STUDIES ON THE INFLUENCE OF PARTICLE SIZE AND LEVEL OF FEED INTAKE ON RUMINAL DIGESTION

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CHAPTER I

INTRODUCTION

Current high costs of grain and other high energy feedstuffs require cattle feeders to obtain maximum utilization of the grain portion of feedlot diets. Research at Oklahoma and other stations has demonstrated the advantages of various grain processing techniques. Several workers have found improved efficiency of feed utilization, and in some cases, increased rate of gain in cattle fed processed grains. Further research indicates site of digestion (ruminal or intestinal) may partially explain improvements in feed efficiency from processing. Grains with high ruminal starch digestibilities tend to produce the best animal performance, perhaps due to limited intestinal digestive capacity. Corn and sorghum are the most frequently fed cereal grains in Oklahoma and the High Plains. Since these grains contain 70-75% starch, the purpose of this study was to more clearly define factors influencing ruminal starch digestion.

Based on present knowledge, such factors as grain particle size, starch alteration, protein solubility, rumen turnover and level of intake would be of major interest. If effects of these factors and their interactions can be more clearly defined, cattle feeders may be able to obtain better utilization of corn and sorghum and ultimately reduce the cost of feedlot grains.

CHAPTER II

REVIEW OF LITERATURE

Structure of Cereal Grains

The corn kernel is a caryopsis or berry, a one-seeded fruit borne on a female inflorescence commonly called the ear. Kernels of dent corn weigh 350 mg on the average (150-600 mg range) and are composed of four major parts: tip cap, pericarp, germ and endosperm (Watson, 1967). The tip cap is a remnant of the organ for attachment of kernel to cob and is structured for rapid absorption of water. The pericarp, also referred to as the hull or bran, is a layer of dead, hollow cells surrounding the kernel. Directly underneath the pericarp is a layer of spongy tube cells, followed by the seed coat or testa and the aleurone. The aleurone is a tough, dense structure comprising 3% of the kernel dry weight. Cell layers from pericarp to aleurone generally fall into the bran or fiber fraction during milling (Watson, 1967).

The germ of embryo generally comprises about 11.5% of kernel dry weight in mature dent corn. Germ is composed of two parts; the scutellum and embryonic axis. The axis makes up about 10% of total germ weight and forms the seedling upon germination. The scutellum on the other hand, is in close proximity to the endosperm and is covered by secretory epithelium. Cells in the epithelial layer of the scutellum secrete enzymes, principally alpha amylase, which diffuse to the endosperm to digest starch for embryonic nourishment. According to Watson

(1967) the total germ component contains 84.3% of the fat, 83.5% of the ash, 65.3% of the sugar and 22.2% of the protein in the whole kernel of dent corn on a dry basis.

Endosperm contains roughly 98.5% of the starch and 73.6% of the protein in whole kernels of dent corn and can loosely be broken down into floury (soft) and horny (hard) regions. Floury endosperm makes up 34% of endosperm weight in dent corn and is characterized by loosely packed starch granules in a thin protein matrix. In contrast, horny endosperm displays tightly packed starch granules with a thick protein matrix and generally has double the protein and lipid content found in floury endosperm (Armstrong, 1972). Chemical composition of corn kernels varies somewhat with variety, but in normal dent corn the starch is usually 27% amylose and 73% amylopectin. Fiber is almost entirely restricted to pericarp which is composed of 40% cellulose and 40% pentoglycan. Sucrose is the major sugar in dent corn (0.9 to 1.9% of whole kernel weight) and is found primarily in the embryo (3/4) and endosperm (1/4). Watson (1967) indicated germ contains 84% of total kernel fat with 98% of the fat in the form of glycerides of fatty acids. Zein is the predominant protein of corn kernels and comprises 60% of endosperm protein in mature dent corn.

Structure of Cereal Grain Starch

Starch of cereal grains is found almost entirely in the endosperm and as discussed previously, endosperm can be loosely divided into floury and horny portions. Starch in horny endosperm is almost all amylopectin while floury endosperm contains both amylose and amylopectin.

Starch is basically an α-D-(1→4) linked glucan with α-D-(1→6) linked branch points and is typically a mixture of amylose and amylopectin. Amylose is a straight chain of glucose units (i.e., no α-D-(1→6) branch points) and is normally 1000 to 2000 D-glucose units in length. Amylopectin, on the other hand, is comprised of short (1→4) chains with numerous (1→6) branch points and has about 19 to 26 D-glucose units per (1→4) chain (Armstrong, 1972).

If starch granules are in a layered or shell structure (Armstrong, 1972) with each layer of uniform thickness, the granule is concentric with the hilum--the nucleus around which successive layers of starch are built up. The hilum will be in an eccentric position if deposition is favored in one direction.

From X-ray crystallographic studies and birefringence it is known that granules contain crystalline regions extending radially from the center and dispersed in a more abundant, amorphous network (Armstrong, 1972). In these highly ordered crystalline regions called crystallites, numbers of parallel polymer chains are held closely together by numerous secondary bond forces in the form of hydrogen bonds. The more abundant amorphous regions comprise a network of randomly distributed polymer chains held loosely together by hydrogen bonding when in sufficient proximity, with associations between molecules in the amorphous regions being weaker than in crystallites.

Starch granules generally contain 10-17% water, most of which is water of crystallization. Drying results in submicroscopic cracks and cavities developing in the granule. Such points of access to the inner granule may facilitate entry of starch degrading enzymes (Armstrong, 1972).

Starch granules are unlike isotropic substances such as glass or plastic and are anistropic since their optical properties are not the same in all directions. According to Armstrong (1972), crystalline regions in the amorphous mass result in double refraction or birefringence. Viewed through a microscope with polarized light a dark interference or Maltese cross occurs, the central point of which is the hilum. This property of birefringence is lost when crystallinity is destroyed.

Gelantinization is a process which can result in loss of birefringence. Due to association of molecules in the granule, starch is insoluable in cold water despite its hydrophilic nature. As water is heated, granules absorb water and swelling begins (Armstrong, 1972). In early stages of heating, swelling is reversible and birefringence is retained. When gelatinization temperance is reached, however, crystallinity is destroyed and birefringence is lost. This change is associated with rupture of secondary hydrogen bonds in the crystallites. No one gelatinization temperature exists, but rather a range of 8 to 10° C over which loss of birefringence occurs. Gelatinization does not necessarily imply rupture of starch granules, but this will occur if heating continues (Armstrong, 1972).

Ruminal and Total Tract Digestion of Cereal Grain Starch and Dry Matter

Several rather complete reviews are available on the subject of ruminal digestion of starch and dry matter (Armstrong and Beever, 1969; Sutton, 1971; and Waldo, 1973) of cereal grains. Thus, this review will not attempt to provide an in-depth literature search, but will

provide a general overview of the subject in question. The reader is referred to the above references for a more detailed consideration.

Under normal conditions the major portion of ingested dry matter disappears from the rumen as a result of microbial fermentation (Annison, 1956). Several factors may influence this percentage and thus total tract dry matter or starch digestion, including grain processing method (whether heat, moisture or simply alteration of particle size), level of intake, roughage type and level and numerous other factors.

Processing Effects

Particle size has been shown by numerous authors to be an important factor governing ruminal and total tract digestion of cereal grains. Atkenson and Beck (1942) investigated the value of grinding sorghum grain for dairy cows. Forty-two percent of whole sorghum grain was recovered in feces compared to 4.8% for coarse ground and 1.5% for fine ground grain. Increasing daily grain intake did not appreciably alter the percent sorghum in feces. Digestibility studies with sorghum at the Kansas station (Smith et al., 1949) are in agreement. Rations consisting of cotton seed meal, sorghum silage and sorghum grain had total tract dry matter digestion coefficients of 48.04% for whole versus 52.34% and 60.19% for coarse and fine ground sorghum, respectively. Moreover, NFE digestion was roughly 14% greater for fine ground than whole grain, possibly indicating greater digestion of starch. Inability to pinpoint level of intake and roughage level in these trials does, however limit conclusions one might draw.

Work of Moe et al. (1973) indicates the importance of physical form in corn grain. Very finely ground corn had higher (68.3% vs. 59.1%) total tract dry matter digestion than whole corn in 54.5% corn rations with dairy cows. Digestion of cell solubles, which would include starch, was much higher for ground than whole corn (81.4% vs. 67.5%). The data indicate that only a slight amount of cracking or alteration of particle size may be adequate, however, as little difference was observed in dry matter and cell soluble digestion of cracked and very finely ground corn rations.

In contrast, White et al. (1972) found grinding did not influence total tract dry matter, NFE, energy or crude protein digestion compared to whole corn. Roughage level in these data, however, was much lower than in the data of Moe et al. (1973). Level of dietary roughage has been shown to have an important effect of lowering ruminal starch digestion of whole corn grain (Cole et al., 1976a) while having little effect on cracked or processed grains (Cole et al., 1976b).

Walker et al. (1973) demonstrated the importance of particle size on ruminal digestion of sorghum. In vitro dry matter disappearance increased as particle size score increased (actual size decreased) and a correlation of .87 was noted between the two parameters. In addition, an experiment with abomasally cannulated steers indicated ruminal and total tract dry matter digestibilities were highest for fine ground grain compared to coarse and intermediate grinds. Wilson et al. (1973) working in a similar vein with corn found that nylon bag dry matter digestion nearly doubled as modulus of fineness was approximately halved. These authors also found whole corn had less total tract organic matter digested than coarse ground corn whether 5 kg or 2.5 kg

hay was fed with grain in dairy cows. Moreover, fine rolled high moisture corn had a greater (70.3%) organic matter digestion than whole high moisture corn (56.8%), indicating the importance of particle size over and above moisture processing.

Processing effects other than particle size have an important impact on ruminal and total digestion of cereal grain starch. McNeill et al. (1971) evaluated site of digestion of sorghum processed by grinding, steam flaking, reconstitution and micronizing. Significantly more starch was digested ruminally by steers fed steam flaked and reconstituted sorghum than by steers fed other treatments. Total tract starch digestion was nearly complete for all treatments. Low levels of intake in this study may have equalized treatment differences in total starch digestion, however.

Hinman and Johnson (1974a) in contrast to McNeill et al. (1971) found no differences in ruminal digestibilities of dry rolled, micronized, ground and steam flaked sorghum. Less starch was digested post-ruminally with dry rolled sorghum, however. In a second study, Hinman and Johnson (1974b) reported no differences in ruminal starch digestibilities between dry rolled and low, medium and high degrees of micronization in 84% sorghum rations. Steaming sorghum under atmospheric conditions and at 3.5 kg/cm^2 resulted in a 5% nonsignificant increase in ruminal starch digestion favoring 3.5kg/cm^2 sorghum (Holmes et al., 1970), indicating the impact of gelatinization on ruminal starch fermentation.

Orskov, et al. (1971) reported approximately 21 to 22% of unprocessed corn starch escaped ruminal fermentation in sheep. Comparing steam flaked, ground and cracked corn (Orskov et al., 1969) in 80%

grain rations with sheep, 14% of starch intake reached the abomasum with cracked corn compared to 5% for steam flaked and 12% for ground corn. Thus gelatinization due to steam flaking is important with corn as well as sorghum.

Beever et al. (1970) compared diets of ground and steam flaked corn and found 78.1% of ground corn starch was digested ruminally vs. 95.7% for flaked corn. Similar results with flaked corn have been reported by other authors (Macrae and Armstrong, 1969 and Nicholson and Sutton, 1969).

Two studies relative to the effects of high moisture processing have also been reported. McKnight et al. (1973) fed heifers a 66% corn ration processed by dry grinding, high moisture grinding and ensiling and treatment of high moisture corn with either of two organic acids. Ruminal digestion of starch was much higher (81.4% vs. 47.3%) for ensiled corn as compared to dry corn. Total tract energy digestibilities were significantly higher for all high moisture diets than for dry corn. Galyean et al. (1976) compared dry rolled, steam flaked and both high moisture ensiled and propionic acid treated corn. Significantly more starch was digested ruminally with high moisture ensiled and steam flaked than dry rolled or high moisture propionic acid treated corn. Propionic acid treated corn was fed in the whole form which may have lowered its ruminal digestion, however. Total tract starch digestion was significantly higher for steam flaked and ensiled corn than for other treatments.

Level of Intake

Increasing the total intake of a pelleted concentrate ration (85% concentrate) in steers by 0.8 to 1 kg per day had no effect on percent

ruminal starch or dry matter digestion (Topps et al., 1968). Macrae and Armstrong (1969) obtained no difference in ruminal starch digestion of sheep fed a rolled barley ration at levels of 25.5 and 16.6 g starch per $kg^{.75}$.

In contrast, Orskov et al. (1969) found increasing starch intake of a rolled barley ration (48% barley) from 19 to 27 g per $kg^{.75}$ reduced ruminal starch digestion from 92.3 to 87.2%. Reduction was only from 95.6 to 93.2% when barley comprised 80% of the ration, however.

Kratchner et al. (1973) found no effect of level of starch intake (ad lib or 80% ad lib) on either ruminal or total tract starch digestion with steam flaked or dry rolled sorghum. Similarly, Waldo et al. (1971) fed increasing amounts of cracked corn (20 to 80%) at a level of 70 g air dry feed per $kg^{.75}$ per day and reported no significant differences in percent ruminal starch digestion. Total tract starch digestion was virtually complete. Moreover, Karr et al. (1966) with steers and Tucker et al. (1968) with sheep fed rations similar to those of Waldo et al. (1971) and observed 30 to 40% starch escaping fermentation in all cases. Total tract digestion was again almost complete.

Total tract digestion studies of Brown et al. (1968) with both sorghum and corn showed no differences in digestion of proximate components, digestible or metabolizable energy as influenced by level of intake. In contrast, Anderson et al. (1959) in two experiments with steers fed mixed forage:concentrate diets found level of intake markedly reduced total tract digestion of dry matter. At one-half maintenance intake, total tract dry matter digestion was 87.5% vs. 74.3% at 2.7 times maintenance intake with 20:80, forage:concentrate rations.

Recent work with dairy cattle confirms the importance of level of intake on starch digestion. Wheeler et al. (1975) conducted studies with dairy cattle fed at maintenance, 2.5 and 3.2 times maintenance intake. Total tract starch digestibility fell from 96.2% to 84.7% as intake increased with 70:30, concentrate:forage rations.

Intestinal Digestion of Cereal Grain Starch

Carbohydrases of the Ruminant Small Intestine

Enzymatic degradation of starch in the ruminant small intestine appears to be similar to degradation in non-ruminants. Enzymes involved in digestion of starch are the amylases and maltases of pancreatic juice and amylase, maltase and oligo-1:6-glucosidase of intestinal mucosa (Armstrong and Beever, 1969). Conflicting results exist regarding deficiencies in activities of ruminant carbohydrases; therefore, each enzyme will be discussed individually to aid clarification.

Amylase

Siddons (1968) has shown pancreatic juices of mature cattle have relatively high amylase activity while young (14-16 wk old) calves have appreciably lower amylase activity. Intestinal activities of this enzyme were similar in both groups. Henschel et al. (1963) infused various carbohydrates into the duodenum of young steers and recovered over 50% of infused wheat starch at the ileum. Addition of amyloglucosidase to starch suspensions reduced recovery to 7%, indicating intestinal amylase activity might be limiting. Larsen et al.

(1956) fed several carbohydrates to 8 to 9 month old calves and measured intestinal digestibility. These authors observed highly variable and low starch digestibilities and hypothesized an inadequate amylase activity as the cause. Differential flow of Cr_2O_3 and ingesta may have been another plausible explanation.

Huber et al. (1961) administered carbohydrates into the omaso-abomasal area of dairy cattle ranging in age from 2 weeks to 2 years and measured blood glucose levels by jugular sample. Starch infusion resulted in only a slight response in blood glucose levels compared to glucose and lactose infusion; however, the response to starch increased with increasing age. The authors suggested a limited amylase activity as the cause for lowered blood glucose response to starch infusion. A more likely cause may have been a much slower rate of hydrolysis of starch than for glucose or lactose and extensive metabolism of glucose from starch by intestinal mucosa and liver with little change in systemic glucose concentrations (Coombe and Smith, 1974).

In contrast to work suggesting limited amylase activities, work of Mayes and Orskov (1974) would indicate the opposite. The authors infused a gelled corn starch solution into the abomasum and recovered samples at the terminal ileum of sheep. Over 50% of alpha-glucosides reaching the ileum were breakdown products of amylase action, indicating utilization of breakdown sugars and not amylase activity may limit intestinal utilization of starch.

Maltase

Several reports of a conflicting nature appear in the literature with regard to ruminant maltase activities. Siddons (1968) has shown

mature cattle and calves have relatively weak maltase activities. Huber et al. (1961) with dairy cattle have shown decreased blood glucose levels when maltose is fed vs. glucose or lactose. Since maltose should be readily hydrolyzed and absorbed, this may indeed indicate a maltase deficiency. Henschel et al. (1963) recovered 30% of duodenally infused maltose at the terminal ileum of steers. Maltase levels present in these studies would only be capable of hydrolyzing 2 g per 2 to 3 hours. Mayes and Orskov (1974) reported the major limitation to starch utilization in sheep small intestines appears to be maltase or the absorption of maltose or glucose by intestinal brush border cells.

In contrast, Hembry et al. (1967) working with sheep concluded since maltase activity was higher in all segments of the digestive tract than amylase or lactase activities, it would not limit intestinal starch digestion. Larsen et al. (1956) found a sharp rise in blood glucose levels of calves following maltose administration and observed both glucose and maltose were readily absorbed in the small intestine.

Other Carbohydrases, Diet and Absorption

Mayes and Orskov (1974) inferred oligo-1.6-glucosidase activity may be deficient in sheep since a considerable amount of carbohydrates reaching the terminal ileum in their study should have been acted upon by this enzyme. Coombe and Siddons (1973) reported its activity was about 50% of maltase activity, but the picture remains unclear regarding this enzyme.

Diet may influence enzyme activities of the small intestine as evidenced by studies of Clary et al. (1969). These workers reported amylase activity of pancreatic juice increased significantly as percent

corn starch in the diet increased and decreased as percent starch decreased. Thus, pre-experimental diet may be a factor to consider in starch utilization studies.

The jejunum appears to be the site of greatest enzymatic starch breakdown, followed by ileum and duodenum. A similar pattern holds true for sites of absorption (Armstrong and Beever, 1969). Microbial activity would also tend to increase in the lower portions of the small intestine. Therefore, a considerable amount of unhydrolyzed starch reaching the lower small intestine might be available for bacterial use with subsequent volatile fatty acid production. Quantitative values of bacterial starch breakdown are unavailable, but work of Mayes and Orskov (1974) indicates as more starch reaches the ileum, pH of ileal digesta falls, presumably due to microbial action.

Processing and Level of Intake Effects

In many studies dealing with processed starch, little starch enters the small intestine as a result of extensive ruminal breakdown. Further, digestion sites generally are not separated (large vs. small intestine). Processed starch should however, be more readily degraded in the small intestine since amylase more readily digests gelatinized starch than raw starch (Lathe and Ruthven, 1956).

Level of starch intake has received little attention and most studies reported in the literature involve experimental animals at or near maintenance intakes. Available work is inconclusive. Topps et al. (1968) and Macrae and Armstrong (1969) reported little effect of increasing intake in sheep on total starch digestion. Kratchner et al. (1973) found no difference in total tract starch digestion when

steers were fed ad lib or 80% ad lib. Conversely, Little et al. (1968) infused quantities of starch ranging from 200 to 600 g per day into the small intestine of steers. Total intestinal starch digestion decreased from 93 to 70% as infusion level increased, indicating a biological limit for starch digestion in ruminant small intestines. Wheeler and Noller (1977) have observed high, negative correlations between fecal pH and fecal starch and a positive correlation between fecal pH and duodenal pH. These authors suggested decreased pH in the duodenum may reduce activity of intestinal alpha-amylase.

Lignin as a Marker in Ruminant Digestion Studies

Lignin has long been used as an internal marker in ruminant digestion studies, but its recovery or more accurately digestibility has often been a point of contention. Should lignin be variably digested its use as a marker, particularly in partioning digestion, would be questionable at best.

Forbes and Garrigus (1948) utilized lignin to study digestibility of pasture forages in steers and wethers. In seven trials with steers, average recovery of lignin was 102±7% and the authors suggested lignin would be an adequate marker for pasture digestion studies. In contrast, Ely et al. (1953) reported lignin digestion coefficients for orchard grass hay cut at 4 stages of maturity to range from 3.8 to 16.0%. Digestion of lignin methoxyl groups (a measure of lignin degradation in the digestive tract) ranged from 20.3 to 31.6%. Stage of maturity of hays had little effect.

Kane et al. (1953) compared chromic oxide, lignin and plant chromagens in digestibility studies with dairy cattle. The authors indicated incomplete recovery of lignin in fecal grab samples made calculation of digestibility from these samples impractical. Total collection methods with lignin consistently resulted in lower dry matter digestion values than with chromic oxide.

Digestion of lignin in several grass species obtained at head just emerged stage varied from 3.0% in timothy to 16.8% in Kentucky bluegrass (Sullivan, 1955). The author concluded that variable digestion of lignin made it unsuitable as an inert reference substance.

Level of intake effects on lignin digestion were investigated by Elam et al. (1962). Sheep fed at 70, 110, and 150% of TDN maintenance requirements had lignin recoveries of 92.1, 90.0 and 88.4%, respectively. In contrast, chromic oxide recovery averaged 100.7% over the three levels of intake.

Considering site of lignin digestion, Johnson et al. (1964) fed pelleted alfalfa hay to wethers and found lignin digestion to be 10%. Rumen samples had slightly higher lignin content than abomasal samples, perhaps indicating ruminal digestion of lignin. Porter and Singleton (1971) found lignin was degraded mainly in the forestomachs and reported lignin digestion of 10%. However, these authors still felt lignin was an adequate marker for estimation of duodenal flow rate. In contrast, Badawy et al. (1958) suggested that although direct proof is lacking, some evidence suggests lignin digestion would be confined to the large intestine.

Balch (1957) used lignin as a marker to partition digestion in dairy cows fed diets ranging from all hay to 24 pounds concentrate

and 2 pounds hay. Samples of digesta were obtained ruminally and digestion of lignin in the total tract ranged from 24.4 to -17.3%. Despite the high and variable digestion coefficients, the author felt lignin adequate for partitioning digestion in ruminants.

It would appear then, that lignin is an analytically undefined entity with variable digestion. Whether lignin digestion is real or merely an analytical problem appears to merit further consideration.

CHAPTER III

NYLON BAG DRY MATTER AND STARCH DIGESTION OF CORN AS INFLUENCED BY PARTICLE SIZE, STEAM FLAKING AND HIGH MOISTURE PROCESSING

Summary

Corn grain was sieved to obtain distinct particle sizes and incubated ruminally in a mature steer using a nylon bag technique. Dry matter and starch disappearance from the bags and chemical analyses (crude protein, acid detergent fiber, soluble nitrogen, gelatinization and starch) were conducted on the original particle sizes. In Experiment I, dry rolled corn (DR) sizes of 6000, 3000, 1500 and 750 microns (μ) were incubated for 2, 4, 6 or 8 hours. Dry matter digestion (DMD) means for size averaged over time indicated little difference between particle sizes of 6000 (4.98%) and 3900 (4.38%) μ sizes. However, DMD increased ($p < .05$) as particle size was halved to 1500 (9.74%) and 750 (18.38%) μ. Starch digestion of DR was similar for 6000 through 1500 μ sizes with 750 μ DR (21.70%) approximately double their average value (9.22%). Experiment II compared steam flaked (SF) and dry ground (DG) at 3000, 1500 and 750 μ sizes for 2, 4, 6 and 8 hours. SF corn had higher ($p < .05$) DMD within each particle size and time than DG; however, an approximate doubling of DMD as size was halfed was not observed in SF corn as in DG. Starch digestion showed patterns similar to DMD. Experiment III concerned evaluation of ground high moisture ensiled corn

(HM) and DG corn. Procedures were the same as Experiment II. DMD of both HM and DG approximately doubled as particle size was halved. Starch digestion increased in HM as size was reduced from 3000 (12.28%) to 1500 (24.02%) to 750 μ (44.34%). Starch digestion patterns in DG were similar to those previously observed. In Experiment IV, incubation of DR 6000 (16.12%), 3000 (19.43%), 1500 (30.83%) and 750 μ (46.32%) sizes for 12 or 24 hours resulted in significant increases ($p < .05$) in DMD as particle size was reduced. Particle size had significant effects on DMD of sorghum grain (Experiment V). This study indicates particle size influences nylon bag DMD of unprocessed corn and milo. Processing corn by steam flaking or high moisture methods resulted in an additional effect on DMD and starch digestion above that observed from particle size alone.

Introduction

Particle size has been demonstrated to have an important effect on cereal grain digestion. Moe et al. (1973) found very finely ground corn has a higher total tract dry matter digestion coefficient (68.3%) than whole corn (59.1%) in 54.5% concentrate diets fed to dairy cows. Walker et al. (1973) has demonstrated the importance of particle size on ruminal digestion of sorghum. In vitro dry matter disappearance increased as particle size decreased, and a correlation of .87 was noted between the two parameters. Wilson et al. (1973) found nylon bag dry matter digestion of corn increased as modulus of fineness decreased.

Processing effects other than particle size have an important effect on extent of digestion. Orskov et al. (1969) compared steam flaked, ground and cracked corn in 80% corn diets fed to sheep.

Fourteen percent of starch intake escaped ruminal fermentation in cracked corn diets compared to 12% for ground and 5% for steam flaked diets. Galyean et al. (1976) reported high moisture ensiled corn had higher ruminal and total tract starch digestibilities than cracked or whole high moisture, propionic acid treated corn.

The purpose of this study was to investigate the influence of particle size and processing (steam flaking and high moisture ensiling) on ruminal dry matter and starch digestion of corn grain, using a nylon bag technique. The effect of particle size on dry matter digestion of dry rolled sorghum grain was also investigated.

Experimental Procedures

Experiment I

Dry rolled corn obtained from the University feed mill was sieved in a set of standard sieves according to procedures outlined by Ensor et al. (1970). Corn retained on sieves with openings of 4000, 2000, 1000 and 500 microns (μ) was separated to provide four distinct particle sizes. Thus, average particle size of the four groups from largest to smallest was 6000, 3000, 1500 and 750 μ. Approximately 5 g of each of the four average particle sizes was weighed into 100 mesh nylon cloth bags. Bags were constructed of parachute material, sewn with nylon thread and were approximately 5 x 12 cm.

Duplicate bags of each particle size, attached to a nylon cord by fishing swivels and anchored by an 80 g lead weight were placed in the rumen of a mature Holstein steer. Four cords, one for each incubation period, were used and incubated for 2, 4, 6 and 8 hours. Thus a total

of 32 bags were utilized, and two observations were obtained for each particle size x time subclass. The procedure described above was replicated to provide a measure of experimental error.

The steer was fed a 62.75% dry rolled corn diet ad libitum and was housed in an individual slotted floor pen. Ingredients in the donor steer diet were as follows: Corn, dent yellow grain (4-02-935); 6% alfalfa, aerial part, dehy ground, mn 15% protein (1-00-022); 14% cotton seed hulls (1-01-599); 10% soybean, seeds, solv-extd ground, mx 7% fiber (5-04-604); 0.1% urea, mn .45% N; 0.5% limestone, ground, mn 33% Ca (6-02-632); 0.5% calcium phosphate, dibasic commercial (6-01-080); 0.15% Aurofac-10 and 0.5% ammonium chloride.

All bags were dried at 100°C and dessicated prior to inclusion of grain samples. Upon completion of incubation, bags were washed thoroughly under tap water and dried 48 hours at 65°C in a forced draft oven, followed by 2 to 4 hours in a 100°C forced draft oven. Bags were then dessicated and weighed to determine dry matter disappearance. Dry matter (DM), crude protein (CP), acid detergent fiber (ADF), soluble nitrogen as a percent of total nitrogen (SN), gelatinization (GEL) and starch content were determined on the original four average particle sizes. Starch content of the grain following incubation was determined so that nylon bag starch digestion could be calculated. SN was determined by extracting grain samples in an "Ohio Aqueous Buffer Solution" (Johnson, 1969) with an ionic strength of 0.14 and a pH of 5.5 using a procedure of Wohlt et al. (1973). GEL was determined as milligrams maltose released after incubation with beta-amylose (Sung, 1969). Starch was measured as α-linked glucose polymers by the enzymatic procedure of MacRae and Armstrong (1968).

Experiment II

Corn obtained from a commercial feedlot was either steam flaked (100°C, atmospheric pressure, approximately 20 minutes) or left in the whole form. Both steam flaked and whole corn were ground through a 6 mm screen in a Wiley mill and sieved as in Experiment I. Resulting average particle sizes were 3000, 1500 and 750 μ with each three particle sizes represented in both dry ground (DG, previously whole) and steam flaked (SF) corn. Nylon bag procedures outlined in Experiment I were conducted on both grains. Thus, each of the four cords had twelve bags, and two observations were obtained for each processing method x particle size x time subclass. As in Experiment I, the procedure was replicated to obtain a measure of experimental error. Additional procedures outlined in Experiment I were conducted, however, the donor steer diet had dry rolled sorghum (4-04-444) substituted for corn grain.

Experiment III

Ground high moisture corn, ensiled in a concrete trench silo, was obtained from a commercial feedlot and sieved to produce average particle sizes of 3000, 1500 and 750 microns (μ) and compared to DG corn used in Experiment II. Procedures and donor steer diet were equivalent to those discussed in Experiment I.

Experiment IV

The four particle sizes of dry rolled corn discussed in Experiment I were incubated ruminally for either 12 or 24 hours to investigate the

influence of extended incubation and particle size on unprocessed corn. Procedures and the donor steer diet were as discussed in Experiment I. Starch disappearance was not determined, however.

Experiment V

Dry rolled sorghum obtained from the University feed mill was sieved as before, and three average particle sizes of 3000, 1500 and 750 μ were obtained. Percentage dry matter of the three particle sizes was 88.39, 88.09 and 88.61%, respectively. Procedures described in Experiment I were followed with the exception that starch disappearance and chemical analyses of the particle sizes were not determined.

Statistical Analysis

Dry matter and starch disappearance data were subjected to analyses of variance procedures assuming a factorial treatment design with replication. Data from Experiment I were analyzed as a 4 x 4 factorial arrangement of treatments with particle size and time as factors. Replication x treatment interaction was taken as experimental error. Experiments II and III were considered as 2 x 3 x 4 factorial treatment arrangement while Experiment V was considered a 3 x 4 arrangement. Replication x treatment was also taken as a measure of experimental error in both Experiments IV and V. Tests of significance among treatment means were accomplished by use of an LSD protected by a preliminary F test.

Results and Discussion

Chemical composition of the corns is shown in Table I. There was little variation in percentage DM within processing methods, but considerable variation among processing methods (e.g. HM vs. SF). Variation for different particle sizes within corn types in crude protein (CP) content is noteworthy, particularly within DR, SF and DG corns. Crude protein varied considerably within SF corn (11.73, 9.45 and 8.64% for SF 750, 1500 and 3000, respectively). ADF also varied considerably within a processing method as did SN and GEL. Moreover, starch content displayed rather extreme variation within processing methods. Variation in chemical composition within processing methods may indicate separation into different components of the corn kernel. That is, more floury or horny endosperm, bran or germ may be included in one particle size as a result of the sieving process. Horny endosperm generally contains more than double the protein found in floury endosperm (Armstrong, 1972, indicating more horny endosperm may be included in particle sizes with higher crude protein contents. In addition, since most fiber in corn kernels is included in the pericarp, particle sizes with higher ADF percentages may contain greater quantities of pericarp, Moreover, high starch content may indicate greater amounts of floury endosperm.

Differences in SN and GEL might influence subsequent DM and starch digestion; however, since all chemical components measured in this study seem to vary with particle size, their effects on dry matter and starch digestion would be confounded with particle size. Thus, any attempt to relate changes in chemical composition to dry matter and

TABLE I

CHEMICAL COMPOSITION OF CORNS
BY PARTICLE SIZE

Corn	Size	DM%	CP%[a]	ADF[a]	SN as % of N[a]	GEL[a] mg	Starch[a] %
DR	6000	88.00	10.52	4.48	13.99	35.81	70.78
	3000	87.35	9.63	2.71	5.84	29.97	85.96
	1500	87.53	9.80	2.37	8.28	22.36	81.47
	750	87.93	10.90	3.38	11.21	38.53	77.90
SF	3000	91.56	8.64	2.27	3.26	213.50	89.14
	1500	91.36	9.45	2.93	3.30	193.73	84.08
	750	92.27	11.73	4.68	6.12	144.99	61.42
DG	3000	85.64	9.58	3.01	5.88	29.75	89.38
	1500	85.48	9.99	3.06	5.31	24.64	83.22
	750	85.89	10.45	3.56	6.59	27.43	74.26
HM	3000	76.65	9.83	3.47	29.62	24.53	76.37
	1500	76.98	9.96	2.93	28.75	27.08	80.01
	750	76.02	9.38	2.20	29.00	24.80	86.89

[a]DM basis.

starch digestion will be strictly speculative in nature and not based on statistical analysis.

Experiment I

Dry matter digestion (DMD) of the four particle sizes is given in Table II. The particle size x time entry of the analysis of variance was judged non-significant ($p > .7339$), thus means are reported averaged over time or size. DMD size means indicate little effect of particle size between 6000 (4.98%) and 3000 (4.38%) μ sizes. However, DMD increased significantly ($p < .05$) as particle size was reduced from 3000 (4.38%) to 1500 (9.74%) to 750 (18.38%) μ. DMD was roughly doubled as average particle size was halved after 3000 μ was reached. Considering this response in another manner, DMD approximately doubled as calculated surface area per unit volume was doubled from 3000 to 750 μ. Wilson et al. (1973) reported similar results with corn grain. Nylon bag dry matter digestion increased as modulus of fineness decreased.

The effect of time averaged over particle size (Table II) indicates significant increases ($p < .05$) as incubation increased from 2 to 8 hours. Magnitude of change with time, however, was not as great as change due to particle size, suggesting at least for short incubation periods particle size has a greater impact on DMD than time.

Particle size x time interaction was also non-significant ($p > .9008$) for starch digestion (Table III) and means are reported averaged over time or size. Particle size appeared to have little effect on starch digestion in 6000, 3000 and 1500 μ sizes; however, starch digestion of 750 μ corn was higher ($p < .05$) than all other sizes.

TABLE II

NYLON BAG DMD OF FOUR PARTICLE SIZES OF DRY ROLLED CORN (SIZE AND TIME MEANS)

Size	Size Means[a]	Time	Time Means[b]
6000	4.98[c]	2	6.65[c]
3000	4.38[c]	4	8.98[d]
1500	9.74[d]	6	9.98[e]
750	18.38[e]	8	11.87[e]

[a]Means averaged over alltimes, standard error of treatment means (SEM) = 0.69.

[b]Means averaged over all sizes, SEM = 0.69.

[cde]Means in a column with different superscripts are significantly different ($p < .05$).

TABLE III

NYLON BAG STARCH DIGESTION OF FOUR PARTICLE SIZES OF DRY ROLLED CORN (SIZE AND TIME MEANS)

Size	Size Means[a]	Time	Time Means[b]
6000	9.25[c]	2	10.65
3000	10.24[c]	4	10.99
1500	8.18[c]	6	14.07
750	21.70[d]	8	13.64

[a]Means averaged over all times, SEM = 2.08.

[b]Means averaged over all sizes, SEM = 2.08.

[cde]Means in column with different superscripts are significantly different ($p < .05$).

Similarity in starch digestion of the three larger particle sizes may be related to variation in chemical composition (Table I); however, the nature of this relationship, if it exists, is unknown. Whatever the case, 750 μ corn (21.70%) had roughly double the starch digestion of the average (9.22%) of the other three sizes. Thus particle size has a significant impact on starch digestion but not as great as its impact on DMD.

Time means (Table III) indicate little influence of time on starch digestion in these data. It would appear logical, however, that incubation periods longer than 8 hours would tend to increase starch digestion.

Experiment II

Table IV shows DMD values for SF and DG corn averaged over size or time. Size x time interaction F value was near unity and non-significant ($p > .8512$) while processing method x size and processing method x time interactions were highly significant ($p < .0025$, $p < .0001$), respectively. Thus comparisons were made between particle size or time means within processing method. An LSD value has been provided for comparisons between processing methods within a size or time. Size means indicate SF corn had significantly ($p < .05$) more DMD within each particle size than DG corn. Galyean et al. (1973) and Orskov et al. (1969) found steam flaked corn had higher ruminal starch digestion than cracked corn in steers and wethers, respectively. In addition, Figroid et al. (1972) found steam flaked sorghum had higher 8 hr nylon bag digestibilities than dry rolled sorghum. Comparison of particle sizes within a processing method indicates significant increases ($p <$

TABLE IV

NYLON BAG DMD OF STEAM FLAKED AND DRY GROUND CORN (SIZE AND TIME MEANS)

Corn	Size Means[d] 3000	1500	750	LSD[f]
SF	23.81[a]	28.51[b]	32.92[c]	2.96
DG	4.68[a]	8.41[b]	17.66[c]	2.96

Corn	Time Means[e] 2	4	6	8	LSD[f]
SF	21.29[a]	24.40[a]	29.27[b]	38.70[c]	3.42
DG	7.79[a]	8.06[a]	10.82[a]	14.33[b]	3.42

[abc]Means in a row with different superscripts are significantly different ($p < .05$).

[d]Means averaged over all times within a corn type, SEM = 1.01.

[e]Means averaged over all sizes within a corn type, SEM = 1.17.

[f]Least significant difference for comparison within columns and rows.

.05) in DMD as particle size was reduced from 3000 (23.81%) to 1500 (28.51%) to 750 (32.92%) μ in SF corn. Particle size comparison within DG corn shows patterns similar to that with DR corn observed in Experiment I. Thus, although particle size is important in SF corn the doubling effect on DMD as particle size is reduced is not observed as in DG corn. Time means (Table IV) indicate more ($p < .05$) DMD was observed at each incubation period with SF corn than DG corn.

Starch digestion is shown in Table V. Particle size x time interaction was non-significant ($p > .5076$) as was processing method x size ($p > .2695$); however, since processing method x time interaction was significant ($p < .0064$) means are reported in the same manner as with DMD for consistency of presentation. Size means suggest more starch ($p < .05$) was digested with SF than DG corn within each particle size, following patterns similar to DMD. GEL (Table I) is much higher for SF corn at all particle sizes than DG corn and may be related to the increase in starch digestion with SF corn. Johnson et al. (1968) observed gelatinization in SF corn as measured by loss of birefringence and found energy retention of SF diets was 6 to 10% greater than dry rolled diets.

Comparing particle sizes within SF corn, starch digestion significantly ($p < .05$) increased from 3000 (30.70%) to 750 (40.76%) μ. Values for DG 3000 (11.46%) and 750 (18.98%) μ sizes indicate particle size had an approximate doubling effect in DG corn. This suggests that particle size may not be as important in increasing the relative magnitude of starch digestion in steam flaked corn as in unprocessed corn. Time means reveal more ($p < .05$) starch was digested with SF corn than DG corn at each incubation period. Moreover, time had a

TABLE V

NYLON BAG STARCH DIGESTION OF STEAM FLAKED AND DRY GROUND CORN (SIZE AND TIME MEANS)

Corn	Size Means[d] 3000	1500	750	LSD[f]
SF	30.70[a]	36.87[b]	40.76[b]	6.33
DG	11.46[a]	12.54[a]	18.98[b]	6.33

Corn	Time Means[e] 2	4	6	8	LSD[f]
SF	26.75[a]	33.60[b]	37.46[b]	46.64[c]	7.31
DG	12.45	11.44	15.10	18.30	7.31

[abc]Means in a row with different superscripts are significantly different ($p < .05$).

[d]Means averaged over all times within a corn type, SEM = 2.16.

[e]Means averaged over all sizes within a corn type, SEM = 2.50.

[f]Least significant difference for comparison within columns or rows.

marked effect on starch digestion within each corn type.

Experiment III

DMD and starch digestion of HM and DG corn (Tables VI and VII) are reported averaged over time or size within processing methods since particle size x time ($p < .8321$) and processing method x time ($p < .5690$) interactions were non-significant. Significantly more ($p < .05$) DMD (Table VI) was noted within each particle size for HM vs. DG. Similar nylon bag results have been observed by Evans and Colburn (1967) with 48 hr digestion periods. Furthermore, comparisons within a processing method indicate that reducing average particle size in both HM and DG corns resulted in significant increases in DMD. An approximate doubling effect on DMD as particle size was halved occurred in both HM and DG corn, in contrast to the absense of this effect in SF corn (Experiment II). Time means indicate no significant effect of time on DMD within HM corn and a significant ($p < .05$) difference in 2 (10.57%) and 8 (19.33%) hour incubations with DG corn. Thus, these data indicate time is far less important than particle size in HM corn, at least for the incubation lengths used in this experiment. More ($p < .05$) DMD was noted within each time for HM compared to DG corn, however.

More starch ($p < .05$) was digested within 1500 and 750 μ sizes for HM than DG corn (Table VII). Again, all interaction F values were less than 1.0, except for processing method x size ($p < .0001$). In contrast to DMD values, greater ($p < .05$) starch digestion occurred in DG (18.96%) than HM (12.28%) corn at the 3000 μ size. As with DMD, an approximate doubling of starch digestion occurred as particle size was halved in HM corn. In contrast, 3000 (18.96%) and 1500 (17.24%) μ DG

TABLE VI

NYLON BAG DMD OF HIGH MOISTURE AND DRY GROUND CORN (SIZE AND TIME MEANS)

Corn	Size Means[d] 3000	1500	750	LSD[f]
HM	16.81[a]	23.81[b]	38.88[c]	4.09
DG	7.86[a]	12.41[b]	22.20[c]	4.09

Corn	Time Means[e] 2	4	6	8	LSD[f]
HM	25.09	25.01	26.36	29.55	4.72
DG	10.57[a]	12.60[a]	14.96[ab]	19.33[b]	4.72

[abc]Means in a row with different superscripts are significantly different ($p < .05$).

[d]Means averaged over all times within a corn type, SEM = 1.40.

[e]Means averaged over all sizes within a corn type, SEM = 1.61.

[f]Least significant difference for comparison within columns or rows.

TABLE VII

NYLON BAG STARCH DIGESTION OF HIGH MOISTURE AND DRY GROUND CORN (SIZE AND TIME MEANS)

Corn	Size Means[d] 3000	1500	750	LSD[f]
HM	12.28[a]	24.02[b]	44.34[c]	5.90
DG	18.96[a]	17.24[a]	26.60[b]	5.90

Corn	Time Means[e] 2	4	6	8	
HM	24.70[a]	25.46[ab]	25.75[ab]	31.61[b]	6.82
DG	18.66[a]	18.51[a]	20.25[ab]	26.32[b]	6.82

[abc]Means in a row with different superscripts are significantly different ($p < .05$).

[d]Means averaged over all times within a corn type, SEM = 2.02.

[e]Means averaged over all sizes within a corn type, SEM = 2.33.

[f]Least significant difference for comparison within columns or rows.

sizes were quite similar with 750 μ (26.60%) being significantly ($p < .05$) higher. An increase in starch digestion of high moisture ensiled corn compared to dry ground corn has been observed by McKnight et al. (1973).

The influence of increased SN in HM corn (Table I) on starch digestion may be important. Galyean et al. (1976) found the in vitro DMD of high moisture ensiled corn was significantly ($p < .05$) greater than dry rolled corn at 3, 6, 9 and 12 hours, presumably due to SN effects. Solubilization of the protein matrix surrounding the starch granule may make starch more accessible to amylolytic bacteria. However, one can only conjecture that higher SN levels influence starch digestion in these data.

Time means (Table VII) show an effect of incubation length on both HM and DG corn similar to that observed with DMD. No significant differences were observed between HM and DG at any incubation period, however.

Experiment IV

Particle size x time interaction was non-significant ($p < .3707$) in this experiment and size and time means (Table VIII) are reported as before. As particle size was reduced from 6000 (16.12%) to 3000 (19.43%) to 1500 (30.83%) to 750 μ (46.32%), DMD increased significantly ($p < .05$). Thus, even with incubation periods extended beyond 8 hours previously studied in Experiment I, particle size had a significant impact on DMD. However, extended incubation tended to eliminate the doubling response that was previously observed. Time effect was also significant. Wilson et al. (1973) also found increased nylon bag

TABLE VIII

EXTENDED INCUBATION NYLON BAG DMD OF FOUR PARTICLE SIZES OF DRY ROLLED CORN (SIZE AND TIME MEANS)

Size	Size Means[a]	Time	Time Means[b]
6000	16.12[c]		
3000	19.43[d]	12	21.96[g]
1500	30.83[e]	24	34.39[h]
750	46.32[f]		

[a]Means averaged over all times, SEM = 2.16.

[b]Means averaged over all sizes, SEM = 1.53.

[cdef]Means in a column with different superscripts are significantly different ($p < .05$)

[gh]Time effect significant ($p < .0002$).

DMD as particle size decreased, even after 48 to 72 hours of incubation.

Experiment V

To determine if the relationships between particle size and DMD described for corn occur in other grains, an experiment was conducted with three particle sizes of sorghum grain (Table IX). Particle size x time interaction was judged highly significant ($p < .0432$) in this experiment, indicating the sizes responded differently depending on incubation length. Within a time, little change occurred between 3000 and 1500 μ sizes with 1500 μ actually lower ($p < .05$) at 4 and 8 hours. In all time periods, however, 750 μ sorghum had higher ($p < .05$) DMD than 3000 and 1500 μ sizes. Figroid et al. (1972) observed similar changes in DMD of sorghum after 8 hr nylon bag incubation. This pattern is similar to the one observed with 6000, 3000 and 1500 μ sizes of DR corn (Experiment I), and indicates particle size has an important effect on DMD once a certain particle size is reached. Walker et al. (1973) studied the effect of particle size on in vitro DMD of sorghum and reported DMD was highly correlated to particle size. It would appear then, that particle size-DMD relationships similar to those observed with corn grain occur in sorghum grain.

These data would indicate that particle size has an important influence on nylon bag dry matter digestion of both corn and sorghum grains. Moreover, processing of corn by steam flaking or high moisture ensiling has an additional effect of increasing dry matter and starch digestion above that observed from particle size alone. Whether rations of reduced particle size would have a higher in vivo ruminal

TABLE IX

NYLON BAG DMD OF THREE PARTICLE SIZES OF SORGHUM

Time	Size[a] 3000	1500	750	LSD[b]
2	8.42[c]	8.07[c]	22.79[d]	4.35
4	15.54[c]	9.66[d]	24.38[e]	4.35
6	15.42[c]	12.12[c]	28.78[d]	4.35
8	20.90[c]	16.32[d]	36.23[e]	4.35

[a]SEM = 1.40.

[b]Least significant difference for comparison within columns or rows.

[cde]Means within a row with different superscripts are significantly different ($p < .05$).

dry matter and starch digestion than those of larger sizes is dependent on other factors. If larger particles are held for longer time periods within the rumen due to omasal filtration mechanisms, allowing time of fermentation to compensate for increased particle size, diets of large particle size may be ruminally digested to the same extent as diets of small grain particle sizes. Thus, additional information relative to particulate outflow rate of grains (and perhaps forages as well) would provide considerable insight into the relationship of particle size and extent of ruminal digestion.

CHAPTER IV

PARTITIONING DIGESTION WITH LIGNIN AS A MARKER

I. INFLUENCE OF CORN PARTICLE SIZE ON SITE AND EXTENT OF DIGESTION IN STEERS

Summary

A 4x4 Latin square design was employed to study site and extent of starch digestion in steers as influenced by corn particle size. Steers (avg. 272 kg) were fitted with permanent rumen cannulae and fed approximately 3.5 kg dry matter in eight portions daily (3 hr intervals). Site of digestion was estimated by withdrawing rumen samples and assuming such samples would be equivalent to abomasal samples if a constant flow of digesta was maintained due to frequent feeding. Ruminal digestion of dry matter (DMD), organic matter (OMD) and starch was estimated by the ratio of lignin in feed to lignin in rumen digesta.

One lot of corn was either ground through 3.18, 4.76 and 7.94 mm screens or left in the whole form and then mixed in 72% corn rations. Geometric mean diameters (microns) of the four grains were as follows: WHOLE (5977.87); 3.18mm (508.94); 4.76mm (587.56) and 7.94mm (832.22).

Digestion of lignin in the total tract was high and variable with WHOLE (53.27%) being different ($p < .05$) from 3.18mm (27.88%)

and 4.76mm (27.99%). Lignin digestion of 7.94mm (38.55%) was different ($p < .05$) from each of the others. Ruminal DMD, OMD and starch digestibilities were considered to be sufficient indicators of trends and not accurate estimates in light of variable lignin digestion and disparities associated with ruminal vs. abomasal sampling. Total tract DMD tended to increase as particle size increased. Ruminal DMD was lower ($p < .05$) for WHOLE (44.87%) than for 3.18mm (62.29%), 4.76mm (63.62%) and 7.94mm (60.78%). Ruminal OMD followed similar patterns. Intestinal DMD and OMD were higher ($p < .05$) for WHOLE than other treatments, indicating the quantitative importance of intestinal digestion in whole corn diets. Total tract starch digestion was lower for WHOLE (88.18%) than for 3.18mm (94.52%), 4.76mm (93.65%) and 7.94mm (93.52%). WHOLE (70.83%) had lower ($p < .05$) ruminal starch digestibilities than ground treatments (avg. 91.72%). Intestinal starch digestion was low and in some cases negative, suggesting ruminal sampling and perhaps variable lignin recovery resulted in overestimation of ruminal starch digestion. Quantities of starch digested in segments of the tract followed patterns similar to digestion coefficients and indicated some alteration of the whole kernel, beyond mastication damage, is needed for ruminal and total tract starch digestion.

Rumen pH and liquid outflow were not significantly influenced by corn particle size. Outflow (L/hr) and dilution rate (%/hr) tended to increase, however, as particle size increased. No significant differences were observed in molar percentages of VFA's across treatment. Fecal pH did not appear to be significantly related to starch content of feces, and dilution rate was not significantly related to molar percent acetate, propionate or butyrate. The value of ruminal

sampling techniques and of lignin as a marker in site of digestion studies with high concentrate diets is discussed.

Introduction

Processing grain by reduction of particle size has been shown to have an important impact on extent of total tract digestion in ruminants. Finely ground sorghum had higher dry matter digestion coefficients than whole and coarsely ground grain fed to steers in a Kansas study (Smith et al., 1949). Walker et al. (1973) observed higher ruminal dry matter digestibility in finely ground sorghum than in intermediate or coarsely ground sorghum. Mehen et al. (1966) reported greater total tract dry matter digestion with fine ground vs. dry rolled sorghum in 77% grain rations fed to steers. In work with corn, Wilson et al. (1973) observed higher total tract organic 2.5 kg hay and 5 kg corn was fed to dairy cows. Galyean (1977a) found particle size influenced nylon bag dry matter and starch digestion in steam flaked and high moisture ensiled corn as well as in unprocessed corn.

The purpose of this study was to examine the site and extent of dry matter and starch digestion in corn-based diets as influenced by corn particle size. The relationship of particle size to rumen pH, volatile fatty acid percentages, and liquid outflow rates was also considered.

Experimental Procedures

Four Brown Swiss x Hereford crossbred steers (avg 272 kg) fitted with permanent, 10.2 cm diameter rumen cannulae and housed in individual metabolism stalls were utilized in a 4x4 Latin square design. A 72%

corn ration (Table X) was fed in 8 portions daily (3 hr intervals) by use of an automatic feeding system.

The four ration treatments differed only in grain particle size. Corn was obtained from the University feed mill and either ground through 3.18, 4.76 and 7.94 mm screens or left in the whole form. Rows of the Latin square were four, 10-day feeding periods while columns were the four steers.

Days 1 through 6 of each period served as an adjustment to new rations, and a total collection of feces was taken on days 7 through 10. Feces was weighed daily and a 10% aliquot retained and refrigerated at 4° C. Aliquots from each of four days were composited, mixed and a sub-sample dried at 65° C for 48 hrs, followed by grinding through a 1 mm screen in a Wiley mill. In an effort to partition ruminal and post-ruminal digestion, 500 ml whole rumen fluid samples were withdrawn on days 9 and 10 of each period by vacuum pump, dried and ground in the same manner as feces samples. The procedure for determining site and extent of digestion is similar to that reported by Balch (1957), except no consideration was given to specifically obtaining samples from the omaso-abomasal orifice. It was felt samples obtained ruminally would be equivalent to abomasal samples if a steady flow of digesta was maintained as a result of frequent feeding. In an effort to measure rumen liquid outflow and rumen volume, chromium ethylene diamine tetraacetic acid (Cr EDTA) was prepared by the method of Binnerts et al. (1968), and 250 ml of solution was dosed ruminally on day 9 of each period. Five hundred ml samples of whole rumen contents were obtained at 4, 7 and 24 hr post-dosing, strained through 4 layers of cheesecloth and approximately 5 ml of 20% sulfuric acid per 100 ml fluid was added to

TABLE X

COMPOSITION OF EXPERIMENTAL DIETS

Ingredient	International Reference No. (IRN)	% D.M. Basis
Corn, dent yellow grain (4)	4-02-935	72
Cotton, seed hulls (1)	1-05-599	16.80
Alfalfa, aerial part, dehy grnd, mn 15% protein (1)	1-00-022	4.80
Soybean, seeds, solv-extd grnd, mx 7% fiber (5)	5-04-604	4.42
Urea, mn 45% N (5)		.64
Salt		.50
Calcium phosphate Dibasic commercial (6)	6-01-080	.40
Calcium carbonate Commercial, mn 38% Ca (6)	6-01-069	.40
Aurofac-50		.02
Vitamin A, palmitate[a] Commercial (7)	7-05-143	.02

[a]3000 IU/gm.

stop microbial action. An additional 500 ml of whole fluid was obtained at the 4 and 24 hr samplings and served as digesta samples for site of digestion estimates discussed previously. Strained fluid from 7 hr samples was analyzed for pH and volatile fatty acids (VFA). VFA procedures have been outlined by Erwin et al. (1961), and a Bendix Series 2500 gas chromatograph was utilized for analysis. Chromium concentration in 4, 7 and 24 hr samples was determined by atomic absorption spectroscopy. This was accomplished by centrifuging strained fluid at 18,000 x g twice, decating the supernatant and aspirating the clear fluid through an Instrumentation Laboratory, Model 253 spectrophotometer. Rumen volume and outflow rates were calculated by regression analysis of the natural logarithm of Cr concentration on time.

Ration samples were obtained on days 7 through 10 of each period, composited and ground in the same manner as fecal and rumen samples. Dry matter, ash, starch and lignin analyses were conducted on rumen, ration and fecal samples. Starch was determined as alpha-linked glucose polymers (MacRae and Armstrong, 1968). Lignin was determined by the permanganate oxidation procedure of Van Soest and Wine (1968). Crude protein content was determined on ration and fecal samples by the Kjeldahl procedure. Particle size of the grains was determined by the method of Ensor et al. (1970). pH of wet fecal contents was also determined with a combination electrode.

Total tract digestibilities of dry matter, organic matter, starch and crude protein were determined by difference between feed and feces. Ruminal dry matter, organic matter and starch digestibilities were determined using lignin as a marker and calculating the ratio of lignin in feed to lignin in rumen samples. Rumen lignin was the average

of the two (4 and 24 hr) digesta samples.

Data were subjected to standard statistical analysis procedures for Latin square designs. Tests of significance among treatment means were accomplished by use of an LSD protected by a preliminary F test.

Results and Discussion

Chemical composition of the diets was similar as would be expected. Dry matter, organic matter, crude protein and lignin percentages on a dry matter basis (respectively) for the four diets were as follows: WHOLE (89.53, 96.27, 11.19, 57.80, 4.09); 3.18mm (89.56, 96.39, 11.81, 55.76, 4.16); 4.76mm (89.53, 96.16, 11.31, 57.99, 4.09) and 7.94mm (89.74, 96.08, 11.44, 56.64, 4.61). Geometric mean diameter and standard deviation of the four diets was: WHOLE (5977.87 $\pm$ 1.03 μ); 3.18mm (508.94 $\pm$ 1.11 μ); 4.76mm (587.56 $\pm$ 1.19 μ) and 7.94 mm (832.22 $\pm$ 1.16 μ) indicating considerable variation in particle size; however, little difference was noted in geometric mean diameter between 3.18 and 4.76mm diets, with 7.94mm being relatively fine, also.

Digestion coefficients as influenced by corn particle size are shown in Table XI. Total tract dry matter digestion (DMD) was similar for all treatments; however, DMD tended to increase as particle size increased. A similar pattern was observed for total tract organic matter digestion (OMD). Ruminal DMD was significantly lower ($p < .05$) for WHOLE (44.87%) than for 3.18mm (62.29%), 4.76mm (63.63%) and 7.94mm (60.78%). Ruminal OMD was similar, although slightly higher in all cases, and was lower ($p < .05$) for WHOLE than for other treatments. Since ruminal DMD and OMD were lower for WHOLE, one would expect the response observed in percentage dry matter digested intestinally. WHOLE

TABLE XI

DIGESTION COEFFICIENTS OF CORN BASED DIETS AS INFLUENCED BY PARTICLE SIZE

Item	Diet				SEM
	3.18mm	4.76mm	7.94mm	WHOLE	
Total DMD, %	75.55	76.12	76.52	77.10	1.07
Total OMD, %	76.07	76.69	77.08	77.43	1.04
Ruminal DMD, %	62.29[a]	63.62[a]	60.78[a]	44.87[b]	2.99
Ruminal OMD, %	64.88[a]	65.71[a]	63.89[a]	47.34[b]	3.20
DM digested intest., %	33.33[a]	34.06[a]	39.72[a]	54.50[b]	3.67
OM digested intest., %	28.85[a]	31.26[a]	35.87[a]	51.99[b]	4.32
Total starch digestion, %	94.52	93.65	93.52	88.18	1.68
Ruminal starch digest., %	92.02[a]	90.18[a]	92.96[a]	70.83[b]	5.02
Starch digest. intest., %	15.93	5.19	-31.11	-2.18	39.62
CP digestion, %	69.23	67.88	67.57	69.56	1.75
Total lignin digest., %	27.88[a]	27.99[a]	38.55[b]	53.27[c]	2.98

[abc]Means in a row with different superscripts are significantly different ($p < .05$)

had higher ($p < .05$) intestinal digestion (54.50%) than other treatments (avg 35.70%). Intestinal OMD paralleled intestinal DMD. As intestinal digestibility calculations are a function of dry matter leaving the rumen and fecal dry matter, their accuracy depends entirely on the accuracy of ruminal DMD and OMD estimates. In this regard, one must consider the high and variable lignin digestion coefficients observed in this study. WHOLE had an apparent total tract lignin digestibility of 53.27% while 3.18mm (27.88%) and 4.76mm (27.99%) were significantly lower ($p < .05$). The lignin digestibility of 38.55% for 7.94mm was different ($p < .05$) from WHOLE and the two smaller particle size diets. The magnitude and variation in lignin digestibility observed in this study would lead one to question the use of lignin as a marker, particularly in cases where site of digestion of concentrate diets is determined. Balch (1957) observed total tract lignin digestibilities ranging from 24.4 to -17.3% in diets of all-hay or up to 24 pounds concentrates and 2 pounds hay with dairy cattle, but still considered lignin a suitable marker for partitioning digestion. Several authors have found lignin digestion coefficients greater than 10% in forage and mixed forage:concentrate diets (Ely et al., 1953; Elam et al., 1962 and Johnson et al., 1964). Whether the extremely high and variable lignin digestibilities observed in this study are due to analytical errors arising from attempts to measure small amounts of lignin in feed or are in fact due to lignin breakdown is unknown. Thus, ruminal digestion coefficients reported in this study should perhaps be regarded as indicators of trends and not accurate estimates. Total tract digestion coefficients should be valid estimates, however, since total feed and fecal collections were used in these calculations.

Starch digestion coefficients (Table XI) indicate a trend for lower total tract starch digestion as particle size increased. Galyean (1977a) reported higher nylon bag starch digestibilities with an average particle size of 750μ than with sizes of 6000, 3000 and 1500μ in unprocessed corn. In the present study, the smallest corn particle size (3.18mm) was roughly 6% higher in total starch digestion than WHOLE (94.52% vs. 88.18%). Level of roughage fed in this study may have reduced starch digestion of WHOLE relative to other treatments as Cole et al. (1976a) have shown total starch digestion increases as roughage level decreases in whole corn diets.

Ruminal starch digestion was high for all treatments and almost equaled total starch digestion in the case of ground corn diets. However, WHOLE (70.83%) was lower ($p < .05$) than 3.18mm (92.02%), 4.76mm (90.18%) and 7.94mm (92.96%). Cole et al. (1976a) have reported a similar ruminal starch digestion value for whole corn in a diet of comparable roughage level. Increasing particle size had little effect on ruminal starch disappearance within ground treatments which may be due to similarity in geometric mean diameters or to selective filtration, allowing large particles to remain in the rumen for longer periods. Nordin and Campling (1976) observed little difference between finely ground (2.5mm) and coarsely ground (3.6mm) corn after 24 hr incubation in nylon bags. It would appear from these estimates, however, that some physical alteration of the whole kernel must occur for maximum ruminal and total tract starch digestion. Starch entering the intestine and digested there was quite low and variable with 7.94mm and WHOLE diets having negative values (-31.11 and -2.18%, respectively). Thus it would appear that sampling ruminally rather than from the

abomasum, resulted in an over-estimation of ruminal starch digestion, causing negative intestinal values. Ruminal and intestinal starch digestibilities, as discussed with DMD and OMD, are probably not accurate estimates but should be sufficient for indicating trends in digestion as influenced by particle size. One might conclude that over-estimates of ruminal starch digestion observed in these data are more likely a result of ruminal sampling techniques than a result of lignin degradation. Since lignin breakdown would be most logically confined to the rumen, any lignin digestion would result in under-estimates of digestion rather than over-estimates. Crude protein digestibility was similar for all treatments, suggesting alteration of physical form has little influence on apparent total tract protein utilization.

Quantities of DM, OM and starch digested are given in Table XII. Dry matter intake was approximately 1.25 times calculated maintenance energy needs, and was similar across treatments. Amounts digested were calculated on the basis of digestion coefficients presented in Table II, and therefore, would be expected to show patterns similar to digestion coefficients. Grams ruminal DMD and OMD were very similar for ground corn diets with WHOLE being significantly ($p < .05$) lower. Dry matter and OM digested intestinally points out intestinal digestion is quantitatively far more important in WHOLE than in 3.18, 4.76 and 7.94mm ground corn diets. Starch intake (avg 2031.06 g) was similar across treatments and grams ruminal starch digestion was approximately 400 g lower for WHOLE than for the average of all ground diets. Again, this suggests the need for some physical alteration of the whole kernel, beyond that incurred during mastication, for maximum ruminal starch digestion. Wilson et al. (1973) found almost no dry matter was digested

TABLE XII

AMOUNTS OF CORN BASED DIETS DIGESTED
AS INFLUENCED BY PARTICLE SIZE

Item	Diet				SEM
	3.18mm	4.76mm	7.94mm	WHOLE	
Intake, DM, gm	3582.20	3581.20	3589.70	3478.80	33.80
Ruminal DMD, gm	2231.52[a]	2278.01[a]	2182.06[a]	1580.59[b]	112.82
Ruminal OMD, gm	2240.27[a]	2262.48[a]	2206.33[a]	1607.61[b]	114.93
Intestinal DMD, gm	474.56[a]	448.09[a]	564.67[a]	1096.82[b]	103.27
Intestinal OMD, gm	382.42[a]	378.42[a]	455.96[a]	980.96[b]	104.11
Fecal DM, gm	876.12	855.10	842.98	801.38	43.73
Fecal OM, gm	826.44	802.65	790.57	760.88	41.77
Intake, starch, gm	1997.48	2076.50	2033.54	2016.73	85.69
Ruminal starch, gm	1842.34[a]	1876.49[a]	1892.36[a]	1467.33[b]	97.23
Intestinal starch, gm	45.47	70.31	8.49	301.07	105.16
Fecal starch, gm	109.67	129.79	132.68	248.33	34.92

[ab]Means in a row with different superscripts are significantly different ($p < .05$)

in whole corn kernels incubated in nylon bags, even after 3 days of incubation. This would suggest starch escaping fermentation in whole corn diets is in the form of whole kernels that excape mastication damage. Considerable quantities of intact kernels in the feces were observed in the present study when corn was fed in the whole form, suggesting intestinal enzymatic attack on kernels escaping ruminal digestion is of a limited nature. Low and variable quantities of starch digested intestinally in this study are no doubt a function of overestimation of ruminal digestion rates, as discussed previously.

Rumen pH (Table XIII) was similar for all treatments with a trend for increasing pH as particle size increased. Fecal pH was significantly higher for WHOLE (6.63) than for 3.18mm (6.33), 4.76mm (6.23) and 7.94mm (6.25). Wheeler and Noller (1977) have reported a high and significant negative correlation between fecal pH and starch in feces. These authors suggested dietary buffers increased intestinal pH to a more favorable range for pancreatic alpha amylase activity and decreased loss of starch in feces on cattle fed high energy rations. The results of this study are not in agreement. When fecal pH was fit as a covariable in a model with steer, period and treatment as independent dummy variables and grams fecal starch or total starch digestion as dependent variables, fecal pH had no significant effect ($p < .70$). In fact, WHOLE had the highest amount of fecal starch and the highest fecal pH in direct contrast to the results of Wheeler and Noller (1977). However, one must recognize fecal pH in this study was not as low as in the earlier work.

Rumen liquid outflow measurements (Table XIII) were not significantly different among treatments, although some noteworthy trends were

apparent. Rumen volume showed no consistent pattern; however, dilution rate (%/hr) and outflow rate (L/hr) tended to increase as particle size increased. WHOLE (1.09) had the highest outflow followed by 7.94mm (0.94), 3.18mm (0.81) and 4.76mm (0.74). Turnover time (days) was essentially the inverse of outflow since rumen volumes were similar. If one is willing to assume particulate and liquid outflow rates of grain based diets are similar, these data would suggest large particles are not held for longer periods in the rumen than smaller particles and may in fact pass more rapidly. The importance of dilution rate and outflow on other factors has been demonstrated by Cole et al. (1976). Microbial protein synthesis per 100 g DM fermented was found to be positively correlated to dilution rate (r = .85) in steers fed high concentrate diets. Isaacson et al. (1975) observed similar trends using continuous culture techniques.

Table XIV gives molar percentages of VFA's as influenced by particle size. No significant differences were noted in molar percentages of any VFA or in total VFA concentration (μM/ml). Molar percent propionate was lower than would be expected with high concentrate diets; however, this trend has been observed previously at this laboratory (Galyean et al., 1976 and Cole et al., 1976), and may be due to utilization of an automatic feeding system. Only small amounts of digesta would be presented to the rumen at a given time using automated feeding, thus reducing fermentable substrate concentration and enhancing salivary flow and buffering.

Hogsdon and Thomas (1975) have observed a relationship between molar percent propionate and dilution rate of the liquid phase. Lower propionate levels were reportedly associated with faster dilution rates.

TABLE XIII

RUMEN PH, FECAL PH AND FLUID OUTFLOW PARAMETERS AS INFLUENCED BY CORN PARTICLE SIZE

Item	Diet				SEM
	3.18mm	4.76mm	7.94mm	WHOLE	
Rumen pH	6.20	6.13	6.25	6.33	0.07
Fecal pH	6.33[a]	6.23[a]	6.25[a]	6.63[b]	0.08
Rumen volume, L	21.70	13.53	15.12	19.41	3.70
Dilution rate, %/hr	4.52	6.54	6.23	5.88	1.13
Outflow, L/hr	0.81	0.74	0.94	1.09	0.12
Turnover, days	1.26	0.95	0.69	0.73	0.29

[ab]Means in a row with different superscripts are significantly different ($p < .05$).

TABLE XIV

MOLAR PERCENTAGES OF RUMINAL VFA'S AS INFLUENCED BY CORN PARTICLE SIZE

Item	Diet				SEM
	3.18mm	4.76mm	7.94mm	WHOLE	
Acetate	61.91	64.97	65.62	61.53	1.76
Propionate	17.88	17.61	18.15	19.18	1.65
Iso-butyrate	1.24	1.09	0.92	1.23	0.05
Butyrate	11.87	9.38	10.13	11.52	1.72
Iso-valerate	3.45	3.59	3.09	3.16	0.41
Valerate	2.06	1.93	1.37	1.93	0.23
Caproate	1.50	1.57	0.72	1.43	0.15
Total VFA (uM/ml)	182.81	188.19	205.82	160.21	24.19

However, in this study dilution rate did not have significant effects on molar percent acetate, butyrate, propionate or total VFA concentration when fit as a covariable in the Latin square model. It should be pointed out, however, that wide ranges in dilution rate similar to those in the work of Hogsdon and Thomas (1975) were not observed in this study. Total concentrations of VFA's are shown in Appendix Table XX.

In conclusion, this study suggests dietary corn particle size has little effect on ruminal, total and intestinal dry matter, organic matter and starch digestion of corn ranging from 508 to 832 microns in geometric mean diameter. Corn in the whole form, however, was significantly lower than ground diets in ruminal dry matter, organic matter and starch digestion, indicating the quantitative importance of intestinal digestion in whole corn diets. One might conjecture similar patterns may hold true for any diet of large particle size. Furthermore, lignin may not be a suitable marker for studies in which site and extent of concentrate diets are investigated, based on its high and variable digestion observed in this study. Sampling ruminally rather than from the abomasum resulted in obvious over-estimates of ruminal starch digestion and this technique is not recommended as it does not appear to provide representative samples in terms of nutrient content.

CHAPTER V

PARTITIONING DIGESTION WITH LIGNIN AS A MARKER. II. INFLUENCE OF LEVEL OF INTAKE ON SITE OF DIGESTION IN STEERS

Summary

Crossbred steers (avg 285 kg), fitted with permanent rumen cannulae and fed an 85% corn diet at 1.00, 1.33, 1.67 and 2.00 times maintenance intake (1.00M, 1.33M, 1.67M and 2.00M, respectively) were employed in a 4 x 4 Latin square design. Site and extent of dry matter, organic matter and starch digestion was estimated by withdrawing rumen samples and assuming such samples would be equivalent to abomasal samples if a constant flow of digesta was maintained due to frequent feeding. Digestion coefficients were calculated on the basis of a lignin in feed to lignin in rumen digesta ratio.

Since digestion coefficients for lignin were high (avg 51.91%), ruminal and intestinal digestion coefficients for dry matter, organic matter and starch were assumed to be adequate indicators of trends but not accurate estimates. Disparities associated with rumen rather than abomasal sampling may have played a large part in problems related to ruminal digestion coefficients (Galyean, 1977b).

Total tract dry matter digestion (DMD) was lower ($p < .05$) for 2.00M (77.64%) and 1.67M (78.89%) than for 1.33M (84.09%) and 1.00M

(85.70%). Ruminal DMD declined from 66.22% (1.00M) to 60.14% (2.00M) as intake doubled, with ruminal organic matter digestion (OMD) following a similar pattern. A trend for less dry matter and organic matter digested intestinally was observed as intake increased from 1.00M (52.58%) to 1.67M (46.75%) and 2.00M (41.40%). Steers fed 1.00M (99.64%) and 1.33M (98.38%) had higher ($p < .05$) total tract starch digestion coefficients than those fed 1.67M (98.80%) and 2.00M (90.41%). Less starch ($p < .05$) was digested ruminally as intake increased from 1.00M (94.48%) to 2.00M (89.64%). Low and negative intestinal starch digestibilities suggest ruminal starch digestion was over-estimated, presumably due to obtaining non-representative samples by ruminal rather than abomasal sampling, as previously mentioned. Grams of nutrients digested in segments of the tract followed trends observed with digestion coefficients. Similarity in grams dry matter and organic matter digested intestinally may suggest a limited starch digestion capacity in that organ.

Rumen pH tended to decrease as intake increased, as did fecal pH. Rumen liquid volume decreased as intake increased from 1.00M (23.58 l) to 2.00M (18.60 l), and dilution rate, outflow rate and turnover time increased with increasing intake. Level of intake did not significantly influence molar percentages or total concentrations of volatile fatty acids (VFA's).

Introduction

A considerable amount of research relative to site of dry matter and starch digestion by cattle fed high concentrate diets has been conducted with research animals at or near maintenance energy intakes. It

would seem logical that increasing level of intake might influence results of such studies. Anderson et al. (1959) found total tract dry matter digestion decreased from 87.5% at one-half calculated maintenance intake to 74.3% at 2.7 times maintenance needs with 80:20, concentrate: forage diets fed to steers. Wheeler et al. (1975) in studies with dairy cattle observed a decrease in total starch digestion of approximately 12% as intake increased from maintenance to 3.2 times maintenance dry matter needs on 71:30, concentrate:forage rations. In contrast, Kratchner et al. (1973) observed no effect of level of starch intake on ruminal or total tract starch digestion with steers fed steam flaked or dry rolled sorghum ad libitum or 80% ad libitum. Similar results have been reported with rolled barley diets fed to sheep (MacRae and Armstrong, 1969).

Thus, the purpose of this research was to further investigate the effect of level of intake on site and extent of dry matter, organic matter and starch digestion in steers fed a corn based diet. Treatment effects on molar percentages of VFA's, rumen pH, liquid outflow and fecal pH, in addition to relationships between fecal pH and starch in feces and liquid dilution rate and volatile fatty acid percentages were considered.

Experimental Procedure

Four Brown Swiss x Hereford steers (avg 285 kg, range 239 to 307 kg), fitted with permanent, 10.2 cm internal diameter rumen cannulae and housed in individual metabolism stalls were utilized in a 4 x 4 Latin square design. A basal, 84% corn diet (Table XV) was fed at multiples of maintenance intake in 8 portions daily (3 hr intervals) by means of an automatic feeding system.

TABLE XV

COMPOSITION OF BASAL DIET

Ingredient	International Reference No. (IRN)	% D.M. Basis
Corn, dent yellow grain (4)	4-02-935	84.00
Cotton, seed hulls (1)	1-01-599	4.80
Alfalfa, aerial part, dehy grnd, mn 15% protein (1)	1-00-022	4.80
Soybean, seeds, solv-extd grnd, mx 7% fiber (5)	5-04-604	4.42
Urea, mn 45% N (5)		.64
Salt		.50
Calcium phosphate, Dibasic commercial (6)	6-01-080	.40
Calcium carbonate, Commercial mn 38% Ca (6)	6-01-069	.40
Aurofac-50		.02
Vitamin A, palmitate[a] Commercial (7)	7-05-143	.02

[a]30,000 IU/gm.

The four treatments were maintenance (1.00M), 1.33 times maintenance (1.33M), 1.67 times maintenance (1.67M) and two times (2.00M) calculated maintenance net energy intakes (NRC, 1970). Maintenance dry matter intakes were calculated for individual steers based upon their weight at the beginning of the trial and were not adjusted for changes in weight during the trial due to the short nature of each feeding period (10 days). Corn in the basal diet was obtained from one lot at the University feed mill and ground through a 4.76 mm screen. The calculated net energy for maintenance of the basal diet was 2.11 Mcal NE_m/kg dry matter. Geometric mean diameter and standard deviation of corn in the basal diet was 714.49 $\pm$ 1.14 microns and ration dry matter, organic matter, crude protein, starch and lignin content, respectively were 89.13%, 96.36%, 12.67%, 67.20% and 3.09%. Rows of the Latin square were four, 10-day feeding periods while columns were the four steers. Sampling and experimental procedures have been reported previously (Galyean, 1977b).

Due to the failure of one steer to consume basal diet at a level of 2.0 times maintenance, one cell of the Latin square was missing. Therefore, data were analyzed by least squares analysis of variance. Standard errors of treatment means were derived from the residual mean square entry of the analysis of variance and based on 3 observations per treatment mean rather than 4. In this respect, standard errors for treatment means are probably more conservative estimates than if derived from the X'X inverse matrix. Tests of significance among treatment means were accomplished by use of an LSD protected by a preliminary F test.

Results and Discussion

Digestion coefficients for dry matter, organic matter, starch and crude protein are shown in Table XVI. Total tract lignin digestibility is noteworthy in that extremely high values (avg 51.91%) were observed, although level of intake had no apparent effect. These results agree with those of Galyean (1977b) who observed lignin digestibilities ranging from 53.27 to 27.88% on 72% corn diets of varying corn particle sizes. Galyean (1977b) discussed the value of lignin as a marker in studies involving site of digestion with high concentrate diets and concluded ruminal and intestinal digestion coefficients observed in their data were sufficient to indicate trends, but were probably not accurate estimates, at least due in part to lignin digestibilities. Similar conclusions would appear to be applicable to data from the present study. Unfortunately, a definitive answer to the question of whether lignin was truly digested or was merely over or under-estimated analytically cannot be drawn from either of these studies. However, one could conclude from results of this study and unpublished data from this laboratory that the value of lignin as a marker in site of digestion studies with high concentrate diets is questionable, due to its variable recovery.

Total tract DMD was lower ($p < .05$) for 2.00M (77.64%) and 1.67M (78.89%) than for 1.33M (84.09%) and 1.00M (85.70%). Total OMD followed a similar pattern, suggesting that as level of intake increases on high concentrate diets, total DMD and OMD decrease markedly. Part of the decrease in total DMD and OMD may be attributable to a trend for lower ruminal DMD and OMD coefficients. Ruminal DMD declined from

TABLE XVI

DIGESTION COEFFICIENTS OF A CORN BASED DIET AS INFLUENCED BY LEVEL OF INTAKE (LEAST SQUARES MEANS)

Item	Level of Intake				SEM
	1.00M	1.33M	1.67M	2.00M	
Total DMD, %	85.70[a]	84.09[a]	78.89[b]	77.64[b]	1.00
Total OMD, %	86.36[a]	84.69[a]	79.45[b]	78.22[b]	1.00
Ruminal DMD, %	66.22	63.27	59.40	60.14	1.94
Ruminal OMD, %	69.92	67.04	63.07	63.12	2.03
DM digested intest., %	52.58	55.98	46.75	41.40	4.82
OM digested intest., %	48.72	51.96	42.29	37.53	5.85
Total starch digestion, %	99.64[a]	98.38[a]	93.80[b]	90.41[b]	1.06
Ruminal starch digest., %	94.48[a]	92.79[ab]	92.36[ab]	89.46[b]	0.92
Starch digest. intest., %	23.00	31.54	-35.09	-25.31	32.44
CP digestion, %	78.86	77.43	74.15	76.79	1.61
Total lignin digest., %	51.61	52.92	51.54	51.58	2.68

[ab]Means in a row with different superscripts are significantly different ($p < .05$).

66.22% (1.00M) to 60.14% (2.00M) as intake doubled. Moreover, as greater quantities of DM and OM bypassed ruminal fermentation, intestinal digestive capacity was apparently overtaxed. A trend for lower percentages of dry matter entering the intestine and digested there was observed as intake increased from 1.00M (52.58%) to 1.67M (46.75%) and 2.00M (41.40%). Thus level of intake apparently influences site and extent of DM and OM digestion, although the data are quite variable in this respect.

Total tract starch digestion, similar to DMD and OMD, decreased as intake increased. Steers fed 1.00M (99.64%) and 1.33M (98.38%) had significantly higher ($p < .05$) starch digestion coefficients than those fed 1.67M (93.80%) and 2.00M (90.41%). Work of Wheeler et al. (1975) is in agreement, yet perhaps more dramatic. Dairy cows fed 70:30, concentrate:forage diets in their studies had lower total starch digestion coefficients as intake increased from maintenance (96.2%) to 3.2 times maintenance (84.7%). Ruminal starch digestion coefficients in the present study reveal less ($p < .05$) starch was digested ruminally when intake increased from 1.00M (94.48%) to 2.00M (89.46%), with 1.33M (94.79%) and 1.67M (92.36%) not significantly different from the two extremes. In contrast, Kratchner et al. (1973) observed no effect of level of intake on ruminal starch digestion of sorghum diets. An indication that ruminal starch digestion coefficients in the present study are overestimates is given by percent intestinal starch digestion. Values of 23.00% (1.00M), 31.54% (1.33M), -35.09% (1.67M) and -25.31% (2.00M) suggest sampling ruminally rather than from the abomasum or duodenum, resulted in overestimates of rumen digestion and obvious under-estimates of intestinal digestion. If trends in this study are

correct, however, level of intake appeared to reduce digestion of starch ruminally, and increase starch entering the intestine. Digestion coefficients for crude protein were not significantly influenced by level of intake, but tended to decrease with increasing intake.

Grams ruminal DMD (Table XVII) increased significantly ($p < .05$) as intake increased from 1.00M (1707.69) to 1.33M (2160.76) to 1.67M (2550.17) to 2.00M (3233.88). This trend would be expected as percent ruminal DMD was similar across treatment and DM entering the rumen increased. Ruminal OMD (gm) followed a like pattern, although values were higher than gms DMD, reflecting mineral secretion, presumably in saliva, anterior to the abomasum and mineral absorption by the intestine. Quantities of DM and OM digested intestinally tended to increase as level of intake increased, however, the differences were non-significant. This points out that although ruminal digestion coefficients were similar across treatment, the far greater quantities of dry matter entering the intestine as intake increased were not effectively digested in that organ. Similarity in quantity of DM digested in the intestine may support the hypothesis of a limited ability for dry matter (principally starch) digestion in ruminant small intestines. Little et al. (1968) infused increasing quantities of starch (200 to 800 g/day) into the small intestine of steers and observed a decrease in percentage starch digestion as infusion level increased, suggesting a biological maximum for intestinal starch digestion. Fecal DM and OM were significantly higher ($p < .05$) in the present study for 1.67M and 2.00M than for 1.33M and 1.00M, in agreement with trends for similar intestinal digestion across level of intake.

TABLE XVII

AMOUNTS OF A CORN BASED DIET DIGESTED AS INFLUENCED BY LEVEL OF INTAKE (LEAST SQUARES MEANS)

Item	Level of Intake				SEM
	1.00M	1.33M	1.67M	2.00M	
Intake, DM, gm	2558.63	3402.51	4273.14	5331.95	115.47
Ruminal DMD, gm	1707.69[a]	2160.76[b]	2550.17[c]	3194.67[d]	89.70
Ruminal OMD, gm	1739.86	2207.68[b]	2160.99[c]	3233.88[d]	96.98
Intestinal DMD, gm	478.94	696.21	816.24	794.21	117.70
Intestinal OMD, gm	387.83	565.12	655.95	637.97	125.10
Fecal DM, gm	367.51[a]	545.54[a]	906.66[b]	1198.31[b]	57.94
Fecal OM, gm	337.68[a]	505.55[a]	850.70[b]	1125.54[b]	54.73
Intake, starch, gm	1724.47	2288.03	2869.30	3496.99	103.91
Ruminal starch, gm	1637.65[a]	2128.13[b]	2656.93[c]	3126.13[d]	54.22
Intestinal starch, gm	46.90	75.13	-24.77	-80.96	51.76
Fecal starch, gm	39.92[a]	84.77[a]	237.15[b]	418.89[c]	37.81

[abcd]Means in a row with different superscripts are significantly different ($p < .05$).

Grams starch digested ruminally was significantly ($p < .05$) affected by level of intake in the same manner as ruminal DMD and OMD. Intestinal digestion (gms) was low and negative in the case of 1.67M and 2.00M, again suggesting site of sampling resulted in poor estimates. It would appear that sampling from an organ of large volume, like the rumen, increases the chance of obtaining a non-representative sample in terms of starch content or other nutrients vs. sampling from an organ of smaller volume like the abomasum. Ruminal sampling in site of digestion studies is probably of limited value, a conclusion also reached by Galyean (1977b). Fecal starch content (gms) increased rather dramatically as level of intake increased from 1.00M (39.92) to 2.00M (448.89). In this respect, fecal starch is probably a better indicator of intestinal starch digestion in this study if one is willing to assume trends observed in ruminal starch digestion are realistic.

Rumen pH (Table XVIII) was not significantly affected by level of intake; however, it tended to decrease as intake increased. Frequent feeding methods used in this study may have equalized potential differences in pH due to intake, compared to more conventional meal feeding systems. Fecal pH was not different across treatments; however, as with rumen pH, it tended to decrease as intake increased from 1.00M (6.40) to 2.00M (5.99). In contrast to the study reported by Galyean (1977b), fecal pH did decrease as starch in feces increased. However, when fitted as a covariable in the Latin square model for grams fecal starch, the effect of fecal pH was non-significant ($p < .5034$). Wheeler and Noller (1977) reported high and significant negative correlations between fecal pH and starch in feces and suggested large quantities of starch entering the intestine are associated with a decrease in pH of

the small intestine, lowering pH below the optimum of pancreatic alpha amylase. The pH curve for this enzyme may allow for considerable variation in pH of digesta without impairment of activity, however. In addition, another plausible explanation for greater fecal starch could be that large quantities of starch bypassing the intestine may not be due to decreased pH optima for alpha amylase but the result of some other failure of starch digestion mechanisms. Lowered pH of intestines and feces may be due to increased microbial activity as a result of increased starch bypass to the intestine with volatile fatty acid production causing a decline in pH.

Rumen liquid outflow estimates (Table XVIII) were not different among treatments. Rumen liquid volume, however, tended to decrease as intake increased from 1.00M (23.58 L) to 1.33M (20.76 L) to 1.67M (22.90 L) to 2.00M (18.60 L), suggesting liquid is forced from the rumen as more dry matter enters. Moreover, saliva flow on high concentrate diets may not be adequate to maintain liquid volume as dry matter intake increases. In line with decreased rumen volume, dilution rate (%/hr) and outflow (L/hr) increased as intake increased. If particulate and liquid outflow are similar on high-concentrate diets, these data would suggest more dry matter (starch) might be flushed to the intestine with increasing intake, perhaps stressing intestinal digestive capacity.

No significant differences were observed in molar percentages of VFA's as intake increased; however, acetate tended to decrease and propionate increase as multiples of maintenance were fed (Table XIX). In contrast to previous work at this laboratory (Galyean, 1977b) the effect of dilution rate when fit as a covariable in the Latin square model for

TABLE XVIII

RUMEN PH, FECAL PH AND FLUID OUTFLOW PARAMETERS AS INFLUENCED BY LEVEL OF INTAKE (LEAST SQUARES MEANS)

Item	Level of Intake				SEM
	1.00M	1.33M	1.67M	2.00M	
Rumen pH	6.28	6.28	6.15	6.00	0.08
Fecal pH	6.40	6.30	6.08	5.99	0.12
Rumen volume, L	23.58	20.76	22.90	18.60	3.11
Dilution rate, %/hr	3.04	3.45	3.95	5.29	0.89
Outflow, L/hr	0.73	0.75	0.82	1.01	0.13
Turnover, days	1.63	1.57	1.50	0.95	0.56

TABLE XIX

MOLAR PERCENTAGES OF RUMINAL VFA'S AS INFLUENCED BY LEVEL OF INTAKE (LEAST SQUARES MEANS)

Item	Level of Intake				SEM
	1.00M	1.33M	1.67M	2.00M	
Acetate	59.60	54.71	53.31	55.76	3.06
Propionate	18.46	22.07	24.53	21.71	2.87
Iso-butyrate	2.01	1.97	1.77	1.47	0.22
Butyrate	10.09	10.69	10.16	12.07	1.04
Iso-valerate	4.90	5.03	4.10	3.99	0.51
Valerate	2.76	2.19	4.47	2.80	0.48
Caproate	2.18	2.41	2.51	2.14	0.35
Total VFA (uM/ml)	128.86	152.54	143.70	150.72	9.66

acetate and propionate, was significant ($p < .0511$ and $p < .0066$, respectively). Acetate tended to increase and propionate to decrease as dilution rate increased. These results agree with those of Hogsdon and Thomas (1975). Total concentration of VFA's are shown in Appendix Table XXI.

In conclusion, this study would suggest that level of intake has significant effects on site and extent of dry matter, organic matter and starch digestion by steers fed high concentrate diets. Studies conducted at maintenance intake may not be representative of feedlot conditions, and more effort should be made to obtain maximum intake in digestion studies. Furthermore, lignin digestion was high (> 50%) in this study in agreement with previous work at this laboratory (Galyean, 1977b). In light of this work, the value of lignin in site of digestion studies dealing with high concentrate diets may be questionable. Sampling ruminally rather than abomasally is not recommended as overestimates of ruminal starch digestion reported in this study may be attributable, in the most part, to sampling method. Researchers should give greater consideration to choice of marker, sampling method and level of intake in future digestion studies with grain based diets.

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APPENDIX

TABLE XX

TOTAL CONCENTRATIONS OF VFA'S AS INFLUENCED BY CORN PARTICLE SIZE[a]

Item	Diet				SEM
	3.18mm	4.76mm	7.94mm	WHOLE	
Acetate	104.12	123.47	134.46	97.82	11.40
Propionate	39.48	32.33	37.19	32.38	7.48
Iso-butyrate	1.85	1.93	1.80	1.73	0.20
Butyrate	26.12	17.77	22.29	18.77	6.48
Iso-valerate	5.77	6.66	6.21	5.04	0.48
Valerate	3.60	3.26	2.43	2.67	0.61
Caproate	1.86	2.77	1.31	1.80	0.25

[a]All values are in uM/ml.

TABLE XXI

TOTAL CONCENTRATIONS OF VFA'S AS INFLUENCED BY LEVEL OF INTAKE (LEAST SQUARES MEANS)[a]

Item	1.00M	1.33M	1.67M	2.00M	SEM
Acetate	76.53	80.33	75.95	82.57	8.32
Proprionate	25.63	39.38	35.88	35.23	7.20
Iso-butyrate	2.41	2.68	2.49	2.21	0.21
Butyrate	12.44	15.89	14.72	17.38	1.69
Iso-valerate	5.97	6.86	5.66	5.87	0.45
Valerate	3.39	4.29	5.43	4.34	0.79
Caproate	2.49	3.07	3.56	3.12	0.64

[a]All values are in uM/ml.

VITA[2]

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