STORAGE AND UTILIZATION OF DISTILLERS AND BREWERS WET GRAINS IN DIETS FOR LACTATING DAIRY COWS

By

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ABSTRACT

STORAGE AND UTILIZATION OF DISTILLERS
AND BREWERS WET GRAINS IN DIETS FOR
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In tests of methods of preserving distillers and brewers wet grains (DWG and BWG), feeding values for cows were compared.

In Experiment I, DWG were treated with NH_3 at 0, 1.57, 3.14, or 5.71% of dry matter (DM), compacted in polyethylene bags, and exposed to aerobic storage at 15.6, 26.7, or 37.8°C for 14 days. Intermediate and high NH_3 reduced temperature increases, mold growth and spoilage, and improved recoveries of dry matter of the stored grains. Increasing storage temperatures hastened appearance of mold growth and spoilage for untreated and low NH_3 grains.

In Experiment II, lactating Holstein cows were fed a basal diet (corn silage, haylage, alfalfa hay, ear corn, and a mineral-vitamin mix) supplemented with DWG (13.78% of total DM) treated with NH_3, or soybean meal (SBM, 10.12% of total DM) for 70 days. Dry matter intakes tended to be lower for cows fed DWG. Although milk yields and composition did not differ significantly between treatments, cows fed DWG produced more milk per kg DM intake than those fed SBM (1.42 vs 1.25). Income over feed costs was greater for DWG than for SBM.

In Experiment Ia, BWG were treated with NH_3 at 0, 2, or 4% of DM. Portions of the treated grains were compacted in plastic pails and exposed

to aerobic storage for 3, 7, or 10 days. From the same treated grains, portions were compacted in polyethylene bags, sealed with twine, and left to ensile for 7, 14, or 28 days. During aerobic storage, water soluble carbohydrates increased in grains treated with high NH_3 but decreased for no and low NH_3. Mold growth and spoilage were inhibited by high NH_3. During ensiling, NH_3 delayed increases of lactate, presumably by restricted fermentation. Butyrate was evident only with no and low NH_3. High NH_3 was most effective in preserving the wet grains during ensiling. Other grains lost their original color and had a strong acid odor.

In Experiment IIa, lactating Holstein cows were fed the basal diet of II supplemented with either 14.3% SBM, 25.6% fresh BWG, 26.3% ensiled BWG, or 14.7% fresh BWG plus .72% urea for 70 days with daily DM intakes of 25.2, 21.9, 20.8, and 22.8 kg. Differences for actual milk (29.3, 29.4, 27.7, and 29.0 kg/day) and milk composition were not significant. Feed efficiencies and income over feed costs were greater for BWG diets than for SBM.

DWG and BWG can be preserved effectively for 2 wk or more when treated with NH_3 at 3 to 4% of DM. NH_3-treated DWG and forms of BWG studied were equal to and more profitable than SBM as protein supplements.

DEDICATION

To the memory of my beloved mother,

Edna W. Johnson, 1922-1980.

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INTRODUCTION

Large quatities of distillers and brewers wet grains are becoming available from the production of alcohol by distilleries, breweries and gasohol plants. Traditionally, these by-products have been utilized in the dried form as feed ingredients for livestock. However, increased costs associated with drying the by-product has caused renewed interest in feeding these feedstuffs in their wet form. On a dry matter basis, the nutritional value of the wet grains is comparable to the dried grains. A key problem in the utilization of these by-products, however, is the rapid molding and spoilage after production, especially during warm weather.

Much of the research with distillers and brewers by-products have focused on protein resistant to ruminal microbial degradation. From available research, it appears that these by-product feeds are more resistant to microbial degradation in the rumen than the commonly fed oilseed meal from soybeans, cottonseed and linseed. Thus, protein of low rumen degradability will result in more post ruminal digestion and usually enhance net amino acid uptake by the host animal. In high producing dairy cows and rapidly growing beef, microbial protein is insufficient to meet protein needs for achieving maximum performance; thus, an increase in protein which will not degrade in the rumen, but will be broken down postruminally is often benificial.

Presently, nonprotein nitrogen (NPN) is well accepted in rations for ruminants. Of the total NPN presently incorporated into ruminant rations, approximately 90% is urea. However, per unit of nitrogen, the cost of ammonia (anhydrous form) ranges from 60 to 70% of the cost of urea. In addition to ammonia furnishing nitrogen for microbial synthesis in the rumen, ammonia treatment has raised energy availability of low quality roughages, increased the true protein of silages and retarded heating and molding of certain feeds, especially during high ambient temperatures.

The research associated with this thesis was designed to ascertain:

1) The influence of added levels of ammonia, exposure time, and storage temperature on the aerobic stability of distillers wet grains (DWG).
2) The value of ammonia-treated DWG as a protein supplement for lactating dairy cows, and return over feed costs of DWG relative to soybean meal.
3) The influence of added levels of ammonia on the aerobic and anaerobic stability of brewers wet grains (BWG).
4) The comparative value of fresh BWG, fresh BWG + urea, ensiled BWG and soybean meal as protein supplements for lactating dairy cows for maintaining milk yields and return over feed costs.

REVIEW OF LITERATURE

This review will examine the information currently available on the preservation of distillers and brewers wet grains, and also evaluate their feeding value in diets of ruminants, principally dairy and beef cattle and sheep.

Distillers Wet Grains

Production

In the conventional beverage distillery, corn is the primary grain used in the distillation process. Grains are ground, slurried with water and cooked to gelatinize starch. The resulting product is then cooled to a temperature suitable for fermentation, innoculated with yeast and allowed to ferment for three to five days. For the commonly used yeast strains, the optimum fermentation conditions are 27 to 35°C at a pH of 3.0 to 5.0 (NAP, 1981). During the fermentation process, starch is converted to alcohol and carbon dioxide. Starch comprises two-thirds of the original corn grain, and therefore, the concentration of nutrients remaining in the by-products is increased three-fold. Whole stillage is the by-product remaining after the distillation of alcohol. This product typically contains 5 to 14% solids. Grains can then be separated from the whole stillage by screening, pressing or centrifugation. The result is isolation of distillers wet grains (DWG) which contains 25-35% solids (NAP, 1981).

Preservation

A key problem in utilization of DWG is rapid molding and spoilage when exposed to air, especially during warm weather. Traditionally, the wet grains are dried after separation from whole stillage in order to facilitate long term storage and movement through normal feed channels. However, with increased petroleum prices the cost of drying the wet grains also increases. Consequently, large distilleries as well as operators of farm sized stills have become interested in alternative methods of usage.

Immediately after separation from whole stillage, fresh DWG is 71^{o}C, contains 65-75% moisture, and has a pH to 3.0 to 4.0. (Klopfenstein and Abrams, 1981; NAP, 1981). Without any preservatives this by-product has a shelf life of approximately 2 to 3 days at summer temperatures (Waller et al; 1982) and 6 to 8 days or longer in cold weather (NAP, 1981). In spite of the low initial pH attempts to store the wet grains for longer than 2 to 3 days in the summer months have resulted in excessive spoilage, mold growth and dry matter losses (Rakshit and Voelker, 1981; Waller et al., 1982). Most workers conclude that a pH 4.2 is the critical value at which silages are well-preserved (Carpintero et al., 1969). However, for high moisture material such as distillers wet grains, it would appear that a lower critical pH is necessary for acceptable preservation. Whittenbury et al., (1967) reported that Clostridia, implicated in DWG deterioration, can tolerate high concentrations of organic acids and hydrogen ions under very wet conditions. Moreover, Zimmer (1971) observed an inverse relationship between dry matter losses associated with the storage of high moisture material and the degree of air-tightness achieved.

Treating fresh DWG with 1% propionic acid (on a dry matter basis) has eliminated mold growth and reduced dry matter losses over a seven-week storage period (Rakshit and Voelker, 1981; Schingoethe et al., 1983). Since mold growth was eliminated by propionic acid treatment, it seems likely that the destruction of organic acids, especially propionate, by other microorganisms is a factor which allows mold growth in untreated-wet grains. Similar results were reported for high moisture grains (Jones and.Stevenson, 1970), grass silage (Daniel et al., 1970) and haylages (Thomas, 1978) treated with propionic acid. In addition to eliminating mold growth and reducing dry matter losses, propionic acid also reduced temperatures in fresh DWG exposed to air (Rakshit and Voelker, 1981; Diallo et al., 1984). Diallo et al. (1984) reported a linear increase in days (16 to 19) to reach 10^{o}C above ambient temperature with increasing levels of added propionic acid, when concentrations of propionate ranged from .3 to .6% of the dry matter.

There is little information available on alkali treatment of fresh DWG. However, in the study reported by Abrams _et al_. (1983), fresh DWG averaging 31.1% dry matter (DM) were treated with 0, 1.0, 2.0 or 3.0g calcium hydroxide/100g DM or with 0, .309, .618 or .926g ammonium hydroxide/100g DM. In addition, the wet grains were also treated with 0 or 5.0g sugar/100g DM and 0 or .3g live microbial silage inoculant (Streptococcus and Lactobacillus)/100g DM. The treated or untreated material were packed tightly in laboratory silos (.946 liter glass jars), sealed and allowed to ensile for 21 days at 39°C. Results from this study showed that no fermentation occurred in the absence of alkali. Such an observation was expected in view of the low pH of fresh DWG (NAP, 1981). However, alkali additions resulted in a butyric acid fermentation with small amounts of acetate, propionate and lactate produced, thereby suggesting the presence of saccharolytic clostridial activity (Whittenbury _et al_., 1967). In addition, sugar addition resulted in delayed decreases in pH for ammonium hydroxide treated grains. According to Rydin _et al_. (1956), added sugar does not always pass through the process of lactic acid fermentation, but may also be transformed into butyric acid, acetic acid, etc. This results in a delayed decrease in pH, thus providing a favorable medium for the growth of putrefactive clostridia (Whittenbury, 1968). Preliminary observations of ammonia-treated DWG exposed to aerobic storage for 9 days at various temperatures, indicated that 4.5% ammonia (on dry matter basis) was effective in retarding temperature increase, spoilage and dry matter losses (Waller _et al_., 1982; Huber _et al_., 1983). In the study reported by Diallo _et al_. (1984), DWG were treated, on a dry matter basis, with .3, .4, .5 or .6% ammonia and subsequently exposed to air for 24 days. Days to reach 10^{o}C above ambient temperature increased with increased ammonia, but were lower than for untreated grains. The ineffectiveness of ammonia in preventing temperature rises was due to the low levels of ammonia applied. Other measures of stability were not reported.

Formaldehyde (F) applied at 2 to 4% by weight of the dry protein content of DWG also prevented spoilage during a 28-day ensiling period (Waller _et al_., 1982). Lower F levels (.3 to .6% of the dry matter) were also effective in preventing temperature increases of DWG exposed to air for 24 days (Diallo _et al_., 1984). A mixture of sodium diacetate and sorbic acid (72% sodium diacetate and 28% sorbic acid) applied at .3 to .6% of the dry matter also prevented increased temperatures of DWG exposed to air for 24 days (Diallo _et al_., 1984).

Field observations of ethanol plants that fail to remove all the ethanol from the grains during the distillation process indicate that the storage life of DWG with at least 0.7% ethanol is longer than that of the wet grains devoid of ethanol (Waller _et al_., 1982).

Feeding Value For Ruminants:

Typical chemical composition values for corn, DWG and soybean meal are shown in Table 1. From these values, it becomes apparent that an energy source (corn) has been converted to a protein source with higher concentrations of acid detergent fiber, fat, calcium and phosphorus. A comparison of DWG with soybean meal reveals lower protein, but higher concentrations of fiber, fat, phosphorus and total digestible nutrients (TDN) in the by-product. The lysine content of protein in distillers grains, however, is less than half that of soy protein (Satter, 1983). On the other hand, methionine supply from distillers grains is distinctly superior to that from soybean meal (Satter, 1983).

The feeding value of DWG for ruminants is based primarily on its ability to resist microbial degradation in the rumen. This resistance is probably due to zein, the major protein in corn, and its by-products, which are not readily degraded by rumen microorganisms (McDonald, 1954). However, under certain feeding regimens depending on stage and type of production, microbial nitrogen is insufficient to optimally furnish the protein needs of the animal. In order to achieve maximum performance, post ruminal digestion of some dietary protein is required (Ørskov, 1978; Huber and Kung, 1981; Owens and Bergen, 1983).

Lactating Dairy Cattle

There is limited information on feeding DWG to dairy

Table 1. Chemical composition of corn, distillers wet grains (DWG) and soybean meal (SBM)[a]

Item	Corn	DWG	SBM
Dry matter (%)	89	25	90
Crude protein (%)	10	30.6	48.9
Acid detergent fiber (%)	1.4	18	7.0
Fat (%)	4.4	8.0	5.2
Calcium (%)	.02	.16	.28
Phosphorus (%)	.30	.79	.75
TDN (%)	88.0	84.0	81.0

[a]All composition values other than TDN were taken from Telplan Program 31 Form 3 (MSU, 1981); TDN values were taken from NRC (1978).

cattle. In the study reported by Schingoethe et al. (1983), a switch-back design consisting of three-4 wk. periods was used to evaluate the feeding value of DWG for lactating dairy cows. In addition to being fed corn silage ad libitum and 3.2 kg alfalfa hay daily, cows were fed either a control concentrate (18.6% crude protein) consisting of corn, oats and soybean meal at 1 kg/2.5 kg milk produced, or 13.6 kg distillers wet grains (22% of total ration dry matter) and a 10.9% crude protein concentrate mix of corn and oats at 1 kg/2.5 kg milk produced in excess of 11 kg milk daily. Cows averaged 12 wks. postpartum at the start of the experimental period. Results from this study revealed no significant differences in milk or milk composition between the two treatment groups. Also, total dry matter consumption and body weight gains were similiar for both groups. Apart from the higher concentration of rumen propionate for cows fed the DWG, rumen volatile fatty acids, pH and ammonia were similar for both groups. The higher concentration of rumen propionate (24.5 vs. 21.9 moles/100 moles VFA) was probably due to miscalculation of the energy value of distillers wet grains, resulting in more total concentrate fed in the diet.

Growing Beef Cattle

Optimum use of preformed protein and maximum use of nonprotein nitrogen have been the goals of ruminant nutritionists and cattle feeders for many years. As previously mentioned, distillers grains are more resistant to microbial degradation in the rumen than are the commonly fed oilseed meals from soybean, cottonseed and linseed (Satter et al., 1977). However, when fed as the sole source of supplemental protein, lack of degradability of the protein in the rumen can result in a deficiency of rumen ammonia for microbial protein synthesis (Taminga, 1979). Since ammonia is the central compound for protein synthesis in the rumen (Huber and Kung, 1981), it seems possible that ammonia can be provided in the rumen by replacing some of the nitrogen in DWG with urea.

This concept was demonstrated in a cattle growth trial reported by DeHaan et al. (1982), where urea provided 50% of the supplemental protein equivalent in rations supplemented with soybean meal or DWG. The control ration of corn silage and corn cobs was supplemented with only urea nitrogen. Daily gains were highest for steers fed DWG and lowest for the urea control. The same trend was observed for feed efficiency. Protein efficiency, defined as gain above the urea control, divided by amount of supplemental protein intake, was higher for steers on the DWG diet.

In a similar growth trial with steers (Abrams et al., 1983), basal diets consisting of corn silage and corn cobs, were supplemented with the following protein supplements: (1) 100% urea; (2) 50, 75 or 100% soybean meal (SBM); (3) 30, 40 or 50% DWG; or (4) 30, 40 or 50% calcium hydroxide ensiled distillers wet grains (EDWG), with urea making up the difference. Preformed protein supplements were combined with the urea supplement to provide incremental levels of test protein, while dietary protein level (11.5%) remained constant. Except for lower gains by steers on the 100% urea supplemented diet, no differences in gains among the steers fed DWG, SBM or EDWG diets were demonstrated. Feed efficiencies followed the same trend. Because the protein in DWG or EDWG was fed one-half the level of SBM protein, steers fed the urea supplemented DWG demonstrated greater protein efficiency than those fed urea supplemented SBM or EDWG. Urea supplemented EDWG was intermediate, probably due to the increased solubilization of nitrogen during ensiling. Assigning a value of 100% to

SBM, the relative values of DWG and calcium hydroxide EDWG were 282% and 142%, respectively.

Finishing Beef Cattle

Once feeder cattle approach the finishing stage, the ability of the rumen to provide protein exceeds the animal's need (NRC, 1984); thus, research in feeding DWG to finishing cattle for its ruminal undegradable nature would not be useful. However, feedlot performance, as measured by feed efficiency and daily gains, was as good or better when fed diets supplemented with DWG than when fed corn-based diets (Farlin, 1981; Firkins et al., 1985). Best performances were obtained with 42.5 to 50.0% of the dietary dry matter as DWG. The fact that DWG contain little or no starch and a higher fiber content suggests that ruminal pH may not have been decreased as much as with diets based on higher amounts of corn, thereby resulting in the improved feedlot performance reported in these trials. However, results from these trials suggest that DWG can be utilized effectively as an energy source by feedlot cattle at levels as high as 50% of dietary dry matter.

Sheep

Studies conducted with sheep in which DWG provided supplementary nitrogen have been limited. However, Abrams et al. (1983) reported that lambs gained faster and more efficiently when DWG and urea provided supplemental nitrogen than when soybean meal and urea or urea alone were protein supplements. Protein efficiencies (gain above the urea control, divided by amount of supplemental protein intake) were also higher for the distillers grain-urea combination (fresh or ensiled with calcium hydroxide). In another study with similar treatment combinations, dry matter intake and digestibility were higher when soybean meal provided the supplemental nitrogen. Generally, if nitrogen were limiting the rumen fermentation, then voluntary feed intake and digestibility would be depressed. From the data reported, rumen ammonia levels at 4 and 6 hours after feeding were lower for lambs receiving DWG compared to those receiving soybean meal.

Brewers Wet Grains

Production

No two breweries produce beer in exactly the same manner. Hence, variation exist both in processing and raw materials used. Nevertheless, the basic cereals used in U.S. breweries today are barley and either corn or rice.

In the brewing process, the cereals are first mashed under carefully controlled conditions in order to convert the starch into more soluble sugars. After mashing is complete, the material is separated into liquid and solid fractions by extraction or filtration. The liquid portion (called "wort") containing the fermentable sugars is added to brew kettles together with hops to produce beer or ale. Brewers wet grains (BWG) is the solid fraction remaining after the "wort" is removed. This fraction may either be fed to animals without further processing or dried to produce brewers dried grains (BDG).

Preservation

A practical concern with the utilization of BWG is their keeping quality during storage and subsequent exposure to air, especially during warm weather. Drying produces a stable product; however, due to costs associated with drying, alternative methods are being investigated.

One method of preservation that has been and is currently being investigated is the application of organic chemicals to the wet grains to prevent spoilage during storage and exposure to air. Chemicals most commonly used are formic acid, propionic acid, formaldehyde and paraformaldehyde. Effects of these preservatives are well documented for high moisture crops (Thomas, 1978) and grains (Jones and Stevenson, 1970; Jones, 1970).

Using test-tube silos (160 ml), Allen and Stevenson (1975) showed that 0.50 and 0.75% formic acid or 0.75% formic-propionic acid mixture, applied on a fresh weight basis, were effective in preserving fresh BWG during 18 days of ensiling. Lower levels resulted in poorly preserved grains containing high levels of acetic acid, butyric acid and ammoniacal nitrogen. Another study (Oleas, 1977) using 250-liter barrels lined with polyvinyl as silos, showed that 2% propionic acid or 1.4% formic acid plus 0.1% paraformaldehyde were effective in preserving fresh BWG during

60 days of ensiling. Ethanol and lactic acid were the main fermentation products. In that study, the barrels were sealed with plastic bags filled with water.

Storing untreated BWG in uncovered piles has resulted in extensive mold growth, discoloration and dry matter losses (Allen _et al_., 1975). However, a 0.40% formic-propionic acid mixture was effective in reducing all aspects of deterioration during a 14-day exposure period. Formic or propionic acid applied at the same level were effective in reducing subsurface deterioration, but were unable to reduce surface spoilage. Sodium chloride, applied at 1.0% of the fresh weight, reduced spoilage in BWG exposed to air (Chase, 1977).

Another method investigated is the application of fermentation stimulants in the form of carbohydrate sources or lactic acid forming bacteria to wet grains. Interest in these methods stems from the fact that most fermentable carbohydrates, which are substrates for lactic acid production, are removed from the grains during brewing. Molasses, the most widely used carbohydrate source, has successfully improved preservation of high moisture crops harvested with levels of low soluble carbohydrates (Dijkstra, 1958; Carpintero _et al_., 1969; McCarrick, 1969). However, when used with BWG, no improvement over untreated wet grains was demonstrated under aerobic or anaerobic conditions (Allen and Stevenson, 1975; Allen _et al_., 1975; Oleas, 1977). Negative results were also reported for lactic acid cultures (Oleas, 1977) with no significant improvement over untreated wet grains. Schoch (1957), however, reduced dry matter losses in BWG ensiled with 10 to 15% dried apple residue.

Feeding Value For Ruminants

As with distillers wet grains, the feeding value of BWG for ruminants is enhanced because protein in the grain resists microbial degradation in the rumen and is available for digestion and absoption in the small intestine. Based on *in vitro* and *in vivo* estimates, the protein in BWG was much more resistant to microbial degradation than that of soybean meal, although not quite as resistant as that of the dried grains (Satter and Whitlow, 1977; Klopfenstein *et al*., 1977; Santos *et al*., 1984). Apart from its high protein content (23-39%) and value, the high fiber (23-62% acid detergent) and energy (66-78% TDN) in BWG make it a valuable component in ruminant rations.

Lactating Dairy Cattle

There is limited information on the relative feeding value of BWG compared to the dried grains in diets for lactating dairy cows. However, the studies by Conrad and Rogers (1977) suggest that BWG are used more efficiently for milk production than the dried grains. Although dry matter intake was depressed when wet grains were fed at 20% of total dietary dry matter, milk production was similar to cows receiving the same dry matter from BDG. In the same report cows fed rewetted BDG at 20% of total dietary dry matter also showed depressed feed intakes, but similar milk production to the group receiving dried grains, suggesting that the moisture in wet grains could limit intake. This is supported by the findings of Davis et al. (1983) who observed a depression in dry matter intake by 1.5, 2.6 and 4.9 kg/cow/day when BWG containing 31% dry matter supplied 20, 30 or 40% of total dietary dry matter.

Murdock et al. (1981), using cows in the first 140 days of lactation, found no differences in milk production, milk composition, rumen volatile fatty acids or rumen pH when BWG (25.6% dry matter) replaced all the supplemental soybean meal and barley in corn silage-alfalfa hay based diets. Brewers wet grains comprised 15 and 30% of total dietary dry matter. The characteristic depression in intake with high levels of BWG in the diet was not observed in this study. However, with cows past peak lactation, Davis et al. (1983) observed depressions in both milk production and dry matter intake when BWG replaced soybean meal and comprised greater than 20% of total dietary dry matter in corn silage-based diets. Surprisingly, cows also lost weight on the diets supplemented with BWG. Weight losses were progressively greater with increasing levels of BWG. No change in body weight was observed for cows on soybean meal supplemented diets. Using cows in a similar stage of lactation, Santos et al. (1984) demonstrated greater absorption of total amino acids from BWG diets than on soybean meal diets. Each test protein supplied approximately 50% of the total dietary protein in corn silage-alfalfa hay based diets. Although milk production was not measured, the results of this study and those reported by Murdock et al. (1981) and Davis et al. (1983) suggest that the feeding of a protein source that is relatively undegradable in the rumen will not always improve milk production.

Beef Cattle and Sheep

Unlike distillers wet grains, there is limited information available on the feeding value of BWG for beef cattle and sheep. The study reported by Linton (1977), comparing the relative feeding value of fresh BWG to stored (untreated) BWG in diets for growing steers, showed no significant differences in cumulative feed intake, weight gains or feed efficiency when fresh or stored grains comprised 25 or 50% of total dietary dry matter. However, the tendency was for better animal performance with the lower level of BWG. Except for a vitamin-mineral supplement (0.23 kg/head/day), corn silage was the only other ingredient in the diets fed.

Available evidence on the feeding value of BWG for sheep is lacking. However, in the report by Satter and Whitlow (1977), sheep were used as models to determine amino acid flow to the intestine from diets in which the major portion of dietary test protein (nitrogen) was provided by Starea (commercial product composed primarily of urea and starch), soybean meal, dried brewers grains or wet brewers grains. The diets were formulated to be isofermentable and to allow adequate ammonia production for maximum microbial growth rates. Results from the study revealed that a larger portion of the brewers grain diets were digested post-ruminally, with the dried grains higher than the wet grains. The flow of amino acids to the intestine was about 30% higher with the brewers grains than other diets. Similar results have been reported by Santos _et al_. (1984) with growing steers.

Summary

Both distillers and brewers wet grains have desirable characteristics that make them suitable components in ruminant rations. However, a practical concern with their utilization is rapid molding and spoilage after production, especially during exposure to air and in warm weather. In order to prolong their keeping quality until they can be effectively incorporated into ruminant diets, several additives such as organic acids, carbohydrates and lactic acid bacteria have been added to wet grains. The effectiveness of organic acids in reducing or preventing spoilage was shown to vary with the level of application and the mixtures used. Carbohydrates and lactic acid bacteria were less effective than organic acids in preventing spoilage.

There is limited information on the relative feeding value of distillers wet grains in diets of dairy cattle. However, available information from beef cattle and sheep studies suggest an equal feeding value to soybean meal when a combination of urea and distillers wet grains supplied the supplemental nitrogen.

Unlike distillers wet grains, few studies have been reported on the relative feeding value of brewers wet grains for beef cattle and sheep. With dairy cattle, some studies indicate an equal feeding value to soybean meal and dried brewers grains, while others indicate a depression in animal performance when fed at more than 20% of dietary dry matter.

Our studies were designed to test various methods of preserving distillers and brewers wet grains, and to determine their feeding value for dairy cows resulting from these methods.

MATERIALS AND METHODS

Distillers Wet Grains

Experiment I. Aerobic Storage

Fresh distillers wet grains (31.2% dry matter), provided by an ethanol producing plant in Michigan, was apportioned into four-50 kg batches to facilitate mixing and application of treatments in a stainless steel feed mixer equipped with a horizontal ribbon mixer. Each batch was treated on a dry matter basis with 0, 1.57, 3.14 or 4.71% ammonia (NH_3) equivalent from ammonium hydroxide (29.4% NH_3). After each treatment, temperatures of the grains were taken with a 20.32cm. temperature probe and samples representative of day zero were frozen immediately. Thereafter, duplicate samples (5.5 kg each) were compacted in double-lined polyethylene bags and exposed to aerobic storage at 15.6, 26.7 or 37.8^{o}C for 4, 9 and 14 days. For the low and intermediate temperatures, constant temperatures were maintained in egg-holding rooms, and a forced-draft incubator was used for the high temperature storage.

Stability Measurements

Changes in temperature and appearance of visible mold in the stored grains were monitored daily. Changes in pH, dry matter, total nitrogen, water soluble nitrogen and ammonia nitrogen were also determined as measures of stability.

To faciliate calculation of dry matter recovery, bags were weighed before and after removal of samples for laboratory analyses. All samples were frozen at - 10°C until analyzed.

Laboratory Analyses

Samples were thawed and ground before analysis in a Hobart chopper. Dry matter was determined on 40g portions of the ground samples by drying in a forced-air oven at 65°C for 48 hours. Aliquots of the ground samples were also used for the determination of total nitrogen by macro-Kjeldahl. Water soluble nitrogen was determined by macro-Kjeldahl on the supernatant after 15g of the ground sample was homogenized with 100 ml distilled water for 2 min. in a Sorvall Omnimixer, strained through four layers of cheesecloth and centrifuged at 27,000x g for 10 min. Ammonia nitrogen was determined on the supernatant by methods described by Chaney and Marbach (1965). The pH was determined on the water extract before centrifugation by using a standard glass electrode connected to a pH meter.

Statistical Analysis

Statistical analysis of the data was according to procedures described by Genstat V (Lawes Agricultural Trust, 1980) and involved partitioning of the main effects and interactions in the analysis of variance into orthogonal polynomial contrasts.

Experiment II. Lactation Trial

Prior to treatment, 20 lactating Holstein cows were fed a complete ration (15% crude protein) consisting of corn silage, alfalfa hay, high moisture ear corn, soybean meal and a vitamin-mineral mix for 14 days. Cows were subsequently blocked for pretreatment milk yield and randomly assigned to one of two treatments for 70 days. Pretreatment milk and milk composition data are in Table 2. At the start of the 70 day trial, cows averaged 79 days in lactation.

Treatments consisted of furnishing the protein supplement in a typical midwestern ration for lactating cows as soybean meal or ammonia (NH_3)-treated distillers wet grains (Table 3). Diets were formulated to be isonitrogenous (15% CP) and isocaloric (1.65 Mcal/kg dry matter). Distillers wet grains was supplied biweekly from the same source as in Experiment 1. Upon arrival, the wet grains were treated on a dry matter basis with 4-5% ammonia equivalent from aqua-ammonia (25% NH_3) mixed in a feed truck equipped with scales and horizontal ribbon mixers, and stored on the concrete floor of a hay barn.

Diets were fed twice daily with a weekly adjustment of the amounts fed to allow for 5-10% refusal. Feeds and feed ingredients were sampled three times per week, and weekly composites frozen at - 10°C until analyzed. Feed intakes and milk production were monitored daily, and

Table 2 . Pre-treatment milk and milk composition data

Variable	Diets[a] SBM	Diets[a] DWG	SE
Milk production (kg/d)			
Actual	30.3	29.7	6.49
Fat-corrected	27.1	27.5	5.78
Solids-corrected	27.7	28.6	6.31
Milk composition (%)			
Fat	3.33	3.52	.390
Protein	2.90	2.97	.181
Solids-not-fat	8.96	9.25	.833

[a]Cows were subsequently placed on diets supplemented with normal soybean meal (SBM) or ammoniated distillers wet grains (DWG).

Table 3 . Ingredient composition of experimental diets

Ingredients	SBM	DWG
	--------% of DM--------	
Corn silage	34.89	36.30
Haylage	9.98	10.38
Alfalfa hay	7.47	7.77
High moisture ear corn	35.55	29.67
Distillers' wet grains (DWG)[a]	---	13.78
Soybean meal (SBM)	10.12	---
Mineral-vitamin mix[b]	2.0	2.10

[a]Treated on a dry matter basis with 4-5% ammonia.
[b]Calcium carbonate, calcium sulfate, dicalcium phosphate, trace mineralized salt and vitamins A (100,000 IU/day) and D (50,000 IU/day).

a daily composite milk sample (AM and PM) was taken for composition analysis biweekly. Cows were weighed for 2 consecutive days during the second and last week of treatment to determine changes in body weight.

Laboratory Analyses

pH, dry matter and total nitrogen determinations of feeds and feed ingredients were as in Experiment 1. Acid detergent fiber (ADF), acid detergent insoluble nitrogen (ADIN) and neutral detergent fiber (NDF) were analyzed according to methods described by Goering and Van Soest (1970). An alpha-amylase preparation was used during NDF analysis to remove interfering carbohydrates (Robertson and Van Soest, 1981). Milk samples were analyzed for fat, protein, lactose and total solids by infrared analysis (A.O.A.C., 1975).

Statistical Analysis

Data were analyzed as a randomized complete block design utilizing Genstat V (Lawes Agricultural Trust, 1980). Treatment means were adjusted by covariance analysis for pretreatment measurements.

Brewers Wet Grains

The brewers wet grains (BWG) used in the following experiments were supplied weekly by Murphy Products Company, Inc., Detroit Michigan.

Experiment Ia. Aerobic and Anaerobic Storage

Upon arrival, fresh BWG (22% dry matter) was apportioned into three-59 kg batches to facilitate mixing and application of treatments in a stainless-steel feed mixer (as used in the DWG stability study). Temperatures of the wet grains were recorded before and immediately after each treatment using the afore-mentioned temperature probes. Each batch was treated on a dry matter basis with 0, 2 or 4% ammonia (NH_3) equivalent from ammonium hydroxide (29.4% NH_3). After each treatment, samples respresentative of day zero were frozen immediately to - $5^{o}C$.

For the aerobic study, approximately 3 kg portions of the treated material were compacted in 3 l plastic pails that were exposed to air at room temperature for 3, 7 or 10 days. Temperature probes were positioned at the 1.5 l level in each pail to monitor daily changes in temperature of the treated grains. Appearance of visible mold was also monitored on a daily basis.

For the anaerobic study, approximately 5.5 kg portions of the treated material were compacted in double-lined polyethylene bags, sealed with twine, packed into a 100 l garbage container and left to ensile at room temperature for 7, 14 or 28 days. In both studies, 2 replicates were made for each treatment and period of storage.

At the specified time intervals, replicates were removed from storage and analyzed immediately for pH, dry matter and total nitrogen. Water soluble extracts were also prepared. For the aerobic samples, all surface mold was discarded and the remaining portion was thoroughly mixed before sampling.

Laboratory Analyses

Determination of pH, dry matter and nitrogen fractions were as described for the DWG experiments. Water soluble carbohydrates and lactic acid were determined on the supernatant (as prepared for water soluble nitrogen) according to methods described by Dubois et al. (1956) and Barker and Summerson (1941), respectively. Concentrations of volatile fatty acids in the samples were determined on the supernatant by gas chromatography. Prior to injection, 1 ml of supernatant was acidified with 0.2 ml of 88% formic acid. Injection volume was 2.5 ul. The column was glass (76.2 cm long with an internal deameter of 4 mm) packed with carbopack C and maintained at 120^{o}C. Inlet port and detector temperatures were maintained at 200^{o}C. A hydrogen flame ionization detector was used, and nitrogen (50 ml/min) was the carrier gas. Peak areas were measured via an electronic integrator and concentrations in samples calculated by comparing to peak areas of standard mixtures of volatile fatty acids.

Statistical Analysis

Data were analyzed as a 2-factor completely randomized design (Gill, 1978a) with the partitioning of the main effects and interactions of the analysis of variance into orthogonal polynomial contrasts using Genstat V (Lawes Agricultural Trust, 1980).

Experiment IIa. Lactation Trial

Thirty-six Holstein cows in early to mid-lactation were originally fed a pretreatment standardization diet (15% crude protein) consisting of corn silage, haylage, high moisture ear corn, soybean meal and a vitamin-mineral premix in a total mixed ration for 2 weeks. Cows were subsequently blocked for pretreatment milk production and randomly assigned to one of four treatments. However, due to the rapid decline in milk production in the first week of treatment for cows on BWG diets, all cows were restandardized for an additional 2 weeks. During this period, fresh and ensiled BWG plus urea were gradually increased in three pretreatment diets (Table 4) which were fed in sequential order for 7, 3 and 4 days. Cows were subsequently blocked for pretreatment milk production (latter 2 weeks) and randomly assigned to one of four treatments for 70 days. Pretreatment milk and milk composition data are in Table 5.

To the basal diet consisting of corn silage, alfalfa hay, high moisture ear corn and vitamin-mineral premix (Table 4) one of the following protein supplements was

Table 4 . Ingredient composition of pre-treatment and experimental diets

Ingredients	Pre-treatment[a]			Experimental[b]			
	(7)	(3)	(4)	SBM	FBWG	EBWG	FBWGU
	% of DM						
Corn silage	25.0	25.0	24.9	24.95	24.69	24.85	25.36
Alfalfa hay	25.0	25.0	24.9	24.78	25.67	25.71	23.63
High moisture ear corn	34.3	35.6	34.1	34.57	22.72	21.85	33.87
Soybean meal	14.3	9.5	7.1	14.31	-----	-----	-----
Fresh brewers wet grains	----	3.0	3.4	-----	25.60	-----	14.65
Ensiled brewers wet grains	----	----	3.7	-----	-----	26.26	-----
Urea	----	.39	.39	-----	-----	-----	.72
Mineral-vitamin mix[c]	1.49	1.59	1:58	1.40	1.32	1.34	2.24

[a]Numbers in parenthesis represent days on diet during pre-treatment.
[b]Soybean meal (SBM), fresh or ensiled brewers wet grains (FBWG, EBWG) and fresh brewers wet grains plus urea (FBWGU).
[c]Dicalcium phosphate, limestone, magnesium oxide, potassium sulfate, trace mineralized salt and vitamins A (100,000 IU/day), D (50,000 IU/day) and E (100 IU/day).

Table 5 . Stage of lactation, milk yield and milk composition of cows on pre-treatment

Item	Diets[a]				
	SBM	FBWG	EBWG	FBWGU	SE
Stage of lactation (Days)	113	141	133	146	63.097
Milk (kg/day)					
Actual	30.85	31.25	30.91	30.55	5.982
Fat-corrected	28.06	29.25	27.25	27.82	5.651
Solids-corrected	29.01	29.86	28.11	28.54	5.717
Composition (%)					
Fat	3.42	3.57	3.21	3.39	.386
Protein	3.49	3.46	3.45	3.49	.225
Lactose	5.02	5.01	5.00	4.97	.268
Solids-not-fat	9.10	9.03	9.01	9.02	.320

[a]Cows were subsequently allotted to experimental diets supplemented with soybean meal (SBM), fresh brewers wet grains (FBWG), ensiled brewers wet grains (EBWG) or fresh brewers wet grains plus urea (FBWGU).

added at the percents listed: soybean meal (SBM), 14.3%; fresh brewers wet grains (FBWG), 25.6%; ensiled brewers wet grains (EBWG), 26.3%; or 14.7% FBWG plus .72% urea (FBWGU). Diets were formulated to be isonitrogenous (15.5% CP) and isocaloric (1.6 Mcal/kg dry matter).

There were two sources of ensiled brewers wet grains. One was ensiled in a fabricated-box structure and fermented for 3 months before feeding. The other was ensiled in a commercial plastic bag silo (Ag-bag) and fermented for 1 month before feeding. Most of the EBWG in the box was used up by the second week of the experimental period. The FBWG was delivered weekly and stored on the concrete floor of a horizontal silo.

Diets were fed twice daily, and the amounts fed adjusted weekly to allow 5-10% refusal. Feed intakes and milk yields were monitored daily. A daily composite milk sample (AM and PM) was taken biweekly for analysis of fat protein, lactose and total solids. Cows were weighed on 2 consecutive days during the second and last week of the experimental period to determine changes in body weight. Complete diets were sampled every other day and weekly composites frozen until analyzed. Fresh and ensiled brewers wet grains were also sampled on alternate days and analyzed individually rather than as composites in order to determine day to day variation in composition associated with exposure to air. Other feed ingredients were sampled biweekly and monthly composites frozen until analyzed.

Laboratory Analyses

Feeds, feed ingredients and milk samples were analyzed as previously described. Ash was determined on air dried samples by standard A.O.A.C. (1984) methods. Rumen undegradable dietary nitrogen was determined on air dried feed samples according to procedures described by Poos et al. (1979), which employed a proteolytic enzyme degradation system.

Statistical Analysis

Animal performance data were analyzed as a split-block repeat-measure design (Gill, 1978b) using Genstat V (Lawes Agricultural Trust, 1980). Treatment means for milk production and milk composition were adjusted by covariance analysis for stage of lactation and other related pretreatment measurements. Bonferroni contrasts (Gill, 1978a) were used to compare differences between dietary means, and a combination of Bonferroni and orthogonal polynomial contrasts (Gill, 1978a) to compare differences in response when there were significant interactions between diets and periods.

RESULTS AND DISCUSSION

Distillers Wet Grains

Experiment I. Stability During Aerobic Storage

Table 6 shows temperature changes of distillers wet grains (DWG) as affected by increasing levels of added ammonia and days of aerobic exposure at three storage temperatures. Changes in temperature in response to the imposed variables were not consistent within or across storage temperatures. Heating, however, as evidenced by increases above storage temperatures, was greater for untreated and low ammonia-treated grains than for those treated with intermediate or high ammonia, especially at the high temperature. For untreated, low, intermediate and high ammonia treatments, peak increases above the high storage temperature averaged (^{o}C) 11.11, 6.66, 3.89 and 2.22, respectively. At intermediate and low storage temperatures, peak increases were less than 10^{o}C, suggesting that heating is not independent of storage temperatures. However, the fact that increases in temperature were decreased with increasing levels of added ammonia suggests that heating could also be due to aerobic microbial metabolism. Other studies have reported similar results for ammonia-treated DWG stored under aerobic conditions

Table 6 . Temperature of distillers wet grains as affected by added levels of ammonia (NH_3) and days of exposure at different storage temperatures

		Storage Temperatures (^oC)								
		15.6			26.7			37.8		
Added NH_3(% DM)[a]/Day[b]	0	4	9	14	4	9	14	4	9	14
0	22.22	16.67	16.12	24.44	24.17	35.83	30.00	40.07	48.89	39.72
1.57	21.11	14.31	22.78	20.56	26.95	33.33	31.11	44.44	38.89	38.89
3.14	20.20	14.44	14.44	19.16	22.78	32.22	29.45	37.50	41.67	36.95
4.71	20.20	14.58	15.00	16.12	23.33	24.72	28.62	37.78	40.00	37.78
Contrasts[c]:			LxL	QdxL	LxQd	CxL	QdxQd	LxC		
Significance:	^oC 15.6		.01	NS[d]	.01	NS	.05	NS		
	26.7		NS	NS	.01	NS	NS	.01		
	37.8		.05	.01	.01	.01	NS	.01		

[ab] Significant interaction for specified storage temperatures, $P < .01$; respective SE are .047, .056 and .101.
[c] Linear (L), quadratic (Qd) and cubic (C) effects of the interaction.
[d] Not significant, $P > .05$.

(Waller et al., 1982; Huber et al., 1983; Diallo et al., 1984). Increased storage temperatures resulted in a general decrease in days to equal or exceed that specified temperature, which were 14, 9 and 4 for low, intermediate and high storage temperatures, respectively.

Changes in appearance of the stored grains during aerobic exposure were not tabulated. However, grains treated with intermediate or high ammonia showed no visible mold growth during aerobic storage at any of the specified temperatures, probably due to the anti-fungal action of ammonia (Bothast et al., 1973; Britt and Huber, 1975; Huber et al., 1979b). In contrast, substantial mold growth during storage was observed for untreated and low ammonia-treated grains, especially at intermediate and high storage temperatures. Similar observations were reported for DWG treated, on a dry matter basis, with less than 3% ammonia (Waller et al., 1982), suggesting that low levels of ammonia might serve as a nitrogen source for fungal growth, since many fungi including most Penicillia and Aspergilli utilize ammonium salts as a nitrogen source (Bothast et al., 1973). Days until appearance of visible mold on the untreated and low ammonia treated grains also decreased with increasing storage temperatures. Average days at low, intermediate and high temperatures for untreated and low ammonia treatments were 10, 11; 7, 6; and 4, 4, respectively.

Dry matter recoveries of DWG as affected by ammonia treatments and storage temperatures after 14 days of aerobic storage are shown in Table 7. As expected, dry matter recoveries decreased with increasing storage temperatures, probably because of dependent temperatures increases in the stored grains (Table 6) resulting in enhanced aerobic microbial activity (Mo and Fyrileiv, 1979). The similarity in recoveries for untreated grains at intermediate and high temperatures cannot be readily explained. However, recoveries were higher with increased ammonia. The improved recoveries with intermediate and high ammonia was probably due to the anti-fungal action of ammonia in suppressing aerobic degradation and consequent heating, as evidenced in Table 6. The relationship between dry matter recovery and the combined effects of increasing ammonia additions and storage temperatures can best be described as being linear by linear, linear by quadratic or quadratic by quadratic.

Ammonia additions to the wet grains resulted in substantially higher initial pH levels than in the untreated grains (Table 8). However, with the exception of the untreated grains, decreases in pH to day 14 were slow and also inversely related to level of ammonia at intermediate and high temperature. This inverse relationship was probably due to the increased efficacy of ammonia at higher concentrations in inhibiting aerobic microbial activity (Bothast et al., 1973; Britt and Huber, 1975) shown to be associated with increases in pH (Mo and Fyrileiv, 1979). For the untreated grains, pH by day 14 at

Table 7 . Dry matter recovery of distillers wet grains as affected by ammonia (NH_3) treatment and storage temperature after 14 days of aerobic storage

	Temp (^oC)[b]	Added NH_3 (% DM)[a]			
		0	1.57	3.14	4.71
		--------------% of initial DM--------------			
	15.6	96.78	97.12	100.00	100.00
	26.7	76.79	86.35	96.72	99.65
	37.8	76.83	78.87	83.80	89.13
Contrasts[c]:	LxL	QdxL	LxQd	CxL	QdxQd
Significance:	.01	NS[d]	.01	NS	.05

[ab]Significant interaction ($P < .01$); SE is 2.154.
[c]Linear (L), quadratic (Qd) and cubic (C) effects of the interaction.
[d]Not significant, $P > .05$.

Table 8 . pH of distillers wet grains as affected by added levels of ammonia (NH_3) and days of exposure at different storage temperatures

		Storage Temperatures (°C)								
		15.6			26.7			37.8		
Added NH_3(% DM)[a]/Day[b]	0	4	9	14	4	9	14	4	9	14
0	4.15	5.50	5.34	4.73	5.30	5.24	7.86	5.70	6.88	7.87
1.57	9.43	9.13	8.58	8.45	8.48	8.98	9.00	8.87	9.20	8.98
3.14	10.20	9.65	9.40	8.89	9.50	8.83	9.05	8.98	8.88	9.18
4.71	10.12	9.95	9.70	9.38	9.95	9.25	8.93	8.93	9.32	8.96
Contrasts[c]:			LxL	QdxL	LxQd	CxL	QdxQd	LxC		
Significance:	°C									
	15.6		.01	.01	.01	.05	.01	.01		
	26.7		.01	.01	.01	.01	.01	.01		
	37.8		.01	.01	.01	.01	.01	.01		

[ab]Significant interaction for specified storage temperatures, $P < .05$; respective SE are .047, .056 and .101.
[c]Linear (L), quadratic (Qd) and cubic (C) effects of the interaction.

low, intermediate and high storage temperatures were 0.58, 3.71 and 3.72 units higher than the initial level of 4.15, which is not surprising in view of the observed temperature increases (Table 6) and the substantial mold growth at intermediate and high storage temperatures. Even with the low pH of 4.73 by day 14 at the low storage temperature, mold growth was not inhibited in untreated grains.

Apart from the initial increases in total nitrogen from ammonia additions at day 0, changes in total nitrogen during aerobic exposure appeared to be influenced mainly by storage temperatures (Table 9). With the untreated grains, total nitrogen increased from 4.07% (of the dry matter) at day 0 to 4.39, 5.23 and 5.83% at day 14 for low, intermediate and high storage temperatures, respectively, probably due to loss of CO_2 during storage. A similar response was observed at low and intermediate storage temperatures for low ammonia-treated grains, but at high temperature total nitrogen decreased from 5.10% at day 0 to 4.19% by day 14, probably due to volatilization of added nitrogen (Fox and Fenderson, 1978). Unlike untreated and low ammonia (except at high temperature)-treated grains, total nitrogen (at day 14) decreased with increasing storage temperatures for grains treated with intermediate or high ammonia, with decreases greater for high than for intermediate ammonia. However, the fact that decreases did not result in concentrations less than 4.07% (total nitrogen of untreated grains at day 0) suggests that the

Table 9 . Total nitrogen in distillers wet grains as affected by added levels of ammonia (NH_3) and days of exposure at different storage temperatures

Added		Storage Temperatures (°C)								
		15.6			26.7			37.8		
NH_3(% DM)[a]/Day[b]	0	4	9	14	4	9	14	4	9	14
	-----------	-----------	-----------	-----------	% of DM	-----------	-----------	-----------	-----------	-----------
0	4.07	3.83	4.26	4.39	3.98	4.45	5.23	4.43	5.11	5.83
1.57	5.10	5.22	5.07	5.25	4.51	5.07	5.67	4.49	4.56	4.19
3.14	6.03	5.55	5.64	5.53	4.94	5.40	5.57	5.35	4.52	4.88
4.71	6.84	6.06	5.90	5.78	6.00	5.51	5.61	5.29	5.16	5.52
Contrasts[c]:			LxL	QdxL	LxQd	CxL	QdxQd	LxC		
Significance:	°C									
	15.6		.01	NS[d]	NS	NS	NS	NS		
	26.7		.01	NS	NS	NS	NS	NS		
	37.8		.01	.01	.01	NS	NS	NS		

[ab]Significant interaction for specified storage temperatures, $P < .01$; respective SE are .168, .240 and .224.
[c]Linear (L), quadratic (Qd) and cubic (C) effects of the interaction.
[d]Not significant, $P > .05$.

decreases were due to volatilization of some of the added nitrogen.

As expected, water soluble nitrogen (WSN) increased with ammonia additions (Table 10). However, during aerobic exposure, increases to day 14 were greater for untreated and low ammonia-grains than for grains treated with intermediate or high ammonia. At low, intermediate and high storage temperatures, WSN increased 2.3-, 21.4- and 24.8 fold for the untreated grains, whereas with low ammonia, concentrations increased 2.8-, 3.5- and 2.4 fold. With intermediate ammonia treatment, concentrations of WSN were lower than the initial levels at low and intermediate temperature, but increased 1.18 fold during exposure at the high temperature. Unlike the other treatments, concentrations of WSN for high ammonia were never higher than initial levels, irrespective of storage temperatures, thus indicating decreased proteolysis with increasing ammonia additions. Similar results were reported for ammonia-treated corn silage (Johnson et al., 1982). The tendency for increases in WSN with intermediate and high ammonia during aerobic exposure was probably due to lower nitrogen retention by unfermented material (Huber et al., 1979a).

Ammonia nitrogen (NH_3-N) concentrations also increased with ammonia additions (Table 11). However, for untreated grains, no NH_3-N (except for trace amounts which were less than 0.001% of the dry matter) was detected at the low storage temperature, but with increasing temperatures

Table 10 . Water soluble nitrogen in distillers wet grains as affected by added levels of ammonia (NH_3) and days of exposure at different storage temperatures

Added		Storage Temperatures (°C)								
		15.6			26.7			37.8		
NH_3(% DM)[a]/Day[b]	0	4	9	14	4	9	14	4	9	14
	% of DM									
0	0.08	0.05	0.05	0.18	0.05	0.21	1.71	0.24	0.96	1.98
1.57	0.83	0.69	0.88	2.31	0.69	1.43	2.89	0.96	1.63	1.97
3.14	1.85	1.08	1.23	1.42	1.30	1.14	1.52	1.08	1.10	2.18
4.71	1.97	1.56	1.61	1.67	1.66	1.39	1.53	1.40	1.17	1.89
Contrasts[c]:			LxL	QdxL	LxQd	CxL	QdxQd	LxC		
Significance:	°C									
	15.6		.01	.01	NS[d]	.01	.01	.01		
	26.7		.01	.01	.01	.01	NS	NS		
	37.8		.01	NS	.01	NS	NS	.01		

[ab]Significant interaction for specified temperatures, $P < .01$; respective SE are .045, .097 and .082.
[c]Linear (L), quadratic (Qd) and cubic (C) effects of the interaction.
[d]Not significant, $P > .05$.

Table 11 . Ammonia nitrogen in distillers wet grains as affected by added levels of ammonia (NH_3) and days of exposure at different storage temperatures

		Storage Temperatures (°C)								
Added		15.6			26.7			37.8		
NH_3(% DM)[a]/Day[b]	0	4	9	14	4	9	14	4	9	14
	% of DM									
0	0.00	0.00	0.00	0.00	0.00	0.05	0.59	0.08	0.27	0.57
1.57 (1.29)[c]	0.50	0.53	0.46	0.94	0.55	0.64	1.23	0.48	0.65	0.84
3.14 (2.58)	0.61	0.65	0.68	0.95	0.68	0.68	0.86	0.66	0.56	0.95
4.71 (3.87)	0.71	0.76	0.84	1.05	0.70	0.89	1.09	0.76	0.60	0.89
Contrasts[d]:			LxL	QdxL	LxQd	CxL	QdxQd	LxC		
Significance:	°C									
	15.6		.01	.01	.05	.01	.01	NS[e]		
	26.7		.01	NS	.01	.01	NS	.01		
	37.8		.01	NS	NS	.05	NS	.01		

[ab]Significant interaction for specified storage temperatures, P < .01; respective SE are .017, .021 and .020.
[c]Added NH_3-N (1.57 x .8225).
[d]Linear (L), quadratic (Qd) and cubic (C) effects of the interaction.
[e]Not significant, P > .05.

(storage), days to exceed initial concentrations were reduced from 9 to 4 days. In addition, although the rate of increase was greater at high than at the intermediate temperature, concentrations were about the same by day 14 (0.57 vs. 0.59% of the dry matter), indicating a catalytic effect of increasing temperatures on deamination activity. With the ammonia treatments, changes in WSN concentrations during aerobic exposure were less consistent across storage temperatures. However, based on the added levels of NH_3-N (1.29. 2.58 and 3.87%) rather than the levels of added ammonia (1.57, 3.14 and 4.71%), essentially all of the increases in NH_3-N for ammonia treatments could be accounted for by increased solubilization of added ammonia, assuming that some ammonia was incorporated into microbial protein and that a portion of such protein was solubilized through proteolysis (Huber et al., 1979a).

Summary

The results from this study have shown that treating fresh distillers wet grains (FDWG) with 3 to 4% (of the dry matter) ammonia not only improves the aerobic stability of the material during increasing ambient temperatures, but also retards heating, eliminates mold growth, improves dry matter recovery and reduces proteolysis and deamination over a two-week period. It was also shown that treating FDWG with 1.6% (of the dry matter) ammonia, results in an unstable material which deteriorated faster than untreated grains.

Experiment II. Lactation Trial

Treating the fresh DWG with 4-5% (of the dry matter) ammonia resulted in adequate preservation of the wet grains during bi-weekly deliveries. No mold growth or extensive temperature increases above ambient temperatures (10-27^{o}C) were observed in the treated grains during storage and feeding out. Dry matter was reduced from 31 to 28% with ammonia treatment, but crude protein was increased from 27 to 39.5% (range was 37-42%) of the dry matter.

Table 12 shows the chemical composition of the diets (Table 3) that were fed during the experimental period. The DWG diet was lower in dry matter, but higher in the detergent fractions (acid detergent insoluble nitrogen, acid detergent fiber and neutral detergent fiber) than the soybean meal (SBM) diet. The greater proportions of corn silage and haylage together with the ammoniated DWG (Table 3) could account for these differences. Differences in crude protein, estimated net energy of lactation (NE_L) and pH between the diets were small.

Dry matter intakes, though not significantly different between groups, tended to be lower for cows fed the DWG diet (Table 13). This was probably due to the higher moisture content and/or the higher detergent fractions of the total diet. Actual, fat and solids corrected milk with their corresponding efficiencies tended to be higher for cows fed the DWG diet, but due to the large associated standard errors, no significant differences were detected

Table 12. Chemical composition of the experimental diets

Item	Diets[a] SBM	DWG
Dry matter (%)	46.1	39.4
Crude protein (% of DM)	14.5	14.4
Acid detergent insol. N (% of total N)	5.9	11.4
Acid detergent fiber (% of DM)	21.5	23.4
Neutral detergent fiber (% of DM)	35.7	42.3
Estimated NE_L (Mcal/kg DM)[b]	1.61	1.64
pH	5.1	5.4

[a]Soybean meal (SBM) and ammoniated-distillers wet grains (DWG).

[b]Net energy of lactation (NE_L) was calculated from Telplan Program 31 Form 3 (MSU, 1981).

Table 13. Dry matter intake, milk yields and persistencies of cows fed the experimental diets

Variable	Diets[a] SBM	DWG	SE	Effect
Dry matter intake (kg/d)	19.4	17.6	3.74	NS[d]
Milk yields (kg/d)[b]				
Actual	27.1	27.9	1.30	NS
Fat-corrected	23.8	24.8	1.10	NS
Solids-corrected	23.9	24.7	1.49	NS
Persistencies (%)[c]				
Actual	90.9	94.3	5.61	NS
Fat-corrected	88.0	91.4	4.69	NS
Solids-corrected	85.8	88.5	6.46	NS

[a]Soybean meal (SBM) and ammoniated distillers wet grains (DWG).
[b]Adjusted by covariance analysis.
[c](Treatment milk/pre-treatment milk)x 100.
[d]Not significant ($P > .05$).

between treatment groups. Similar results were reported by Schingoethe et al. (1983) for cows (averaging 84 days postpartum) fed DWG or SBM supplemented diets.

Milk composition, feed efficiency and average daily gains of cows fed the experimental diets are in Table 14. Milk fat, protein and solids-not-fat were not significantly different between treatment groups. The tendency, however, for lower milk protein for cows on the DWG diet was probably due to a greater proportion of the dietary nitrogen being tied up as acid detergent insoluble nitrogen (Table 12). Similar results were reported for distillers dried grains and extruded and heat-treated soybean meal (Kung et al., 1983; Satter and Stehr, 1984), all of which undergo some degree of heating. However, cows fed the DWG diet were more efficient ($P < .05$) in converting feed to milk. This improved feed efficiency was probably due to more efficient utilization of the nutrients in the DWG diet than those in the SBM diet, since there were no significant treatment differences in average daily gains.

Table 15 shows the profitability of the experimental diets. The costs of the various feed ingredients used in the diets are representative of the prices paid by farmers in Michigan in 1985. Based on these prices and actual milk yield and feed intake data, lower feed costs and greater income over feed costs were realized when ammoniated DWG supplied the supplemental protein to basal diets. Both DWG and SBM supplied approximately 40% of the dietary protein.

Table 14. Milk composition, feed efficiency and average daily gains of cows fed the experimental diets

Variable	Diets[a] SBM	DWG	SE	Effect
Milk composition (%)[b]				
Fat	3.19	3.27	.166	NS[c]
Protein	3.17	3.07	.102	NS
Solids-not-fat	8.55	8.63	.537	NS
Feed efficiency				
(kg 4%-FCM/kg DM)	1.25	1.42	.139	.05
Average daily gain (kg/d)	.41	.42	.251	NS

[a] Soybean meal (SBM) and ammoniated distillers wet grains (DWG).
[b] Adjusted by covariance analysis.
[c] Not significant ($P > .05$).

Table 15. Economic evaluation of the experimental diets

Item	Diets[a]	
	SBM	DWG
Milk yield (kg/day)[a]	27.1	27.9
Milk income ($/cow/day)[b]	7.17	7.38
Feed intake (kg DM/day)	19.38	17.56
Feed price ($/M ton DM)[c]	126.09	113.94
Feed cost ($/cow/day)	2.44	2.00
Income over feed costs ($/cow/day)	4.73	5.38

[a]Soybean meal (SBM) and ammoniated distillers wet grains (DWG).
[b]Adjusted by covariance analysis.
[c]Milk price @ $12/cwt or $0.2646¢/kg.
[d]Price of ration ingredients ($/ton): corn silage, 22; alfalfa hay, 80; corn 107 ($3.00/bushel); haylage, 35; soybean meal, 160; distillers wet grains, 30; anhydrous ammonia, 250; vitamin-mineral mixes, 300.

Summary

Ammonia treatment of fresh DWG (at 4-5% of the dry matter) provided adequate preservation of the wet grains during feeding out of bi-weekly deliveries. The treated grains (DWG) were equal to soybean meal in maintaining milk yields and milk composition of cows fed these supplements to supply approximately 40% of their dietary protein in total-mixed diets. Dry matter intakes on the DWG diet were 1.8 kg lower than on the SBM diet, but cows were more efficient in converting feed to milk. In addition, economic evaluation of the diets showed a greater return over feed costs when DWG supplied the supplemental protein to total-mixed diets consisting of corn silage, haylage, alfalfa hay, high-moisture ear corn and a vitamin-mineral mix.

Brewers Wet Grains

Experiment Ia.

Stability During Aerobic and Anaerobic Storage

An interaction ($P \leq .05$) between ammonia additions and days of aerobic or anaerobic (ensiling) storage was indicated by the analysis of variance for each response variable in this study, with the exception of dry matter (anaerobic only - Table 17) and acetic acid (aerobic and anaerobic - Table 21) for which main effect means are presented in addition to the treatment combination means.

Changes in temperature and dry matter of brewers wet grains (BWG) during aerobic storage are shown in Table 16.

Ammonia treatment of the wet grains (at day 0) resulted in an immediate decrease of the initial temperature (52.22 vs. 41.11 and 41.67°C). Since the initial temperature of the wet grains is a residual effect of the brewing process, it is theorized that the mechanism of action of ammonia in decreasing this initial temperature might be through a cooling effect of ammonia on the wet grains, rather than the anti-fungal action of ammonia (Bothast et al., 1973; Britt and Huber, 1975) in suppressing aerobic microbial activity and thus preventing temperature increases. However, though inconsistent, temperatures decreased during storage with less drastic differences between treatments, probably due to adequate compaction of the wet grains in the plastic containers. Ambient temperatures fluctuated between 22 and 29°C.

Initial dry matter concentrations of BWG appeared less affected by ammonia additions (Table 16), probably due to volatilization of some of the added ammonia during dry matter determination by oven drying (Fox and Fenderson, 1978). However during aerobic exposure, dry matter decreases (% of initial dry matter) to day 10 averaged 3.7, 4.3 and 1.7% for untreated, low and high ammonia treatments.

Mold growth and spoilage data for BWG during aerobic storage are not tabulated. However, these were evident only in the untreated and low ammonia-treated grains. Visible mold was evident in the untreated grains at day 6,

Table 16. Changes in temperature and dry matter of brewers wet grains as affected by ammonia (NH_3) treatment and days of aerobic exposure

Added		Temperature(°C)				Dry matter(%)			
NH_3(% DM)[a]/Days[b]		0	3	7	10	0	3	7	10
0		52.22	23.71	29.30	27.22	21.36	21.79	21.06	20.58
2		41.11	23.24	27.78	25.84	21.28	21.84	21.08	20.37
4		41.67	22.59	27.23	26.66	21.27	21.55	21.51	20.90
Contrasts[c]:				LxL	QdxL	LxQd	QdxQd	LxC	QdxC
Significance:	Temperature			.01	.01	.01	.01	.01	.01
	Dry matter			.01	.05	NS[d]	NS	.05	NS

[ab]Significant interaction between added levels of ammonia and days of aerobic exposure - $P < .01$ and .05 for temperature and dry matter, respectively; respective SE are .373 and .138.
[c]Linear (L), quadratic (Qd) and cubic (C) effects of the interaction (levels of NH_3 x days).
[d]Not significant - $P > .05$.

and in the low ammonia-treated grains at day 9. Of the 18 cm of compacted grains, spoilage by day 10 averaged 2.5, 1.3 and 0 cm from the surface for untreated, low and high ammonia-treated grains. The delayed spoilage with increased ammonia was due to the anti-fungal action of ammonia. Bothast et al. (1973) reported that both external and infecting molds and yeast were eliminated in high moisture corn treated with .5 or 2% of the dry matter as ammonia.

Changes in dry matter concentrations of BWG during ensiling are shown in Table 17. Dry matter concentrations of the wet grains were more stable during ensiling than when subjected to aerobic exposure. This difference could be attributed to the decreased oxygen tension in the ensiled grains. Zimmer (1971) demonstrated an inverse relationship between dry matter losses associated with the storage of high moisture material and the degree of air-tightness achieved. However, despite the improved stability under anaerobic (ensiling) conditions, low ammonia addition resulted in decreased dry matter concentrations when compared to untreated and high ammonia treatments. Since the interaction between ammonia treatments and days of aerobic exposure was not significant, the relationship between dry matter changes and ammonia additions can best be described as being quadratic ($P < .01$). Day effect was not significant, as indicated by the contrasts.

Table 18 shows the pH changes of BWG during aerobic

Table 17. Dry matter of brewers wet grains as affected by ammonia (NH_3) treatment and days of ensiling

Added NH_3(% DM)[a]/Days[b]		0	7	14	28	NH_3 effect[c]
		%				
0		21.36	21.47	21.66	21.05	21.39
2		21.28	21.10	20.84	20.16	20.85
4		21.27	21.20	21.35	21.55	21.34
Day effect[d]:		21.31	21.26	21.29	20.92	
Contrasts[e]:			L	Qd	C	
Significance:	NH_3 effect		NS	.01	-	
	Day effect		NS[f]	NS	NS	

[ab]Interaction was not significant - $P > .05$; SE is .307.
[cd]Respective SE are .154 and .177.
[e]Linear (L), quadratic (Qd) and cubic (C) effects of NH_3 levels or day effect.
[f]Not significant - $P > .05$.

storage and ensiling. As was expected, ammonia treatments increased initial pH values (5.10 vs. 9.69 and 10.03, for untreated, low and high ammonia treatments), but resulted in delayed decreases in pH during aerobic storage and ensiling. Under aerobic storage, initial pH of untreated grains decreased to a low of 4.07 by day 7, followed by a slow increase to 4.40 by day 10. These changes were probably associated with the production and destruction of organic acids. With the ammonia treated grains, pH values by day 10 were still alkaline (8.20 and 9.62 for low and high ammonia treatments), presumably due to the higher initial pH and restricted fermentation. For the ensiled grains, pH values for the untreated grains decreased to a low of 3.68 within the first week of ensiling, followed by a slow increase to 4.46 by day 28. The low pH of 3.68 suggests that fermentation was complete by the first week of ensiling; whereas, the increased pH thereafter could be associated with clostridial activity (Mo and Fyrileiv, 1979). For the ammonia treatments, pH decreases were greater with low than with high ammonia; average decreases from days 0 to 28 were 4.11 and 0.35 pH units, respectively, indicating greater fermentative activity with low ammonia. The pH values for the untreated ensiled grains are in agreement with those reported by Allen and Stevenson (1975) for untreated BWG ensiled in laboratory silos.

Ammonia treatments resulted in delayed increases in

Table 18. pH of brewers wet grains as affected by ammonia (NH_3) treatment and days of aerobic exposure or ensiling

Added			Aerobic			Ensiled	
NH_3(% DM)[a]/Days[b]	0	3	7	10	7	14	28
	--------	--------	--------	% of DM	--------	--------	--------
0	5.10	4.27	4.02	4.40	3.68	4.16	4.46
2	9.69	9.80	9.10	8.20	9.12	6.81	5.58
4	10.03	10.20	9.83	9.62	9.95	9.88	9.68
Contrasts[c]:		LxL	QdxL	LxQd	QdxQd	LxC	QdxC
Significance:	Aerobic	NS[d]	.01	.01	.01	NS	NS
	Ensiled	NS	.01	.01	.01	.01	.01

[ab]Significant interaction between added levels of ammonia and days of aerobic exposure or ensiling - $P < .01$;respective SE are .087 and .127.

[c]Linear (L), quadratic (Qd) and cubic (C) effects of the interaction (levels of NH_3 x days.

[d]Not significant - $P > .05$.

lactic acid production (Table 19). Lactic acid concentrations at day 0 for untreated, low and high ammonia-treated grains were: 1.02, 0.51 and 0.66% of the dry matter. The reason for the lower concentrations in the ammonia-treated grains is not apparent. However, during aerobic storage lactic acid production peaked at day 7, with concentrations greater for the untreated than the ammonia-treated grains (3.43 vs. 0.70 and 0.83%), probably due to the lower pH levels in the untreated grains (Table 18). At the end of the aerobic storage period (day 10), lactic acid concentrations were 52, 70 and 68% of the levels observed at day 7 for untreated, low and high ammonia treatments, suggesting greater stability for ammonia treatments under aerobic exposure. During ensiling, lactic acid production also peaked at day 7 with concentrations of lactic acid higher in the untreated than in the ammonia treated grains (3.53 vs. 0.76 and 0.70%). This difference is also related to the more favorable pH levels in the untreated grains (Table 18) for lactic acid producing bacteria. However, with increasing days of ensiling lactic acid decreased, with concentrations at day 28 averaging 8.2, 14.5 and 71.4% of the levels observed at day 7, again suggesting greater stability for ammonia treatments during extended periods of ensiling (or anaerobic storage). The lower lactic acid levels with ammonia treatment are characteristic of ammonia additions in excess of 1% of the dry matter (Britt and Huber, 1975; Huber _et al_., 1979a; Johnson _et al_, 1982).

Table 19. Lactic acid in brewers wet grains as affected by ammonia (NH_3) treatment and days of aerobic exposure or ensiling

Added			Aerobic			Ensiled		
NH_3(% DM)[a]/Days[b]		0	3	7	10	7	14	28
		---	---	---	% of DM	---	---	---
0		1.02	2.26	3.43	1.79	3.53	0.64	0.29
2		0.51	0.55	0.70	0.49	0.76	0.69	0.11
4		0.66	0.79	0.83	0.57	0.70	0.52	0.50
Contrasts[c]:			LxL	QdxL	LxQd	QdxQd	LxC	QdxC
Significance:	Aerobic		.01	.01	.01	.01	.01	.05
	Ensiled		.05	NS[d]	.01	NS	.01	.01

[ab]Significant interaction between added levels of ammonia and days of aerobic exposure or ensiling - $P < .01$; respective SE are .118 and .410.
[c]Linear (L), quadratic (Qd) and cubic (C) effects of the interaction (levels of NH_3 x days).
[d]Not significant - $P > .05$.

Residual water soluble carbohydrates (WSC) concentrations were higher in the ammonia-treated than the untreated grains (Table 20). Average concentrations at day 0 for untreated, low and high ammonia treatments were: 6.39, 8.42 and 7.19% of the dry matter. The increased concentrations with ammonia treatments were probably due to the action of ammonia in hydrolyzing hemicellulose (Harbes _et al._, 1982). During aerobic storage, WSC concentrations decreased in untreated and low ammonia-treated grains, but increased in grains treated with high ammonia probably due to decreased utilization for lactic acid production coupled with the residual action of ammonia in hydrolyzing hemicellulose. The rapid decrease in concentration of WSC for low ammonia treatment between days 7 and 10 is surprising, since there was no corresponding increase in lactic acid production (Table 19), which suggests that the rapid decrease could be due to the production of other organic acids (Whittenbury _et al._, 1967), or to utilization by aerobic microbes (Mo and Fyrileiv, 1979). During ensiling, WSC concentrations in the untreated grains were lower than the initial concentration, probably due to partial utilization for lactic acid production. However, as was observed during aerobic storage, there was a tendency for small increases in WSC in the untreated grains, which was probably due to the slow hydrolysis of hemicelluose by organic acids at the lower pH (Woolford, 1972; Ohyama and Masaki, 1977). For low

Table 20. Residual water soluble carbohydrates in brewers wet grains as affected by ammonia (NH_3) treatment and days of aerobic exposure or ensiling

Added NH_3 (% DM)[a]/Days[b]		0	Aerobic 3	Aerobic 7	Aerobic 10	Ensiled 7	Ensiled 14	Ensiled 28
		% of DM						
0		6.39	1.90	2.06	2.24	1.04	1.07	1.21
2		8.42	7.11	7.16	3.81	7.77	4.84	2.67
4		7.19	7.37	9.82	10.07	9.76	9.75	7.86
Contrasts[c]:			LxL	QdxL	LxQd	QdxQd	LxC	QdxC
Significance:	Aerobic		.01	.01	.01	.01	NS[d]	NS
	Ensiled		.01	.01	.01	.01	.01	.01

[ab] Significant interaction between added levels of ammonia and days of aerobic exposure or ensiling - $P < .01$; respective SE are .222 and .179.

[c] Linear (L), quadratic (Qd) and cubic (C) effects of the interaction (levels of NH_3 x days).

[d] Not significant - $P > .05$.

ammonia-treated grains, WSC concentrations decreased with increasing days of ensiling, which is indeed surprising in view of the decreasing concentrations of lactic acid with increasing days of ensiling (Table 19). The pH decline (Table 18) between days 14 and 28 was optimum for lactic acid producing bacteria, which suggests that lactic acid was produced, but was converted to other organic acids by clostridial microbes (Mo and Fyrileiv, 1979). For the grains treated with high ammonia, WSC concentrations increased to day 7 (7.19 to 9.76%), then showed a slight tendency to decrease with increasing days of ensiling.

Acetic acid concentrations were generally higher during ensiling than during aerobic storage (Table 21). The interactions between ammonia treatments and days of aerobic storage or ensiling were not significant. However, during aerobic storage, acetic acid production increased, with concentrations greater in the ammonia-treated than in the untreated grains. The reason for the greater concentrations with ammonia treatments is not apparent, but it can be speculated that there was probably greater stimulation of the homofermentative acetic acid bacteria (Mo and Fyrileiv, 1979) which are known to produce acetic acid from ethanol in the presence of oxygen. Ethanol production was not measured in our study, but its presence in BWG was confirmed by Oleas (1977). For the ensiled grains, acetic acid production also increased with increasing days of ensiling, but concentrations were greater for untreated and low

Table 21. Acetic acid in brewers wet grains as affected by ammonia (NH_3) treatment and days of aerobic exposure or ensiling

Added NH_3 (% DM)[a]/Days[b]	0	Aerobic 3	Aerobic 7	Aerobic 10	NH_3 effect[c]	Ensiled 7	Ensiled 14	Ensiled 28	NH_3 effect[d]
	----------	----------	----------	----------	% of DM	----------	----------	----------	----------
0	0.82	0.92	1.12	1.63	1.12	1.03	2.42	2.58	1.72
2	1.13	1.33	1.54	1.68	1.42	1.78	1.89	2.48	1.82
4	1.08	1.18	1.25	1.65	1.29	1.27	1.71	1.90	1.49
Day effect[e]:	1.01	1.14	1.30	1.65		1.36	2.01	2.32	
Contrasts[f]:			L	Qd	C		L	Qd	C
Significance: NH_3 effect			.05	.01	-		NS[g]	NS	-
Day effect			.01	.05	NS		.01	NS	NS

[ab] Interaction between added levels of ammonia and days of aerobic exposure or ensiling was not significant - $P > .05$; respective SE are .115 and .306.
[cd] Respective SE are .058 and .153.
[e] SE are .067 and .177 for aerobic and ensiled day effect, respectively.
[f] Linear (L), quadratic (Qd) and cubic (C) effects of NH_3 levels or day effect.
[g] Not significant - $P > .05$.

ammonia treated grains than for grains treated with high ammonia. This difference was probably due to the restricted fermentation with high ammonia treatment, since acetic acid production is usually looked upon as a normal process during ensiling (Whittenbury et al., 1967).

Except for butyric and acetic acids, no other volatile fatty acids were detected in any of the sampled grains. Butyric acid values are not tabulated. However, under aerobic storage, butyric acid was detected at day 10 only in the untreated grains; the concentration was 0.16% of the dry matter. For the ensiled grains, butyric acid was detected only for the low ammonia treatment. At day 14, concentrations averaged 0.33% of the dry matter, but increased to 4.18% by day 28. The fact that lactic acid (Table 19) and WSC (Table 20) concentrations decreased drastically during the same time interval, suggests that they were utilized by saccharolytic clostridia (Whittenbury et al., 1967; Mo and Fyrileiv, 1979), thus resulting in the production of butyric acid.

Changes in total nitrogen (N) concentrations during aerobic storage and ensiling are shown in Table 22. As was expected, initial concentrations were increased with ammonia additions; average concentrations were 4.41, 5.45 and 6.65% of the dry matter for untreated, low and high ammonia treatments. During aerobic storage, average N concentrations by day 10 were 10.7, 12.5 and 5.6% greater than initial concentrations at day 0. These differences could be attributed to

Table 22. Total nitrogen in brewers wet grains as affected by ammonia (NH_3) treatment and days of aerobic exposure or ensiling

Added NH_3(% DM)[a]/Days[b]		0	Aerobic 3	Aerobic 7	Aerobic 10	Ensiled 7	Ensiled 14	Ensiled 28
		--------	--------	--------	% of DM	--------	--------	--------
0		4.41	4.53	4.67	4.88	4.51	4.66	4.86
2		5.45	5.63	5.78	6.13	5.80	6.04	6.62
4		6.65	6.73	6.54	7.02	6.30	6.89	6.94
Contrasts[c]:			LxL	QdxL	LxQd	QdxQd	LxC	QdxC
Significance:	Aerobic		NS[d]	.05	NS	NS	.05	NS
	Ensiled		NS	.01	NS	NS	.05	.05

[ab]Significant interaction between added levels of ammonia and days of aerobic exposure or ensiling - $P < .01$; respective SE are .086 and .113.
[c]Linear (L), quadratic (Qd) and cubic (C) effects of the interaction (levels of NH_3 x days).
[d]Not significant - $P > .05$.

the dry matter losses as reflected in the decreases in dry matter percent (Table 16). For the ensiled grains, total nitrogen also increased during ensiling. By day 28, average concentrations for untreated, low and high ammonia treatments were 10.2, 21.5 and 4.4% greater than initial concentrations. The greater increase in total nitrogen concentration for low ammonia-treated grains was probably due to the increased production of butyric acid which often results in large losses of dry matter (Mo and Fyrileiv, 1979).

Water soluble nitrogen (WSN) concentrations also increased initially with ammonia additions (Table 23). Initial concentrations for untreated, low and high ammonia treatments were 3.28, 33.09 and 46.42% of total nitrogen. During aerobic storage and ensiling WSN concentrations increased in the untreated grains, suggesting increased proteolysis of the grain protein. Average WSN concentrations by day 10 and 28 for aerobic storage and ensiling, respectively, were 18.9 and 71.6% greater than the initial concentration. For the ammonia-treated grains, there was no consistent change in WSN concentrations during either storage system, the reasons for which are not apparent.

Ammonia additions resulted in initial increases in ammonia nitrogen (NH_3-N) concentrations (Table 24). For untreated, low and high ammonia, 0.79, 7.79 and 8.50% of total nitrogen (or .04, .42 and .56% of the dry matter) was NH_3-N. During aerobic storage, NH_3-N increased with concentrations by day 10 averaging 3.48, 17.22 and 21.01%

Table 23. Water soluble nitrogen in brewers wet grains as affected by ammonia (NH_3) treatment and days of aerobic exposure or ensiling

Added NH_3 (% DM)[a]/Days[b]		0	Aerobic 3	Aerobic 7	Aerobic 10	Ensiled 7	Ensiled 14	Ensiled 28
		--------	--------	--------	% of total N	--------	--------	--------
0		3.28	2.43	3.63	3.90	4.10	4.94	5.63
2		33.09	31.26	33.57	28.25	31.38	29.40	38.15
4		46.42	43.85	44.57	44.09	50.00	45.73	48.58
Contrasts[c]:			LxL	QdxL	LxQd	QdxQd	LxC	QdxC
Significance:	Aerobic		.05	.01	NS	.01	NS	.05
	Ensiled		NS[d]	NS	NS	.01	.05	NS

[ab] Significant interaction between added levels of ammonia and days of aerobic exposure or ensiling - $P < .01$; respective SE are .802 and 1.252.
[c] Linear (L), quadratic (Qd) and cubic (C) effects of the interaction (levels of NH_3 x days).
[d] Not significant - $P > .05$.

Table 24. Ammonia nitrogen in brewers wet grains as affected by ammonia (NH_3) treatment and days of aerobic exposure or ensiling

Added			Aerobic			Ensiled		
NH_3 (% DM)[a]/Days[b]		0	3	7	10	7	14	28
		-------	-------	-------	% of total N	-------	-------	-------
0		0.79	0.44	1.27	3.48	0.56	0.65	1.74
2		7.79	9.77	18.08	17.22	17.92	24.67	25.23
4		8.50	10.25	23.78	21.01	16.43	19.31	25.82
Contrasts[c]:			LxL	QdxL	LxQd	QdxQd	LxC	QdxC
Significance:	Aerobic		.01	NS[d]	.01	NS	.01	NS
	Ensiled		.01	.01	NS	.01	.05	.05

[ab]Significant interaction between added levels of ammonia and days of aerobic exposure or ensiling - $P < .01$; respective SE are .860 and .656.

[c]Linear (L), quadratic (Qd) and cubic (C) effect of the interaction (levels of NH_3 x days).

[d]Not significant - $P > .05$.

of total nitrogen (or .17, 1.05 and 1.48% of the dry matter). During ensiling, NH_3-N also increased with concentrations by day 28 averaging 1.74, 25.23 and 25.82% of total nitrogen (or .08, 1.67 and 1.79% of the dry matter). Based on the levels of added NH_3-N (0, 1.65 and 3.29%) rather than added NH_3 (0, 2 and 4%), it is suggested that the increases with time in NH_3-N with high ammonia treatment were due to solubilization of added N which was initially bound in the water insoluble form (Huber et al., 1979a). For the low ammonia treatment, it seems reasonable to conclude that the increases in NH_3-N were due to solubilization of added N plus deamination of amino acids (Johnson et al., 1982). Whereas, with the untreated grains, the increases in NH_3-N could only be due to deamination of amino acids. Moreover, this activity appeared to be greater during aerobic storage than during ensiling, probably because of a slower decline in pH during aerobic exposure (Table 18).

At the end of the ensiling period, untreated and low ammonia-treated grains had a bleached-like appearance and a strong acid smell. Whereas, with the high ammonia-treated grains, there was still the brown appearance (from ammonia treatment) and a strong ammonia odor.

Summary

The results obtained in this study, indicate that the mechanism influencing the stability of BWG during aerobic and anaerobic storage is complex and not determined by a single factor. However, greater quality retention was shown for anaerobic (ensiling) than aerobic stroage. Also, the high ammonia treatment (4% of the DM) was most effective in preserving the quality of the wet grains during aerobic and anaerobic storage.

Experiment IIa. Lactation Trial

In the following tables, the abbreviations SBM, FBWG, EBWG and FBWGU will be used to designate the diets supplemented with soybean meal, fresh brewers wet grains, ensiled brewers wet grains and fresh brewers wet grains plus urea, respectively.

Table 25 shows the average chemical composition of diets fed during the experiment. Dry matter content of the brewers wet grains (BWG) diets were generally lower than the SBM diet. Also, acid detergent fiber was higher in FBWG and EBWG diets than in SBM and FBWGU diets. These differences were due to the relative proportions of BWG added to the diets (Table 4). Diets were formulated to be isonitrogenous (15.5% CP); however, crude protein (CP) analysis for the respective diets were 0.77, 0.13, 0.39 and 0.27 units lower than intended. In addition, a greater proportion of the dietary protein (N x 6.25) was acid detergent insoluble nitrogen in the BWG diets than the SBM diet.

Chemical composition of fresh and ensiled BWG fed during the experiment is in Table 26. Mean values were similar for fresh and ensiled grains. However, the increase in

Table 25. Chemical composition of experimental diets

Item	Diets[a]			
	SBM	FBWG	EBWG	FBWGU
Dry matter (%)	60.0	45.97	45.08	49.75
Acid detergent fiber (% DM)	23.21	26.74	26.33	23.78
Crude protein (% DM)	14.73	15.37	15.11	15.23
ADIN (% total nitrogen)[b]	3.5	8.7	10.6	6.7
ASH (% DM)	6.31	7.20	6.54	6.66
Net energy (Mcal/kg DM)[c]	1.63	1.58	1.57	1.58

[a]Normal soybean meal (SBM), fresh brewers wet grains (FBWG), ensiled brewers wet grains (EBWG) and fresh brewers wet grains plus urea (FBWGU).
[b]Acid detergent insoluble nitrogen.
[c]Net energy of lactation was calculated from Telplan Program 31 Form 3 (MSU, 1981).

Table 26. Chemical composition of fresh and ensiled brewers wet grains during the experimental period[a]

Item	Brewers wet grains			
	Fresh	SD	Ensiled[b]	SD
pH	4.79	.47	4.17	.24
	-------------------% of DM-------------------			
Dry matter	26.50	1.33	27.00	.94
Nitrogen	5.05	.36	5.02	.35
Acid detergent fiber	29.06	2.09	27.05	2.29
Acid detergent insoluble N	.70	.13	.71	.13
ASH	4.79	.42	4.97	.56
Ammonia nitrogen	.047	.036	.028	.011
Lactic acid	2.58	1.57	3.42	2.05
Water soluble carbohydrates	3.15	2.50	1.41	.57

[a]Seventy days.
[b]Ensiled in a commercial plastic bag silo for 31 days.

lactic acid, and the decrease in pH and soluble carbohydrates indicate that some fermentation occurred during storage. The pH and ammonia nitrogen values of the ensiled grains were within accepted critical limits (a pH of less than or equal to 4.2 and NH_3-N less than or equal to 8% of total N) suggested for satisfactory preservation (Carpintero et al., 1969).

Percent undegraded (in vitro) and water soluble nitrogen of the diets fed during the experiment are shown in Table 27. Undegraded nitrogen at zero hours, which corresponds to the solubilization of the more rapidly degradable fraction of the feed protein, was highest for FBWG (69.5%) and least for FBWGU (55.8%). These differences could be accounted for by the relative proportions of water-soluble nitrogen (% of total N) in the respective diets. However, after 1 hour of incubation, which is closely related to the relative rate of degradation in the rumen (Poos et al., 1979), undegraded nitrogen values were 43.2, 40.3, 33.8 and 28.6% of the total N for EBWG, FBWG, FBWGU and SBM diets, respectively. These values are in agreement with in vivo estimates of undegradable protein in diets supplemented with soybean meal or brewers wet grains (Santos et al., 1984).

Analysis of variance for dry matter intakes indicated an interaction ($P < .01$) for diet by period. In view of this, period means for the different treatments are presented

Table 27. Undegraded and water soluble nitrogen in the experimental diets

Items	Undegraded nitrogen[a]			Water-soluble nitrogen
	0	.5	1	
	% of total N			
Soybean meal	62.79	37.64	28.56	25.33
Standard deviation (SD)	3.52	2.45	.54	2.50
Fresh brewers wet grains	69.45	48.46	40.28	24.24
SD	5.04	.31	1.20	3.78
Fresh brewers wet grains + urea	55.76	39.41	33.77	35.29
SD	4.54	1.16	.60	1.81
Ensiled brewers wet grains	65.99	50.95	43.16	23.86
SD	3.77	.70	.99	1.99

[a]Determined by Ficin assay at 0, .5 and 1 hr incubation times (Poos _et al_, 1979).

(Table 28). Dry matter intakes were consistently higher for cows fed the SBM diet. This trend was reflected by a greater difference in linearity ($P < .005$) when SBM was compared with FBWG + FBWGU or with EBWG. The higher intakes on FBWGU than the FBWG diet (though not significant) probably resulted from replacing approximately 45% of the fresh brewers wet grains with its crude protein equivalent from urea and its energy from corn (Table 4). The relatively lower intakes on the EBWG diet probably resulted from more of the dietary protein being tied up as acid detergent insoluble nitrogen (Table 25), coupled with the low level of water soluble nitrogen in the diet (Table 27). When the unavailable protein (ADIN) was subtracted from the total protein, the remaining dietary protein for EBWG was 13.5% of the dry matter which was below recommended requirements (NRC, 1978). Although dry matter intakes were relatively lower on the brewers wet grains diets, others (Murdock et al., 1981) have reported similar intakes for cows fed brewers wet grains at 15 or 30% of dietary dry matter. However, depressed intakes were reported for cows fed greater than 30% of dietary dry matter as brewers wet grains (Davis et al., 1983).

Table 29 shows milk yields, persistencies and milk composition of cows fed the experimental diets. The interaction for diet by period was not significant, thus only treatment means are presented. There were no significant

Table 28. Dry matter intake of cows fed the experimental diets

Diets[a]	Period (weeks)[b]				
	1	2	3	4	5
	kg/Day				
Soybean meal (D1)	25.08	24.69	24.80	25.18	26.39
Fresh brewers wet grains (D2)	23.23	21.67	21.76	21.70	21.19
Ensiled brewers wet grains (D3)	22.24	21.32	20.73	19.66	20.02
Fresh brewers wet grains+urea (D4)	24.55	23.11	22.74	22.49	20.84
Contrasts[c]	L	Qd	C	Qr	
D2 vs. D4	NS[d]	NS	NS	NS	
D1* vs. D2+D4	.005	NS	NS	NS	
D2 vs. D3	NS	NS	NS	NS	
D1* vs. D3	.005	NS	NS	NS	

[ab]Significant interaction between diets and periods, $P < .01$; SE is 1.42.
[c]Linear (L), quadratic (Qd), cubic (C) and quartic effects of periods.
[d]Not significant, $P > .05$.
*Greater difference in linearity.

Table 29. Milk yields, persistencies and milk composition of cows fed the different protein supplements[a]

Item	Diets[b]				SE
	SBM	FBWG	EBWG	FBWGU	
Milk (kg/day)[c]					
Actual	29.28	29.37	27.72	29.00	2.20
Fat-corrected	26.16	25.57	25.56	25.95	2.56
Solids-corrected	26.18	25.67	25.36	25.88	2.45
Persistency[d]					
Actual	95.6	94.6	88.5	93.3	8.1
Fat-corrected	94.4	90.6	89.8	92.5	9.7
Solids-corrected	92.1	88.4	86.5	89.3	9.1
Composition (%)[c]					
Fat	3.33	3.17	3.39	3.33	.295
Protein	3.27	3.20	3.23	3.16	.12
Lactose	4.71	4.71	4.64	4.74	.13
Solids-not-fat	8.64	8.58	8.55	8.55	.22

[a] Interaction between diet and period was not significant, $P > .05$.
[b] Soybean meal (SBM), fresh or ensiled brewers wet grains (FBWG, EBWG) and fresh brewers wet grains plus urea (FBWGU); contrasts FBWG vs. FBWGU-NS; SBM vs. FBWG + FBWGU-NS; FBWG vs. EBWG-NS; SBM vs. EBWG-NS. NS - not significant, $P > .05$.
[c] Adjusted by covariance analysis.
[d] Persistency = (treatment milk/pre-treatment milk) x 100.

treatment differences in yields of milk, 4% fat-corrected milk (FCM) and solids-corrected milk (SBM). Also, treatment differences for persistencies and milk composition were not significant.

Body weight gains and feed efficiencies are shown in Table 30. The interaction for diet by period was not significant ($P > .05$), thus only treatment means are presented. Body weight gains and feed efficiencies (milk/kg DM intake) were numerically greater for the FBWG and EBWG than the other diets, but differences were not significant for any of the contrasts (shown in the foot-notes). The reason for the improved feed efficiencies for cows fed the BWG diets is not apparent. However, since the cows were weighed only during the second and last week of the experiment, any losses in body weight in support of milk production could have been negated by greater gains (in body weight) between the second and last week of the experiment. Davis et al. (1983) reported improved feed efficiency for cows fed brewers wet grains diets, but the apparent increase in feed efficiency, compared to cows fed the soybean meal diet, was accounted for by losses in body weight.

The relative profitability of the different protein supplements is presented in Table 31. Income over feed costs was lowest for SBM and greatest for FBWGU. The reason for the higher profit on the brewers grains diets compared to SBM, was the lower cost of protein supplement and the

Table 30. Body weight gains and feed efficiencies of cows fed the different protein supplements

Item	Diet[a]				
	SBM	FBWG	EBWG	FBWGU	SE
Initial body weight (kg)	613	640	612	608	----
Body weight gain (kg/day)[b]	.070	.276	.129	.036	.081
Feed efficiency[c]					
Fat-corrected milk (kg)[b]	1.06	1.21	1.19	1.13	.21
Solids-corrected milk (kg)[b]	1.07	1.21	1.18	1.12	.22

[a]Soybean meal (SBM), fresh brewers wet grains (FBWG), ensiled brewers wet grains (EBWG) and fresh brewers wet grains plus urea (FBWGU).

[b]Contrasts: FBWG vs. FBWGU-NS; SBM vs. FBWG + FBWGU-NS; FBWG vs. EBWG-NS; SBM vs. EBWG-NS. NS - not significant ($P > .05$).

[c]Feed efficiency = milk/dry matter intake (kg).

Table 31. Influence of protein supplement on income over feed costs of experimental diets

Item	Diets			
	SBM	FBWG	EBWG	FBWGU
Milk yield (kg/day)[a]	29.28	29.37	27.72	29.00
Milk income ($/cow/day)[b]	7.75	7.77	7.33	7.67
Feed intake (kg DM/day)	25.23	21.91	20.79	22.75
Feed price ($/M Ton DM)[c]	122.09	116.62	106.69	95.04
Feed cost ($/cow/day)	3.08	2.56	2.43	2.16
Income over feed costs ($/cow/day)	4.67	5.21	4.90	5.51

[a]Adjusted for pre-treatment milk yields.
[b]Milk price @ $12/cwt or $0.2646¢/kg.
[c]Price of ration ingredients ($/ton): corn silage, 22; alfalfa hay, 80; corn, 107 ($3.00/bushel); soybean meal, 160; fresh and ensiled brewers wet grains, 30; vitamin-mineral mixes, 300.

higher feed intake on the SBM diet with only small differences in milk yields. Even if intakes had been equal for all the diets, the SBM diet would have still been the least profitable. An additional savings was also realized when part of the fresh brewers grains was replaced with urea and ear corn. However, a different set of feed prices could have changed the relative rankings of the diets, but these were the prices the author felt as representative of the feeds in Central Michigan during 1985.

<u>Summary</u>

The results obtained in this study indicate that the forms of brewers wet grains studied were equal as replacements for soybean meal in diets for lactating dairy cows. In addition, greater income over feed cost were realized on the brewers wet grains than on the soybean meal diet.

GENERAL SUMMARY

The results obtained from the DWG stability study indicated that the wet grains can be effectively preserved for 2 weeks or more when treated with ammonia at 3 to 4% of its dry matter. At these application rates, dry matter recovery was improved, mold growth and spoilage were inhibited and temperature increases above storage temperature were reduced. For untreated and low ammonia treated grains, mold growth and spoilage increased with increasing storage temperature. Based on the intensity of mold growth during storage, the average shelf life of untreated and low ammonia-treated grains was 11, 7 and less than 4 days at low (15.6°C), intermediate (26.7°C) and high (37.8°C) storage temperatures.

In the DWG lactation trial, ammonia-treated DWG (4-5% of the DM) were equal to soybean meal in maintaining milk yields and milk composition of cows fed these supplements to supply approximately 40% of the dietary protein in total mixed diets. Intakes were higher on the SBM diet, but cows fed DWG were more efficient in converting feed to milk. In addition, greater income over feed cost was realized on the DWG diet.

For the aerobic and anaerobic (ensiling) stability studies with BWG, the results obtained indicated that BWG can be effectively preserved during aerobic storage and ensiling when treated with 4% of its dry matter as ammonia. Visible mold growth was evident in the untreated grains at day 6 and in the low ammonia (2% of DM) grains at day 9, during aerobic storage. Subsurface spoilage was also greater for untreated than low ammonia grains during aerobic storage. At the end of the ensiling period, high ammonia grains retained its original color, but had a strong odor of ammonia; whereas, with untreated and low-ammonia treated grains, the original color had faded and there was a strong acid odor.

In the BWG lactation trial, the forms of brewers wet grains studied were equal to soybean meal in diets for lactating dairy cows. By reducing the amount of brewers wet grains fed, and replacing its crude protein equivalent with urea and its energy with corn, greater dry matter intakes were achieved. Ensiled brewers wet grains in the diets of lactating dairy cattle resulted in about 7% lower dry matter intakes than fresh brewers wet grains. A longer adaptation period to the diet probably might have improved intakes. However, ensiling can be practiced as a method for storage of brewers wet grains. In addition, incorporation of brewers wet grains into diets for lactating dairy cows should be a gradual process to allow cows to adjust to the new diets.

The fresh brewers grains were more profitable than soybean meal, and ensiled grains were intermediate. Use of urea with fresh brewers grains resulted in greatest income over feed costs, and appears to be a desirable alternative to soybean meal as a protein supplement for high producing dairy cows.

Future studies are needed to clarify the mechanism of action of ammonia in increasing the soluble carbohydrates in brewers wet grains during aerobic storage and ensiling. Such studies will be useful in evaluating the potential digestibility and energy value of the wet grains for ruminants.

APPENDICES

APPENDIX TABLE 1

Butyric acid in brewers wet grains as affected by ammonia treatment and days of aerobic exposure or ensiling[a]

Added			Aerobic			Ensiled		
NH_3(% DM)/Days		0	3	7	10	7	14	28
		---	---	---	% of DM	---	---	---
0	Rep. 1.[b]	0	0	0	0.15	0	0	0.13
	Rep. 2.	0	0	0	0.16	0	0	0.00
2	Rep. 1.	0	0	0	0	0	0.45	3.78
	Rep. 2.	0	0	0	0	0	0.21	4.58
4	Rep. 1.	0	0	0	0	0	0	0
	Rep. 2.	0	0	0	0	0	0	0

[a]Raw data.
[b]Two replicates (Rep.) per treatment.

APPENDIX TABLE 2

An example of an analysis of variance with orthogonal polynomial contrasts[a]

Variate = pH[b] Source of Variation	df	Sum of Squares	Mean Square	F-value	Significance of F
Treatment	2	124.75	62.37	3891.28	P <.01
Linear	1	122.27	122.27	7627.86	P <.01
Quadratic	1	2.48	2.48	154.70	P <.01
Day	3	10.07	3.36	209.32	P <.01
Linear	1	9.90	9.90	617.72	P <.01
Quadratic	1	0.15	0.15	9.48	P <.01
Cubic	1	0.01	0.01	0.76	NS[c]

Appendix Table 2 (cont'd)

Variate = pH Source of Variation	df	Sum of Squares	Mean Square	F-value	Significance of F
Trt. by Day	6	14.71	2.45	152.95	$P < .01$
Lin. by Lin.	1	0.004	0.004	0.27	NS
Quad. by Lin.	1	11.92	11.92	743.74	$P < .01$
Lin. by Quad.	1	0.85	0.85	53.09	$P < .01$
Quad. by Quad.	1	0.70	0.70	43.87	$P < .01$
Lin. by Cub.	1	0.19	0.19	12.04	$P < .01$
Deviations	1	1.03	1.03	64.65	$P < .01$
Residual	12	0.19	0.02		
TOTAL	23	149.724	6.51		

[a]Two-factor Completely Randomized Design.
[b]The pH of brewers wet grains.
[c]Not significant, $P > .05$.

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