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NELSON, Delbert Kent, 1939-
EFFECT OF DIETARY ENERGY CONCENTRATION
AND LEVEL ON DIGESTION IN THE BOVINE
GASTROINTESTINAL TRACT.

Iowa State University, Ph. D., 1968
Physiology

University Microfilms, Inc., Ann Arbor, Michigan

EFFECT OF DIETARY ENERGY CONCENTRATION
AND LEVEL ON DIGESTION IN THE BOVINE GASTROINTESTINAL TRACT

by

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A Dissertation Submitted to the
Graduate Faculty in Partial Fulfillment of
The Requirements for the Degree of
DOCTOR OF PHILOSOPHY

Major Subject: Animal Nutrition

Approved:

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1968

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INTRODUCTION

The effect of diet on digestion in the total gastrointestinal tract of ruminants has been thoroughly studied. These results, however, do not reveal the effect of diet on digestion in the various segments of the tract. Some research has examined the effect of diet on rumen digestion, but information regarding the lower tract, posterior to the rumen, is only fragmentary. Several studies have shown that the lower tract is an important site of nutrient absorption for some rations. In the few studies which have compared different diets, a variety of conclusions have been reached.

It is known that the physical form of the diet and plane of nutrition both affect rate of passage, and it is probable that rate of passage affects degree of digestion in various segments of the tract. The particle size and chemical structure of the ration undoubtedly affect the solubility of the nutrients and influence the degree of rumen and lower tract digestion. Yet in most experiments physical form, chemical structure and plane of nutrition have been confounded, and the effect of any one of these on digestion was difficult to measure.

Since most of the past research has the shortcomings mentioned, I felt a study was needed where ration effects could be controlled.

In the present experiments, physical form of the diet was held constant, while energy concentration and plane of nutrition were varied. The effect of these two factors on digestion in various segments of the tract was examined.

REVIEW OF LITERATURE

A technique common to a gastrointestinal absorption study is sampling from ligated segments of the tract of recently sacrificed animals. Its main disadvantages are that the animal can be used only once and digestion at time of slaughter may not represent average digestion for all segments of the tract. Another common technique involves surgically installing fistulae or re-entrant cannulae along the digestive tract from which samples may be taken. This allows for sampling from one animal on several different rations and at different times after feeding. Both methods have been adequately described and discussed by McGilliard (1961) and will not be reviewed further here. McGilliard (1961) has also thoroughly reviewed the various marker techniques used in this type of experiment.

Since the data in the literature are from various species on various rations and refer to different segments of the digestive tract, dry matter will be examined first to give a general view of the digestion in the various segments. The term digestion will be used to infer disappearance of the nutrient being discussed.

Dry Matter Digestion

Most of the dry matter digestion occurs in the forestomach of ruminants. Gray et al. (1958a) sacrificed sheep at various times after eating, used lignin as a marker, and found that average forestomach dry matter digestion coefficients were 34% and 60% for diets of straw and

alfalfa hay, respectively. In both cases this represented nearly all the dry matter digested in the tract.

Sampling from a rumen fistula, Hale et al. (1947) estimated that 85% of the digestible dry matter of a hay diet was digested in the rumen of a cow. Hogan (1957) and Hogan and Phillipson (1960) used sheep with re-entrant duodenal and ileal cannulae and determined digestibility at these points by total collection and re-introduction. With diets of hay and concentrate, 70% of the digestible dry matter was digested in the stomach while 11% was digested in the small intestine and 19% in the large intestine. A unique method of measuring duodenal flow electromagnetically and sampling intermittently was used by Ridges and Singleton (1962). The technique facilitated flow measurement without removal of digesta. Their data indicated that 58% of the digestible dry matter disappeared in the stomach of goats on a hay and concentrate ration.

McGilliard (1961) made total collections and re-introduction at a duodenal re-entrant cannula. As the diet changed progressively from all hay to 30% hay and 70% concentrate, the percentage of dry matter digested in the stomach decreased while the coefficients of digestibility of residues entering the intestines increased. Sineshchekov (1965) reported similar results and indicated that intestinal digestion can increase 1.5 to 2 times if roughages are replaced by concentrates. McGilliard (1961), however, observed that complete replacement of all roughage with concentrate reversed the trend and increased the total digestion coefficient,

the stomach digestion coefficient, and the percent of digestible dry matter digested in the stomach.

Comparing two levels of fiber fed to young calves, Yang and Thomas (1965) found that reducing fiber increased rumen digestion coefficients as well as the percentage of digestible dry matter digested by the rumen. Rogerson (1958) fed various ratios of roughage and concentrate to sheep and, after slaughter, determined digestibility by use of the lignin ratio technique. He found that as concentrate was increased, rumen dry matter digestibilities increased, but intestinal digestion coefficients also increased. Consequently, as concentrate increased the percent of the digestible dry matter digested by the rumen decreased.

Balch (1957) measured digestibility in the ruminoreticulum by sampling near the reticulo-omasal opening. Using the lignin ratio technique he found digestibility coefficients to range from 26-62%, depending upon the ration. Like Rogerson (1958) he found that increasing the concentrate portion of the diet increased ruminoreticular digestion of dry matter, but unlike Rogerson (1958) he observed a simultaneous increase in the percent of digestible dry matter digested in the ruminoreticulum.

Campling et al. (1961) measured organic matter rather than dry matter. They observed little difference in the percentage of digestible organic matter absorbed in the rumen of cows on hay or straw, even though the ruminal digestion coefficient for the hay was higher. Organic matter digestion observed by McGilliard (1961), Ridges and Singleton (1962) and Yang and Thomas (1965) was similar to their dry matter digestion

except the values generally were higher for the upper part of the tract due to the secretion of inorganic nutrients into the abomasum which lowered the dry matter digestion values.

A negative dry matter digestibility was observed in the abomasum or proximal intestine by several groups of workers, (Harris and Phillipson, 1962; McGilliard, 1961; Rogerson, 1958; Yang and Thomas, 1965) mainly because of the ash added into the tract at this point. Endogenous nitrogen is also added in the abomasum and duodenum, but its effect on digestibility will be discussed later.

Carbohydrate Digestion

Carbohydrate is the most prevalent nutrient in forages and grains, consequently it provides the major portion of digestible energy in ruminant rations. The carbohydrates of livestock feeds are normally classified as nitrogen-free-extract and crude fiber. The carbohydrates which compose each of these classes will be discussed separately, even though some carbohydrates may fall into both of these poorly-defined groups.

Nitrogen-free-extract (NFE) is not a chemical classification but a solubility classification. It contains sugars, starches, hemicellulose, and the soluble portion of cellulose and pentosans.

The intestinal absorption of many NFE components has been extensively studied in calves and other young ruminants. Nipple pail feeding or introduction of glucose, galactose, or lactose into the omasum or a duodenal fistula has resulted in high digestibility values (Henschel et al.,

1963; Huber et al., 1961b; Morrill, 1963) and positive responses in blood reducing sugar (Dollar and Porter, 1957; Huber et al., 1961a; Huber et al., 1961b; Larsen et al., 1956; Velu et al., 1960). Maltose, however, does not greatly increase blood reducing sugars in very young calves (Dollar and Porter, 1957; Huber et al., 1961a; Velu et al., 1960), but maltase activity increases in the intestine as the calf matures (Dollar and Porter, 1957). Huber et al. (1961a) introduced maltose into the omasum and observed an increase in blood reducing sugars up to 50 days of age, but the response dropped thereafter. Dollar and Porter (1957) observed that maltose caused blood reducing sugar increases at 9 weeks of age while Larsen et al. (1956) and Huber et al. (1961b) found large increases beyond 6 months of age.

Some studies have shown no increase in blood reducing sugar upon ingestion of sucrose (Dollar and Porter, 1957; Huber et al., 1961a; Velu et al., 1960). However, changes in blood reducing sugars may not reflect true sucrose digestion. Henschel et al. (1963) observed that 38% of the sucrose introduced into a duodenal fistula disappeared in the small intestine. Morrill (1963) observed a similar average of 41% and speculated that sucrose was metabolized to volatile fatty acids.

Starch, like sucrose, does not cause appreciable responses in blood reducing sugar when introduced into the small intestine (Dollar and Porter, 1957; Huber et al., 1961a; Larsen et al., 1956). However, work by Huber et al. (1961b) indicated 80%-89% of their dietary starch was utilized in the intestine. A similar value was obtained by Henschel et al. (1963)

when soluble starch digestion was measured between the proximal duodenum and ileum of 4 to 6 month old steers.

Even though the intestine can absorb soluble carbohydrates, some research indicates that little, if any, digestible carbohydrates escape the fermentation in the rumen (Hale et al., 1947; Phillipson and McAnally, 1942; Schambye, 1951). Undoubtedly the forestomach is the major site of soluble carbohydrate digestion, but such exclusive digestion by the forestomach has not been born out by other research. McGilliard (1961) found that 75% to 90% of the digestible NFE was assimilated in the stomach. Similar values were observed by Ridges and Singleton (1962). Kameoka (1964) demonstrated that the forestomach of a goat digested 88% of the digestible sugars in the ration.

Trials conducted by Rogerson (1958) and Freer and Campling (1963) showed that increasing the concentrate portion of the diet, increased the rumen digestion coefficient for NFE. Rogerson (1958) also noticed that this caused an increase in intestinal digestion of the dietary NFE. Similar trends for starch were recently published by Karr et al. (1966). They observed that post-ruminal digestion of starch played a greater role in starch utilization as concentrate levels increased. However, they observed a limitation in the amount of starch which could be utilized in the small intestine and attributed this to a possible lack of amylolytic activity. Henschel et al. (1963) observed a similar limitation but estimated that there is enough amylase in the small intestine of steers to hydrolyze 120 grams of starch in 2 or 3 hours. Wright et al. (1966) suspect that starch hydrolysis is very rapid in sheep small

intestine since starch values in the intestine were much lower than those of the abomasum.

Most of the above work indicates that the small intestine plays an important role in soluble carbohydrate utilization. A quantitative estimate of intestinal carbohydrate digestion was made by Fries and Conner (1960). They observed that nearly 300 g of glucose may be absorbed per day from the small intestine of a 225 kg steer, and this may account for about 10% of the total absorbed energy.

Little soluble carbohydrate is utilized beyond the posterior ileum in ruminating sheep and cattle except in cases of very high concentrate feeding (Bolin et al., 1960; Karr et al., 1966; Rogerson, 1958). As mentioned earlier much sucrose and starch escape digestion in the small intestine of calves on milk replacer diets containing these carbohydrates. Data of Huber et al. (1961b) suggest that sucrose and starch are partially hydrolyzed to monosaccharides by the microbes in the lower tract. Morrill (1963) found that 44% of the dietary sucrose was digested in the large intestine. Xenoulis (1967) further confirmed sucrose digestion in the large intestine by injecting uniformly labeled C^{14} -sucrose into the cecum of milk-fed calves. An average of 77% of the C^{14} did not appear in the feces. That which did appear was incorporated into microbial protein and lipid.

Crude fiber, unlike soluble carbohydrate, is not digested in the small intestine. Cellulose, the main component of this class is insoluble and immune to the attack of digestive enzymes. However, breakdown of cellulose occurs in the ruminoreticulum and the large intestine where it undergoes microbial degradation to volatile fatty acids and

microbial carbohydrate. Spravcev (1963) clearly demonstrated the cellulose digesting ability of the rumen and cecum of sheep by placing cellophane strips in both compartments. The breakdown of cellophane was greater in the cecum than the rumen, 11.68% compared to 8.17%. Previously Hale et al. (1947) had found a similar value (11.6%) for the lower tract but observed that 78% of the digested cellulose disappeared in the rumen. Gray (1947) fed hay and straw to sheep and calculated the percent digestible cellulose absorbed by the rumen, cecum and colon to be 70, 17, and 13, respectively. A similar diet was fed by Goodall and Kay (1965) to sheep with ileal cannulae, but little, if any, cellulose disappeared between the ileum and rectum.

McGilliard (1961) fed several rations composed of alfalfa hay and corn to a steer and found no digestion of crude fiber below the duodenum. He did observe that as dietary concentrate was increased from 0 to 70% the stomach digestion coefficients of crude fiber decreased from 55.2 to 13.7% and consequently total digestion coefficients decreased. Yang and Thomas (1965) and Balch (1957) also noticed that concentrate additions reduced rumen digestion of crude fiber. Head (1961) speculates that addition of starch to the diet lowers the pH of the rumen which in turn affects the sensitive cellulose digesting micro-organisms.

Campling et al. (1961) compared hay and straw and found the rumen digestion coefficient for fiber to be higher for the hay due to the greater quantity of non-digestible lignin in the crude fiber of the straw.

Volatile Fatty Acid Absorption

The study of carbohydrate utilization is not limited to measuring the quantitative changes of the digesta passing along the tract. Since most of the digestible carbohydrates are fermented to volatile fatty acids (VFA), largely, acetate, propionate and butyrate, the measurement of VFA in digesta or in blood draining the various segments of the tract gives a picture of the mode of energy absorption by the ruminant.

Quantitative measurements of digesta clearly show that VFA concentrations correspond to sites of microbial activity, namely, ruminoreticulum, cecum and colon (Badawy et al., 1958). Probably half of the VFA produced in the ruminoreticulum are absorbed before the digesta pass to the omasum (Johnston et al., 1961).

In spite of its small size, the omasum has a great internal surface area because of the structure of its folds or leaves. Badawy et al. (1958c) made an extensive study of this unusual organ and found that 77% of the VFA entering the omasum was absorbed. Many other researchers have also observed that nearly all of the VFA entering the omasum was absorbed before the digesta entered the abomasum (Boyne et al., 1956; Conrad et al., 1956; Elsdon et al., 1946; Huber and Moore, 1964; Johnston et al., 1961; Ward et al., 1961; Yang and Thomas, 1965).

A small but constant level of VFA does pass to the abomasum (Masson and Phillipson, 1952; Phillipson and McAnally, 1942). However, it is doubtful if abomasal absorption of VFA is significant, even though some absorption has been observed by Ash (1961) and Yang and Thomas (1965).

The concentration of VFA in the small intestine is very low, but a secondary rise in concentration comparable to the level observed in the rumen occurs in the large intestine (Elsden et al., 1946; Huber and Moore, 1964; Ward et al., 1961). Barcroft et al. (1943) measured the VFA content of blood draining the digestive tract of sheep and observed that the cecum, like the rumen, reticulum and omasum, produced elevated levels of blood VFA.

There is little information concerning the effect of diet on the absorption of VFA along the tract. Kumeno and Nishimatsu (1963) have shown that preparation of rations as a gruel rather than as pellets results in increased excretion of VFA in the feces of calves. Concentrate feeding appears to increase the level of total VFA in the cecum. However, the differences are very small (Packett et al., 1966).

Nitrogen Digestion

Unlike other absorbed nutrients, nitrogen may not be beneficially utilized by the ruminant. This is due to the sequence of events which occur in the rumen and liver.

All proteins which enter the rumen undergo proteolysis to peptides and amino acids by the rumen micro-organisms. Possibly some of these amino acids are absorbed from the rumen (Cook et al., 1965) or incorporated into bacterial protein, but most, along with the peptides and some non-protein nitrogen compounds, are deaminated with the formation of ammonia. Ammonia is the main soluble nitrogen source for microbial protein synthesis, but it is also readily absorbed by the rumen epithelium from

which it is carried by the venous system to the liver. The liver metabolizes the ammonia to urea. Some of this blood urea is returned to the rumen via the saliva or passes through the rumen wall. Most of the blood urea, unfortunately, is excreted in the urine and this loss of digestible nitrogen makes the term nitrogen digestibility misleading. This sequence of events is thoroughly reviewed by Chalmers (1961), McGilliard (1961) and Blackburn (1965).

Rumen nitrogen losses can be affected by level of dietary nitrogen, concentration of dietary nitrogen, and solubility of the nitrogenous compounds. Solubility is probably the most critical factor in rumen nitrogen losses. Chalmers (1961) has clearly shown that heating of protein supplements reduces the solubility of the protein and consequently reduces rumen ammonia levels. Cows on diets of pure alfalfa hay have been shown to have rumen losses of nitrogen as high as 86% because of the high solubility of the alfalfa protein (Hale et al., 1947). Kameoka (1962) fed N^{15} labeled orchard grass to goats, and nearly 42% of the N^{15} was absorbed in the forestomach. This loss decreased to 16% when solubility was reduced by cutting and drying the grass. In this study Kameoka (1962) also measured urea nitrogen losses in the forestomach. When N^{15} urea was fed with barley and oats 33% of the N^{15} was recovered at the omasum. When N^{15} urea was fed with starch-poor feed only 7% of the isotope was recovered.

The importance of starch or other soluble carbohydrate in converting rumen ammonia to bacterial nitrogen is reviewed by Blackburn (1965). This was first demonstrated by Mills et al. (1942) when they added starch

to a urea-timothy hay diet. The result was a reduced rumen ammonia and an increased rumen protein level. Kameoka and Morimoto (1962) demonstrated this same effect by measuring nitrogen losses orad to the omasum and found that starch addition to rations of equal nitrogen content resulted in lower losses of rumen nitrogen. They also observed that as they increased the nitrogen content of the diet, the disappearance of nitrogen from the rumen increased. Hogan (1965) also observed that grazing sheep had higher stomach losses of nitrogen when daily intakes of nitrogen increased and when nitrogen percent of the forage increased. Similar results have been observed by Ridges and Singleton (1962).

Rations which are low in nitrogen, such as straw, may result in no stomach loss of nitrogen (Gray, 1958b). In some cases where nitrogen intake has been extremely low, the amount of nitrogen passing the duodenum has been greater than the daily intake (Harris, 1962; McGilliard, 1961; Struckov, 1965). This added nitrogen apparently originates from salivary urea, urea from the rumen wall (Blackburn, 1965) and from abomasal and duodenal secretions of nitrogenous compounds. Badawy et al. (1958a) determined that most of these secretions are non-protein nitrogen. The bile nitrogen was also mainly non-protein.

These workers also observed that the increase in nitrogen in the proximal small intestine was not as great in cannulated live sheep as it was in slaughtered sheep. This supported their earlier work (Badawy et al., 1957) which indicated that the shock of a captive bullet killer

caused shedding of the epithelium into the lumen of the gut, thus increasing the nitrogen content in the duodenum.

Below the pylorus, nitrogen absorption is probably very similar to that of non-ruminants. Kameoka (1964) introduced both soybean meal and casein into the abomasum of goats and observed results similar to non-ruminants. The digesta nitrogen which enters the abomasum is chiefly microbial protein and consequently is highly digestible. Harris and Phillipson (1962) found that 62-69% of the nitrogen passing the duodenum was absorbed below that point. McGilliard observed a wider range of 63-78%. To date there is no significant information on the digestion of nitrogen in the large intestine.

EXPERIMENTAL

Experiment I

Procedure

Animals Two 7-month-old Holstein steers, weighing approximately 160 kg, were used for the experiment. To obtain access to the reticulo-omasal opening, a large rumen fistula, about 12 cm in diameter, was established by a one-stage operation in the left paralumbar fossa of each steer. A Plexiglas cannula with a removable cap was placed in the fistula immediately upon completion of the surgery. About 2 weeks following the rumen fistula surgery, a re-entrant duodenal fistula was established by the method of Conner et al. (1957). An ileal fistula was established in the same operation.

Feed and water were withheld from the steers for 48 hours prior to surgery. The operative area was closely clipped, scrubbed with bactericidal soap solution and rinsed with alcohol.

Anesthesia was induced by intravenous injection of 3% Surital sodium.¹ The animal was placed on the operating table in the left lateral recumbancy. The trachea was immediately intubated, and the endotracheal tube was attached to a closed-circuit anesthesia machine. Anesthesia was maintained with Fluothane.²

The re-entrant duodenal fistula was placed about 10 cm from the pylorus. The two stainless steel cannulae were approximately 8 cm

¹ Parke-Davis Co., Detroit, Michigan.

² A halothane anesthetic, Ayerst Laboratories, New York.

apart and were connected by two Plexiglas elbows joined together by tygon tubing (Figures 1 and 2). Figure 3 shows the external location of the preparation.

An ileal fistula was established during the same operation within 5 cm of the ileocecal junction. The techniques used were similar to those used for each duodenal cannula. The external location of the cannula is shown in Figure 3.

Before closure of the abdominal incision, two Polyotic¹ oblets were placed in the peritoneal cavity. Polyotic¹ powder was dusted on the incision after surgery and several times weekly, thereafter. To prevent general infection, 5 ml of an antibiotic combination² was injected intramuscularly on the day of surgery and at weekly intervals thereafter.

Three other steers were prepared for the experiment, but due to surgical or post-surgical complications it was necessary to sacrifice them. Because of the unusual location of the tract in one steer, a Holstein, it was impossible to establish the re-entrant duodenal fistula. This steer was sacrificed in midsurgery. An Ayrshire steer suffered a nerve injury in the left front leg during surgery and was unable to walk or stand. Consequently post-surgical recovery was poor and the steer's condition deteriorated to the point that sacrifice was deemed necessary after the third week. The other steer, not used,

¹Tetracycline hydrochloride, American Cyanamid Co., Princeton, New Jersey.

²400,000 units penicillin in dihydrostreptomycin solution, 0.5 mg/2cc, Corvett Division of Eli Lilly and Co., Indianapolis, Indiana.

Figure 1. Unassembled cannula which was used in re-entrant duodenal and ileal fistulae

The stem (7.6 cc long, 1.3 cm inside diameter, 1.6 cm outside diameter) and flange (3.8 cm long, 1 cm internal radius) were constructed from surgical stainless steel; the elbow and collars were made of Plexiglas

Figure 2. One-half of assembled re-entrant cannula

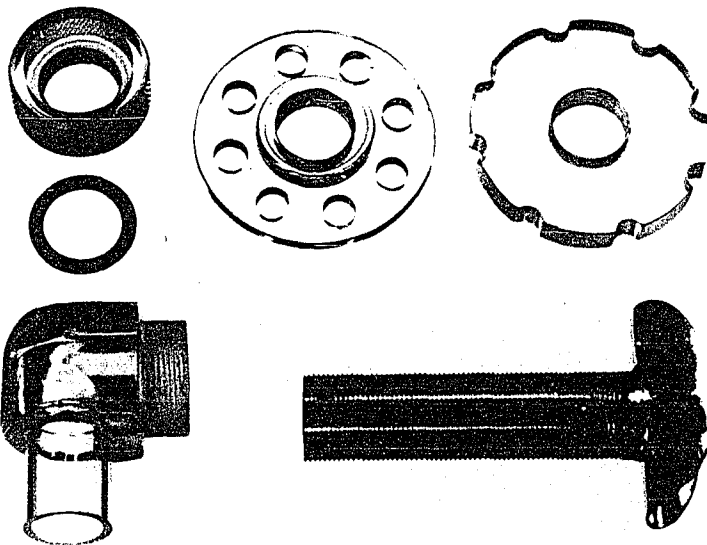
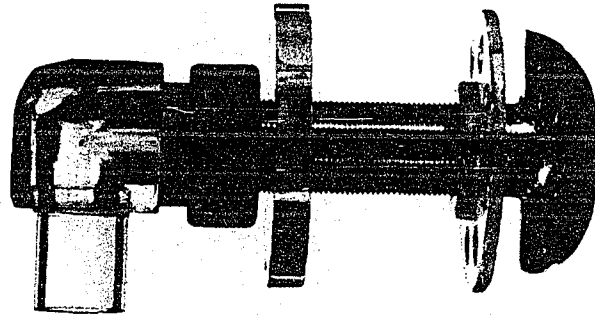


Figure 3. The re-entrant duodenal and ileal cannulae in situ



recovered from surgery but later suffered from closure of the duodenal fistula and was subsequently sacrificed.

Rations and feeding The digestible energy concentration of the diet was varied by incorporating three levels of ground corn cobs (30%, 20% and 10%) into the rations. The respective energy concentrations are expressed as low, medium and high. The compositions of the rations are shown in Table 1. Dehydrated alfalfa pellets and molasses were added to the rations to increase palatability. The rations were fed at a rate of two times the digestible energy (D.E.) required for maintenance calculated by using $D.E. = 76 \text{ kcal} \times w^{0.75}$ where w is body weight in pounds.

Microthene MN710¹, a non-absorbable polyethylene powder was added to each daily allowance as a digestibility marker. This was added at a rate of 5% of the diet and mixed with the ration by hand until the distribution appeared to be homogeneous.

Each animal was fed each ration for 15 days, 10 days during pretrial and 5 days during collections. During the pretrial, the ration was fed twice daily, 1/2 at each feeding. During the collection period, 1/12 of the ration was fed at 6:00 AM and 1/12 at 2 hr intervals thereafter until 8:00 PM when the remaining 5/12 of the ration was fed.

Sample collection and handling Samples from the digestive tract were collected at 2 hr intervals (1 hr after feeding) from 7:00 AM to 7:00 PM. The rumen sampling technique of Balch (1957) was used. The

¹U.S.I. Chemicals, Chicago, Illinois. Particle size of 50 mesh, density .916.

Table 1. Composition of rations

| | <u>Energy concentration</u> | | |
|-----------------------------|------------------------------|--------|------|
| | Low | Medium | High |
| | <u> % </u> | | |
| Ground corn | 48.5 | 58.8 | 69.1 |
| Ground corn cobs | 30.0 | 20.0 | 10.0 |
| Dehy. alfalfa pellets | 5.0 | 5.0 | 5.0 |
| Molasses | 10.0 | 10.0 | 10.0 |
| Urea | 3.5 | 3.2 | 2.9 |
| Dical. phosphate | 2.0 | 2.0 | 2.0 |
| Salt | 1.0 | 1.0 | 1.0 |
| Vitamin-mineral premix | 78g | 78g | 78g |
| Aurofac 10 ^a | 46g | 46g | 46g |
| Calculated D. E. Kcal/kg | 2759 | 2924 | 3089 |

^a

Chlortetracycline 10g/pound, American Cyanamid Co., Princeton,
New Jersey.

cap of the rumen cannula was removed, and a 50 ml bottle with the top closed was held near the reticulo-omasal opening. The bottle was opened, allowed to fill, closed, and removed from the rumen. It was assumed that the fluid collected would represent digesta leaving the rumen. A drop of saturated HgCl_2 was added to stop fermentation. This sample was used to determine digestion coefficients.

Immediately after the rumen sample was taken, a 50 ml sample was drawn from the oral elbow of the duodenal re-entrant fistula by removing the tubing for 10 to 20 sec. Stomach digestibility coefficients were calculated from this sample.

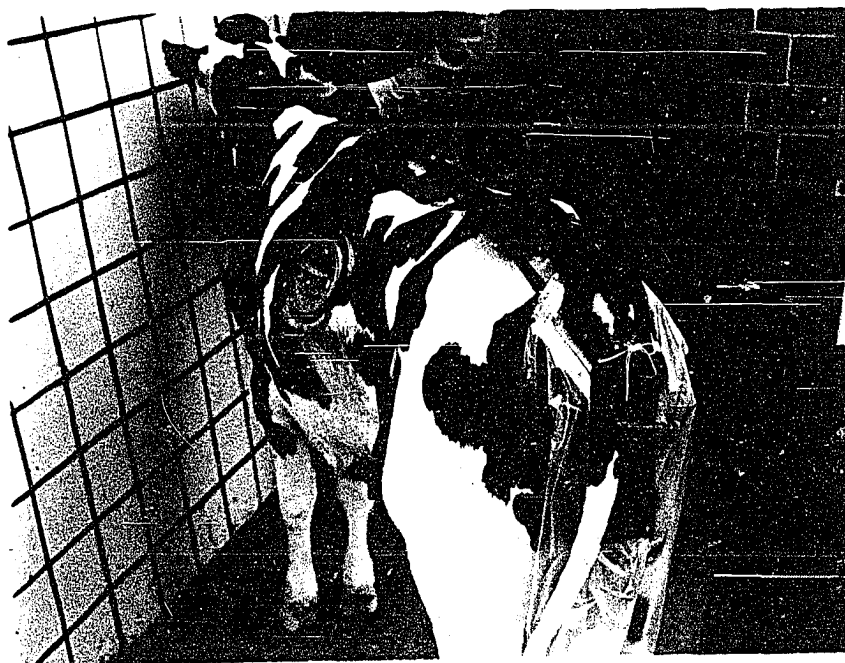
Sampling from the ileal cannula was not successful. In the case of one animal, the digesta were too dry to pass through the cannula. The digesta were fluid in the other animal, but flow was very irregular and insufficient. Consequently, ileal collections were discontinued.

Digesta samples were allowed to accumulate in the refrigerator throughout the day. At the end of the day, these were added to a large plastic bottle holding the frozen samples from the previous days; thus a composite for the 5-day collection period ultimately was attained. The containers were then returned to the freezer.

All feces were saved from the animals during the 5-day collection. The collection harness and bag are shown in Figure 4. The bags were emptied frequently by untying a string which bound the bottom of the bag. Daily collections were stored in a refrigerator after a few thymol crystals were added to each to prevent mold. At the end of the 5-days, the feces

Figure 4. Leather harness and plastic bag used to collect feces

21b



were combined and mixed by use of an industrial potato masher. A sample was taken and frozen for later analysis.

The rumen and duodenum samples were ground while still frozen. The sample in the plastic bottle was reduced to small pieces by use of a large hammer. The bottle then was cut away, and the frozen pieces were placed in a meat grinder which had been cooled with dry ice. Dry ice was frequently added to the grinder to keep it well chilled. When the total sample had been ground, it was thoroughly mixed in a large plastic bag and returned to the freezer while still in the fine granular form. Small portions were taken from several places in the bag when needed for analysis.

Fecal samples were ground and sampled simultaneously by drilling holes in the frozen feces with a high speed power drill and a one-inch wood bit. This was done only as the feces were needed for analysis.

Chemical analysis Ground frozen digesta and fecal samples were used for analysis in all cases but two, energy and ash. Analysis of feed was done on ground air-dry samples.

Dry matter Determination of dry matter on feed, ruminal, duodenal and fecal samples was done by drying for 24 hours in a Virtis¹ freeze drier at 0.3 mm of mercury and 25°C.

Polyethylene Polyethylene content of rumen, duodenal, and fecal samples was determined on the same samples used for dry matter.

¹Virtis Research Equipment, Gardiner, New York.

This was done according to the procedure of Chandler et al. (1964). During the digestion with nitric acid, profuse foaming occurred; therefore a few drops of octyl alcohol were added as an antifoamant.

Water soluble carbohydrates The term soluble carbohydrate usually refers to monosaccharides and disaccharides. Deriaz (1961) claimed that his "soluble carbohydrate" determination included these sugars and also starch. All samples were analyzed by this method and the resulting values were low, suggesting that starch was not fully extracted. Further investigation revealed that corn starch could not be quantitatively recovered by the method. Therefore, the soluble carbohydrate values represent only the portion of the sugars and starch which are soluble in hot water.

Acid detergent fiber and lignin Van Soest's (1963) acid detergent method was used for fiber and lignin determinations. The technique was modified by using octyl alcohol as an antifoamant rather than decalin. The decalin caused the polyethylene in the digesta and fecal samples to dissolve, then cluster around particles during digestion. Consequently these particles were not digested. Octyl alcohol did not cause this clumping.

Energy Since wet digesta and fecal samples were not combustible, it was necessary to dry large samples in the Virtis freeze dryer mentioned earlier. The energy was then determined with a Parr 1230 oxygen bomb calorimeter and a 2601 adiabatic controller.¹

¹ Parr Instrument Company, Moline, Illinois.

Ash The same dried material prepared for energy determinations was used for ash determinations. Each sample (weighing about 2 g) was weighed into a porcelain crucible and placed in a muffle furnace, preheated to 600°C, for 15 hrs; then it was cooled in a desiccator for one hr and weighed.

Organic matter Ash was subtracted from dry matter to obtain organic matter.

Total nitrogen The total nitrogen of all material was determined by the procedure outlined by Meeker and Wagner (1933) with the following modifications: 1) Hengar¹ granules were used as a catalyst instead of K_2SO_4 and $-CuSO_4$, 2) frozen digesta and feces were weighed into Whatman extraction thimbles which were then dropped into the flasks, 3) the ammonia was distilled into 2% boric acid and titrated with 0.1N HCL instead of H_2SO_4 , 4) methyl red-methylene blue was used as the indicator rather than methyl red.

Urea and ammoniacal nitrogen The urea and ammoniacal nitrogen method of AOAC (1965) was modified by using decahydronapthalene as an antifoamant and by collecting ammonia in 2% boric acid and titrating with 0.1N HCL to a methyl red-methylene blue end point.

Computation of digestion coefficients Polyethylene and lignin were used as non-digestible markers to determine extent of digestion in

¹ Hengar Company, Philadelphia, Pennsylvania.

each portion or segment of the tract. The following general equation was used:

$$\% \text{ nutrient digested} = 100 - \left(100 \times \frac{\text{marker in feed } (\%)}{\text{marker in feces } (\%)} \times \frac{\text{nutrient in feces } (\%)}{\text{nutrient in feed } (\%)} \right)$$

Expression of digestion . Digestion in and along the tract was expressed three ways: 1) progression of digestion along the tract expressed as coefficient of digestion for the ruminoreticulum, total stomach and total tract, 2) coefficients of digestion for undigested residues passing through the omasum-abomasum and the intestine, where the residue is considered as feed for that segment, 3) percentage of total digestion occurring in the various segments; namely, ruminoreticulum, omasum-abomasum and intestine.

Experimental design The trial was analyzed as a 2 x 3 factorial where animals were one factor and rations the other. Significant differences between means were determined by Duncan's new multiple range test as prescribed by Harter (1960).

Results and discussion

Selection of a non-digestible marker A reference material must possess various characteristics before it can be considered reliable. These are: 1) It is palatable and does not alter the normal digestive process. 2) It is not absorbed to any significant degree. 3) The reference substance and nutrient under investigation move together through the gastrointestinal tract. Many of the commonly used markers do not have these characteristics. Polyethyleneglycol (PEG), because

of its water soluble nature, lends itself well to tracing the flow of water and solutes along the digestive tract, but it does not move through the tract at the same rate as dry matter residues, and a variable diurnal excretion pattern is observed in the feces (Corbett et al., 1958; Sperber et al., 1953). This variation in PEG concentration in the digesta or feces would result in erroneous digestion coefficients derived from analysis of "grab" samples.

Chromic oxide also exhibits a variable diurnal excretion pattern (Balch et al., 1957; Corbett et al., 1958; Elam et al., 1959), especially when included in high concentrate rations (Bloom et al., 1957). Consequently, neither PEG nor chromic oxide were considered as suitable markers for this experiment.

Recently, polyethylene powder with a particle size of 50 mesh and a density of .916 was tested by Chandler et al. (1964). They found that this marker was not absorbed, did not effect palatability and was distributed in the tract in a manner similar to dry matter.

Polyethylene powder was used as a reference material in Experiment I, but recovery was not satisfactory. For three 5-day collection periods, recoveries ranged from 53.3% to 66.0% for one steer and from 54.9 to 89.6 for the other. The average recovery was only 64.8%. The losses of polyethylene undoubtedly occurred during rumen sampling when some digesta were lost. Since the specific gravity of microthene is low, it would rise to the dorsal parts of the rumen and would be highly concentrated in the lost material.

Lignin also possesses the characteristics of a reliable reference substance. Since it is a natural constituent of feeds, it does not affect palatability or normal digestive processes, and it moves down the tract together with the undigested nutrients. However, some researchers have questioned whether lignin can pass through the tract without being digested since several experiments have resulted in poor lignin recovery in the feces (Adolph et al., 1947; Ely et al., 1953; Hale et al., 1940; Hale et al., 1947; Kane et al., 1953; Salo, 1958; Sullivan, 1955).

The method of lignin analysis can greatly affect lignin recovery. Since lignin is not a definite chemical entity, accurate determination is somewhat speculative. Ellis et al. (1946) experimented with a variety of drying temperatures and analytical procedures. From this they derived a method which gave minimum lignin values which were highly repeatable. A variation of this method has been used by Kane et al. (1953); however, their "true" lignin digestibility results were less satisfactory than "crude" lignin results obtained by the method of Ellis et al. (1946). Balch et al. (1954) compared several lignin analyses for the purpose of accurately determining digestibility. They found the method of Armitage et al. (1948) to be the most satisfactory for all types of feedstuffs. Rogerson (1958) also found this method to be reliable on hay or concentrates. Another method of lignin determination has been proposed by Salo (1957); however, it gives satisfactory recovery values for mature hay but not hay cut in the early growth stage.

The influence of feedstuff on lignin recovery has also been observed by other researchers. Adolph et al. (1947) found that alfalfa lignin is more digestible from morning cuttings than afternoon cuttings. Comparing lignin digestibility of orchard grass hay cut at four stages of maturity, Ely et al. (1953) found the lignin of immature hay to be less digestible than the more mature hays. In contrast, Sullivan (1955) observed that lignin digestibility decreased as the maturity of timothy hay progressed. Salo (1958) found similar results for timothy-clover hay.

In spite of a variety of feedstuffs analyzed and analytical methods used, lignin has proved to be a satisfactory marker in many experiments. Crampton and Maynard (1938) fed grass to a steer and rabbits and recovered 99% of the lignin. Ellis et al. (1946) studied recoveries from grass diets of sheep and observed values of 96% to 104%. Using the method of Ellis et al. (1946), Forbes and Garrigus (1948) measured average recoveries of 102% for steers on various pasture forages. Nearly complete recoveries of lignin have been observed when various hays were fed to sheep (Forbes et al., 1946; Gray, 1947; Rogerson, 1958) and when alfalfa hay and silage were fed to dairy cows (Kane et al., 1950).

"Lignin recovery does not seem to be affected by addition of concentrates. Swift et al. (1947) measured digestion coefficients of high concentrate sheep rations using total collection and lignin-ratio techniques. They found that the methods gave nearly equal results. Rogerson (1958) observed similar results when corn alone was fed to sheep. Using cows, Ellis et al. (1946) recovered 102% of the lignin in a hay-concentrate

diet. Balch et al. (1954) compared various methods of lignin determinations on various diets fed to cows and found that method had little effect on recovery of lignin when concentrates were added to the diet. Their average recovery for various ratios of hay to concentrate was 101%. Recently, Johnson (1965) used the method of Van Soest (1963) to determine lignin in feed and feces of steers on a fattening ration of corn. His recovery was 103%.

Since nearly all of the literature indicated that lignin recovery was satisfactory for high concentrate diets and because all other common reference materials had serious shortcomings, lignin was chosen as the reference material for this experiment.

The average lignin recovery for Experiment I was 90.7%. This value may not be low because of the loss of some of the lignin from the tract during sampling.

Progressive dry matter digestion The coefficients of digestion for progressive digestion along the tract are given in Table 2. These values are graphically represented in Figures 5, 6, 7 and 8. The pattern of dry matter digestion shown in Figure 5 reflects the digestion pattern of all nutrients. For all rations, ruminoreticular dry matter digestion coefficients were between 70% and 80%. The subsequent total stomach coefficients dropped 15% to 20%, but intestinal digestion raised the coefficients back near the level of the ruminoreticulum. This general pattern has been observed by other researchers. Balch (1957) did not measure total stomach digestion but observed that ruminoreticular digestion represented nearly all of the total digestion in cattle. Similarly,

Table 2. Progressive digestion along the tract expressed as coefficients of digestion for each portion at different energy concentrations

| | | | Energy concentration | | |
|------------------------------------|-----------------|------|----------------------|--------------------|-------------------|
| Portion of tract | | | Low | Medium | High |
| | | | % | | |
| Dry matter | Ruminoreticulum | (RR) | 73.3 ^a | 78.1 ^b | 80.1 ^b |
| | Total stomach | (TS) | 56.7 | 58.0 | 62.4 |
| | Total tract | (TT) | 74.6 ^c | 76.0 ^c | 83.0 ^d |
| Energy | | (RR) | 69.9 ^a | 75.2 ^b | 77.5 ^b |
| | | (TS) | 53.6 | 55.6 | 60.1 |
| | | (TT) | 70.8 ^c | 71.9 ^c | 80.5 ^d |
| Organic matter | | (RR) | 73.9 ^a | 78.8 ^b | 80.8 ^b |
| | | (TS) | 59.2 | 60.2 | 64.4 |
| | | (TT) | 74.9 ^c | 76.5 ^c | 83.5 ^d |
| Water soluble carbohydrate | | (RR) | 86.1 | 88.6 | 87.6 |
| | | (TS) | 80.0 | 75.9 | 79.0 |
| | | (TT) | 91.2 | 88.8 | 92.5 |
| Fiber | | (RR) | 46.7 ^B | 49.9 ^B | 36.2 ^A |
| | | (TS) | 46.5 ^b | 41.7 ^{ab} | 32.2 ^a |
| | | (TT) | 47.6 | 46.2 | 39.6 |
| Ash | | (RR) | 63.6 | 66.7 | 67.8 |
| | | (TS) | 15.8 | 16.8 | 25.2 |
| | | (TT) | 70.4 | 67.5 | 72.2 |
| Total nitrogen | | (RR) | 73.5 | 78.0 | 76.4 |
| | | (TS) | 54.4 | 52.8 | 55.9 |
| | | (TT) | 80.1 | 80.6 | 82.5 |
| Urea and NH ₃ -nitrogen | | (RR) | 94.8 | 95.6 | 96.2 |
| | | (TS) | 93.1 | 94.8 | 95.3 |
| | | (TT) | 98.2 | 98.7 | 98.3 |

Values with different small (ab) superscripts are significantly different ($P < 0.1$).

Values with different small (cd) superscripts are significantly different ($P < 0.25$).

Values with different capital (AB) superscripts are significantly different ($P < 0.01$).

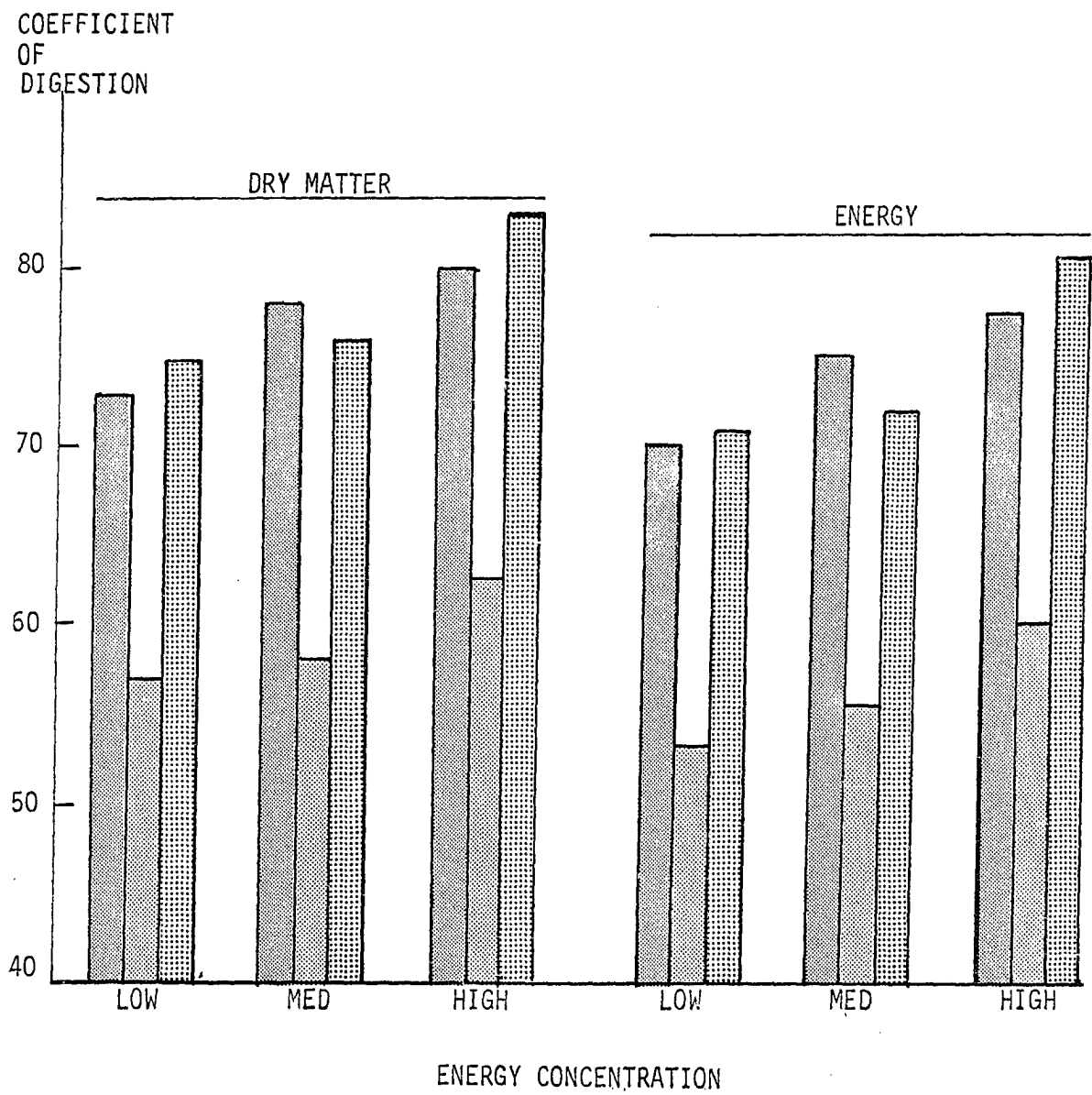
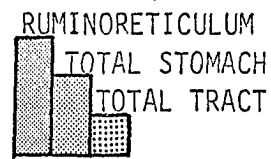


Figure 5. Progressive digestion of dry matter and energy

RUMINORETICULUM
TOTAL STOMACH
TOTAL TRACT

COEFFICIENT
OF
DIGESTION

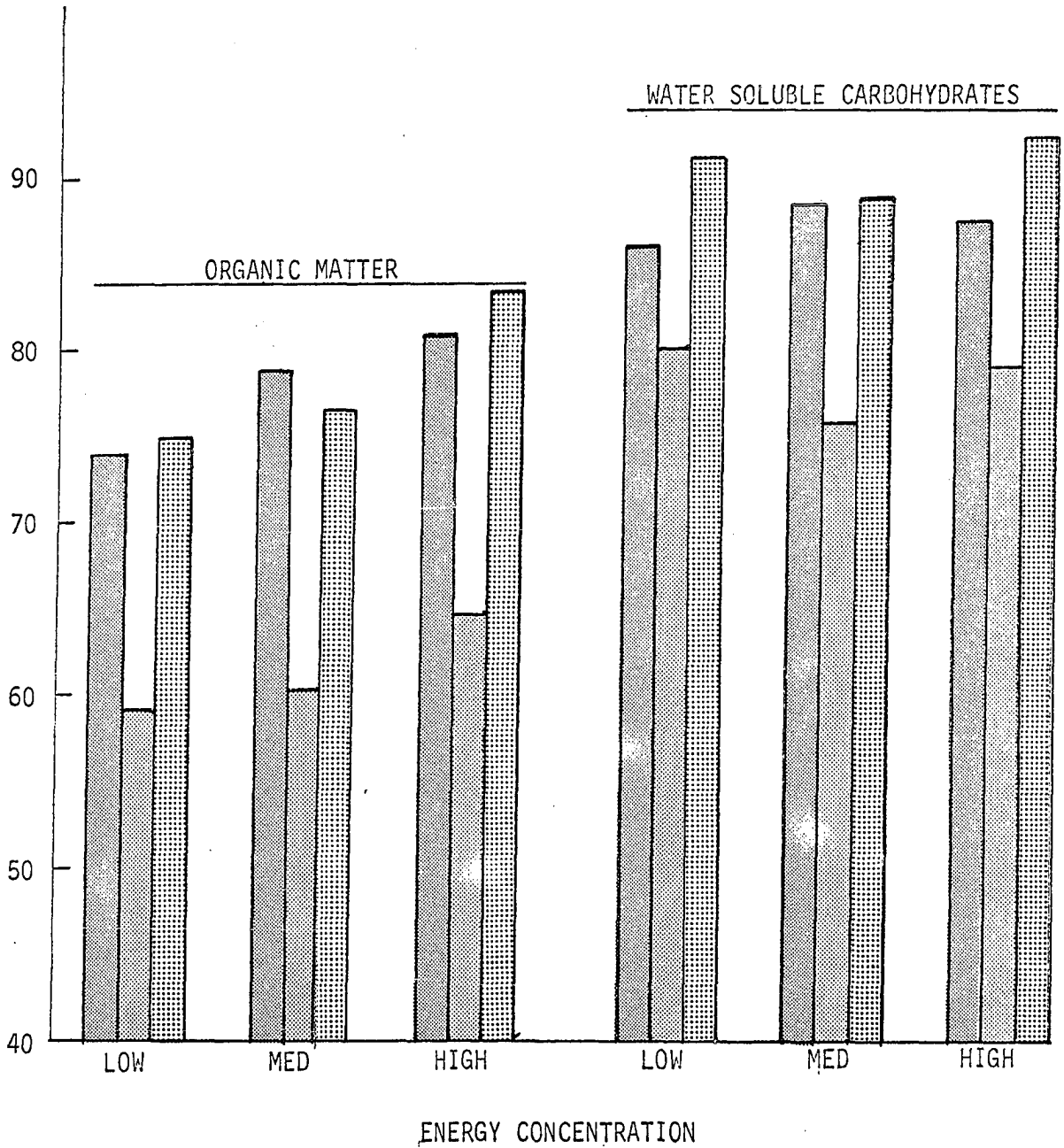


Figure 6. Progressive digestion of organic matter and water soluble carbohydrate

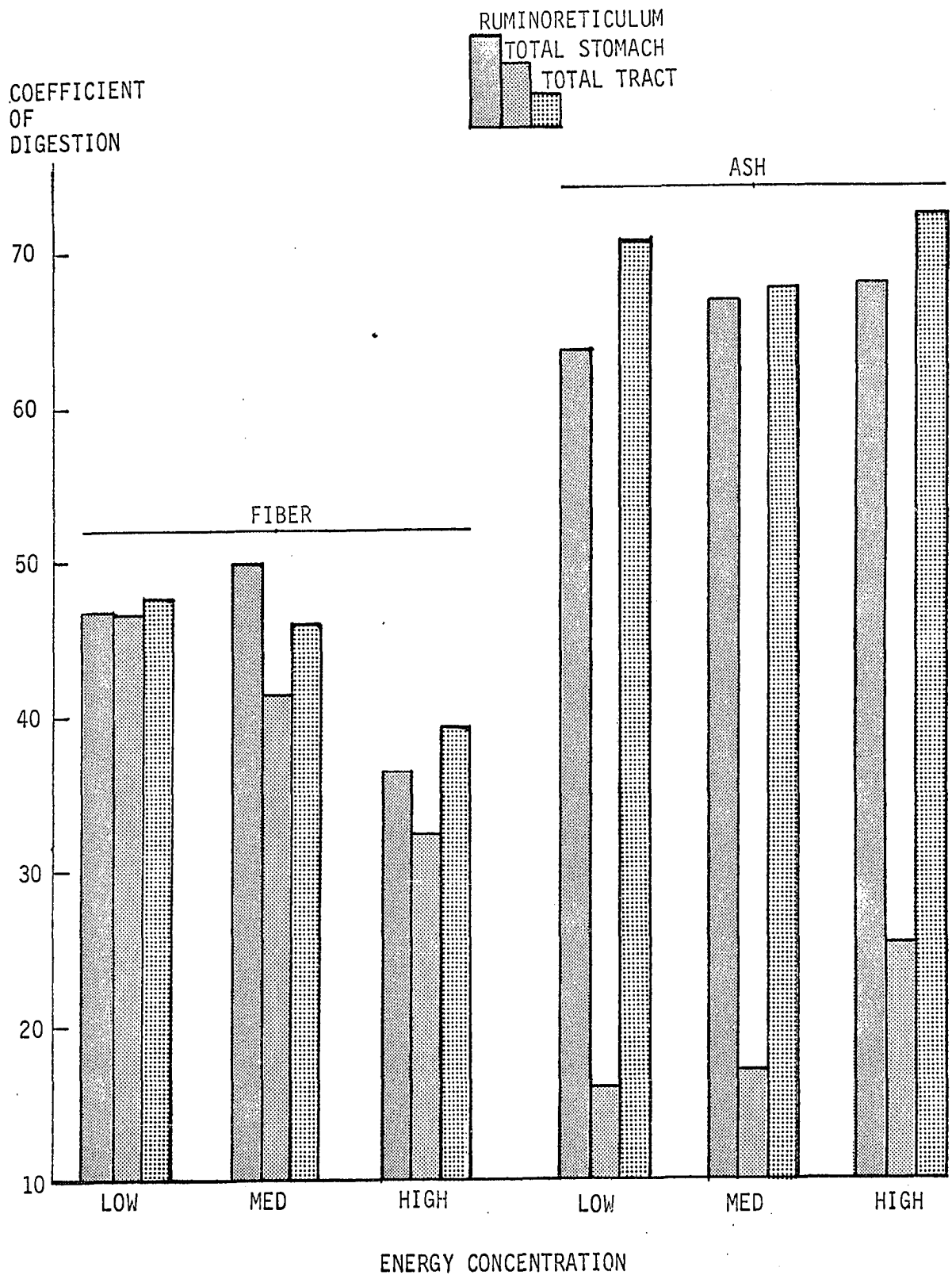
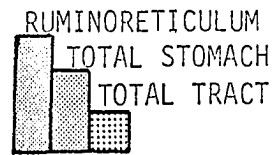


Figure 7. Progressive digestion of fiber and ash



COEFFICIENT
OF
DIGESTION

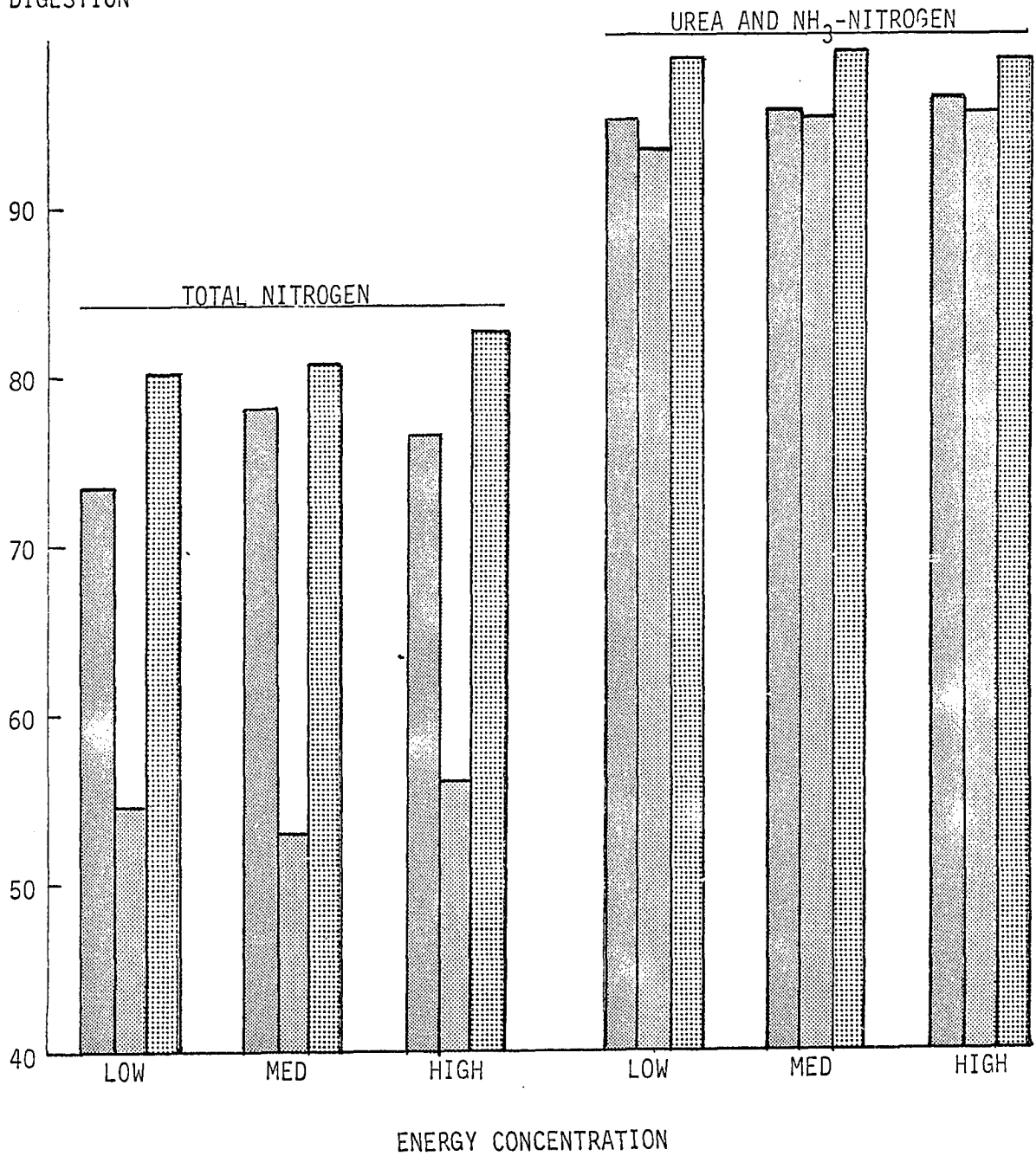


Figure 8. Progressive digestion of total nitrogen and urea and NH₃-nitrogen

Gray et al. (1958a) found that the forestomach of sheep digested nearly all of the digestible dry matter.

The low digestion coefficient for the stomach, as compared to the ruminoreticulum, resulted from endogenous additions of dry matter between the reticulo-omasal opening and the duodenal fistula. Most, if not all, occurred in the abomasum and the two or three inches of duodenum orad to the fistula. This is clearly expressed in Table 3 where digestion of the dry matter residues entering the omasum-abomasum were negative and ranged from -62.3% to -93.2%. Further examination of the table reveals that concentrations of all the nutrients increased in the omasum-abomasum, and those which contributed most were ash and nitrogen. These same nutrients were the major additions into the abomasum and small intestine of sheep sacrificed by Rogerson (1958). Large ash additions into this area have also been observed by McGilliard (1961) and Yang and Thomas (1965).

Intestinal dry matter digestion approached or exceeded dry matter additions of the omasum-abomasum; consequently total digestion coefficients were nearly the same or slightly greater than ruminoreticular digestion coefficients. McGilliard (1961), Rogerson (1958) and Yang and Thomas (1965) have reported similar intestinal digestion.

Effect of energy concentration on dry matter digestion As energy concentration of the diet was increased, the coefficients of digestion of dry matter increased in the ruminoreticulum (Table 2). The coefficient for the low energy diet was significantly lower than the others ($P < 0.1$).

Table 3. Coefficients of digestion in various segments for the residues entering that segment

| | | | Energy Concentration | | |
|------------------------------------|-----------------|------|----------------------|-------------------|-------------------|
| | | | Low | Medium | High |
| | | | % | | |
| Dry matter | Omasum-abomasum | (OA) | -62.3 | -93.2 | -88.4 |
| | Intestine | (I) | 41.5 ^C | 42.1 ^C | 54.9 ^D |
| Energy | | OA | -54.1 | -79.8 | -61.2 |
| | | I | 37.2 ^C | 35.7 ^C | 51.5 ^D |
| Organic matter | | OA | -56.3 | -88.4 | -84.8 |
| | | I | 38.5 ^C | 39.8 ^C | 54.1 ^D |
| Water soluble carbohydrate | | OA | -43.1 | -127.0 | -71.3 |
| | | I | 52.2 ^C | 48.4 ^C | 64.7 ^d |
| Fiber | | OA | -0.4 | -17.1 | -6.2 |
| | | I | 5.6 ^c | 7.5 ^c | 11.0 ^d |
| Ash | | OA | -132.8 | -149.3 | -127.6 |
| | | I | 64.2 | 60.9 | 60.7 |
| Total nitrogen | | OA | -71.3 | -114.6 | -86.4 |
| | | I | 55.6 | 58.2 | 58.3 |
| Urea and NH ₃ -nitrogen | | OA | -33.0 | -22.5 | -20.9 |
| | | I | 73.2 | 76.1 | 60.4 |

Values with different small (cd) superscripts are significantly different ($P < .25$).

Values with different capital (CD) superscripts are significantly different ($P < .05$).

Higher energy concentration also appeared to increase total stomach digestion coefficients, but the differences were not significant and may have been partially influenced by differences in ruminoreticular digestion. The high energy coefficient for the total tract was higher ($P < 0.25$) than the other total tract values.

It is possible that secretions into the abomasum nullified some of the ruminoreticular digestive differences. These secretions, expressed in Table 3 as negative digestion coefficients for residues passing through the omasum-abomasum, show no relationship to the energy concentration.

Intestinal digestion of dry matter residues was affected by energy concentration. The dry matter digestion coefficient for the high energy diet was significantly greater than that of the other diets ($P < 0.05$). Generally, it can be concluded that an increase in energy concentration will cause greater dry matter digestion in the ruminoreticulum and in the intestine, the result being an increase in total digestion. Since digestion increased in the upper and lower portions of the tract, the percentage of total dry matter digestion occurring in the various segments did not change significantly (Table 4).

The results of this experiment are similar to those obtained by Rogerson (1958) in that he also observed increases in ruminoreticular and intestinal dry matter digestion when energy in the diet was increased. Balch (1957), on the other hand, only observed an increase in ruminoreticular digestion and not lower tract digestion when concentrate

Table 4. Percentage of total digestion occurring in various segments of the tract

| | | | Energy concentration | | |
|------------------------------------|-----------------|------|----------------------|--------|-------|
| Segment of tract | | | Low | Medium | High |
| | | | % | | |
| Dry matter | Ruminoreticulum | (RR) | 98.3 | 102.9 | 96.6 |
| | Omasum-abomasum | (OA) | -22.3 | -26.7 | -21.5 |
| | Intestine | (I) | 24.0 | 23.8 | 24.9 |
| Energy | RR | | 98.8 | 104.8 | 96.3 |
| | OA | | -23.1 | -27.7 | -21.8 |
| | I | | 24.3 | 22.9 | 25.5 |
| Organic matter | RR | | 98.8 | 103.1 | 96.8 |
| | OA | | -19.7 | -24.4 | -19.8 |
| | I | | 20.9 | 21.3 | 23.0 |
| Water soluble carbohydrates | RR | | 94.5 | 99.9 | 94.8 |
| | OA | | -6.5 | -14.2 | -9.3 |
| | I | | 12.0 | 14.3 | 14.5 |
| Fiber | RR | | 98.3 | 108.2 | 91.3 |
| | OA | | -0.3 | -18.2 | -10.0 |
| | I | | 1.9 | 10.0 | 18.7 |
| Ash | RR | | 90.4 | 99.0 | 93.7 |
| | OA | | -68.6 | -74.2 | -59.5 |
| | I | | 78.2 | 75.2 | 65.8 |
| Total nitrogen | RR | | 91.7 | 96.8 | 93.3 |
| | OA | | -24.1 | -31.6 | -26.2 |
| | I | | 32.4 | 34.8 | 32.9 |
| Urea and NH ₃ -nitrogen | RR | | 96.6 | 96.9 | 97.7 |
| | OA | | -1.7 | -0.8 | -0.8 |
| | I | | 5.1 | 3.9 | 3.1 |

was increased in the diet of a cow. In contrast, McGilliard (1961) observed that proportionately less dry matter was digested in the stomach as concentrate was increased up to 70% of the diet. This energy increase had an opposite effect on the residues passing through the intestine, namely an increase in digestibility. This last observation agrees with the results of my experiment and the trials of Rogerson (1958).

It must be kept in mind that Balch (1957), McGilliard (1961) and Rogerson (1958) changed the physical nature of their rations as they increased the energy, and the effects of these physical changes cannot be compared. Yang and Thomas (1965) on the other hand made an attempt to control the physical nature of their calf rations while varying the fiber content. As they increased fiber, they lowered digestible dry matter and digestible organic matter. In this respect their experiment was similar to mine. Unlike my experiment, their calves were allowed to consume feed ad libitum, and digesta were obtained by sacrificing the calves, ligating the tracts and sampling from the segments. In agreement with the results of my experiment, they observed a direct relationship between rumen digestion coefficients and energy content of the diet. But contrary to my results, they observed that residues of the high energy diets were digested to a lesser degree in the intestine. The differences in results may lie with the ration and feeding or the methods used to procure samples.

Effect of energy concentration on energy and organic matter

digestion The actual digestible energy values of the low, medium and

high energy rations were 2869, 2875, and 3251 kcal per kg, respectively, not greatly different than the predicted values shown in Table 1.

As would be expected, the diets had the same effect on energy and organic matter digestion as they did on dry matter digestion. The patterns of digestion were so similar that significant differences were repeated for the ruminoreticular and total tract digestion coefficients (Table 2) and the intestinal digestion coefficients for passing residue (Table 3). McGilliard (1961) and Yang and Thomas (1965) examined organic matter digestion; their dry matter and organic matter digestion patterns were also very similar. Since the relationship between my organic matter data and theirs is essentially the same as that discussed for dry matter, the discussion will not be repeated here.

Effect of energy concentration on carbohydrate digestion The digestible water soluble carbohydrate (WSC) concentration was 18.5%, 21.5% and 25.1% for the low, medium and high energy rations respectively. However, energy concentration had no influence on the WSC coefficients of digestion for the ruminoreticulum, stomach or total tract (Table 2).

Freer and Campling (1963) and Rogerson (1958), on the other hand, showed that increasing the concentrate portion of the diet increased ruminoreticular digestion of NFE. McGilliard's (1961) data, like mine, showed no dietary effect on total stomach digestion of soluble carbohydrates.

Table 3 indicates that diet did affect WSC digestion of residues in the intestine. The value for the high energy diet was greater than

the other diets, ($P < 0.25$), suggesting that some soluble carbohydrates may escape rumen fermentation when high concentrate rations are fed. Other researchers have observed the same diet effect on NFE digestion in the intestine (McGilliard, 1961; Rogerson, 1958).

Table 4 lists the proportion of digestible WSC digested in the various segments of the tract. The ruminoreticular values are very high, 94.5% to 99.9%. Hale *et al.* (1947) also observed a high value (100.2%) for NFE digestion in the rumen. McGilliard (1961), Ridges and Singleton (1962) and Sineshchekov (1965) observed lower values (near 80%) for the total stomach. Their values would be expected to be lower because of endogenous secretion of carbohydrate into the abomasum in the form of hexosamine.

Energy concentration had a much greater influence on fiber digestion than on WSC digestion. The fiber digestion coefficient for the ruminoreticulum was much lower for the high energy diet than for the other diets (Table 2). This difference influenced the digestion coefficients for total stomach and the total tract, but the total tract coefficients were not significantly different.

Several other researchers have also observed that ruminoreticular or stomach digestion of crude fiber decreased as dietary concentrate increased (Balch, 1957; McGilliard, 1961; Yang and Thomas, 1965). This reduction in digestion may have been caused by the addition of starch which ferments rapidly and reduces the pH of the rumen. This increased acidity subsequently affects the sensitive cellulose digesting organisms (Head, 1961).

The reduced ruminoreticular digestion of fiber in the high energy diet was accompanied by a significant increase in digestion of fiber passing through the intestine (Table 3). Freer and Campling (1963) and Rogerson (1958) observed the same pattern. The intestinal digestion of fiber evidently occurred in the large intestine where cellulose fermentation has been demonstrated by Gray (1947) and Hale et al. (1947).

Table 2 and Table 3 suggest that fiber is secreted into the omasum-abomasum, which is an impossibility. The "negative digestion" results from the technique which unavoidably included some endogenous ash and protein in acid-detergent fiber (Colburn and Evans, 1967). Negative digestion values for fiber in the abomasum has also been observed by Yang and Thomas (1965) who used the same technique.

Effect of energy concentration on ash digestion Energy had no influence on ash digestion in any segment of the tract. However, the experiment did confirm that large amounts of ash are secreted into the abomasum (Table 3). McGilliard (1961), Rogerson (1958) and Yang and Thomas (1965) also measured large secretions. McGilliard (1961) observed greater ash secretions for high concentrate diets, while Yang and Thomas (1965) observed the greatest ash secretion for high fiber diets. The differences between rations and methods used in their experiments do not make them comparable.

Effect of energy concentration on total nitrogen and urea and ammoniacal nitrogen digestion Total nitrogen and urea and ammoniacal

nitrogen (urea and $\text{NH}_3\text{-N}$) digestion will be discussed together because urea represented a large part of the dietary nitrogen.

Energy concentration had no significant effect on the digestion of either of these nutrients. The nature of the diets or feeding practice caused a large, unexpected absorption of nitrogen from the ruminoreticulum for all rations (Table 2 and Figure 8). The factors which cause rumen absorption of nitrogen are thoroughly discussed in the review of literature.

The major causes of ruminoreticular nitrogen absorption were probably the large percentage of dietary nitrogen in the form of urea (51% to 61%) and the solubility of the urea. The conversion of this urea nitrogen to ammonia and the subsequent absorption of ammonia must have occurred very rapidly; the urea and $\text{NH}_3\text{-N}$ levels in the ruminoreticulum 1 hr after feeding were almost nil. This rapid conversion and absorption may have been caused not only by the solubility of urea but by the low plane of nutrition and the frequent feeding techniques. Because of these latter two factors, only 8.2-11.1 g of nitrogen entered the rumen every two hours; the rumen easily metabolized much of this to ammonia and absorbed it within a short period of time. Rapid metabolism of urea was also observed by Pearson and Smith (1943) when they fed 40 g of urea to a steer and were unable to detect urea in the digesta 1 hr after feeding.

The ammonia nitrogen absorbed from the ruminoreticulum is converted to urea by the liver and is excreted in the urine except for a small

amount returned to the rumen via saliva and through the rumen wall. In this experiment ruminoreticular losses nearly equalled total digestion (Table 2). Consequently, very little nitrogen could have been retained on any of the diets unless substantial amounts of amino acids were absorbed from the rumen. Nitrogen retention was not determined in the experiment, but body weight changes were observed. Body weight changes were sometimes negative and averaged less than 0.5 kg gain per day during collection periods.

Experiment II

Procedure

Animals The two steers used in Experiment I were again used for Experiment II. Prior to the initiation of Experiment II both steers developed infections around their ileal cannulae which caused subsequent sloughing of the cannulae. Once the cannulae had been expelled, the lesions healed rapidly and completely.

Ration and feeding The rations were the same as those used in Experiment I, shown in Table 1. To study the effects of energy level as well as energy concentration, all rations were fed at 1.0 and 1.5 times maintenance requirement for digestible energy.

Each steer was fed each ration for 30 days; 15 days at 1.0 times maintenance and 15 days at 1.5 times maintenance. At each level of energy, 10 days were allowed for adjustment, and samples were subsequently collected for 5 days. Aside from these changes, feeding was the same as in Experiment I.

Sample collection and handling Since some digesta had been lost during rumen sampling in Experiment I, and the effect of this loss on digestibility at different levels of energy could not be predicted, ruminoreticular samples were not taken.

Duodenal and fecal samples were collected and handled the same as in Experiment I.

Chemical analysis Duodenal digesta and feces were analyzed for dry matter, acid detergent fiber and lignin, energy, ash, organic matter, total nitrogen and urea and $\text{NH}_3\text{-N}$, using the same procedure as in Experiment I.

Expression of digestion Digestion coefficients were computed the same as in Experiment I using lignin as the digestibility marker. Digestion along the tract was expressed in three ways: 1) progression of digestion along the tract expressed as coefficient of digestion for the total stomach and the total tract, 2) coefficient of digestion for residues passing through the intestine, 3) percentage of total digestion occurring in the total stomach and the intestine.

Experimental design The trial was analyzed as a 2 x 3 factorial, where energy level was one factor and energy concentration the other. Since the steers were used repeatedly for all treatments, they were treated as blocks. Significant differences between means were determined by Duncan's new multiple range test as prescribed by Harter (1960).

Results and Discussion

During the delay between Experiment I and Experiment II, ad libitum feed consumption decreased for both steers to the extent that they would

not consume 2 times maintenance requirement of D.E. It was speculated at the time that scar tissue was developing around the flanges of the duodenal cannulae and restricting flow of digesta. After the experiment, necropsy revealed massive scar tissue around the flanges of the oral cannulae in both steers. The effect of flow restriction on digestion in the various segments may have had little or no influence on differences between rations. However, since the effect could not be measured, the results of Experiment II should be viewed with some reservations.

Effect of energy concentration and energy level on dry matter digestion Energy concentration had no significant effect on average dry matter coefficients of digestion for the total stomach or total tract (Table 5). These values were very similar to comparable values of Experiment I (Table 2).

The digestion coefficients for dry matter residues passing through the intestine (Table 6) were lower than those of Experiment I (Table 3), but response to energy concentration was similar. However, energy concentration had no significant effect on intestinal dry matter digestion. Energy concentration also failed to influence the percentage of dry matter digestion occurring in the stomach or intestine (Table 7). The stomach dry matter digestion represented about 80% of the total, a value which is somewhat higher than the 70% observed by Hogan (1957) for sheep and considerably higher than the 43% observed by McGilliard (1961) for a 70% concentrate-30% hay diet of a steer. The difference between my experiment and theirs may have been the physical nature of the roughage fed, ground

Table 5. Coefficients of digestion for the total stomach (TS) and total tract (TT) at different energy concentrations and energy levels

| Energy concentration | | Low | | Medium | | High | | Average | |
|----------------------|-----|-------------------|------|--------------------|------|--------------------|------|-------------------|-------------------|
| Energy level | | 1.0 | 1.5 | 1.0 | 1.5 | 1.0 | 1.5 | 1.0 | 1.5 |
| % | | | | | | | | | |
| Dry matter | TS | 57.8 | 61.6 | 53.2 | 65.6 | 58.2 | 66.6 | 56.4 | 64.6 |
| | Avg | 59.7 | | 59.4 | | 62.4 | | | |
| | TT | 75.0 | 70.0 | 77.0 | 77.1 | 76.6 | 78.9 | 76.2 | 75.3 |
| | Avg | 72.5 | | 77.0 | | 77.8 | | | |
| Energy | TS | 54.5 | 60.5 | 49.9 | 64.3 | 55.7 | 63.9 | 53.3 | 62.9 |
| | Avg | 57.7 | | 57.1 | | 59.8 | | | |
| | TT | 71.1 | 66.1 | 72.8 | 73.5 | 72.8 | 74.8 | 72.2 | 71.5 |
| | Avg | 68.6 | | 73.1 | | 73.8 | | | |
| Organic matter | TS | 61.2 | 64.5 | 58.1 | 68.9 | 62.0 | 68.7 | 60.4 | 67.4 |
| | Avg | 62.9 | | 63.5 | | 65.4 | | | |
| | TT | 75.2 | 70.8 | 77.3 | 78.2 | 77.8 | 79.5 | 76.8 | 76.2 |
| | Avg | 73.0 | | 77.7 | | 78.7 | | | |
| Fiber | TS | 46.8 | 51.9 | 29.1 | 38.1 | 27.2 | 16.6 | 34.4 | 35.5 |
| | Avg | 49.4 ^A | | 33.6 ^{BD} | | 21.9 ^{BE} | | | |
| | TT | 51.3 | 48.6 | 31.6 | 41.5 | 28.2 | 16.7 | 37.1 ^D | 35.6 ^E |
| | Avg | 49.9 ^A | | 36.6 ^B | | 22.5 ^C | | | |

Values with different capital (DE) superscripts are significantly different ($P < 0.05$).

Values with different capital (ABC) superscripts are significantly different ($P < 0.01$).

Table 5. (Continued)

| Energy concentration | | Low | | Medium | | High | | Average | |
|--------------------------------|-----|------|------|--------|------|-------|------|---------|------|
| Energy level | | 1.0 | 1.5 | 1.0 | 1.5 | 1.0 | 1.5 | 1.0 | 1.5 |
| <hr/> | | | | | | | | | |
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| <hr/> | | | | | | | | | |
| Ash | TS | -6.1 | 9.5 | -28.8 | 6.7 | -6.6 | 30.6 | -13.8 | 15.6 |
| | Avg | 1.7 | | -11.05 | | 11.95 | | | |
| | TT | 71.4 | 55.5 | 71.8 | 56.3 | 57.1 | 69.1 | 66.8 | 60.5 |
| | Avg | 63.4 | | 64.3 | | 63.1 | | | |
| <hr/> | | | | | | | | | |
| Nitrogen | TS | 50.8 | 44.3 | 36.3 | 58.3 | 48.5 | 63.7 | 45.2 | 55.4 |
| | Avg | 47.6 | | 47.3 | | 56.1 | | | |
| | TT | 78.4 | 70.4 | 79.6 | 79.1 | 74.0 | 82.9 | 77.3 | 77.5 |
| | Avg | 74.4 | | 79.4 | | 78.5 | | | |
| <hr/> | | | | | | | | | |
| Urea and NH ₃ -N | TS | 93.4 | 94.1 | 89.2 | 91.4 | 92.6 | 92.6 | 91.7 | 92.7 |
| | Avg | 93.8 | | 90.3 | | 92.6 | | | |
| | TT | 97.9 | 97.2 | 98.1 | 98.2 | 97.6 | 98.6 | 97.9 | 98.0 |
| | Avg | 97.6 | | 98.2 | | 98.1 | | | |

corn cobs vs. long hay. The average percentage of digestion occurring in the total stomach and intestines in Experiment II (80% and 20% respectively) were not greatly different than the average percentages of Experiment I (76% and 24% respectively).

Energy level, like energy concentration, failed to significantly affect dry matter digestion in the stomach, even though the 1.5 level appeared to be higher. This difference did not influence total tract

Table 6. Coefficients of digestion of residues entering the intestine

| Energy concentration | Low | | Medium | | High | | Average | |
|--------------------------------|------|------|--------|------|------|------|-------------------|-------------------|
| Energy level | 1.0 | 1.5 | 1.0 | 1.5 | 1.0 | 1.5 | 1.0 | 1.5 |
| <hr/> | | | | | | | | |
| % | | | | | | | | |
| <hr/> | | | | | | | | |
| Dry matter | 40.9 | 21.9 | 44.6 | 32.0 | 40.8 | 36.9 | 42.1 | 30.3 |
| | Avg | 31.4 | | 38.3 | | 38.9 | | |
| Energy | 36.3 | 14.5 | 39.8 | 24.8 | 36.1 | 30.1 | 37.4 | 23.1 |
| | Avg | 25.4 | | 32.3 | | 33.1 | | |
| Organic matter | 35.9 | 18.1 | 40.7 | 28.9 | 39.1 | 34.4 | 38.6 | 27.1 |
| | Avg | 27.0 | | 34.8 | | 36.7 | | |
| Fiber | 8.1 | -7.0 | 8.0 | 5.3 | 1.4 | -0.3 | 5.8 | -0.7 |
| | Avg | 0.6 | | 6.7 | | 0.6 | | |
| Ash | 72.0 | 49.3 | 69.4 | 51.4 | 50.2 | 53.0 | 63.8 ^a | 51.2 ^b |
| | Avg | 60.5 | | 60.4 | | 51.6 | | |
| Nitrogen | 56.1 | 46.4 | 57.6 | 49.4 | 41.5 | 52.2 | 51.7 | 49.3 |
| | Avg | 51.3 | | 53.5 | | 46.9 | | |
| Urea and NH ₃ -N | 68.3 | 51.0 | 79.4 | 77.8 | 64.2 | 79.5 | 70.5 | 69.4 |
| | Avg | 59.7 | | 78.4 | | 71.9 | | |

Values with different superscripts are significantly different (P < 0.1).

values in that the latter values were nearly equal. The stomach difference was nullified by digestion occurring in the intestine (Table 6) which was apparently, but not significantly, greater for the 1.0 level of maintenance. The percentage of total digestion (Table 7) occurring in the stomach appeared to be greater for the 1.5 level than 1.0 level, but again the differences were not significant.

Effect of energy concentration and energy level on energy and organic matter digestion Energy concentration and energy level affected these components much the same as they did dry matter. Since the effects were so similar, and no significant differences did occur, the discussion will not be repeated.

Effect of energy concentration and energy level on fiber digestion As indicated in Table 5, fiber digestion was significantly affected by energy concentration. As energy concentration increased, fiber digestion in the total stomach decreased. This agrees with the results of Experiment I and the results obtained by other researchers (Balch, 1957; McGilliard, 1961; Yang and Thomas, 1965). Undoubtedly, the difference in digestion occurred in the ruminoreticulum.

The fiber digestion values were also different for the total tract, but this was due primarily to the influence of stomach digestion rather than intestinal digestion since there were no significant differences in intestinal fiber digestion (Table 6). Likewise, there was no significant effect of energy concentration on percentage of fiber digestion in the stomach or intestine (Table 7). Table 7 indicates that a very

Table 7. Percentage of total digestion occurring in total stomach (TS) and the intestine (I)

| Energy concentration | | Low | | Medium | | High | | Average | |
|----------------------|-----|-------|-------|--------|------|-------|------|---------|------|
| Energy level | | 1.0 | 1.5 | 1.0 | 1.5 | 1.0 | 1.5 | 1.0 | 1.5 |
| <hr/> | | | | | | | | | |
| <hr/> | | | | | | | | | |
| % | | | | | | | | | |
| <hr/> | | | | | | | | | |
| Dry matter | TS | 76.9 | 88.3 | 68.8 | 84.9 | 75.8 | 84.4 | 73.8 | 85.9 |
| | Avg | 82.6 | | 76.9 | | 80.1 | | | |
| | I | 23.1 | 11.7 | 31.2 | 15.1 | 24.2 | 15.6 | 26.2 | 14.1 |
| | Avg | 17.4 | | 23.2 | | 19.9 | | | |
| Energy | TS | 76.5 | 91.7 | 68.1 | 87.2 | 76.4 | 85.5 | 73.7 | 88.1 |
| | Avg | 84.1 | | 77.7 | | 81.0 | | | |
| | I | 23.5 | 8.3 | 31.9 | 12.8 | 23.6 | 14.5 | 26.3 | 11.9 |
| | Avg | 15.9 | | 22.4 | | 19.1 | | | |
| Organic matter | TS | 81.3 | 91.2 | 75.0 | 88.0 | 79.6 | 86.5 | 78.6 | 88.6 |
| | Avg | 86.3 | | 81.5 | | 83.1 | | | |
| | I | 18.7 | 8.8 | 25.0 | 12.0 | 20.4 | 13.5 | 21.4 | 11.4 |
| | Avg | 13.8 | | 18.5 | | 17.0 | | | |
| Fiber | TS | 91.2 | 107.0 | 93.7 | 91.1 | 96.5 | 96.6 | 93.8 | 98.2 |
| | Avg | 99.1 | | 92.4 | | 96.6 | | | |
| | I | 8.8 | -7.0 | 6.3 | 8.9 | 3.5 | 3.4 | 6.2 | 1.8 |
| | Avg | 0.9 | | 7.6 | | 3.5 | | | |
| Ash | TS | -7.1 | 19.7 | -47.6 | 2.4 | -13.3 | 43.4 | -22.7 | 21.8 |
| | Avg | 6.3 | | -22.6 | | 15.1 | | | |
| | I | 107.1 | 80.3 | 147.6 | 97.6 | 113.3 | 56.6 | 122.7 | 78.2 |
| | Avg | 93.7 | | 122.6 | | 85.0 | | | |

Table 7.(Continued)

| Energy concentration | | Low | | Medium | | High | | Average | |
|--------------------------------|-----|------|------|--------|------|------|------|---------|------|
| Energy level | | 1.0 | 1.5 | 1.0 | 1.5 | 1.0 | 1.5 | 1.0 | 1.5 |
| <hr/> | | | | | | | | | |
| <hr/> | | | | | | | | | |
| % | | | | | | | | | |
| <hr/> | | | | | | | | | |
| Nitrogen | TS | 64.8 | 63.6 | 44.1 | 73.1 | 65.1 | 76.7 | 58.0 | 71.1 |
| | Avg | 64.2 | | 58.6 | | 70.9 | | | |
| | I | 35.3 | 36.4 | 55.9 | 26.9 | 34.9 | 23.3 | 42.0 | 28.9 |
| | Avg | 35.9 | | 41.4 | | 29.1 | | | |
| <hr/> | | | | | | | | | |
| Urea and NH ₃ -N | TS | 95.4 | 96.8 | 90.9 | 93.1 | 94.9 | 93.9 | 93.7 | 94.6 |
| | Avg | 96.1 | | 92.0 | | 94.4 | | | |
| | I | 4.6 | 3.2 | 9.1 | 6.9 | 5.1 | 6.1 | 3.2 | 5.4 |
| | Avg | 3.9 | | 8.0 | | 5.6 | | | |

high percentage of fiber digestion occurred in the stomach, probably in the ruminoreticulum as demonstrated in Experiment I (Table 4).

Energy level had no influence on stomach digestion of fiber, but total tract digestion was significantly greater ($P < 0.05$) for the 1.0 level than for the 1.5 level. Intestinal digestion of residual fiber influenced the total digestion, even though the intestinal values were not significantly different (Table 6).

Effect of energy concentration and energy level on ash digestion

Coefficients of digestion of ash fluctuated greatly between animals and among rations; consequently there were no significant differences among energy concentrations for total stomach or total tract (Table 5).

More ash was added at the abomasum than was absorbed from the total stomach of one steer. It was of such magnitude that several of the average coefficients for the total stomach were negative. McGilliard (1961) also observed negative ash digestion coefficients for the stomach, some exceeding -100% for high concentrate diets.

The intestines absorbed an average of 57% of the residual ash (Table 6). This represented nearly 100% of all ash absorption (Table 7).

Differences due to energy level were rendered non-significant due to the animal variation except for coefficients of digestion for ash residue passing through the small intestine (Table 6). Here significantly more ($P < 0.10$) residual ash was digested in the intestine at the 1.0 level than at the 1.5 level. This response was probably caused by the amount of endogenous ash present and not a direct effect of the rations.

Effect of energy concentration and energy level on total nitrogen and urea and NH_3 -N digestion Neither concentration nor level of energy had any significant effect on digestion of nitrogen. Coefficients of digestion of nitrogen were about 50% for the stomach (Table 5), which is not greatly different from that observed for Experiment I (Table 2), indicating a large quantity of nitrogen absorption from the ruminoreticulum. The factors which caused ruminoreticular absorption of nitrogen in Experiment I undoubtedly affected nitrogen absorption in this experiment. Intestinal digestion coefficients (Tables 3 and 6) and the percentages of nitrogen digested in the intestine (Tables 4 and 7) were similar for the two experiments.

Urea and $\text{NH}_3\text{-N}$ digestion, like total nitrogen digestion, was unaffected by energy concentration or level. Nearly all of the urea and $\text{NH}_3\text{-N}$ had been absorbed as ammonia or incorporated into microbial protein before the digesta reached the duodenum (Table 5). The small amount of urea and $\text{NH}_3\text{-N}$ present in the duodenal digesta may have been partially endogenous. This was subsequently absorbed in the intestine (Table 6). Again, digestion coefficients were not greatly different than those observed for Experiment I.

General Discussion

Generally, the results of these experiments indicate that as energy concentration increases, digestion of dry matter increases in both the ruminoreticulum and the intestine. These increases would be expected since there were more digestible nutrients in the higher energy rations. However, an individual examination of most of these nutrients revealed that energy concentration does not greatly affect the degree of their digestion along the tract. In the one exception, fiber, digestion in the ruminoreticulum was greater for low energy diets. When energy concentration was increased, ruminoreticular digestion of fiber was depressed. Head (1961) has speculated on the factors which affect ruminoreticular fiber digestion but further study is warranted.

The experiments revealed that ruminoreticular digestion accounts for the major portion of total digestion for the nutrients examined. This finding supports most of the results in the literature. Large quantities of endogenous organic and inorganic matter were added to

the tract in the abomasum; however they were not as large as those observed by McGilliard (1961).

Nitrogen absorption in the ruminoreticulum was much greater than had been expected. The factors which could have caused this are discussed but the exact causes are unknown. This experience points out the need for more study on the fate of non-protein nitrogen in the rumen. It would be well to employ N^{15} -labeled urea to determine the degree of urea nitrogen incorporated into microbial protein or ammonia, and to determine the ultimate sites of absorption and secretion.

Energy level had no major effects which could be attributed to energy level alone. Had there been a greater spread between the levels of energy intake, differences in patterns of digestion might have been observed.

The techniques used in these experiments simulated normal conditions as much as possible so that the most accurate information could be obtained. Sampling from fistulae of live animals rather than sampling from the ligated tract of sacrificed animals not only enabled collection of digesta representative of digesta in an intact live animal, but also allowed the repeated use of the animals on several diets.

Composition of digesta, particularly in the upper part of the tract, varies from one feeding to the next. This variation in composition makes it difficult for the researcher to obtain an "average" sample. In these experiments, feeding and sampling were done frequently during the day to reduce the diurnal variation in composition of the digesta collected.

Frequent feeding may not represent normal feeding practices, but it did enable me to more accurately compare several rations under similar feeding conditions.

One problem in these experiments was changing energy concentration while attempting to maintain physical nature of the diet. In most of the other research discussed, energy concentration was altered by feeding various ratios of concentrate and hay.

Since the proportion of long hay in the diet affects rate of passage, the results of those experiments may be a reflection of differences in rate of passage rather than differences in energy concentration. In my experiments I attempted to minimize dietary effect on rate of passage by feeding ground corn cobs as the roughage rather than hay. Ground corn cobs are physically more similar to ground corn than most roughages; however, particle size of the ground cob was not identical to that of the ground corn. Thus, it must be recognized that rate of passage may have varied between rations and had some influence on the location of digestion in the tract. A possible means of avoiding such physical difference would be to fine grind and pellet all rations. Pelletizing would also enable the use of a greater variety of feeds at wider ratios while maintaining similar rates of passage. An attempt should be made to reduce the amount of non-functional tract which exists in the use of the re-entrant duodenal fistula. This improvement would undoubtedly allow the animal to consume more normal amounts of feed. More detailed information on absorption of specific nutrients could be obtained with the use of isotopes.

Because of the limitations of these experiments, one should be cautious in making inferences about a population. Intake, ration combinations and number of animals were all limited. In addition to this, few of the differences among rations were highly significant ($P < 0.01$).

It must be kept in mind that the limitations were unavoidable due the nature of the experiment. Fistulated animals of this type are costly to prepare and have a limited period of usefulness; consequently, the number of animals and the number of rations to which they can be exposed are limiting factors.

These experiments were different from most of the other research discussed in that physical nature of the diet was not a major variable. Consequently, effect of rate of passage were minimized, and my results should reflect more clearly the effects of energy concentration and energy level.

SUMMARY

Two 7-month-old Holstein steers were fitted with ruminal and re-entrant duodenal fistulae for use in these experiments. Three urea supplemented corn-corn cob rations containing different energy concentrations were fed to each steer. Ground corn cobs were selected as the roughage source of the diets to minimize the physical difference between rations.

Possible diurnal variation in composition of digesta was avoided by frequent feeding and collection of digesta.

In Experiment I, digesta were sampled at the reticulo-omasal opening and the re-entrant duodenal fistula. All feces were collected. Digestion of dry matter, energy, organic matter, water soluble carbohydrates, fiber, ash, total nitrogen and urea and $\text{NH}_3\text{-N}$ were determined for the ruminoreticulum, omasum-abomasum and intestine. Lignin was used as the marker.

Between 70% and 80% of the dry matter was digested in the ruminoreticulum. This represented about 100% of the total dry matter digestion. Total stomach digestion coefficients were 15% to 20% below those of the ruminoreticulum, indicating that dry matter was added to the digesta by the abomasum. The amount of dry matter digested in the intestine was similar to the amount added by the abomasum. This general pattern was observed for all nutrients measured.

As energy concentration increased, ruminoreticular digestion of dry matter, energy and organic matter increased and intestinal digestion of

the residues of these nutrients increased. Similarly, intestinal digestion of residual water soluble carbohydrate increased at the high concentration of energy, suggesting that some soluble carbohydrates may escape rumen fermentation at this concentration.

Ruminoreticular digestion of fiber was inversely related to energy concentration. This depressing effect of high energy diets of ruminoreticular fiber digestion was sufficiently great that it influenced total stomach digestion coefficients. In contrast, intestinal digestion of residual fiber was higher at the high energy concentration than at the low.

Energy concentration had no influence on ash digestion in the tract, but the experiment did reveal that large quantities of ash are added by the abomasum.

Nitrogen and urea and $\text{NH}_3\text{-N}$ digestion were unaffected by energy concentration. The results of the experiment suggest that much urea nitrogen was rapidly converted to ammonia and was absorbed from the rumen. Ruminoreticular nitrogen digestion represented more than 90% of the total nitrogen digestion. This extremely high rumen absorption of nitrogen was probably caused by the high urea content of the rations and the frequency of feeding.

The same rations were fed in Experiment II at two levels to study the effect of energy level as well as energy concentration on digestion along the tract. Frequency of feeding was the same as in Experiment I. Ingesta were sampled from the re-entrant duodenal fistula, and all feces

were collected. Digestion was determined for the total stomach and intestine.

Digestion coefficients of all nutrients were similar to those observed for Experiment I. Energy concentration affected total stomach and intestinal digestion of all nutrients much the same as it did in the first experiment. However, few of the effects were significant due to animal variation. As energy concentration increased, total stomach digestion of fiber decreased. This difference in stomach digestion influenced total digestion of fiber similarly.

Energy level had no significant effect on digestion of any nutrients except fiber and ash. Fiber was digested in the total tract to a greater degree at the low level of energy. The intestinal digestion of residual ash was also greater at this low level.

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ACKNOWLEDGMENTS

I wish to express my appreciation to Dr. N. L. Jacobson for his assistance in planning and conducting these experiments. For his critical evaluation of this thesis during its preparation, I am especially grateful.

These experiments would not have been possible had it not been for the skillful performance of surgery on experimental animals by Dr. A. D. McGilliard.

I am also grateful to John Peters for his laboratory assistance and sincere interest in this work.