	8.15	-0.51
10% Added Fat	13.48	13.59	14.97	1.38
15% Added Fat	18.60	19.90	21.45	1.55
<u>90% Concentrate Ration</u>				
0% Added Fat	3.02	3.20	4.58	1.38
5% Added Fat	8.35	7.52	8.17	0.05
10% Added Fat	14.96	15.06	13.94	-1.12
15% Added Fat	19.70	19.34	19.09	-0.25

a. Chloroform-methanol-hydrochloric acid lipid extract minus chloroform-methanol lipid extract.



those determined by the commonly reported ether extract method may have little meaning.

Another important consideration is the quantity of lipid ingested. As expected, lipid ingested per unit of body weight increased as the level of dietary fat increased (Table 10). In addition, with but one exception, as the concentrate level increased, rations containing equivalent quantities of fat were consumed at a reduced rate per 100 lb of body weight (Table 12). Cattle, when assigned to the 15 percent fat, highest concentrate ration, actually consumed less lipid per unit of body weight than did animals eating 10 percent fat rations at any concentrate level. As concentrate level of the diets increased, lipid ingested per unit of body weight decreased (Table 11). This occurred as a function of reduced feed intake at higher concentrate levels.

From analyses of variance Tables 13 and 14, ration concentrate level appeared to exert a greater influence on the digestibilities of ration components than did fat level. Concentrate level influenced ( $P < .01$ ) digestibilities of ration dry matter, gross energy, crude protein, acid detergent fiber and lipid, while fat influenced ( $P < .05$ ) only dry matter, gross energy, and lipid digestibilities. Percent dietary lipid affected ( $P < .05$ ) total digestible nutrients and influenced ( $P < .01$ ) feed consumption and fecal soaps expressed as a percent of feces or ingested lipid (Tables 27 and 28). Ration concentrate level also affected ( $P < .01$ ) the latter four parameters.

Table 10. Grams lipid ingested and percents apparently digested and excreted plus percent fecal soaps excreted by level of added fat per animal per 100 lb body weight. Experiment I.

% Added Fat	Ingested, g <sup>a</sup>	Apparently Digested, g <sup>a</sup>	Apparently Digested, % <sup>a</sup>	Excreted	
				Lipid, % <sup>b</sup>	Fecal Soap, % <sup>c</sup>
0	195	133	68.2 <sup>d</sup>	21.9	9.9 <sup>d</sup>
5	350	234	67.1 <sup>d,e</sup>	15.1	17.8 <sup>e</sup>
10	549	315	57.7 <sup>f</sup>	12.0	30.3 <sup>f</sup>
15	600	372	60.9 <sup>e,f</sup>	10.2	28.9 <sup>f</sup>

a. Chloroform-methanol-hydrochloric acid, extracted lipid.

b. Chloroform-methanol extracted lipid as a percent of ingested lipid.

c. Chloroform-methanol-hydrochloric acid lipid extract minus chloroform-methanol lipid extract, as a percent of ingested lipid.

d,e,f. Means with different superscripts within columns 4 and 6 are significantly different ( $P < .05$ ).

Table 11. Grams lipid ingested and percents apparently digested and excreted plus percent fecal soaps excreted by concentrate level per animal per 100 lb body weight. Experiment I.

% Concen- trate	Ingested, g <sup>a</sup>	Apparently Digested, g <sup>a</sup>	Apparently Digested, % <sup>a</sup>	Excreted	
				Lipid, % <sup>b</sup>	Fecal Soap, % <sup>c</sup>
60	489	274	57.3 <sup>d</sup>	17.4	25.2 <sup>d</sup>
75	463	319	69.0 <sup>e</sup>	13.0	18.0 <sup>e</sup>
90	320	198	64.1 <sup>e</sup>	14.0	21.9 <sup>d,e</sup>

a. Chloroform-methanol-hydrochloric and extracted lipid.

b. Chloroform-methanol extracted lipid as a percent of ingested lipid.

c. Chloroform-methanol-hydrochloric acid lipid extract minus chloroform-methanol lipid extract, as a percent of ingested lipid.

d,e. Means with different superscripts within columns 4 and 6 are significantly different ( $P < .05$ ).

Table 12. Grams lipid ingested and percents apparently digested and excreted plus percent fecal soaps excreted per animal per 100 lb body weight. Experiment I.

Item	Ingested, g <sup>a</sup>	Apparently Digested, g <sup>a</sup>	Apparently Digested, % <sup>a</sup>	Excreted	
				Lipid, % <sup>b</sup>	Fecal Soap, % <sup>c</sup>
<u>60% Concentrate Ration</u>					
0% Added Fat	213	128	60.2 <sup>d, e</sup>	27.6	12.2 <sup>d</sup>
5% Added Fat	473	315	66.5 <sup>d</sup>	16.3	17.2 <sup>d</sup>
10% Added Fat	622	315	50.7 <sup>e</sup>	13.8	35.5 <sup>e</sup>
15% Added Fat	647	337	52.0 <sup>e</sup>	11.9	36.1 <sup>e</sup>
<u>75% Concentrate Ration</u>					
0% Added Fat	209	151	72.0	19.4	8.6 <sup>d</sup>
5% Added Fat	323	218	67.8	15.4	16.8 <sup>d, e</sup>
10% Added Fat	574	360	62.7	10.6	26.7 <sup>f</sup>
15% Added Fat	744	547	73.7	6.4	19.9 <sup>e, f</sup>
<u>90% Concentrate Ration</u>					
0% Added Fat	163	119	72.5 <sup>d</sup>	18.7	8.8 <sup>d</sup>
5% Added Fat	255	170	67.2 <sup>d, e</sup>	13.5	19.3 <sup>e</sup>
10% Added Fat	452	270	59.8 <sup>e</sup>	11.6	28.6 <sup>e, f</sup>
15% Added Fat	409	233	57.1 <sup>e</sup>	12.2	30.7 <sup>f</sup>

a. Chloroform-methanol-hydrochloric acid extracted lipid.

b. Chloroform-methanol extracted lipid as a percent of ingested lipid.

c. Chloroform-methanol-hydrochloric acid lipid extract minus chloroform-methanol lipid extract, as a percent of ingested lipid.

d,e,f. Means with different superscripts within columns 4 and 6 within each concentrate level are significantly different ( $P < .05$ ).

Table 13. Analyses of variance of ration digestibilities by added fat levels within concentrate level. Experiment I.

Source of Variation	df	M e a n   S q u a r e s				Lipid <sup>a</sup>
		Dry Matter	Gross Energy	Crude Protein	Acid Detergent Fiber	
% Concentrate	2	701.62 <sup>**</sup>	660.58 <sup>**</sup>	447.36 <sup>**</sup>	880.57 <sup>**</sup>	380.04 <sup>**</sup>
% Fat, 60% Concentrate	3	33.88	24.45	45.61	61.18	154.29 <sup>*</sup>
% Fat, 75% Concentrate	3	27.63	36.46	100.71 <sup>*</sup>	181.42	72.12
% Fat, 90% Concentrate	3	63.36 <sup>*</sup>	105.91 <sup>**</sup>	46.55	143.86	141.33 <sup>*</sup>
Error	22	17.05	20.52	32.26	94.70	42.12

\*. (P<.05).

\*\*.. (P<.01).

a. Chloroform-methanol-hydrochloric acid extracted lipid.

Table 14. Analyses of variance of ration digestibilities by concentrate levels within added fat level. Experiment I.

Source of Variation	df	M e a n   S q u a r e s				Lipid <sup>a</sup>
		Dry Matter	Gross Energy	Crude Protein	Acid Detergent Fiber	
% Fat	3	61.37*	90.52*	24.44	18.56	193.21*
% Concentrate, 0% Fat	2	270.93**	256.96**	241.89**	311.59	146.40
% Concentrate, 5% Fat	2	110.59**	146.75**	84.09	139.43	0.53
% Concentrate, 10% Fat	2	121.12**	98.76*	15.63	22.50	94.84
% Concentrate, 15% Fat	2	301.03**	274.70**	363.79**	994.63**	384.79**
Error	22	17.05	20.52	32.26	94.70	44.03

\*. (P<.05).

\*\*.. (P<.01).

c. Chloroform-methanol-hydrochloric acid extracted lipid.

Dry Matter and Gross  
Energy Digestibilities

Increasing the level of dietary fat depressed ( $P<.05$ ) dry matter and gross energy digestibilities within the 90 percent concentrate rations (Table 15). Within this concentrate level digestibilities of 0 percent fat rations were greater ( $P<.05$ ) than for 10 and 15 percent rations. Summed across concentrate levels, digestibilities of the 0 and 5 were similar and somewhat higher than those for the 10 and 15 percent fat rations. At all levels of fat, digestibilities were lower ( $P<.05$ ) for 60 than 75 and 90 percent concentrate diets (Table 16). This was also true for the summation across fat levels. The depressing influence of fat on digestibilities was greatest for 90 and least for 75 percent concentrate rations.

That added dietary fat decreases dry matter digestibility has been confirmed with both cattle (1,48,147,177) and sheep (24,42). Added fat has also been shown to depress ration gross energy digestibility (1,147,177). However Esplin et al. (53) reported a nonsignificant increase ( $P<.05$ ) in dry matter and gross energy digestibilities with the addition of 4 percent fat to a 30 percent alfalfa hay diet containing 1 percent dicalcium phosphate. Characteristic of an emerging pattern is the report by Swift et al. (161) in which 3.7 percent corn oil increased and 7.1 percent corn oil decreased dry matter and gross energy digestibilities compared to the control ration. Substantiating Swift's observation are reports by Bujisse (28) and Dijkstra (46).

Table 15. Dry matter and gross energy digestion coefficients by added fat levels within concentrate level. Experiment I.

% Concentrate	Ration <sup>a</sup> Constituent	% Added Fat			
		0	5	10	15
60	DM	67.5	70.0	65.8	62.0
	GE	66.4	66.4	63.8	60.4
75	DM	79.8	79.1	74.4	81.4
	GE	78.4	77.9	71.7	79.5
90	DM	86.2 <sup>b</sup>	82.8 <sup>b, c</sup>	78.2 <sup>c</sup>	75.8 <sup>c</sup>
	GE	84.6 <sup>b</sup>	80.8 <sup>b, c</sup>	75.0 <sup>c, d</sup>	71.0 <sup>d</sup>
Means	DM	77.8 <sup>b</sup>	77.3 <sup>b, c</sup>	72.8 <sup>d</sup>	73.1 <sup>c, d</sup>
	GE	76.4 <sup>b</sup>	75.0 <sup>b, c</sup>	70.2 <sup>d</sup>	70.3 <sup>c, d</sup>

a. DM = dry matter, GE = gross energy.

b, c, d. Means with different superscripts within row are significantly different ( $P < .05$ ).



Table 16. Dry matter and gross energy digestion coefficients by concentrate levels within added fat level. Experiment I.

% Added Fat	Ration <sup>a</sup> Constituent	% Concentrate		
		60	75	90
0	DM	67.5 <sup>b</sup>	79.8 <sup>c</sup>	86.2 <sup>c</sup>
	GE	66.4 <sup>b</sup>	78.4 <sup>c</sup>	84.6 <sup>c</sup>
5	DM	70.0 <sup>b</sup>	79.1 <sup>c</sup>	82.8 <sup>c</sup>
	GE	66.4 <sup>b</sup>	77.9 <sup>c</sup>	80.8 <sup>c</sup>
10	DM	65.8 <sup>b</sup>	74.4 <sup>c</sup>	78.2 <sup>c</sup>
	GE	63.8 <sup>b</sup>	71.7 <sup>c</sup>	75.0 <sup>c</sup>
15	DM	62.0 <sup>b</sup>	81.4 <sup>c</sup>	75.8 <sup>c</sup>
	GE	60.4 <sup>b</sup>	79.5 <sup>c</sup>	71.0 <sup>d</sup>
Means	DM	66.3 <sup>b</sup>	78.7 <sup>c</sup>	80.8 <sup>c</sup>
	GE	64.2 <sup>b</sup>	76.8 <sup>c</sup>	77.8 <sup>c</sup>

a. DM = dry matter, GE = gross energy.

b,c,d. Means with different superscripts within row are significantly different ( $P < .05$ ).

### Crude Protein Digestibility

The magnitude of the influence of dietary fat on apparent crude protein digestibility appeared less than was the case for dry matter and gross energy digestibilities. Increasing the level of dietary fat increased ( $P < .05$ ) apparent crude protein digestibility for the 15 percent fat, 75 percent concentrate ration (Table 17). Summed across concentrate levels, digestion coefficients for the various fat levels were not different ( $P > .05$ ). Increasing ration concentrate level increased ( $P < .05$ ) crude protein digestibility within 0 and 15 percent fat rations (Table 18). Digestion coefficients for 75 and 90 percent concentrate rations (summed across fat levels) were higher ( $P < .05$ ) than for the lowest concentrate ration.

This is somewhat at variance with the work of Albin and Durham (1) and Dijkstra (46) who reported a depressing influence of fat on apparent crude protein digestibility. In addition, several workers (20, 28, 48, 177) found fat to be without effect on apparent crude protein digestibility, although in general high roughage diets were used. However, Esplin et al. (53) and Page et al. (140) have reported a favorable, though not statistically significant, response to fat additions. As with dry matter and gross energy, Swift et al. (161) found an increase in apparent crude protein digestibility with 3.7 percent added corn oil and a decrease with 7.1 percent.

### Acid Detergent Fiber Digestibility

Added dietary fat did not influence ( $P > .05$ ) acid detergent fiber digestibility (Table 19). There was no noticeable pattern in response

Table 17. Crude protein digestion coefficients by added fat levels within concentrate level. Experiment I.

% Concentrate	% Added Fat			
	0	5	10	15
60	54.7	61.5	62.7	56.3
75	65.0 <sup>a</sup>	70.6 <sup>a,b</sup>	67.2 <sup>a</sup>	78.2 <sup>b</sup>
90	72.6	72.1	65.2	65.3
Means	64.1	68.1	65.0	66.6

a,b. Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 18. Crude protein digestion coefficients by concentrate levels within added fat level. Experiment I.

% Added Fat	% Concentrate		
	60	75	90
0	54.7 <sup>a</sup>	65.0 <sup>b</sup>	72.6 <sup>b</sup>
5	61.5	70.6	72.1
10	62.7	67.2	65.2
15	56.3 <sup>a</sup>	78.2 <sup>b</sup>	65.3 <sup>a</sup>
Means	58.8 <sup>a</sup>	70.3 <sup>b</sup>	68.8 <sup>b</sup>

a,b. Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 19. Acid detergent fiber digestion coefficients by added fat levels within concentrate level. Experiment I.

% Concentrate	% Added Fat			
	0	5	10	15
60	35.0	40.4	43.0	33.4
75	52.8	55.0	48.2	66.5
90	52.6	50.3	44.1	36.8
Means	46.8	48.6	45.1	45.6

Table 20. Acid detergent fiber digestion coefficients by concentrate levels within added fat level. Experiment I.

% Added Fat	% Concentrate		
	60	75	90
0	35.0	52.8	52.6
5	40.4	55.0	50.3
10	43.0	48.2	44.1
15	33.4 <sup>a</sup>	66.5 <sup>b</sup>	36.8 <sup>a</sup>
Means	38.0 <sup>a</sup>	55.6 <sup>b</sup>	46.0 <sup>a</sup>

a,b. Means with different superscripts within row are significantly different ( $P < .05$ ).

to fat additions to 60 and 75 percent concentrate levels, but for the 90 percent level acid detergent fiber digestibility decreased ( $P>.05$ ) with each increase in dietary fat. Increasing ration concentrate level increased ( $P<.05$ ) acid detergent fiber digestibility only within the 15 percent fat level for the 75 percent concentrate ration (Table 20). Digestibilities at each fat level were lower ( $P>.05$ ) for 60 than 75 and 90 percent concentrate rations.

The lack of response of acid detergent fiber digestibility to dietary fat (with the one exception noted) was surprising. The overwhelming evidence reported in the literature indicates that a substantial depression in crude fiber digestibility occurred upon the addition of fat to the diet (20,28,48,140,177), and this was perhaps a greater depression than occurred with any other ration component. Most of this work was conducted employing relatively high fiber, low fat diets, which makes even more interesting the observation that fat influenced acid detergent fiber digestibility for only the 90 percent concentrate rations. Even though crude fiber and acid detergent fiber are not equivalent, this result was not expected.

#### Lipid Digestibility

Increasing ration lipid content decreased ( $P<.05$ ) digestion coefficients at the 10 and 15 percent fat levels for 60 and 90 percent concentrate rations (Table 21). Within the highest fat level lipid digestibility of the 75 percent concentrate ration was greatest ( $P<.05$ ) as shown in Table 22. Summed across fat levels, lipid digestibilities

Table 21. Chloroform-methanol-hydrochloric acid lipid digestion coefficients by added fat levels within concentrate level. Experiment I.

% Concentrate	% Added Fat			
	0	5	10	15
60	60.2 <sup>a,b</sup>	66.5 <sup>a</sup>	51.5 <sup>b</sup>	52.0 <sup>b</sup>
75	72.1	67.7	62.7	73.7
90	72.5 <sup>a</sup>	67.2 <sup>a,b</sup>	59.7 <sup>b</sup>	57.4 <sup>b</sup>
Means	68.2 <sup>a</sup>	67.2 <sup>a,b</sup>	58.0 <sup>c</sup>	61.0 <sup>b,c</sup>

a,b. Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 22. Chloroform-methanol-hydrochloric acid lipid digestion coefficients by concentrate levels within added fat level. Experiment I.

% Concentrate	% Concentrate		
	60	75	90
0	60.2	72.1	72.5
5	66.5	67.7	67.2
10	51.5	62.7	59.7
15	52.0	73.7 <sup>b</sup>	57.4 <sup>a</sup>
Means	57.5 <sup>a</sup>	69.1 <sup>b</sup>	64.2 <sup>b</sup>

a,b. Means with different superscripts within row are significantly different ( $P < .05$ ).

for the two highest concentrate rations were greater ( $P < .05$ ) than for 60 percent concentrate rations.

The trend toward lower lipid digestibility with increasing fat level reported here is in conflict with the bulk of the literature (20, 28, 53, 147, 161). However, this can be readily explained by considering the difference between the commonly reported ether extract and chloroform-methanol-hydrochloric acid extract utilized in this experiment. Hydrochloric acid hydrolyzes fecal soaps into mineral and fatty acid constituents, permitting measurement of lipid associated with fecal soaps by extraction with lipid solvents. Examination of the data in Table 23 reveals that fecal samples of animals fed added fat, especially at higher levels, contained large quantities of fecal soaps (up to 24 percent by weight). In addition, larger feed lipid values were obtained by chloroform-methanol-hydrochloric acid extraction than by ether extract (Table 9). Consideration of these two factors, but particularly fecal soap lipid, in digestibility calculations resulted in lower lipid digestion coefficients.

#### Total Digestible Nutrients

The addition of digestible crude protein, ether extract  $\times 2.25$ , crude fiber, and nitrogen-free extract is the commonly accepted method of calculating total digestible nutrients. However, it is apparent from the previous discussion on lipid digestibility that the use of ether extract for the purpose of calculating total digestible nutrients of high fat rations will yield erroneous results. For this reason, it is suggested that the total digestible nutrients formula be modified,

Table 23. Percent ether extract, chloroform-methanol extract, chloroform-methanol-hydrochloric acid extract, and fecal soaps of feces. Experiment I.

Item	Ether Extract	Chloroform- methanol Extract	Chloroform- methanol- hydrochloric Acid Extract	Fecal Soaps <sup>a</sup>
<u>60% Concentrate Ration</u>				
0% Added Fat	2.35	3.44	4.96	1.52
5% Added Fat	3.06	5.13	10.54	5.42
10% Added Fat	3.62	5.30	19.82	14.52
15% Added Fat	4.00	6.20	24.80	18.60
<u>75% Concentrate Ration</u>				
0% Added Fat	1.95	4.69	6.77	2.08
5% Added Fat	2.72	6.02	12.87	6.84
10% Added Fat	3.51	6.22	21.87	15.66
15% Added Fat	3.94	7.32	30.16	22.85
<u>90% Concentrate Ration</u>				
0% Added Fat	5.73	6.24	9.15	2.91
5% Added Fat	6.14	6.42	15.20	8.78
10% Added Fat	7.71	7.40	25.93	18.53
15% Added Fat	9.16	9.59	33.69	24.11

a. Chloroform-methanol-hydrochloric acid lipid extract minus chloroform-methanol lipid extract.



replacing ether extract with lipid determined by an acid containing solvent such as the one used in this experiment. Table 24 contains means of total digestible nutrients calculated according to this modified formula, with acid detergent fiber replacing crude fiber. This latter substitution is not without question, as nitrogen-free extract digestion coefficients for the two highest fat levels several times exceeded 100 (Table 63, Appendix). Because of the aforementioned changes in the total digestible nutrients formula, it was considered advantageous to calculate and use total digestible nutrients from the following relationship:

$$\frac{\text{TDN}}{\text{lb}} = \frac{\text{kilocalories digestible energy/pound}}{2000}$$

The means of total digestible nutrients calculated in the latter manner in general agree quite well with those calculated by the modified formula, although they are on the average several units lower (Table 25). However, when individual means were placed in order of magnitude, a number of differences were noted between total digestible nutrients calculated by the two methods.

Increasing the level of diet fat increased ( $P < .05$ ) total digestible nutrients at the 75 percent concentrate level for the 15 percent fat ration (Tables 25 and 27). When summed across concentrate levels, total digestible nutrients were greater ( $P < .05$ ) for 15 than 0 percent fat rations. For each fat level total digestible nutrients were greater ( $P < .05$ ) for 75 and 90 than 60 percent concentrate rations (Tables 26 and 28). Within the 15 percent fat level the ration with the greatest

Table 24. Total digestible nutrients by added fat levels within concentrate levels.<sup>a,b</sup> Experiment I.

% Concentrate	% Added Fat				Means
	0	5	10	15	
60	67.3	74.4	70.4	71.7	71.0
75	80.9	83.2	88.0	98.4	86.4
90	87.4	86.7	87.0	87.0	87.0
Means	78.5	81.4	80.1	85.7	

a. TDN = digestible protein + digestible chloroform-methanol-hydrochloric acid lipid times 2.25 + digestible acid detergent fiber + digestible nitrogen-free extract.

b. Data not subjected to statistical analysis.

Table 25. Total digestible nutrients by added fat levels within concentrate levels.<sup>a</sup> Experiment I.

% Concentrate	% Added Fat			
	0	5	10	15
60	64.2	69.9	70.4	70.1
75	77.7 <sup>b</sup>	83.5 <sup>b</sup>	81.1 <sup>b</sup>	95.1 <sup>c</sup>
90	84.3	85.9	87.0	84.6
Means	75.4 <sup>b</sup>	79.8 <sup>b,c</sup>	79.5 <sup>b,c</sup>	83.3 <sup>c</sup>

a. TDN = digestible energy (kcal) per lb ration divided by 2000.

b,c. Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 26. Total digestible nutrients by concentrate levels within added fat levels.<sup>a</sup> Experiment I.

% Added Fat	% Concentrate		
	60	75	90
0	64.2 <sup>b</sup>	77.7 <sup>c</sup>	84.3 <sup>c</sup>
5	69.9 <sup>b</sup>	83.5 <sup>c</sup>	85.9 <sup>c</sup>
10	70.4 <sup>b</sup>	81.1 <sup>c</sup>	87.0 <sup>c</sup>
15	70.1 <sup>b</sup>	95.1 <sup>c</sup>	84.6 <sup>d</sup>
Means	68.7 <sup>b</sup>	84.3 <sup>c</sup>	85.5 <sup>c</sup>

a. TDN = digestible energy (kcal) per lb ration divided by 2000.

b,c,d. Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 27. Analyses of variance of several parameters by concentrate levels within added fat level. Experiment I.

Source of Variation	df	M e a n   S q u a r e s			
		TDN <sup>a</sup>	Feed Consumed <sup>b</sup>	Fecal <sup>c</sup> Soaps	Fecal <sup>d</sup> Soaps
% Concentrate	2	1015.30**	0.94**	36.08**	148.41**
% Fat, 60% Concentrate	3	27.20	0.47*	187.33**	453.19**
% Fat, 75% Concentrate	3	171.65**	0.26	246.74**	167.37**
% Fat, 90% Concentrate	3	4.62	0.39*	263.81**	299.34**
Error	22	26.40	0.11	3.14	25.82

\*. (P<.05).

\*\*. (P<.01).

a. Total digestible nutrients, calculated from digestible energy.

b. Feed consumed as a percent of body weight.

c. Fecal soaps as a percent of feces.

d. Fecal soaps as a percent of ingested lipid.

Table 28. Analyses of variance of several parameters by added fat levels within concentrate level. Experiment I.

Source of Variation	df	M e a n   S q u a r e s			
		TDN <sup>a</sup>	Feed Consumed <sup>b</sup>	Fecal Soaps <sup>c</sup>	Fecal Soaps <sup>d</sup>
% Fat	3	94.25*	0.70**	690.55**	830.15**
% Concentrate, 0% Fat	2	316.00**	0.90**	1.47	11.92
% Concentrate, 5% Fat	2	189.60**	0.12	6.81	3.70
% Concentrate, 10% Fat	2	211.60**	0.24	12.85*	65.05
% Concentrate, 15% Fat	2	472.34**	0.40*	24.94**	203.58**
Error	22	26.40	0.11	3.14	25.82

\*. (P<.05).

\*\*.. (P<.01).

a. Total digestible nutrients, calculated from digestible energy.

b. Feed consumed as a percent of body weight.

c. Fecal soaps as a percent of feces.

d. Fecal soaps as a percent of ingested lipid.

( $P < .05$ ) total digestible nutrients content was the 75 percent concentrate ration.

The results of this experiment are somewhat at variance with the work of Bohman and Lesperance (20), who added fat to an alfalfa hay ration, and Esplin et al. (53), who added fat to a 70 percent concentrate ration. Both papers reported an increase ( $P < .05$ ) in total digestible nutrients, whereas an increase ( $P < .05$ ) at only one fat and concentrate level was noted here.

#### Fecal Soaps as a Percent of Feces

Within every concentrate level, fecal soaps increased ( $P < .05$ ) with each increase in diet fat level (Table 29). Increasing ration concentrate level also increased fecal soap excretion, although this was only significant ( $P < .05$ ) for 10 and 15 percent fat levels (Table 30). Summing across fat levels, each increase in concentrate level also increased ( $P < .05$ ) fecal soap excretion. The concomitant increase in fecal soaps, as a percent of fecal dry matter, with dietary fat level found here is in agreement with the work of Bohman and Lesperance (20), Esplin et al. (53), and Roberts and McKirdy (147).

#### Fecal Soaps as a Percent of Ingested Lipid

Fecal soap excretion increased as the level of dietary fat increased, with but a single exception (Table 31). Within each concentrate level fecal soap excretion was lower ( $P < .05$ ) for 0 than 10 and 15 percent fat rations. Fecal soap values summed across concentrate levels were lower ( $P < .05$ ) for the 0 than 5 percent level, both being

Table 29. Fecal soaps<sup>a</sup> as a percent of fecal dry matter by added fat levels within concentrate level. Experiment I.

% Concentrate	% Added Fat			
	0	5	10	15
60	1.52 <sup>b</sup>	5.42 <sup>c</sup>	14.52 <sup>d</sup>	18.60 <sup>e</sup>
75	2.08 <sup>b</sup>	6.85 <sup>c</sup>	15.66 <sup>d</sup>	22.85 <sup>e</sup>
90	2.91 <sup>b</sup>	8.79 <sup>c</sup>	18.53 <sup>d</sup>	24.11 <sup>e</sup>
Means	2.17 <sup>b</sup>	7.02 <sup>c</sup>	16.24 <sup>d</sup>	21.85 <sup>e</sup>

a. Chloroform-methanol-hydrochloric acid lipid extract minus chloroform-methanol lipid extract.

b, c, d, e. Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 30. Fecal soaps<sup>a</sup> as a percent of fecal dry matter by concentrate levels within added fat level. Experiment I.

% Added Fat	% Concentrate		
	60	75	90
0%	1.52	2.08	2.91
5%	5.42	6.84	8.78
10%	14.52 <sup>b</sup>	15.66 <sup>b, c</sup>	18.53 <sup>c</sup>
15%	18.60 <sup>b</sup>	22.85 <sup>c</sup>	24.11 <sup>c</sup>
Means	10.01 <sup>b</sup>	11.86 <sup>c</sup>	13.58 <sup>d</sup>

a. Chloroform-methanol-hydrochloric acid lipid extract minus chloroform-methanol lipid extract.

b, c, d. Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 31. Fecal soaps<sup>a</sup> as a percent of ingested lipid by added fat levels within concentrate level. Experiment I.

% Concentrate	% Added Fat			
	0	5	10	15
60	12.27 <sup>b</sup>	17.23 <sup>b</sup>	35.53 <sup>c</sup>	36.07
75	8.63 <sup>b</sup>	16.80 <sup>b,c</sup>	26.67 <sup>d</sup>	19.90 <sup>c,d</sup>
90	8.80 <sup>b</sup>	19.30 <sup>c</sup>	28.63 <sup>c,d</sup>	30.73 <sup>d</sup>
Means	9.93 <sup>b</sup>	17.81 <sup>c</sup>	30.34 <sup>d</sup>	28.93 <sup>d</sup>

a. Chloroform-methanol-hydrochloric acid lipid extract minus chloroform-methanol lipid extract.

b,c,d. Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 32. Fecal soaps<sup>a</sup> as a percent of ingested lipid by concentrate levels within added fat level. Experiment I.

% Added Fat	% Concentrate		
	60	75	90
0	12.27	8.63	8.80
5	17.23	16.80	19.30
10	35.53	26.67	28.63
15	36.07 <sup>b</sup>	19.90 <sup>c</sup>	30.73 <sup>b</sup>
Means	25.25 <sup>b</sup>	18.00 <sup>c</sup>	21.87 <sup>b,c</sup>

a. Chloroform-methanol-hydrochloric acid lipid extract minus chloroform-methanol lipid extract.

b,c. Means with different superscripts within row are significantly different ( $P < .05$ ).



lower than values for the two highest fat levels. The effect of concentrate level was to reduce ( $P < .05$ ) fecal soap excretion for the 75 percent concentrate ration within the 15 percent fat level (Table 32). No information was found in the literature regarding this parameter.

It is interesting to note that fecal soaps expressed as a percent of feces (Table 29) or as a percent of ingested lipid (Table 31) followed the consistent trend of increasing fecal soap values with increasing diet fat levels. Fecal soap values, expressed as a percent of feces, increased ( $P < .05$ ) for each increase in ration concentrate level, when summed across fat levels. However, when expressed as a percent of ingested lipid, fecal soap excretion increased ( $P < .05$ ) only through the 10 percent fat level; the 10 and 15 percent fat levels were not different ( $P > .05$ ) from each other.

#### Calcium Digestibility

Apparent calcium digestion coefficients indicated that calcium excretion increased (digestibility decreased) as fecal soap excretion increased, for 60 and 75 percent concentrate rations; however, this relationship did not hold for 90 percent concentrate rations (Table 33). A spot check of the fecal soap fraction indicated that calcium was the major mineral present, along with sodium, potassium, and magnesium. In support of these observations, Grainger and Stroud (69) and Grainger et al. (68) noticed that corn oil decreased the apparent digestibility of calcium and increased the excretion of fecal soaps by lambs. Davison and Woods (43,44), however, found no significant change in the fecal

Table 33. Apparent calcium digestion coefficients.<sup>a</sup> Experiment I.

Steer Number	Treatment	% Concentrate		
		60	75	90
13	0% Added Fat	37.3	81.1	27.1
18		43.0	45.7	38.8
23		<u>36.1</u>	<u>43.3</u>	<u>44.6</u>
		Average	38.8	40.0
16	5% Added Fat	27.8	43.3	34.6
17		38.5	-	-
22		<u>28.8</u>	<u>26.7</u>	<u>41.3</u>
		Average	31.7	35.0
15	10% Added Fat	32.0	33.9	14.4
20		27.8	36.2	25.6
24		<u>24.5</u>	<u>22.9</u>	<u>13.2</u>
		Average	28.1	31.0
14	15% Added Fat	1.6	33.3	0
19		32.5	53.2	22.3
21		<u>31.2</u>	<u>49.5</u>	<u>51.5</u>
		Average	21.8	45.3

a. Data not subjected to statistical analysis.

excretion of calcium when 5 percent corn oil was added to high roughage lamb rations.

#### Fecal Energy

Fecal gross energy (kcal per g fecal dry matter) increased with increases in both ration fat and concentrate levels (Table 59, Appendix). Similar observations were made by Bohman and Lesperance (20) and Roberts and McKirdy (147).

#### Rumen pH

Addition of fat to either constant or ad libitum fed diets was without effect ( $P > .05$ ) except for the summation across constant feed concentrate levels, resulting in a lower ( $P < .05$ ) rumen sample pH for 15 than 0, 5, and 10 percent fat rations (Tables 34 and 36). There, however, was a definite trend toward lowered pH with both regimens for increasing fat level. There was also a consistent decline in rumen sample pH with increasing concentrate level (Tables 35 and 37). For constant feed, 0 and 15, and for ad libitum feed, 0 percent fat level, pH values were lower ( $P < .05$ ) for 90 than 60 percent concentrate rations. When pH values were summed across fat levels, pH was lower ( $P < .05$ ) for 90 than 60 and 75 percent concentrate rations for both constant and ad libitum periods. In general, pH of rumen samples were slightly higher for constant than for ad libitum feed periods.

Putnam et al. (143) also observed a decline in rumen pH upon addition of fat to the diet, but Brethour et al. (24) noticed no diurnal change in rumen pH when corn oil was fed.

Table 34. Rumen sample pH for constant feed intakes by added fat levels within concentrate level. Experiment I.

% Concentrate	% Added Fat			
	0	5	10	15
60	6.78	6.72	6.77	6.28
75	6.80	6.62	6.35	6.07
90	6.05	6.08	6.00	5.52
Means	6.54 <sup>a</sup>	6.47 <sup>a,b</sup>	6.37 <sup>a,b</sup>	5.96 <sup>c</sup>

a,b,c. Means with different superscripts within row are significantly different ( $P < .25$ ).

Table 35. Rumen sample pH for constant feed intakes by concentrate levels within added fat level. Experiment I.

% Added Fat	% Concentrate		
	60	75	90
0	6.78 <sup>a</sup>	6.80 <sup>a</sup>	5.05 <sup>b</sup>
5	6.72	6.62	6.08
10	6.77	6.35	6.00
15	6.28 <sup>a</sup>	6.07 <sup>a,b</sup>	5.52 <sup>b</sup>
Means	6.64 <sup>a</sup>	6.46 <sup>a</sup>	5.91 <sup>b</sup>

a,b. Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 36. Rumen sample pH for ad libitum feed intakes by added fat levels within concentrate level. Experiment I.

% Concentrate	% Added Fat			
	0	5	10	15
60	6.67	6.73	6.08	6.15
75	6.45	6.40	6.28	6.07
90	5.80	6.02	5.83	5.70
Means	6.31	6.39	6.07	5.97

### Volatile Fatty Acids

Level of dietary fat exerted less influence on rumen sample volatile fatty acid concentrations than did ration concentrate level (Tables 67 through 70, see Appendix). Increasing dietary fat level (summed across concentrate levels) promoted a general, though nonsignificant ( $P > .05$ ) decrease in acetic acid concentration while propionic acid levels increased ( $P < .05$ ) in one step and leveled off for both constant and ad libitum feeding periods (Tables 38 and 39). Within the 60 percent rations (constant feed), acetic acid values were lower ( $P < .05$ ) for 10 and 15 than 0 and 5 percent fat levels, but propionic acid concentrations were greater ( $P < .05$ ) for 10 and 15 than 0 and 5 percent fat levels (Table 40). For the same (60 percent) concentrate level fed ad libitum acetic acid levels were lower ( $P < .05$ ) for 10 and 15 than for 0 percent fat levels, while propionic acid levels were again opposite and higher ( $P < .05$ ) for 10 and 15 than for 0 percent fat levels (Table 41). Thus, it is apparent that dietary fat shifted volatile fatty acid concentrations in favor of propionic acid at the expense of acetic acid. Within 90 percent concentrate ad libitum rations propionic acid levels increased ( $P < .05$ ) at the 5 and 10 percent fat levels. No other trends were apparent for acetic and propionic acids.

Each increase in the proportion of ration concentrates (summed across fat levels) decreased ( $P < .05$ ) acetic and increased ( $P < .05$ ) propionic acid concentrations, with but one exception, for both regimens (Tables 42 and 43). Within 0 and 5 percent fat levels for both regimens increasing ration concentrates decreased ( $P < .05$ ) acetic and increased

Table 37. Rumen sample pH for ad libitum feed intakes by concentrate levels within added fat level. Experiment I.

% Added Fat	% Concentrate		
	60	75	90
0	6.67 <sup>a</sup>	6.45 <sup>a,b</sup>	5.80 <sup>b</sup>
5	6.73	6.40	6.02
10	6.08	6.28	5.83
15	6.15	6.07	5.70
Means	6.41 <sup>a</sup>	6.30 <sup>a</sup>	5.84 <sup>b</sup>

a,b. Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 38. Volatile fatty acid molar percentages<sup>a</sup> of rumen fluid samples for constant feed intakes by added fat level. Experiment I.

% Added Fat	Volatile Fatty Acids					
	C <sub>2</sub>	C <sub>3</sub>	iso-C <sub>4</sub>	C <sub>4</sub>	iso-C <sub>5</sub>	C <sub>5</sub>
0	56.7	29.1 <sup>b</sup>	1.1	9.8	1.5	1.7
5	54.5	29.5 <sup>b</sup>	1.8	10.4	2.6	1.2
10	51.0	35.1 <sup>c</sup>	1.0	9.5	1.9	1.1
15	51.0	35.7 <sup>c</sup>	1.1	9.4	1.4	1.4

a. Molar percent = moles per 100 moles.

b,c. Means with different superscripts within column are significantly different ( $P < .05$ ).

Table 39. Volatile fatty and molar percentages<sup>a</sup> of rumen fluid samples for ad libitum feed intakes by added fat level. Experiment I.

% Added Fat	Volatile Fatty Acids					
	C <sub>2</sub>	C <sub>3</sub>	iso-C <sub>4</sub>	C <sub>4</sub>	iso-C <sub>5</sub>	C <sub>5</sub>
0	53.2	32.0 <sup>b</sup>	1.0	10.3 <sup>b</sup>	1.4	2.0
5	50.3	38.8 <sup>c</sup>	0.9	7.0 <sup>c</sup>	1.3	1.7
10	48.1	39.3 <sup>c</sup>	0.9	8.8 <sup>b,c</sup>	1.2	1.5
15	49.4	36.7 <sup>b,c</sup>	1.1	9.2 <sup>b,c</sup>	1.6	2.0

a. Molar percent = moles per 100 moles.

b,c. Means with different superscripts within column are significantly different ( $P < .05$ ).

Table 40. Volatile fatty acid molar percentages<sup>a</sup> of rumen fluid samples for constant feed intakes by added fat levels within concentrate level. Experiment I.

Item	% Added Fat			
	0	5	10	15
<u>60% Concentrate Ration</u>				
C <sub>2</sub>	66.4 <sup>b</sup>	62.5 <sup>b</sup>	52.0 <sup>c</sup>	52.9 <sup>c</sup>
C <sub>3</sub>	18.7 <sup>b</sup>	21.7 <sup>b</sup>	31.5 <sup>c</sup>	33.3 <sup>c</sup>
iso-C <sub>4</sub>	1.3	0.9	1.3	1.5
C <sub>4</sub>	11.9	12.1	10.2	9.4
iso-C <sub>5</sub>	0.9	1.5	2.2	1.9
C <sub>5</sub>	0.9	1.1	1.3	1.1
<u>75% Concentrate Ration</u>				
C <sub>2</sub>	60.4	51.9	54.2	56.2
C <sub>3</sub>	25.2	34.7	32.0	33.8
iso-C <sub>4</sub>	1.3 <sup>b</sup>	3.2 <sup>c</sup>	1.1 <sup>b</sup>	1.0 <sup>b</sup>
C <sub>4</sub>	9.4	7.8	9.5	6.7
iso-C <sub>5</sub>	2.0	1.4	2.3	1.3
C <sub>5</sub>	1.8	1.1	1.1	1.0
<u>90% Concentrate Ration</u>				
C <sub>2</sub>	43.2	49.1	46.8	43.9
C <sub>3</sub>	43.5	32.2	41.7	40.0
iso-C <sub>4</sub>	1.0	1.3	0.8	0.9
C <sub>4</sub>	8.2	11.2	8.8	12.1
iso-C <sub>5</sub>	1.7 <sup>b</sup>	5.0 <sup>c</sup>	1.1 <sup>b</sup>	1.2 <sup>b</sup>
C <sub>5</sub>	2.4 <sup>b</sup>	1.2 <sup>c,d</sup>	0.9 <sup>d</sup>	2.0 <sup>b,c</sup>

a. Molar percent = moles per 100 moles.

b,c,d. Means with different superscripts within row are significantly different (P<.05).

Table 41. Volatile fatty acid molar percentages<sup>a</sup> of rumen fluid samples for ad libitum feed intakes by added fat levels within concentrate level. Experiment I.

Item	% Added Fat			
	0	5	10	15
<u>60% Concentrate Ration</u>				
C <sub>2</sub>	63.8 <sup>b</sup>	58.2 <sup>b</sup>	50.5 <sup>c</sup>	53.5 <sup>c</sup>
C <sub>3</sub>	22.1 <sup>b</sup>	28.7 <sup>b</sup>	36.8 <sup>c</sup>	35.4 <sup>c</sup>
iso-C <sub>4</sub>	0.7	0.6	0.6	0.7
C <sub>4</sub>	11.6	9.9	9.4	7.5
iso-C <sub>5</sub>	0.6	1.1	1.0	1.3
C <sub>5</sub>	1.2	1.4	1.6	1.5
<u>75% Concentrate Ration</u>				
C <sub>2</sub>	50.5	51.0	52.0	49.4
C <sub>3</sub>	36.6	38.6	34.8	36.8
iso-C <sub>4</sub>	1.2	1.2	1.0	1.5
C <sub>4</sub>	8.2	6.4	9.1	8.5
iso-C <sub>5</sub>	1.5	1.2	1.4	1.8
C <sub>5</sub>	2.0	1.5	1.5	2.0
<u>90% Concentrate Ration</u>				
C <sub>2</sub>	45.3	41.8	41.8	45.3
C <sub>3</sub>	37.5 <sup>b</sup>	49.0 <sup>c</sup>	46.2 <sup>c</sup>	37.8 <sup>b</sup>
iso-C <sub>4</sub>	1.0	1.0	1.1	1.2
C <sub>4</sub>	11.2 <sup>b</sup>	4.7 <sup>c</sup>	8.0 <sup>b,c</sup>	11.5 <sup>b</sup>
iso-C <sub>5</sub>	2.0	1.4	1.2	1.8
C <sub>5</sub>	3.0	2.2	1.6	2.5

a. Molar percent = moles per 100 moles.

b,c. Means with different superscripts within row are significantly different (P<.05).



Table 42. Volatile fatty acid molar percentages<sup>a</sup> of rumen fluid samples for constant feed intakes by concentrate level. Experiment I.

% Concentrate	V o l a t i l e F a t t y A c i d s					
	C <sub>2</sub>	C <sub>3</sub>	iso-C <sub>4</sub>	C <sub>4</sub>	iso-C <sub>5</sub>	C <sub>5</sub>
60	58.4 <sup>b</sup>	26.3 <sup>b</sup>	1.2	10.9	1.6	1.1
75	55.7 <sup>b</sup>	31.4 <sup>c</sup>	1.6	8.3	1.8	1.2
90	45.8 <sup>c</sup>	39.3 <sup>d</sup>	1.0	10.1	2.2	1.0

a. Molar percent = moles per 100 moles.

b,c,d. Means with different superscripts within column are significantly different ( $P < .05$ ).

Table 43. Volatile fatty acid molar percentages<sup>a</sup> of rumen fluid samples for ad libitum feed intakes by concentrate level. Experiment I.

% Concentrate	V o l a t i l e F a t t y A c i d s					
	C <sub>2</sub>	C <sub>3</sub>	iso-C <sub>4</sub>	C <sub>4</sub>	iso-C <sub>5</sub>	C <sub>5</sub>
60	56.5 <sup>b</sup>	30.8 <sup>b</sup>	0.7 <sup>b</sup>	9.6	1.0 <sup>b</sup>	1.4
75	50.7 <sup>c</sup>	36.7 <sup>c</sup>	1.2 <sup>c</sup>	8.0	1.5 <sup>c</sup>	1.7
90	43.5 <sup>d</sup>	42.6 <sup>d</sup>	1.1 <sup>c</sup>	8.8	1.6 <sup>c</sup>	2.3

a. Molar percent = moles per 100 moles.

b,c,d. Means with different superscripts within column are significantly different ( $P < .05$ ).

( $P < .05$ ) propionic acid concentrations (Tables 44 and 45). The same trends were evident for 10 and 15 percent fat levels but they were significant ( $P < .05$ ) only for acetic acid within 15 percent constant feed rations.

Upon examination of the volatile fatty acid data, one finds a small but rather consistent trend for acetic, isobutyric, butyric, and isovaleric acid levels to be higher for constant feed periods, while propionic and valeric acids tended to be present in larger proportions for ad libitum feed periods. However, in general, corresponding values were similar.

Esplin et al. (53) reported little change in molar proportions of the various volatile fatty acids upon addition of 4 percent fat to a 70 percent concentrate ration. This is in agreement with the work reported here, in that few differences in the proportions of volatile fatty acids were noted between 0 and 5 percent added fat rations. Putnam et al. (143) reported that oil additions to all concentrate diets increased the proportion of propionate at the expense of acetate, which also concurs with the results of this experiment.

Correlations (calculated from residual sums-of-squares and cross products) between volatile fatty acid concentrations for both regimens revealed that molar percents (moles per 100 moles) acetic acid were negatively correlated ( $P < .01$ ) with molar percents of propionic and valeric acids (Tables 46 and 47). For constant feed periods propionic acid concentrations were negatively correlated ( $P < .01$ ) with isovaleric and positively correlated with valeric acid levels. For ad libitum feed periods

Table 44. Volatile fatty acid molar percentages<sup>a</sup> of rumen fluid samples for constant feed intakes by concentrate levels within added fat level. Experiment I.

Item	% Concentrate		
	60	75	90
<u>0% Added Fat Ration</u>			
C <sub>2</sub>	66.4 <sup>b</sup>	60.4 <sup>b</sup>	43.2 <sup>c</sup>
C <sub>3</sub>	18.7 <sup>b</sup>	25.2 <sup>b</sup>	43.5 <sup>c</sup>
iso-C <sub>4</sub>	1.3	1.3	1.0
C <sub>4</sub>	11.9	9.4	8.2
iso-C <sub>5</sub>	0.9	2.0	1.7
C <sub>5</sub>	0.9 <sup>b</sup>	1.8 <sup>b,c</sup>	2.4 <sup>c</sup>
<u>5% Added Fat Ration</u>			
C <sub>2</sub>	62.5 <sup>b</sup>	51.9 <sup>c</sup>	49.1 <sup>c</sup>
C <sub>3</sub>	21.7 <sup>b</sup>	34.7 <sup>c</sup>	32.2 <sup>b,c</sup>
iso-C <sub>4</sub>	0.9 <sup>b</sup>	3.2 <sup>c</sup>	1.3 <sup>b</sup>
C <sub>4</sub>	12.1	7.8	11.2
iso-C <sub>5</sub>	1.5 <sup>b</sup>	1.4 <sup>b</sup>	5.0 <sup>c</sup>
C <sub>5</sub>	1.1	1.1	1.2
<u>10% Added Fat Ration</u>			
C <sub>2</sub>	52.0	54.2	46.8
C <sub>3</sub>	31.5	32.0	41.7
iso-C <sub>4</sub>	1.3	1.1	0.8
C <sub>4</sub>	10.2	9.5	8.8
iso-C <sub>5</sub>	2.2	2.3	1.1
C <sub>5</sub>	1.3	1.1	0.9
<u>15% Added Fat Ration</u>			
C <sub>2</sub>	52.9 <sup>b</sup>	56.2 <sup>b</sup>	43.9 <sup>c</sup>
C <sub>3</sub>	33.3	33.8	40.0
iso-C <sub>4</sub>	1.5	1.0	0.9
C <sub>4</sub>	9.4	6.7	12.1
iso-C <sub>5</sub>	1.9	1.3	1.2
C <sub>5</sub>	1.1	1.0	2.0

a. Molar percent = moles per 100 moles.

b,c. Means with different superscripts within row are significantly different ( $P < .05$ ).

Table 45. Volatile fatty acid molar percentages<sup>a</sup> of rumen fluid samples for ad libitum feed intakes by concentrate level within added fat levels. Experiment I.

Item	% Concentrate		
	60	75	90
<u>0% Added Fat Ration</u>			
C <sub>2</sub>	63.8 <sup>b</sup>	50.5 <sup>c</sup>	45.3 <sup>c</sup>
C <sub>3</sub>	22.1 <sup>b</sup>	36.6 <sup>c</sup>	37.5 <sup>c</sup>
iso-C <sub>4</sub>	0.7	1.2	1.0
C <sub>4</sub>	11.6 <sup>b</sup>	8.2	11.2
iso-C <sub>5</sub>	0.6 <sup>b</sup>	1.5 <sup>c</sup>	2.0 <sup>c</sup>
C <sub>5</sub>	1.2	2.0	3.0
<u>5% Added Fat Ration</u>			
C <sub>2</sub>	58.2 <sup>b</sup>	51.0 <sup>b</sup>	41.8 <sup>c</sup>
C <sub>3</sub>	28.7 <sup>b</sup>	38.6 <sup>c</sup>	49.0 <sup>d</sup>
iso-C <sub>4</sub>	0.6 <sup>b</sup>	1.2 <sup>c</sup>	1.0 <sup>b,c</sup>
C <sub>4</sub>	9.9 <sup>b</sup>	6.4 <sup>b,c</sup>	4.7 <sup>c</sup>
iso-C <sub>5</sub>	1.1	1.2	1.4
C <sub>5</sub>	1.4	1.5	2.2
<u>10% Added Fat Ration</u>			
C <sub>2</sub>	50.5 <sup>b</sup>	52.0 <sup>b</sup>	41.8 <sup>c</sup>
C <sub>3</sub>	36.8 <sup>b</sup>	34.8 <sup>b</sup>	46.2 <sup>c</sup>
iso-C <sub>4</sub>	0.6	1.0	1.1
C <sub>4</sub>	9.4	9.1	8.0
iso-C <sub>5</sub>	1.0	1.4	1.2
C <sub>5</sub>	1.6	1.5	1.6
<u>15% Added Fat Ration</u>			
C <sub>2</sub>	53.5	49.4	45.3
C <sub>3</sub>	35.4 <sup>b</sup>	36.8 <sup>c</sup>	37.8 <sup>c</sup>
iso-C <sub>4</sub>	0.7 <sup>b</sup>	1.5 <sup>c</sup>	1.2 <sup>c</sup>
C <sub>4</sub>	7.5	8.5	11.5
iso-C <sub>5</sub>	1.3	1.8	1.8
C <sub>5</sub>	1.5	2.0	2.5

a. Molar percent = moles per 100 moles.

b,c,d. Means with different superscripts within row are significantly different (P<.05).

Table 46. Correlations of rumen sample volatile fatty acids<sup>a</sup> for constant feed periods.<sup>b</sup> Experiment I.

Variable	C <sub>3</sub>	iso-C <sub>4</sub>	C <sub>4</sub>	iso-C <sub>5</sub>	C <sub>5</sub>
C <sub>2</sub>	-.890**	0.006	-.245	0.373	-.673**
C <sub>3</sub>	...	-.051	-.118	-.530**	0.548**
iso-C <sub>4</sub>	...	...	-.327	0.212	0.064
C <sub>4</sub>	...	...	...	-.156	0.068
iso-C <sub>5</sub>	...	...	...	...	0.016

\*\* (P<.01).

a. Expressed as molar percent (moles per 100 moles).

b. Correlations calculated from residual sums-of-squares and cross products.

Table 47. Correlations of rumen sample volatile fatty acids<sup>a</sup> for ad libitum feed periods.<sup>b</sup> Experiment I.

Variable	C <sub>3</sub>	iso-C <sub>4</sub>	C <sub>4</sub>	iso-C <sub>5</sub>	C <sub>5</sub>
C <sub>2</sub>	-.852**	0.141	-.329	0.388	-.722**
C <sub>3</sub>	...	-.301	-.193	-.431*	0.373
iso-C <sub>4</sub>	...	...	0.202	0.322	-.142
C <sub>4</sub>	...	...	...	-.118	0.589**
iso-C <sub>5</sub>	...	...	...	...	-.317

\*, (P<.05).

\*\*, (P<.01).

a. Expressed as molar percent (moles per 100 moles).

b. Correlations calculated from residual sums-of-squares and cross products.

propionate levels were negatively correlated ( $P < .01$ ) with isovalerate, and butyric acid was positively correlated ( $P < .01$ ) with valeric acid levels.

#### Digestible Energy Intake

Digestible energy intake ( $y$ ) as a function of dietary lipid levels ( $x$ ), within each concentrate level, was studied with multiple regression analysis for power functions and sums of power functions through the fourth power to determine the dietary fat level at maximum digestible energy intake per 100 lb of body weight. The limited number of observations and substantial experimental variation precluded the possibility of determining whether these curves were exponential, logarithmic, or polynomial in nature, and the polynomial form was accepted.

For 60 percent concentrate rations, the equation of best fit (by the least squares method) was  $y = 4651.7 - 0.00846x^4$ . The regression was significant ( $P < .01$ ), as shown in Table 71, and the coefficient of  $x$  was different ( $P < .01$ ) from zero. (Tables 57 through 83 appear in the Appendix.) The coefficient of determination,  $r^2$ , was 0.71 and the simple correlation,  $r = -.84$ , between dietary fat level and digestible energy intake was significant ( $P < .01$ ). Maximum digestible energy intake, 4652 kcal per 100 lb of body weight for the 5-day collection period was calculated to occur at 0 percent dietary lipid. However, examination of Figure 1 shows that essentially no difference (11 kcal) in digestible energy intake was calculated to occur between 0 and 6 percent dietary lipid.

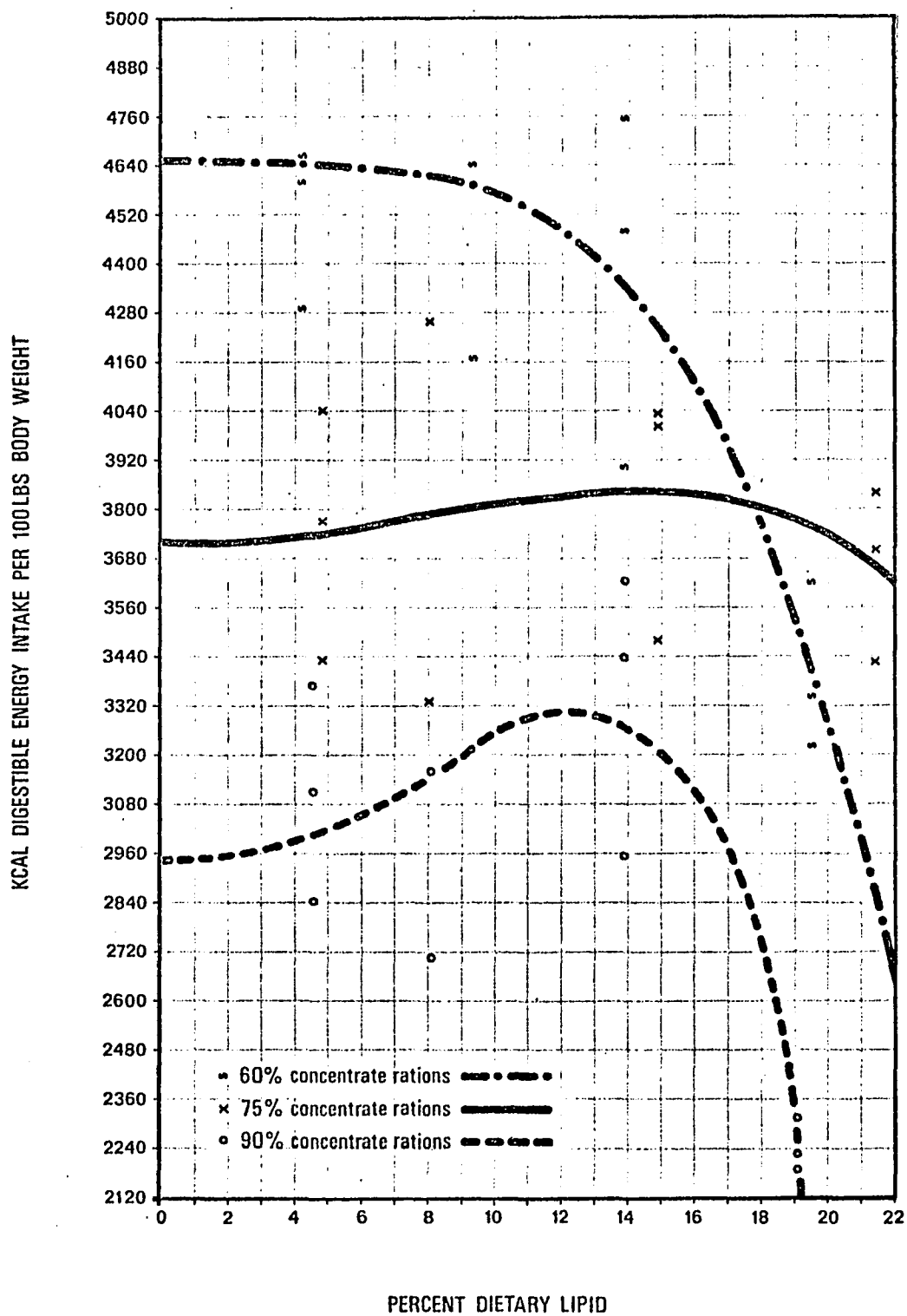


Figure 1. Digestible energy intake as a function of dietary lipid.



Although the quartic function best fit the data, first and second power power functions and sums of the three power functions fit the data nearly as well. Since more than one curve adequately explained these data, care should be exercised in the interpretation of the data around the fourth power expression offered. Each of the three curves achieved its maximum at  $x = 0$ , implying that the highest level of digestible energy consumption would have occurred at 0 percent dietary lipid.

For 75 percent concentrate rations the equation of best fit was  $y = 3727.1 - 0.00277x^4 + 1.12x^2$ . However, regression coefficients were not different ( $P > .05$ ) from zero and were therefore rejected. The regression was not significant ( $P > .05$ ), as is shown in Table 71, and the coefficient of determination was 0.55. Maximum digestible energy intake was calculated to occur at 14.22 percent dietary lipid. The value was 3840 kcal per 100 lb of body weight for the 5-day collection period--much lower than the corresponding figure of 4652 kcal for 60 percent concentrate rations. A graph of the data is presented in Figure 1.

The equation of best fit for 90 percent concentrate rations was  $y = 2952.9 - 0.043x^4 + 0.72x^3$ . Both coefficients were different ( $P < .05$ ) from zero and were therefore accepted. The regression was significant at the 1 percent level of probability (Table 71), and the coefficient of determination was 0.74. Maximum digestible energy intake per 100 lb of body weight was calculated to occur at 12.59 percent dietary lipid, and for the 5-day collection period the value was 3309 kcal, considerably lower than the values of 4652 and 3840 kcal for 60 and 75 percent

rations, respectively. As Figure 1 shows, digestible energy intake declined rather rapidly on either side of the maximum level. The previously mentioned fact that two of three steers consuming the 0 percent fat ration dramatically reduced their feed intake must be considered when interpreting this data.

Because these data were obtained during collection periods and therefore during restricted feeding, their validity may be questioned. However, feeding levels were approximately 90 percent of ad libitum intakes, and therefore differences in digestible energy intake between treatments may be real, although absolute values may be somewhat misleading.

### Discussion

A serious fault of the experimental design, dictated by the objectives of another study, was the confounding of ration concentrate level and time. This precluded a "clean" appraisal of the influence of concentrate level on the parameters examined. While time periods involved for each concentrate level were relatively short, and one would seem justified in biologically overlooking this, there is still the problem of carry-over effects from ration to ration which cannot be assessed. These facts should be considered when interpreting the data reported in this experiment.

In general, the performance data reported here parallels that found in the literature, lending credibility to these results. As in the literature, one problem associated with the feeding of fats, as far

as performance was concerned, appeared to be decreasing feed intake with increasing level of dietary fat.

As dietary fat level increased, lipid ingested per unit of body weight increased, while as ration concentrate level increased, the quantity of lipid ingested per 100 lb of body weight decreased. The latter was a function of reduced feed intake at higher concentrate levels. Similarly, the quantity of ingested lipid for the various fat levels was considerably reduced with each increase in ration concentrate level. For example, cattle consuming the 60 percent concentrate, 5 percent fat ration actually ingested more lipid per unit of body weight than did cattle consuming 90 percent concentrate rations at any fat level. Within each fat level, 90 percent concentrate rations resulted in the lowest quantity of ingested lipid per unit of body weight. Yet cattle, when fed this concentrate level most often, and to the greatest extent, exhibited decreased digestibilities of ration components and depressed performance. Thus the quantity of ingested lipid did not appear to be the factor limiting these two parameters. The factor responsible seemed intrinsically related to concentrate, or its converse, roughage level of the diet. This conclusion is supported by the data of Nieman (138) which showed that only gram-positive microorganisms are susceptible to the action of small amounts of fatty acids. Hungate (92) reported that the maximum proportion of such organisms in the rumen digesta occurs with animals fed high grain rations. Additional supporting data were reported by Cameron and Hogue (32) who observed a substantial depression of food consumption and rate of gain

when sheep fed 15 percent corn oil were given 10 rather than 50 and 90 percent roughage diets.

If this conclusion is valid, perhaps the common commercial feedlot practices of feeding a constant proportion of fat, irrespective of diet, or of increasing the proportion of dietary fat to 4 or 5 percent as the grain content of the diet increases, should be revised. Indeed, it appears from the results of this work and the supporting data that these practices should be altered so that a larger proportion of lipid is added to low rather than high concentrate diets. That ration concentrate level should be a consideration in the use of feeding fats for ruminants bears further study.

Lipid digestibility and excreted lipid as a percent of ingested lipid declined with increasing fat level, while fecal soap excretion as a percent of ingested lipid increased. Lipid digestibility and fecal soap excretion were in fact highly correlated ( $P < .01$ ),  $r = -.941$  (calculated from residual sums-of-squares and cross products).

Throughout the study it appeared that ration concentrate level had as much influence as dietary fat level on the variables studied. Digestibilities for the 75 percent concentrate rations were consistently superior to those for other concentrate levels, which bears further investigation.

The substantial reduction in feed consumed at both high concentrate and fat levels undoubtedly biased the digestion coefficients reported. If all rations had been fed at the same proportion of body weight, digestibilities of high concentrate and high fat diets would

have probably been lower with respect to the other diets than reported.

## Experiment II

### General Considerations

The range of feed intake attained in this experiment was not as large as desired--only 0.92 to 2.04 times the calculated maintenance energy requirement (Table 48). Average daily gains were quite variable and, as would be expected, were closely related to level of feed intake. Gains for the highest level of intake were normal. The feed intake range was not expanded into submaintenance levels, because it has been shown (12, 57, 58, 71, 97) that the feed intake, digestibility relationship below maintenance apparently differs from that above maintenance. Differences in the amount of feed ingested between and among the two replications within a period or particular level of intake precluded valid summation across either periods or feed intake levels. Thus, for each digestibility parameter, comparing means of periods or levels of intake was questionable, and analysis of variance based on the Latin-square experimental design was not possible. Instead, level of feed consumption (x) was handled as a continuous variable and digestion coefficients (y) were regressed on this in order to determine the influence of level of feed intake on digestibility parameters.

Feed consumption as a percent of body weight also shows that maximum feed intake (2.28 percent) was low (Table 72). The narrow range of feed intakes achieved in this experiment was of fundamental importance in that polynomial, exponential and logarithmic expressions

Table 48. Level of energy consumption times the calculated maintenance level of nutrition. Experiment II.<sup>a</sup>

Level of Intake <sup>b</sup>	P e r i o d s				Means
	1	2	3	4	
A	0.98 (1) <sup>c</sup>	1.01 (2)	0.97 (3)	0.96 (4)	0.97
	0.98 (5)	0.92 (8)	0.98 (6)	0.97 (7)	
B	1.33 (2)	1.36 (1)	1.28 (4)	1.22 (3)	1.30
	1.30 (6)	1.28 (7)	1.26 (5)	1.34 (8)	
C	1.58 (3)	1.63 (4)	1.58 (1)	1.44 (2)	1.54
	1.49 (7)	1.64 (6)	1.53 (8)	1.45 (5)	
D	1.91 (4)	2.04 (3)	1.74 (2)	1.73 (1)	1.80
	1.74 (8)	1.88 (5)	1.66 (7)	1.75 (6)	

a. Ninety percent dry matter basis.

b. Approximate levels of energy consumption: A = 1.0, B = 1.3, C = 1.5, and D = 1.8 times maintenance.

c. Heifer number.

of linear, quadratic, cubic or quartic order behave generally in the same manner over the range of  $x$  equals 1 to 2, the range of feed intake studied. For this reason, it was not possible to fit the data with a specific mathematical expression from which the influence of feed intake on digestibility parameters could be calculated. Therefore it was assumed that polynomial expressions best fit the data, and linear through quartic expressions of this kind were calculated in order to compare characteristics of these expressions in relation to the data.

Each of the power functions fitted to dry matter, gross energy, and crude protein digestibility data by the least squares method, first through fourth powers, explained the data equally well. Each accounted for approximately an equal proportion of the variation ( $r^2$ ), had a significant correlation coefficient ( $P < .01$ ) not different ( $P > .05$ ) from other correlations, and had a significant regression ( $P < .01$ ). Regression coefficients were different ( $P < .01$ ) from zero, lending to validity of the equations. Because of the similarity in correlation coefficients, the addition of any or all of the power functions to other power functions produced nonsignificant improvements ( $P > .05$ ) in estimation regressions. Thus it was impossible to determine the mathematical relationship between level of feeding and digestibilities for the three digestibility parameters studied. This made prediction beyond the data to higher values of feed consumption precarious, since the equations fitting the data respond differently at higher values of  $x$ .

### Dry Matter Digestibility

Dry matter digestibility declined rather consistently from lowest to highest levels of feed intake, and the mean magnitude of this change was approximately 4.5 percentage units (Table 49). Increments of change in digestibility when feed intake increased from 1 to 2 times maintenance were calculated from the four expressions to be -5.4 to -6.3 percentage units (Table 50).

Feed intake for the lowest feeding level ranged from 0.92 to 1.01, with a mean of 0.97 times maintenance (Table 48). Mean dry matter digestibility at the lowest feeding level was 85.4 percent (Table 45). For a feed intake of 0.97 times maintenance, digestibility calculated from the linear expression (Table 50) was less than 80 percent, while for the three curvilinear expressions, digestibilities were approximately 85 percent, very close to the determined value. At the highest level of feed intake, which varied between 1.73 and 2.04, with a mean of 1.81 times maintenance (Table 48), dry matter digestibility averaged 80.9 percent (Table 49). Digestibility for the 1.81 times maintenance level of feed intake was calculated from the linear polynomial (Table 50) to be 75.3 percent, but again the curvilinear expressions all gave values almost identical to that observed for the highest feeding level. This evidence provided strong support for the theory which defines the relationship between feed intake and dry matter digestibility as curvilinear. This is in agreement with the work of Leaver et al. (105) and Blaxter and Wainman (15), who found this relationship for organic matter to be curvilinear and is in disagreement



Table 49. Dry matter digestion coefficients. Experiment II.

Level of Intake <sup>a</sup>	P e r i o d s				Means
	1	2	3	4	
A	83.2 (1) <sup>b</sup>	84.8 (2)	84.9 (3)	87.1 (4)	85.4
	84.8 (5)	87.2 (8)	86.0 (6)	85.5 (7)	
B	82.4 (2)	83.8 (1)	80.3 (4)	86.3 (3)	83.6
	81.2 (6)	79.1 (7)	85.6 (5)	87.9 (8)	
C	82.3 (3)	80.2 (4)	84.1 (1)	86.2 (2)	83.1
	84.2 (7)	80.3 (6)	83.6 (8)	87.5 (5)	
D	77.9 (4)	80.0 (3)	81.5 (2)	82.9 (1)	80.9
	79.1 (8)	81.0 (5)	83.5 (7)	81.5 (6)	

a. Approximate levels of energy consumption: A = 1.0, B = 1.3, C = 1.5, and D = 1.8 times maintenance.

b. Heifer number.

Table 50. Characteristics of expressions fitting dry matter digestibility data. Experiment II.

Item	Order of Expression			
	x	x <sup>2</sup>	x <sup>3</sup>	x <sup>4</sup>
Expression	$y = -5.38x + 85.04$	$y = 1.92x^2 + 87.30$	$y = -0.87x^3 + 86.09$	$y = -0.42x^4 + 85.45$
Coefficient of determination-r <sup>2</sup>	0.398	0.409	0.411	0.404
Correlation coefficient-r	-.631**	-.640**	-.641**	-.636**
Increment of change <sup>a</sup>				
Expression	b	2cx+c	3dx <sup>2</sup> +3dx+d	4ex <sup>3</sup> +6ex <sup>2</sup> +4ex+e
Percentage units	-5.4	-5.8	-6.1	-6.3
% Relative change <sup>b</sup>	-6.3	-6.6	-7.2	-7.3
ANOVA	df	Mean Squares		
Regression	1	93.07**	95.56**	95.99**
Residual	30	4.68	4.60	4.59

\*\*, (P<.01).

a. Change in digestibility (y) for change in feed intake (x) from 1 to 2.

b. Change in digestibility for x equals 1 to 2 based on a, the expression constant.

with Brown (27), who reported that a linear relationship existed for the dry matter of both 50 and 80 percent grain diets.

Data reported here are in agreement with the literature in that dry matter digestibility and feed consumption were negatively associated (2,11,19,27,49,50,59,67,126,135,151,167). The 4.5 percentage unit decline in digestibility observed for less than a twofold increase in feed intake, and the mean calculated decline of approximately 5.9 percentage units for a doubling of feed consumption from the maintenance level are somewhat higher than values reported in the literature. Truter and Louw (167) noted a decline of 2.5 percentage units when the intake of a 50 percent concentrate diet increased from maintenance to twice this level. Forbes et al. (59) reported the mean decline in digestibilities of 50 and 66 percent concentrate diets, when feed consumption increased from 1 to 2 times maintenance, was 3.1 percentage units. Brown (27) doubled feed intake from the maintenance level and noted 3.8 and 2.0 percentage unit decreases in dry matter digestibilities for 80 and 50 percent concentrate rations, respectively. The greater decreases in digestion coefficients observed here may perhaps be explained by the fact that corresponding values in the literature were obtained with lower than the 90 percent concentrate diet employed here. Leaver et al. (105), Brown (27) and Bloom et al. (19) reported greater depressions in digestibilities of dry or organic matter as the proportions of concentrates in the diet increased.

### Gross Energy Digestibility

Mean gross energy digestion coefficients decreased 5.0 percentage units from the lowest to the highest levels of feed consumption (Table 51). When feed intake was doubled to 2 times maintenance, gross energy digestibility was calculated to decrease by -6.0 to -7.1 percentage units (Table 52).

For feed intakes at the low feeding level (mean equal to 0.97 times maintenance, Table 48), gross energy digestibility averaged 84.0 percent (Table 51). Digestibilities calculated for 0.97 times maintenance from linear and curvilinear expressions (Table 52) fitted to the data were within 1 percentage unit of determined values. For the highest level of feed intake, which averaged 1.81 times maintenance (Table 48), mean gross energy digestibility was 79.0 percent (Table 51). Digestibilities calculated from linear and curvilinear expressions (Table 52) for the 1.81 times maintenance level of energy nutrition were all within 1 percentage unit of that observed for the highest feeding level. Although the cubic function fit the data better than did other expressions, any differences were minute, making assessment of the mathematical relationship between feed consumption and gross energy digestibility impossible. Forbes et al. (57) studied this relationship for a 50 percent concentrate diet and found it to be curvilinear in nature.

Although the lack of mathematical definition was unfortunate, calculated decreases in gross energy digestibility were similar for all expressions fitted. The observation that gross energy digestibility

Table 51. Gross energy digestion coefficients. Experiment II.

Level of Intake <sup>a</sup>	P e r i o d s				Means
	1	2	3	4	
A	81.3 (1) <sup>b</sup>	83.5 (2)	82.4 (3)	85.7 (4)	84.0
	85.1 (5)	85.5 (8)	84.5 (6)	84.0 (7)	
B	82.2 (2)	81.9 (1)	78.6 (4)	84.7 (3)	82.1
	81.6 (6)	77.6 (7)	83.7 (5)	86.4 (8)	
C	82.2 (3)	78.5 (4)	81.3 (1)	84.6 (2)	82.1
	82.9 (7)	79.3 (6)	82.0 (8)	86.1 (5)	
D	71.4 (4)	79.4 (3)	79.4 (2)	80.7 (1)	79.0
	79.0 (8)	80.0 (5)	82.5 (7)	79.6 (6)	

a. Approximate levels of energy consumption: A = 1.0, B = 1.3, C = 1.5, and D = 1.8 times maintenance.

b. Heifer number.

Table 52. Characteristics of expressions fitting gross energy digestibility data. Experiment II.

Item	Order of Expression			
	x	x <sup>2</sup>	x <sup>3</sup>	x <sup>4</sup>
Expression	$y = -5.96x + 90.17$	$y = 2.15x^2 + 86.25$	$y = -0.98x^3 + 84.92$	$y = -0.48x^4 + 85.06$
Coefficient of determination-r <sup>2</sup>	0.383	0.401	0.410	0.410
Correlation coefficient-r	-.619**	-.633**	-.640**	-.639**
Increment of change <sup>a</sup>				
Expression	b	2cx+c	3dx <sup>2</sup> +3dx+d	4ex <sup>3</sup> +6ex <sup>2</sup> +4ex+e
Percentage units	-6.0	-6.4	-6.9	-7.1
% Relative change <sup>b</sup>	-6.6	-7.5	-8.2	-8.4
ANOVA	df	Mean Squares		
Regression	1	114.10**	119.42**	121.99**
Residual	30	6.12	5.94	5.85

\*\* (P<.01).

a. Change in digestibility (y) for change in feed intake (x) from 1 to 2.

b. Change in digestibility for x equals 1 to 2 based on a, the expression constant.

declined with increasing levels of feed consumption is in agreement with a substantial proportion of the literature (15,16,57,58,59,67,112,141). The 5.0 percentage unit decline in digestibility determined for less than a twofold increase in feed intake and the mean calculated decline of 6.6 percentage units for doubling of feed consumption to 2 times the maintenance level were higher than the only value found in the literature. Blaxter and Wainman (15) observed that gross energy digestibility of a 33 percent concentrate diet declined 1.4 percentage units when feed intake was doubled to 2 times maintenance. Moe, Tyrrell, and Reid (128) reported ration total digestible nutrients declined 23 relative percent when the intake of feed for an unspecified concentrate level increased from 1 to 6 times maintenance. Explanation of the greater depression in gross energy digestibility here may again be involved with the higher concentrate ration utilized.

#### Crude Protein Digestibility

Mean apparent crude protein digestibility decreased 8.9 percentage units from the lowest to highest level of feed intake (Table 53). Decreases in digestibility when feed intake was raised from 1 to 2 times maintenance were calculated from the four expressions to be 10.3 to 11.6 percentage units.

Feed intake at the lowest level averaged 0.97 times maintenance (Table 48), and the mean crude protein digestibility for this level was 71.3 percent (Table 53). Digestibilities calculated for 0.97 times maintenance from linear and curvilinear expressions (Table 54) fitted to the data by the method of least squares were within about 1 percentage

Table 53. Crude protein digestion coefficients. Experiment II.

Level of Intake <sup>a</sup>	P e r i o d s				Means
	1	2	3	4	
A	67.0 (1) <sup>b</sup>	72.0 (2)	67.5 (3)	76.1 (4)	71.3
	75.3 (5)	73.9 (8)	69.1 (6)	69.3 (7)	
B	66.8 (2)	68.7 (1)	60.4 (4)	71.2 (3)	66.5
	66.2 (6)	58.2 (7)	67.5 (5)	76.3 (8)	
C	64.8 (3)	59.5 (4)	67.5 (1)	71.4 (2)	66.9
	67.7 (7)	62.2 (6)	67.2 (8)	75.2 (5)	
D	58.1 (4)	60.8 (3)	59.5 (2)	62.2 (1)	62.4
	62.6 (8)	64.1 (5)	67.8 (7)	63.5 (6)	

a. Approximate levels of energy consumption: A = 1.0, B = 1.3, C = 1.5, and D = 1.8 times maintenance.

b. Heifer number.



Table 54. Characteristics of expressions fitting crude protein digestibility data. Experiment II.

Item	Order of Expression				
	x	x <sup>2</sup>	x <sup>3</sup>	x <sup>4</sup>	
Expression	y = -10.31x + 81.25	y = -3.65x <sup>2</sup> + 74.32	y = -1.63x <sup>3</sup> + 71.96	y = -0.78x <sup>4</sup> + 70.73	
Coefficient of determination-r <sup>2</sup>	0.386	0.388	0.381	0.367	
Correlation coefficient-r	-.621**	-.623**	-.617**	-.606 <sup>II</sup>	
Increment of change <sup>a</sup>					
Expression	b	b	2cx+c	3dx <sup>2</sup> +3dx+d	4ex <sup>3</sup> +6ex <sup>2</sup> +4ex+e
Percentage units	-10.3	-11.0	-11.4	-11.6	
% Relative change <sup>b</sup>	-12.7	-15.5	-15.8	-16.4	
ANOVA	df	Mean Squares			
Regression	1	341.41**	342.94**	337.23**	325.04**
Residual	30	18.11	18.06	18.25	18.66

\*\* (P<.01).

a. Change in digestibility (y) for change in feed intake (x) from 1 to 2.

b. Change in digestibility for x equals 1 to 2 based on a, the expression constant.

unit of the determined value. At the highest level of feed intake, which averaged 1.81 times maintenance (Table 48), mean crude protein digestibility was 62.4 percent (Table 53). Digestibilities calculated from the linear and curvilinear expressions (Table 54) for the 1.81 times maintenance feeding level) were within 0.5 percentage unit of the determined digestibility for the highest feeding level. The data were best fit by the quadratic expression, although differences between it and other expressions in this regard were so small as to be inconsequential.

The data did not permit the assignment of a definite mathematical expression to the relationship between feed intake level and crude protein digestibility. That increased feed intake depresses crude protein digestibility concurs with the literature (16,19,49,57,58,59,67,126,135,151,167,180). The mean calculated depression in protein digestibility of about 11.1 percentage units was much larger than the value of 5.2 percentage units reported by Truter and Louw (167) with a 50 percent concentrate diet under the same feeding regimen. Crude protein digestibility declined to a greater extent than did dry matter and gross energy digestibilities. This is in agreement with Watson et al. (180), who observed that crude protein digestibility was depressed to a greater extent than was any other ration constituent. Similarly, Forbes et al. (57,58) reported that depressed digestibility of the ration was due mainly to nitrogen-free extract and crude protein fractions.

## Discussion

Feed consumption was limited to such a narrow range as to preclude mathematical definition of the relationship between level of feed consumption and ration component digestibilities. However, the apparent trend (especially for dry matter digestibility) was for these relationships to be curvilinear in nature.

The depressing influence of increasing level of feed intake on the digestibilities of high concentrate rations as noted in this work and in the literature, within the limits studied, appears unassailable.

## Experiment III

### Digestibility Studies

Diet crude protein level affected ( $P < .01$ ) the apparent digestibilities of dry matter, gross energy, and crude protein (Table 55). Dry matter and gross energy digestibilities were approximately 5 percentage units greater for the highest than lowest protein level diets (Table 56). Apparent crude protein digestibility increased approximately 20 percentage units from low to high protein diets, and this was the largest increase of the three ration components examined. For each component studied there was a consistent trend toward higher digestion coefficients with increasing crude protein content of the ration, although a leveling off was noted between the two highest protein levels.

Linear regression, with ration crude protein level as the independent variable and dry matter digestibility as the dependent variable, generated the mathematical relationship  $y = 69.29 + 0.76x$ . Thus for each increase of 1 percentage unit in ration crude protein, dry matter

Table 55. Analyses of variance of ration digestibilities.  
Experiment III.

Source of Variation	df	M e a n   S q u a r e s		
		Dry Matter	Gross Energy	Crude Protein
% Protein	3	15.88**	21.64**	296.84**
Error	12	1.60	2.68	7.54

\*\*<sub>1</sub>. ( $P < .01$ ).

Table 56. Digestion coefficients by level of ration protein.  
Experiment III.

Source of Variation	P r o t e i n   L e v e l <sup>a</sup>			
	A	B	C	D
Dry Matter	76.8 <sup>b</sup>	78.8 <sup>c</sup>	80.7 <sup>d</sup>	81.2 <sup>d</sup>
Gross Energy	75.6 <sup>b</sup>	77.5 <sup>b</sup>	79.4 <sup>c</sup>	80.9 <sup>c</sup>
Crude Protein	55.1 <sup>b</sup>	62.5 <sup>c</sup>	70.4 <sup>d</sup>	74.5 <sup>d</sup>

a. Approximate percent protein in ration: A = 10, B = 12, C = 15, and D = 16.

b,c,d. Means with different superscripts within row are significantly different ( $P < .05$ ).

digestibility increased by 0.76 percentage units. The simple correlation between these variables was 0.82 ( $P < .01$ ). To increase diet protein level, soybean oil meal was substituted for milo, and since dry matter digestibilities of the two feedstuffs are comparable (133), improvement in dry matter digestibility for higher protein diets appeared to result from an enhancement of the digestive process by additional protein. Improved dry matter digestibility with increasing protein content of the diet has been previously reported (30,63,98,184,185). Also concurring are Kay, Bowers, and G. McKiddie (96) and French, Glover, and Duthie (62), both of whom reported that dry matter digestibility increased, but at a decreasing rate, as protein content of the diet increased.

The linear mathematical relationship between diet crude protein level (independent variable) and gross energy digestibility (dependent variable) was  $y = 67.76 + 0.78x$ , similar to the corresponding expression for dry matter. Gross energy digestibility increased 0.78 percentage units for each 1 percentage unit increase in ration protein. The simple correlation between the two variables was 0.82 ( $P < .01$ ), identical to that between dry matter and ration protein. As was the case for dry matter, gross energy digestibilities of soybean oil meal and milo are comparable (133), so that improvement in gross energy digestibility of the higher protein (soybean oil meal added) rations appeared to be the result of improved digestive powers derived from the additional protein.

The equation of the regression line between percent ration crude protein ( $x$ ) and apparent crude protein digestibility ( $y$ ) was

$y = 26.34 + 2.90x$ . Apparent crude protein digestibility increased by 2.90 percentage units for each 1 percentage unit increase in ration crude protein. The simple correlation between these two factors was 0.96 ( $P < .01$ ). While soybean oil meal protein is more digestible than that of milo (133), this difference is relatively small and could not account for a substantial proportion of the improvement in apparent crude protein digestibility of the higher protein (soybean oil meal added) diets. This implied that additional dietary protein increased the efficiency of the digestive process. That increasing ration protein improves crude protein digestibility has been previously reported (63,96,98,185). French et al. (62), using a large quantity of world data, related  $y$ , apparent crude protein digestibility, to  $x$ , the percentage crude protein of feed dry matter, by the equation  $y = 70 \log x - 15$ . This equation also generally fit the data reported in this study. It was also noted that crude protein digestibility decreased at an increasing rate at higher levels of ration protein.

#### Metabolic Fecal Nitrogen Determinations

Data from this experiment and Experiment II were examined 10 ways with multiple regression analysis (linear through quartic) in an effort to establish metabolic fecal nitrogen levels by extrapolation to zero nitrogen intake and to determine the mathematical relationship between fecal nitrogen and dry matter intake and excretion. Equations calculated from the data were assumed to be polynomial in nature.

1. Total grams of nitrogen excreted (dependent variable) were regressed on total grams of nitrogen consumed (independent variable).

None of the power functions fitted to the data, first through the fourth power, or sums of these power functions, resulted in a significant regression ( $P > .05$ ). All simple correlations were less than 0.12, and were not different ( $P > .05$ ) from zero or from each other ( $P > .05$ ). The equation of best fit by the least squares method was  $y = 270.49 + 2.32x - 0.0034x^2$ . Regression coefficients were not different ( $P > .05$ ) from zero, and the coefficient of determination was only 14.95 percent. In addition, fecal nitrogen calculated by extrapolation to zero nitrogen intake was considerably less than zero, which was improbable. Therefore this regression equation was rejected.

This method was employed by Titus (166), who determined that the mathematical relationship between nitrogen excretion and consumption was  $y = 184.95 + 0.1447x$ , giving a metabolic fecal nitrogen value of 0.63 g nitrogen per 100 g food ingested. A possible explanation for the curvilinear response observed here, as opposed to that observed by Titus (166), is the employment of ration protein levels between 10 and 16 percent. French et al. (62) noted that increasing the level of dietary protein in higher protein diets resulted in increasing apparent crude protein digestibility at an ever decreasing rate. Titus (166), on the other hand, utilized ration protein levels of 5 to 13 percent, where linearity might more likely be expected.

2. When grams nitrogen per 100 g feed dry matter was used as the independent variable and grams fecal nitrogen per 100 g feed dry matter as the dependent variable, none of the first through fourth power power functions or sums of these power functions fitted to the

data resulted in a significant regression ( $P > .05$ ). Negative correlations were not different from zero ( $P > .05$ ), although the highest correlation, for  $x^4, y$  was  $r = -.45$ , which approached significance at the 5 percent level of probability. The expression  $y = 0.418 + 0.2099x - 0.0060x^4$  best fit the data, as shown by the coefficient of determination of 36.46 percent. The fourth power regression coefficient was significant ( $P < .05$ ), although the linear was not ( $P > .05$ ).

Extrapolated to zero nitrogen intake, fecal nitrogen per 100 g feed dry matter consumed was 0.42 g, approximately 20 relative percent lower than the value determined by the linear extrapolation of Hironaka, Bailey, and Kozub (87). They did, however, find that a quartic degree polynomial, not suitable for extrapolation, significantly reduced the variation of the dependent variable. The results reported here agree with the work of Hironaka et al. (87) in that fourth degree polynomial expressions best fit the data, and linear regression equations indicated small negative slopes for the regression lines. Mitchell and Bert (124), working with rats, Bosshardt and Barnes (21) studying mice, and Blaxter and Mitchell (14) using ruminants all reported that the relationship between fecal nitrogen per 100 g ingested dry matter and nitrogen content of the feed was linear.

3. Using grams nitrogen per 100 g feed dry matter as the independent variable and grams fecal nitrogen per 100 g fecal dry matter as the dependent variable, first through fourth power power functions were all found to fit the data satisfactorily. Each regression and its coefficient was significant ( $P < .01$ ), and simple correlation coefficients,



0.70 (x,y) through  $0.63(x^4,y)$ , were different ( $P < .01$ ) from zero but not different ( $P > .05$ ) from each other. Coefficients of determination progressively decreased ( $P > .05$ ) for the higher order expressions. The linear regression  $y = 2.20 + 0.609x$  described the line of best fit as determined by the coefficient of determination, 49.59 percent. No further improvement ( $P > .05$ ) in regression sums-of-squares occurred when higher degree polynomials were used. This is in agreement with the value of 2.39 mg per gram feed dry matter intake reported by Ellis et al. (51). Hironaka et al. (87) also found a linear relationship,  $y = 1.12 + 0.255x$  between the same variables, although the magnitude of the calculated fecal nitrogen excretion at zero nitrogen intake was about one-half that obtained in this work. The simple correlation  $r = 0.70$  between percent feed nitrogen and fecal nitrogen per 100 g fecal dry matter was considerably higher ( $P > .05$ ) than  $r = -.45$  between percent feed nitrogen and fecal nitrogen per 100 g feed dry matter consumed. Hironaka et al. (87) also reported a closer relationship between feed and fecal nitrogen when expressed on the basis of fecal dry matter.

4. With grams nitrogen intake per 100 g fecal dry matter as the independent variable and grams fecal nitrogen per 100 g fecal dry matter as the dependent variable, first through third power power functions were found to satisfactorily fit the data. Each regression and its coefficient were significant ( $P < .01$ ). The simple correlations, 0.77 (x,y) through  $0.68(x^3,y)$  were different ( $P < .01$ ) from zero but not ( $P > .05$ ) from each other. The linear regression equation  $y = 2.63 + 0.0827x$  had the highest coefficient of determination, 50.21 percent,

and no further improvement ( $P > .05$ ) in estimation of regression was obtained by sums of power functions. These results are not in agreement with those of Hironaka et al. (87), who found that a quadratic polynomial curve best fit the data and that at zero nitrogen intake, fecal nitrogen per 100 g fecal dry matter was approximately one-third that reported in this work. However, there is agreement in that both variables increased concomitantly, and correlations were higher when nitrogen intake and excretion were expressed on the basis of fecal dry matter excretion rather than on the basis of dry matter intake. Correlations were intermediate between the above two when nitrogen intake was expressed as a function of dry matter intake and nitrogen excretion was expressed as a function of fecal dry matter. Fecal nitrogen excretion has been shown to be more closely related to dry matter excretion than ingestion (51, 117, 134).

5. With grams digestible nitrogen per 100 g dry matter intake as the independent variable and grams fecal nitrogen per 100 g fecal dry matter as the dependent variable, first and second power functions adequately fit the data. Both regressions and regression coefficients were significant ( $P < .01$ ) and simple correlations,  $r = 0.66$  and  $0.63$ , respectively, were different ( $P < .01$ ) from zero but not ( $P > .05$ ) from each other. Sums of power functions did not improve ( $P > .05$ ) the regression sums-of-squares. The linear regression equation  $y = 2.75 + 0.54x$  enjoyed the best fit according to the coefficient of determination  $43.73$ , versus  $39.90$  percent for the quadratic function.

Extrapolated to zero digestible nitrogen intake, fecal nitrogen per 100 g fecal dry matter was 2.75 g.

6. Using grams digestible nitrogen per 100 g fecal dry matter as the independent variable and the same dependent variable as for number 5, grams fecal nitrogen per 100 g fecal dry matter, first and second power power functions again satisfactorily fit the data. Both regressions and regression coefficients were significant ( $P < .01$ ) and simple correlations,  $r = 0.72$  and  $0.67$ , respectively, were different ( $P < .01$ ) from zero but not ( $P > .05$ ) from each other. Sums of power functions did not improve ( $P > .05$ ) the estimate of regression. The linear regression equation  $y = 2.91 + 0.084x$  described the line of best fit as determined by the coefficient of determination  $0.52$ , versus  $0.45$  for the quadratic polynomial. By extrapolation to zero grams digestible nitrogen intake per 100 g fecal dry matter excreted, grams fecal nitrogen per 100 g fecal dry matter was estimated to be  $2.91$  g.

While the simple correlations  $r = 0.72$  determined here and  $0.66$  determined in number 5 for polynomials best fitting the data were not different ( $P > .05$ ) from each other, they do substantiate the trend observed in numbers 2, 3, and 4 that fecal nitrogen excretion was more closely related to fecal dry matter than to dry matter intake.

7. When grams undigested nitrogen per 100 g feed dry matter was used as the independent variable and grams fecal nitrogen per 100 g fecal dry matter was used as the dependent variable, a significant regression ( $P < .05$ ) could not be fitted to the data.

8. To further define the relationship between fecal nitrogen excretion and dry matter intake and excretion, grams dry matter intake was considered as the independent and grams nitrogen excreted as the dependent variable. The equation  $y = -2021.1 + 0.1830x - 0.000004x^2$  best fit the data as shown by the coefficient of determination of 39.59 percent. The regression was significant ( $P < .05$ ); however, neither regression coefficient was different ( $P > .05$ ) from zero, and the negative constant predicted fecal nitrogen at zero dry matter intake (that portion of metabolic fecal nitrogen not attributable to food passage through the gut) to be approximately -9 g per 100 g of food dry matter consumed.

9. The independent variable used was changed from grams dry matter intake, as in number 8, to grams dry matter excreted, and grams nitrogen excreted was again used as the dependent variable. The linear expression  $y = 100.2 - 0.139x$  described the line of best fit according to the coefficient of determination, 39.07 percent, and the lowest ( $P > .05$ ) standard error of the dependent variable. Both the regression and regression coefficient were significant ( $P < .01$ ), and the correlation between  $x$  and  $y$  was 0.62 ( $P < .01$ ). Second through fourth power power functions also fit the data, though progressively less well. Their regressions and regression coefficients were significant ( $P < .05$ ), but the coefficients of determination progressively decreased, and standard errors of  $y$  increased. Correlation coefficients ranged from a high of 0.62 ( $x^2, y$ ) to a low of 0.59 ( $x^4, y$ ), and all were different ( $P < .01$ ) from zero, but not from each other ( $P > .05$ ). There was no

further improvement ( $P > .05$ ) in regression sums-of-squares when sums of power functions were employed. The linear equation constant predicted that the constant fraction of metabolic fecal nitrogen, that portion not attributable to food passage through the gut, was 2.1 g per 100 g of excreted dry matter.

10. Level of feed intake data (Experiment II), in which a constant proportion of protein but different quantities of dry matter were fed, was used here in a further attempt to evaluate that portion of metabolic fecal nitrogen not attributable to the passage of food through the gut. Grams feed dry matter was used as the independent and grams nitrogen excreted as the dependent variable. However, first through fourth power power functions were significant ( $P < .01$ ), with significant ( $P < .01$ ) regression coefficients and correlations, ranging from 0.91 (x,y) to 0.87 ( $x^4$ ,y). From the four equation constants, the constant portion of metabolic fecal nitrogen was calculated to be -0.18 to 0.44 g nitrogen per 100 g feed dry matter consumed. Although the linear equation best fit the data, quadratic through quartic equations fit nearly as well, precluding precise mathematical assessment.

### Discussion

Mitchell (121) has pointed out that metabolic nitrogen of the feces perhaps should not be considered in protein digestibility calculations because it largely represents a wastage of nitrogen consequent upon the digestion of feed. On the other hand, he indicated that it is important to differentiate between metabolic fecal nitrogen and feed nitrogen of the feces, because they result from the operation of

different factors, and probably vary quantitatively in response to different conditions, in addition to most likely being qualitatively different. In the determination of biological values, which are based upon absorbed nitrogen, it is necessary to make allowances for metabolic nitrogen of the feces.

Metabolic fecal nitrogen plays an important part in determining ruminant protein requirements, according to Blaxter and Mitchell (14). For the young, rapidly growing animal, some 25 percent of the metabolizable protein requirement is used for replacement of the metabolic fecal loss, while for the mature animal, nearly 70 percent of the total protein requirement is determined by this factor.

Several considerations are necessary when attempting to establish metabolic fecal nitrogen by the extrapolation method. Although apparent crude protein digestibility may change over the range of nitrogen intake being investigated, approximately 20 percentage units in this case, the regression equation fully accounts for this variable. Another potential variable is the change in dry matter digestibility over the range of nitrogen intake studied, which was approximately 4.5 percentage units in this case. If fecal nitrogen excretion is computed on the basis of a unit weight of fecal dry matter excretion, the dry matter digestibility error will be incorporated in the metabolic fecal nitrogen calculation. For an experiment such as this one, in which the animals were fed at a constant proportion of body weight, it may be more accurate to compute feed and fecal nitrogen on a 5-day collection period basis, instead of per 100 g feed or fecal dry matter. However,

the magnitude of such an error should be relatively small, and since fecal nitrogen excretion appears closely related to dry matter intake (121,149,150) and fecal dry matter excretion (51,87,117,134), computing fecal nitrogen excretion on the basis of feed or fecal dry matter is more meaningful.

One assumes with the extrapolation method that the rate of change in apparent crude protein digestibility is the same between zero and the lowest level of nitrogen intake, as between the lowest and highest levels of nitrogen intake studied. This assumption seems especially precarious when mathematically predicting the change between 0 and 1.1 percent dietary nitrogen, because this relationship apparently hasn't been established. It may also be equally invalid to utilize only high protein diets, where apparent crude protein digestibility may increase at a decreasing rate (62), to extrapolate to zero nitrogen intake in order to calculate metabolic fecal nitrogen.

Apparently the relatively small number of observations and somewhat narrow range of protein intakes, coupled with considerable experimental variation, made it generally impossible to accurately define the mathematical relationship between feed and fecal nitrogen. This precluded meaningful extrapolation to zero nitrogen intake for the purpose of establishing a metabolic fecal nitrogen value. Of the several ways of relating feed and fecal nitrogen for the purpose of metabolic fecal nitrogen calculations, it appears from the literature (51, 87,117,134) and the results of this work, that fecal nitrogen excretion is more closely related to fecal dry matter excretion than to dry matter intake.

## SUMMARY

### Experiment I

An evaluation of the addition of 0, 5, 10, and 15 percent animal fat to 60, 75, and 90 percent concentrate rations was undertaken because of the dearth of information available regarding the addition of large amounts of fat to high concentrate cattle rations.

Feed consumption as a percent of body weight decreased ( $P < .05$ ) with both increasing dietary fat and concentrate levels. Within concentrate levels, increasing diet fat level decreased feed intake and average daily gain, with one exception, and generally increased feed required per unit of gain.

Increasing fat level of the diet depressed ( $P < .05$ ) dry matter and gross energy digestibilities within 90 percent concentrate diets, and increasing ration concentrate level increased ( $P < .05$ ) these digestibilities at each fat level. Within 75 percent concentrate rations, an increase in fat level increased ( $P < .05$ ) apparent crude protein digestibility, while for 0 and 15 percent fat levels, increasing ration concentrate percentage increased ( $P < .05$ ) crude protein digestibility. Added fat level did not influence ( $P > .05$ ) acid detergent fiber digestibility, but within the 15 percent fat level, digestibility was greater ( $P < .05$ ) for the 75 percent concentrate ration. Increasing diet lipid content decreased ( $P < .05$ ) chloroform-methanol-hydrochloric acid lipid digestibility at the 10 and 15 percent fat levels for 60 and 90 percent concentrate rations. Within the 15 percent fat level, lipid



digestibility of the 75 percent concentrate ration was greatest ( $P < .05$ ). Increasing the level of added fat appeared to have a more unfavorable effect on the digestibilities of 90 percent than lower concentrate rations.

As concentrate level increased, rations containing equivalent proportions of fat were consumed at a reduced rate per 100 lb body weight. Within each fat level, 90 percent concentrate rations resulted in the lowest quantity of ingested lipid per unit body weight. Yet, cattle when consuming this diet, most often and to the greatest extent, exhibited decreased digestibilities of ration components and depressed performance. Thus the limiting factor did not appear to be the quantity of ingested lipid, but rather a lipid X concentrate or roughage level interaction.

Total digestible nutrients calculated from digestible energy were on the average 2 units lower than when calculated by a modified formula utilizing acid detergent fiber and chloroform-methanol-hydrochloric acid lipid digestibilities. Increasing the level of added fat increased ( $P < .05$ ) total digestible nutrients for the 15 percent fat, 75 percent concentrate ration. For each fat level, ration total digestible nutrients were greater ( $P < .05$ ) for 75 and 90 than for 60 percent concentrate rations. Fecal soaps (chloroform-methanol-hydrochloric acid lipid extract minus chloroform-methanol lipid extract), expressed as a percent of fecal dry matter, increased ( $P < .05$ ) with each increase in diet fat level for all concentrate levels, and increasing ration concentrate level increased ( $P < .05$ ) fecal soaps for 10 and 15 percent fat

diets. Within each concentrate level, fecal soap excretion, expressed as a percent of ingested lipid, was lower ( $P < .05$ ) for 0 than for 10 and 15 percent fat rations. For the 75 percent concentrate ration with 15 percent added fat, fecal soap excretion (based on ingested lipid) decreased ( $P < .05$ ). For the 90 percent concentrate ration with 15 percent added fat, fecal soap levels reached 24 percent of fecal dry matter and 31 percent of ingested lipid by weight. Thus the importance of including fecal soap lipid in lipid digestibility and total digestible nutrient calculations is apparent.

Apparent calcium digestibility decreased (excretion increased) as fecal soap excretion increased for 60 and 75 percent concentrate rations. Fecal gross energy (kcal per g fecal dry matter) increased with increases in both ration fat and concentrate levels. Fat level of constant and ad libitum fed diets was without effect ( $P > .05$ ) on rumen sample pH. The pH values were lower ( $P < .05$ ) for 90 than for 60 percent concentrate rations containing 0 percent fat. Limiting intake of the 90% concentrate rations containing 15% fat also decreased rumen sample pH as compared to the 60% concentrate ration containing 15% fat.

Fat level of the diet exerted less influence on rumen sample volatile fatty acid concentrations than did ration concentrate level.

For constant feed periods, acetic acid concentrations were lower ( $P < .05$ ) and propionic acid levels were greater ( $P < .05$ ) for fat levels of 10 and 15 than 5 and 0 percent when 60 percent concentrate rations were fed. With ad libitum intakes of 60 percent concentrate rations, acetic acid concentrations were lower ( $P < .05$ ), and propionic

acid levels were greater ( $P < .05$ ) for 10 and 15 than 0 percent fat rations. Increasing ration concentrate level decreased ( $P < .05$ ) acetic acid and increased ( $P < .05$ ) propionic acid concentrations for 0 and 5 percent fat levels. Acetic, isobutyric, butyric, and isovaleric acid levels tended to be greater for constant feed periods, and propionic and valeric acid concentrations tended to be greater for ad libitum feed periods. For both constant and ad libitum feed periods, molar percents of acetic acid were negatively correlated ( $P < .01$ ) with those of propionic and isovaleric acids. Propionic acid proportions for both regimens were negatively correlated ( $P < .05$ ) with isovaleric, and for constant feed periods positively correlated ( $P < .01$ ) with valeric acid concentrations. For ad libitum feed periods, butyric and valeric acid levels were positively correlated ( $P < .01$ ).

Limited data regarding the influence of dietary fat level on digestible energy intake showed that added fat to 10 percent of the diet had little depressing effect on digestible energy intake, while the 15 percent fat level dramatically reduced it. Digestible energy consumption drastically declined with each increase in ration concentrate level.

#### Experiment II

A 90 percent concentrate cattle ration was fed between approximately 1 and 2 times the calculated maintenance energy requirements to evaluate, by multiple regression analysis, the influence of level of feed intake on apparent ration component digestibilities.

The narrow range of feed intakes attained was of fundamental importance in that polynomial, exponential, and logarithmic expressions of linear through quartic order generally behave in the same manner over the range of  $x$  equals 1 to 2, the range of feed intakes studied. For this reason, expressions were assumed to be polynomial in nature.

Each of the power functions (first through fourth powers) fitted to the data by the least squares method explained the data for the three digestibility parameters equally well. Each expression accounted for approximately an equal proportion of the variation ( $r^2$ ), had a significant correlation ( $P < .01$ ) not different ( $P > .05$ ) from other correlations, and had a significant regression ( $P < .01$ ). Regression coefficients were different ( $P < .01$ ) from zero, lending to validity of the equations. Summing power functions produced nonsignificant improvements ( $P > .05$ ) in estimation of regressions. Thus it was not possible to determine the mathematical relationship between level of feeding and the apparent digestibility parameters studied. This made prediction beyond the data to greater levels of feed consumption precarious, as power functions fitting the data respond quite differently at higher values of  $x$ .

Apparent dry matter, gross energy, and crude protein digestibilities declined rather consistently from the mean low (0.97 times maintenance) to the mean high (1.81 times maintenance) level of feed intake. The observed decline in dry matter digestibility over this range was 4.5 percentage units. Calculated from the fitted expressions, changes in dry matter digestibility for increasing feed intakes from 1

to 2 times maintenance were -5.4 to -6.3 percentage units. Digestibilities calculated from the curvilinear equations were much closer to determined values than were values from the linear expression, thereby lending support to the theory defining the relationship between feed intake and dry matter digestibility as curvilinear.

Over the range of mean feed intakes, gross energy digestibility declined 5.0 percentage units. Calculated from the fitted expressions, changes in gross energy digestibility for increasing feed intakes from 1 to 2 times maintenance were -6.0 to -7.1 percentage units. Digestibilities calculated from the linear and curvilinear expressions were equally close (1 percentage unit) to the determined values.

The measured difference in crude protein digestibility over the range of mean feed intakes was 8.9 percentage units, the largest digestibility decline observed. Changes in crude protein digestibility by raising feed intake from 1 to 2 times maintenance were calculated for the fitted expressions to be -10.3 to -11.6 percentage units. Again digestibilities calculated from the linear and curvilinear expressions were equally close (1 percentage unit) to the determined values.

### Experiment III

Four 90 percent concentrate rations varying in crude protein level from approximately 10 to 16 percent were fed to cattle to determine the influence of diet crude protein level on the apparent digestibilities of ration components. Metabolic fecal nitrogen was calculated by extrapolating regression lines fitted to the data by multiple

regression analysis, for first through fourth order equations, to zero nitrogen intake.

Increasing diet crude protein level increased ( $P < .01$ ) apparent digestibilities of dry matter, gross energy, and crude protein by approximately 5, 5, and 20 percentage units, respectively. From linear regressions the calculated increases in digestibility per 1 percentage unit increase in diet crude protein were 0.76, 0.78, and 2.90 percentage units, respectively.

When grams fecal nitrogen per 100 g feed dry matter were regressed on grams nitrogen per 100 g feed dry matter, the expression  $y = 0.418 + 0.2099x - 0.0060x^4$  best fit the data, although the regression was not significant ( $P > .05$ ). Fecal nitrogen at zero nitrogen intake was 0.42 g per 100 g feed dry matter. For grams fecal nitrogen per 100 g fecal dry matter regressed on grams nitrogen per 100 g feed dry matter, the significant regression ( $P < .01$ )  $y = 2.20 + 0.609x$  was the equation of best fit. Regressing grams fecal nitrogen per 100 g fecal dry matter on grams nitrogen intake per 100 g fecal dry matter, the significant regression equation ( $P < .01$ )  $y = 2.63 + 0.0827x$  had the highest coefficient of determination. Correlation was higher ( $P > .05$ ) when nitrogen intake and excretion were expressed on the basis of fecal dry matter rather than on dry matter intake. Values were intermediate when nitrogen intake and excretion were expressed on the basis of dry matter intake and fecal dry matter, respectively.

With grams fecal nitrogen per 100 g fecal dry matter regressed on grams digestible nitrogen per 100 g dry matter intake, the

equation ( $P < .01$ )  $y = 2.75 + 0.54 x$  described the line of best fit. The same dependent variable as above was also regressed on grams digestible nitrogen per 100 g fecal dry matter. The linear regression equation  $y = 2.91 + 0.084x$  was significant ( $P < .01$ ) and best fit the data.

The regression of total grams nitrogen excreted on total grams nitrogen consumed yielded a nonsignificant regression ( $P > .05$ ). A significant regression ( $P < .05$ ) could not be fitted to the data when grams fecal nitrogen per 100 g fecal dry matter were regressed on grams undigested nitrogen per 100 g feed dry matter.

To evaluate the constant fraction (not attributable to food passage) of metabolic fecal nitrogen, grams nitrogen excreted were regressed on grams dry matter intake. At zero nitrogen intake fecal nitrogen was calculated to be -9 g per 100 g dry matter consumed. The same dependent variable as above was also regressed on grams dry matter excreted, and the constant fraction of metabolic fecal nitrogen was calculated to be 2.1 g per 100 g of excreted dry matter. Using level of feed intake data (Experiment II), grams nitrogen excreted were regressed on grams feed dry matter. The four power functions fit the data equally well, and as a result the constant fraction metabolic fecal nitrogen calculations ranged from -0.18 to 0.44 g nitrogen per 100 g feed dry matter. Several inherent pitfalls in the extrapolation method of determining metabolic fecal nitrogen are pointed out.

## APPENDIX

### RAW STATISTICAL DATA



Table 57. Grams dry matter consumed during collection periods.  
Experiment I.

Steer Number	Treatment	% Concentrate Ration		
		60	75	90
13	0% Added Fat	31,671	32,993	26,984
18		35,630	32,993	26,984
23		31,676	37,117	33,923
16	5% Added Fat	32,071	32,408	27,178
17		32,071	-	-
22		32,071	28,357	24,897
15	10% Added Fat	28,545	28,694	26,162
20		28,545	28,694	27,042
24		28,545	28,694	26,040
14	15% Added Fat	18,555	23,655	13,924
19		18,555	20,620	13,924
21		18,555	22,765	13,452

Table 58. Grams fecal dry matter excreted during collection periods.  
Experiment I.

Steer Number	Treatment	% Concentrate Ration		
		60	75	90
13	0% Added Fat	11,445	6,253	4,078
18		9,740	6,407	3,177
23		10,794	8,278	4,947
16	5% Added Fat	10,228	5,930	5,260
17		9,649	-	-
22		9,007	6,669	3,721
15	10% Added Fat	8,468	6,481	6,016
20		10,563	7,019	5,064
24		10,260	8,568	6,157
14	15% Added Fat	8,563	5,101	3,889
19		6,466	3,614	3,991
21		6,136	3,771	2,158

Table 59. Kilocalories gross energy per gram of feces. Experiment I.

Steer Number	Treatment	% Concentrate Ration		
		60	75	90
13	0% Added Fat	4.399	4.701	4.827
18		4.334	4.585	5.000
23		4.474	4.704	4.917
16	5% Added Fat	4.905	5.174	5.451
17		4.873	-	-
22		5.832	4.861	4.978
15	10% Added Fat	5.156	5.561	5.773
20		5.087	5.554	6.035
24		5.180	5.414	5.811
14	15% Added Fat	5.281	5.940	6.501
19		5.376	5.679	6.165
21		5.363	5.864	6.200

Table 60. Percent crude protein of feces. Experiment I.

Steer Number	Treatment	% Concentrate Ration		
		60	75	90
13	0% Added Fat	17.49	23.14	26.12
18		20.95	24.55	28.63
23		20.32	24.12	28.97
16	5% Added Fat	17.76	19.35	21.09
17		19.82	-	-
22		17.99	20.05	22.05
15	10% Added Fat	17.14	18.16	22.39
20		15.81	16.28	21.07
24		14.31	16.35	21.47
14	15% Added Fat	16.19	17.39	20.16
19		15.28	15.62	21.89
21		15.98	16.70	19.60

Table 61. Percent and detergent fiber of feces. Experiment I.

Steer Number	Treatment	% Concentrate Ration		
		60	75	90
13	0% Added Fat	54.21	41.09	41.14
18		55.68	44.08	39.75
23		54.30	40.18	36.74
16	5% Added Fat	54.85	43.79	39.42
17		52.87	-	-
22		53.72	43.42	41.87
15	10% Added Fat	47.12	41.44	30.74
20		49.34	41.53	30.66
24		47.32	40.26	29.44
14	15% Added Fat	46.76	34.77	25.81
19		44.28	37.82	33.47
21		44.39	36.60	29.32

Table 62. Percent nitrogen-free extract of feed. Experiment I.

% Concentrate Ration	% Added Fat			
	0	5	10	15
60	46.79	41.64	33.47	32.40
75	57.66	52.60	46.05	39.28
90	65.27	59.67	57.06	49.91

Table 63. Nitrogen-free extract digestion coefficients. Experiment I.

Steer Number	Treatment	% Concentrate Ration		
		60	75	90
13	0% Added Fat	92.1	95.1	97.6
18		99.0	96.2	98.5
23		95.2	92.5	96.8
16	5% Added Fat	96.5	96.1	96.3
17		97.0	-	-
22		98.0	93.0	97.2
15	10% Added Fat	100.0 <sup>a</sup>	97.0	94.9
20		98.5	93.6	96.9
24		95.7	91.1	94.9
14	15% Added Fat	100.0 <sup>a</sup>	97.8	97.0
19		100.0 <sup>a</sup>	97.5	98.3
21		100.0 <sup>a</sup>	97.8	97.9

a. Exceeded 100 percent digestibility.

Table 64. Percent ash of feces and feed. Experiment I.

Steer Number	Treatment	% Concentrate Ration		
		60	75	90
<u>Feces</u>				
13	0% Added Fat	13.66	12.82	13.34
18		16.39	12.66	13.83
23		13.64	10.58	11.10
16	5% Added Fat	12.36	10.34	9.77
17		12.79	-	-
22		14.44	10.68	12.91
15	10% Added Fat	18.94	10.66	10.32
20		15.10	9.15	10.48
24		14.25	8.67	11.15
14	15% Added Fat	17.96	10.53	12.28
19		16.53	13.05	9.83
21		18.39	11.93	11.64
<u>Feed</u>				
	0% Added Fat	7.88	5.69	4.69
	5% Added Fat	7.33	5.01	4.96
	10% Added Fat	9.44	5.49	4.37
	15% Added Fat	8.37	5.02	5.22



Table 65. Analyses of variance of rumen pH for added fat levels within concentrate level. Experiment I.

Source of Variation	<u>Ad Libitum Feed</u>		<u>Constant Feces</u>	
	df	Mean Squares	df	Mean Squares
% Concentrate	2	1.01**	2	1.49**
% Fat, 60% Concentrate	3	0.34	3	0.17
% Fat, 75% Concentrate	3	0.09	3	0.30
% Fat, 90% Concentrate	3	0.04	3	0.20
Error	23	0.14	21	0.12

\*. (P&lt;.05).

II. (P&lt;.01)

Table 66. Analyses of variance of rumen pH for concentrate levels within added fat level. Experiment I.

Source of Variation	<u>Ad Libitum Feed</u>		<u>Constant Feces</u>	
	df	Mean Squares	df	Mean Squares
% Fat	3	0.32	3	0.61**
% Concentrate, 0% Fat	2	0.61*	2	0.55*
% Concentrate, 5% Fat	2	0.30	2	0.27
% Concentrate, 10% Fat	2	0.15	2	0.36
% Concentrate, 15% Fat	2	0.17	2	0.47*
Error	23	0.15	21	0.12

\*. (P&lt;.05).

\*\*. (P&lt;.01).

Table 67. Analyses of variance of volatile fatty acids during constant feed intake periods for added fat levels within concentrate level. Experiment I.

Source of Variation	df	M e a n   S q u a r e s					
		C <sub>2</sub>	C <sub>3</sub>	iso-C <sub>4</sub>	C <sub>4</sub>	iso-C <sub>5</sub>	C <sub>5</sub>
% Concentrate	2	458.88**	454.35**	1.20	18.98	1.07	0.74
% Fat, 60% Concentrate	3	152.44**	155.35**	0.15	5.41	1.01	0.07
% Fat, 75% Concentrate	3	34.27	51.71	2.37*	5.38	0.65	0.41
% Fat, 90% Concentrate	3	17.69	54.89	0.12	9.75	7.09**	1.13*
Error	21	24.10	24.72	0.53	6.72	1.24	0.32

\*. (P<.05).

\*\*.. (P<.01).

Table 68. Analyses of variance of volatile fatty acids during constant feed intake periods for concentrate levels within added fat level. Experiment I.

Source of Variation	df	M e a n   S q u a r e s					
		C <sub>2</sub>	C <sub>3</sub>	iso-C <sub>4</sub>	C <sub>4</sub>	iso-C <sub>5</sub>	C <sub>5</sub>
% Fat	3	67.01	101.67*	0.92	1.45	2.23	0.63
% Concentrate, 0% Fat	2	435.41**	496.09**	0.07	10.69	1.05	1.63*
% Concentrate, 5% Fat	2	127.35*	121.09*	3.29**	12.07	8.51*	0.01
% Concentrate, 10% Fat	2	32.62	74.39	0.16	1.13	1.02	0.10
% Concentrate, 15% Fat	2	122.82*	41.79	0.29	22.14	0.43	0.88
Error	21	24.10	24.72	0.53	6.72	1.24	0.32

\*. (P<.05).

\*\*.. (P<.01).

Table 69. Analyses of variance of volatile fatty acids during ad libitum feed intake periods for added fat levels within concentrate level. Experiment I.

Source of Variation	df	M e a n   S q u a r e s					
		C <sub>2</sub>	C <sub>3</sub>	iso-C <sub>4</sub>	C <sub>4</sub>	iso-C <sub>5</sub>	C <sub>5</sub>
% Concentrate	2	473.98**	396.81**	1.07**	7.52	1.16**	2.27
% Fat, 60% Concentrate	3	100.69*	137.69**	0.02	8.41	0.22	0.07
% Fat, 75% Concentrate	3	3.61	7.24	0.09	4.16	0.15	0.25
% Fat, 90% Concentrate	3	11.03	88.31*	0.03	24.08**	0.38	1.01
Error	23	28.07	24.00	0.07	4.61	0.24	0.85

\*. (P<.05).

\*\*.. (P<.01).

Table 70. Analyses of variance of volatile fatty acids during ad libitum feed intake periods for concentrate levels within added fat level. Experiment I.

Source of Variation	df	M e a n   S q u a r e s					
		C <sub>2</sub>	C <sub>3</sub>	iso-C <sub>4</sub>	C <sub>4</sub>	iso-C <sub>5</sub>	C <sub>5</sub>
% Fat	3	41.63	95.91*	0.09	15.67*	0.27	0.50
% Concentrate, 0% Fat	2	272.82**	224.56**	0.19	10.37	1.51**	2.35
% Concentrate, 5% Fat	2	161.72**	248.31**	0.31*	18.47*	0.04	0.46
% Concentrate, 10% Fat	2	91.38	110.71*	0.24	1.69	0.12	0.01
% Concentrate, 15% Fat	2	50.43	4.25	0.42*	12.60	0.27	0.70
Error	23	28.08	24.01	0.07	4.61	0.24	0.85

\*, (P<.05).

\*\*, (P<.01).

Table 71. Analyses of variance of digestible energy intake by ration concentrate level: Experiment I.

Source of Variation	Degrees of Freedom and Mean Squares					
	60% Concentrate Ration		75% Concentrate Ration		90% Concentrate Ration	
Regression	1	2,945,630**	1	26,382	1	914,366**
Residual	10	123,001	9	112,271	9	78,600
Total	11		10		10	

\*\*, (P<.01).

Table 72. Level of feed consumption as a percent of body weight.  
Experiment II.<sup>a</sup>

Level of Intake <sup>b</sup>	P e r i o d s			
	1	2	3	4
A	1.13 (1) <sup>c</sup>	1.14 (2)	1.08 (3)	1.05 (4)
	1.14 (5)	1.01 (8)	1.10 (6)	1.01 (7)
B	1.53 (2)	1.52 (1)	1.44 (4)	1.33 (3)
	1.55 (6)	1.31 (7)	1.39 (5)	1.46 (8)
C	1.84 (3)	1.93 (4)	1.73 (1)	1.54 (2)
	1.52 (7)	1.88 (6)	1.65 (8)	1.55 (5)
D	2.24 (4)	2.28 (3)	1.90 (2)	1.82 (1)
	1.98 (8)	2.11 (5)	1.75 (7)	1.91 (6)

a. Ninety percent dry matter basis.

b. Approximate feed consumption as a percent of body weight:  
A = 1.1, B = 1.4, C = 1.7, D = 2.0.

c. Heifer number.

Table 73. Grams dry matter consumed during collection periods.  
Experiment II.

Heifer Number	P e r i o d s			
	1	2	3	4
1	10,698	15,919	20,162	24,240
2	14,589	11,440	22,082	19,491
3	16,534	23,879	11,521	15,688
4	19,452	18,904	14,879	11,886
5	10,214	21,889	15,362	19,491
6	12,888	17,909	11,039	21,866
7	23,138	19,852	23,042	14,260
8	11,806	20,334	17,112	17,112



Table 74. Grams fecal dry matter excreted during collection periods.  
Experiment II.

Heifer Number	P e r i o d s			
	1	2	3	4
1	1,794	2,516	3,209	4,137
2	2,561	1,736	4,083	2,632
3	2,928	4,764	1,740	2,149
4	4,302	3,740	2,928	1,531
5	1,557	4,170	2,207	2,443
6	2,424	3,524	1,546	4,044
7	3,663	4,147	3,799	2,065
8	4,071	1,509	3,334	2,069

Table 75. Kilocalories gross energy per gram of feces. Experiment II.

Heifer Number	P e r i o d s			
	1	2	3	4
1	4.843	5.074	5.097	4.867
2	4.399	4.933	4.855	4.828
3	4.366	4.673	5.055	4.809
4	4.601	4.918	4.734	4.773
5	4.235	4.761	4.945	4.773
6	4.251	4.764	4.824	4.747
7	4.702	4.660	4.622	4.751
8	4.341	4.951	4.768	4.846

Table 76. Percent crude protein of feces. Experiment II.

Heifer Number	P e r i o d s			
	1	2	3	4
1	25.58	23.88	27.29	29.34
2	29.61	22.76	29.26	27.49
3	25.83	24.25	28.78	27.83
4	24.61	25.27	26.91	24.57
5	21.06	23.26	30.21	26.20
6	25.40	23.66	29.52	26.10
7	26.24	27.71	26.11	28.04
8	23.23	26.25	27.70	25.91

Table 77. Grams dry matter consumed during collection periods.  
Experiment III.

Level of Intake <sup>a</sup>	P e r i o d s			
	1		2	
A	21,424	(18) <sup>b</sup>	22,996	(17)
	25,472	(23) <sup>c</sup>	23,954	(21)
B	23,609	(19)	23,671	(20)
	21,642	(22)	22,132	(23)
C	21,701	(17)	22,728	(18)
	21,701	(20)	24,662	(24)
D	23,314	(21)	25,367	(19)
	23,620	(24) <sup>d</sup>	24,001	(22)

a. Approximate percent protein in ration: A = 10, B = 12, C = 15, and D = 16.

b. Steer number.

c. Steer 23 consumed a 9.95 percent protein ration.

d. Steer 24 consumed a 16.56 percent protein ration.

Table 78. Grams fecal dry matter excreted during collection periods.  
Experiment III.

Level of Intake <sup>a</sup>	P e r i o d s			
	1		2	
A	4,958	(18) <sup>b</sup>	5,235	(17)
	5,752	(23) <sup>c</sup>	5,790	(21)
B	4,634	(19)	5,045	(20)
	4,433	(22)	5,176	(23)
C	4,150	(17)	4,058	(18)
	4,066	(20)	5,260	(24)
D	4,548	(21)	4,915	(19)
	4,073	(24) <sup>d</sup>	3,639	(22)

a. Approximate percent protein in ration: A = 10, B = 12, C = 15, and D = 16.

b. Steer number.

c. Steer 23 consumed a 9.95 percent protein ration.

d. Steer 24 consumed a 16.56 percent protein ration.

Table 79. Dry matter digestion coefficients. Experiment III.

Level of Intake <sup>a</sup>	P e r i o d s			
	1		2	
A	76.9	(18) <sup>b</sup>	77.2	(17)
	77.4	(23) <sup>c</sup>	75.8	(21)
B	80.4	(19)	78.7	(20)
	79.5	(22)	76.6	(23)
C	80.9	(17)	82.1	(18)
	81.3	(20)	78.7	(24)
D	80.5	(21)	80.6	(19)
	82.8	(24) <sup>d</sup>	84.8	(22)

a. Approximate percent ration protein: A = 10, B = 12, C = 15, and D = 16.

b. Steer number.

c. Steer 23 consumed a 9.95 percent protein ration.

d. Steer 24 consumed a 16.56 percent protein ration.

Table 80. Kilocalories gross energy per gram of feces. Experiment III.

Level of Intake <sup>a</sup>	P e r i o d s			
	1		2	
A	4.6105	(18) <sup>b</sup>	4.4568	(17)
	4.6344	(23) <sup>c</sup>	4.4860	(21)
B	4.7406	(19)	4.5646	(20)
	4.6611	(22)	4.5628	(23)
C	4.5630	(17)	4.6325	(18)
	4.6456	(20)	4.7141	(24)
D	4.7054	(21)	4.6720	(19)
	4.7331	(24) <sup>d</sup>	4.6357	(22)

a. Approximate percent protein in ration: A = 10, B = 12, C = 15, and D = 16.

b. Steer number.

c. Steer 23 consumed a 9.95 percent protein ration.

d. Steer 24 consumed a 16.56 percent protein ration.

Table 81. Gross energy digestion coefficients. Experiment III.

Level of Intake <sup>a</sup>	P e r i o d s			
	1		2	
A	75.6	(18) <sup>b</sup>	76.3	(17)
	75.7	(23) <sup>c</sup>	74.7	(21)
B	78.6	(19)	77.8	(20)
	78.0	(22)	75.6	(23)
C	79.9	(17)	81.0	(18)
	80.0	(20)	76.8	(24)
D	78.9	(21)	79.4	(19)
	81.5	(24) <sup>d</sup>	84.0	(22)

a. Approximate percent ration protein: A = 10, B = 12, C = 15, and D = 16.

b. Steer number.

c. Steer 23 consumed a 9.95 percent protein diet.

d. Steer 24 consumed a 16.56 percent protein diet.

Table 82. Percent crude protein of feces. Experiment III.

Level of Intake <sup>a</sup>	P e r i o d s			
	1		2	
A	18.15	(18) <sup>b</sup>	22.26	(17)
	17.86	(23) <sup>c</sup>	19.78	(21)
B	23.34	(19)	22.04	(20)
	21.43	(22)	21.49	(23)
C	23.56	(17)	25.73	(18)
	22.03	(20)	21.35	(24)
D	22.43	(21)	21.70	(19)
	25.73	(24) <sup>d</sup>	25.00	(22)

a. Approximate percent protein in ration: A = 10, B = 12, C = 15, and D = 16.

b. Steer number.

c. Steer 23 consumed a 9.95 percent protein ration.

d. Steer 24 consumed a 16.56 percent protein ration.



Table 83. Crude protein digestion coefficients. Experiment III.

Level of Intake <sup>a</sup>	P e r i o d s			
	1		2	
A	56.5	(18) <sup>b</sup>	50.8	(17)
	59.5	(23) <sup>c</sup>	53.6	(21)
B	61.2	(19)	64.1	(20)
	62.9	(22)	61.6	(23)
C	70.2	(17)	69.2	(18)
	72.7	(20)	69.5	(24)
D	70.4	(21)	76.0	(19)
	73.2	(24) <sup>d</sup>	78.4	(22)

a. Approximate percent ration protein: A = 10, B = 12, C = 15, and D = 16.

b. Steer number.

c. Steer 23 consumed a 9.95 percent protein ration.

d. Steer 24 consumed a 16.56 percent protein ration.

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