UTILIZATION OF WASTE POTATO STARCH IN UREA CONTAINING LIQUID SUPPLEMENTS

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CHAPTER I

INTRODUCTION

In recent years, interest in liquid supplements has increased significantly in the formula feed industry. They allow the livestock feeder an acceptable method of incorporating non-protein nitrogen (NPN) into cattle rations. Liquid supplements offer diversified feeding applications which include: 1. mixing with a complete ration, 2. as a top dressing, and 3. free choice. It is usually easier to handle and store liquid than dry supplements.

Major problems with liquid supplements are product uniformity, overconsumption and stability. Many liquid supplements on the market today undergo a layering effect when stored for several days. This segregation is not always visually detectable and requires analytical techniques to measure its occurance. Product instability is a result of ingredients and additives in the formula breaking down during extended storage periods. During extreme cold weather, many liquid supplements require heated storage to protect them from freezing. Overconsumption is a concern of every livestock feeder when employing a free-choice feeding technique.

The potato chip industry is facing an immediate perplexing environmental problem of disposing of waste streams. Potato starch is one of the major streams that could be used in the liquid supplement industry as an energy source for ruminants.

The purpose of this research was to adapt a hydrothermal cooking process to produce a liquid supplement that would utilize the potato starch waste stream. Urea was used as the source of NPN. Investigations were

specifically designed to evaluate the effects of storage and of the hydrothermal processing of potato liquid supplements. Liquid supplements using potato starch waste and corn as the carbohydrate sources were compared with an *in vitro* fermentation technique and for animal acceptability.

CHAPTER II

LITERATURE REVIEW

Potato Starch Waste

The potato chip industry continues to increase its production in the United States. Potato chip processing starts with whole white potatoes that are washed, peeled, and sliced. These potato slices are washed with jets of high pressure water to remove starch and slivers from the cut surfaces. Following another washing, the slices are leached in hot water (71). This leaves the wash water containing a considerable amount of potato starch.

The first permanent water pollution act was issued in 1948. The next two decades saw several other acts and amendments passed. The most comprehensive program to date is known as the Federal Water Pollution Act Amendments of 1972. Laws, enforced by the Environmental Protection Agency, placed strict guidelines on the disposal of waste water from industrial plants (83).

Potato processing waste water has a high biochemical oxygen demand value, causing it to be subject to the regulations set by the Environmental Protection Agency. The Netherlands and Northern areas in the United States have investigated methods for land disposal of potato processing streams (22,27). The land disposal method of relieving the waste problem of the potato industry has been shown to be an economic detriment (22).

The potato starch has nutritive value and can be removed by allowing the starch to settle out, centrifugation, or by filtering the waste stream. The wash water could then be purified and reused. This type of recovery could supply the feed industry with a source of energy.

On a dry weight basis, potato tuber contains 65 to 80% starch. Table 1 shows ranges for a proximate analysis of white potatoes (67). The ranges

are due to differences in potato varieties and geographic location. The starch is calorically the most important nutritional component of potatoes. Table 2 shows the amylose and amylopectin content of white potatoes (26). The physical and chemical properties of this starch often dictate the operating conditions for processing. The nonstarchy polysaccharides, such as cellulose and pectic substances, occur as components of the cell wall (67).

Chemical and Physical Properties of Starch

Nutritionally, starch is of great importance to the feed industry as a source of energy. An abundance of starch can be found stored in green plants as minute granules (40). Starch is also stored in the caryopsis of cereal grains and the tuber material of potatoes. An understanding of the chemical and physical properties of starch granules can be an important factor in developing processing techniques applicable to the feed industry.

Starch exists in a proteinaceous matrix as granules in the underground stem of potatoes, or the endosperm of kernals, and differs according to origin. Granular size depends on the plant species and may range from .5 to 100 μ in diameter. The shape of the granule may be ellipsoidal, polygonal, or spherical depending upon origin. The rice granule is polygonal and varies from 3 to 8 microns. Corn has polygonal and spherical granules that range from 5 to 25 microns (28). The average size of the potato starch granule is 35 to 40 μ and ranges from 15 to 120 μ in diameter. The small granules are spherical, while the larger granules are oval and irregular (14,28).

Near the center of a starch granule, a hilum can be observed with a microscope. The hilum is believed to be the nucleus about which the granule has been enlarged by apposition (28). Starch granules exhibit birefringence as evidenced by the maltese cross shown under a polarized light (28,50).

TABLE 1. PROXIMATE ANALYSIS OF WHITE POTATOES[A]

	Range (% as is)	Average (% as is)
Water (% wet basis)	63.2-86.9	77.5
Total solids	13.1-36.8	22.5
Protein (N X 6.25)	.7-4.6	2.0
Fat	.02-.96	.1
Carbohydrate		
Total	13.3-30.53	19.4
Fiber (crude)	.17-3.48	.6
Ash	.44-1.9	1.0

[A]Schwimmer, S. and H. K. Burr. 1959. Structure and chemical composition of the potato tuber. In Potato Processing. Talburt, W. F. and O. Smith. (67).

TABLE 2. ANALYSIS OF POTATO STARCH[A]

Dry matter (%)	21.0
Specific gravity (g/cc)	1.082
Starch (%)	14.5
Amylose (%)	3.9
Amylose (% of starch)	27.0
Amylopectin (%)	10.6
Amylopectin (% of starch)	73.0

[A]Eipeson, W. E. and K. Paulus. 1973. Investigations on some chemical constituents of potatoes and their influence on the behavior during canning. Potato Research 16:270. (26).

Striation marks are concentrical lines that are arranged around the hilum. Striations develop during granular syntheses and indicate a rhythmic variation occurring during deposition of starch. A diurnal effect has been suggested as the cause for this rhythmic variation. However, potato starch striations are not the consequence of a diurnal effect, even though they are the most vividly expressed of the starches (29).

The chemical structure of starch is comprised of repeating glucose units that can be divided into two major fractions: a linear fraction (amylose), and a branched fraction (amylopectin) (28). The fractions were first separated using a physiochemical method (63). Fractionation methods using precipitation were later developed (63,66).

Methylation of starch, followed by acid hydrolysis, was later used to produce methyl esters of glucose. The structure of amylose was shown to be a straight chain polymer linked by α-1,4 glucosidal bonds (8). The methylated products of amylopectin indicated the presence of α-1,6 and α-1,4 glucosidal linkage, which allows for the branched structure (35).

The straight chain fraction of starch is not believed to vary extensively in length as a result of starch origin. Amylose contains several thousand glucopyranose molecules linked by α-1,4 bonds, which allows starch to attain various conformations in solution. Starch in solution, in the presence of a complexing agent, allows the chain to exist in a helical conformation by twisting of the α-1,4 bonds. This helical structure is thought to be responsible for the characteristic blue color with iodine (32,61).

Each glucose unit, except the terminal units of amylose, contain one primary and two secondary hydroxyl groups. The nonreducing end of the chain has one primary and three secondary hydroxyl groups. The reducing end has one aldehydic reducing group, one primary, and two secondary hydroxyl groups (29).

A colormetric determination, using congo red, can be used to determine the amylose content of starch. The affinity of the dye for carbohydrate is reduced by the presence of branching. The binding affinity of congo red for amylose seems to be independent of chain length (17).

The precision of determining the ratio of amylose to amylopectin can be increased by potentiometrically titrating free iodine in an iodine:potassium iodide solution of starch. Many starches are comprised of 75 to 85% amylopectin, however, values from 20 to 100% have been reported (23). Potato starch is approximately 73% of the branched chain carbohydrate (26).

The degree of branching in amylopectin depends on starch origin (34). Potato starch amylopectin has a relatively low degree of branching (58), containing only 4 to 5% α-1,6 bonds (23). The amylopectin chain length can vary greatly. The molecular weight is difficult to measure, but has been estimated as high as one million (29,32).

The amylopectin fraction of potato starch contains phosphate esters. One phosphate group occurs every 212 to 273 anhydroglucose units. On hydrolysis, potatoes produce a measurable amount of glucose-6-phosphate, indicating the phosphate is on the primary alcohol. The ability to separate amylopectin from amylose in potato starch by electrodialysis is probably because of these phosphate groups (86).

As the tuber grows, new granules are continuously formed. Most of the phosphate is incorporated into the starch during earlier stages of granule development. Phytic acid is incorporated in the starch in the latter stages of development. Therefore, larger granules, which are developed first, contain slightly less phosphate than smaller granules on a weight basis (7). The average size granule of 40 μ contains .06 to .09% phosphorus (31).

Amylolytic enzymes are used to hydrolyze starch molecules into oligosaccharides, maltose and glucose. Two major types of amylases that degrade

starch are α-amylase and β-amylase. The enzyme α-amylase is an endoenzyme (60) that splits the α-1,4 linkage of the starch chain at random, while the α-1,6 bonds go unaffected. The result of α-amylase action in a starch slurry is a decrease in viscosity, loss of iodine staining power, and an increase in reducing power. Beta-amylase cleaves the α-1,4 bonds in a stepwise fashion, producing maltose from the nonreducing end of the chain. The action stops three or four glucose units before an α-1,6 branch point is encountered. This leaves a residue of a high molecular weight dextrin intact (32).

The surface of cereal starch granules are readily attacked by bacterial α-amylase. This penetration of enzyme occurs at pits or fissures naturally occurring in starch granules (30,46). In contrast, the surface of potato starch granules is very resistant to the action of amylases. Enzyme hydrolysis of potato starch has shown a selective pattern in attacking the starch granules. There seems to be no preference as to size of the granule hydrolyzed first (46). Researchers have presented many theories on amylolytic degradation of potato starch.

A protective membrane, that differs in chemical composition from the internal granule, has been suggested by some investigators for the behavior of potato starch degradation. Microscopic observations have clearly shown that the outer membrane is nonexistent in cereal starches and probably does not exist in potato starch (7,45).

The rate of enzyme activity in a starch solution can be increased by raising the temperature of the solution. Extending the temperature from 30 to 50 C and doubling the concentration of bacterial α-amylase, potato starch was degraded at approximately the same rate as corn starch at the lower temperature and enzyme level. Extensive potato starch solubilization was noted before granular erosion occurred. The corn starch, treated at

the lower temperature and enzyme level, exhibited extensive fragmentation and granular erosion. These results suggest that the enzyme susceptibility to starch is not related to the swelling or solubilization patterns of the starch when heated in water (46).

Other studies by the same investigator (46) showed that the enzyme susceptibility of potato starch can be increased by drying without heat. Neither the x-ray defraction pattern of the starch, nor the type of crystallinity were shown to be related to rate of enzymatic attack. The conclusion drawn from this study (46) agreed with Badenhuizen (7) that pores or fissures exist in the cereal starches that are not found in tuber starch (25,46). These cavities possibly do not exist because of the high degree of hydration that allows the granule to be compact and the surface area to remain smooth (40,45). From these results, it was noted at an AACC meeting (6) that action of amylases on starch granules is directly related to the botanical source.

Native starch granules are relatively insoluble in cold water, however, as the temperature is increased, the phenomenon of gelatinization occurs. Gelatinization is best described as "the irreversible rupture of the native, secondary bond forces in the crystalline regions of the starch granule" (68). Gelatinization is responsible for physical, chemical, and biological changes in the properties of starch (70).

The temperature at which initial loss of birefringence occurs is the lower limit of the gelatinization temperature range. Microscopists identify the top of the range as complete gelatinization, which is identified by the loss of all birefringence from the starch granules (49). This range can vary from 8 to 10 C and differs according to starch origin (28). The gelatinization temperature range for corn is 62 to 70 C and is 56 to 67 C for potatoes (29).

When water is used as the solvent, the gelatinization temperature of starch granules can be lowered by the addition of an agent that breaks hydrogen bonding. Examples are salts, alkali and urea (28,68).

The swelling behavior of starch is controlled by the extent and kind of association within the granule. The degree of association is dependent on size, shape and ratio of amylose to amylopectin (44). Starch's affinity for water is attributed to its abundance of hydroxyl groups. At room temperature, water absorption first causes reversible swelling. With continued heating, hydrogen bonding holding the starch granule together weakens. Swelling tangentially to many times its original size, maximum hydration is finally reached (29). A low pressure area develops around the hilum causing the granule to rupture and collapse. This causes cleavage of secondary bonds between polymer molecules, therefore, allowing a rapid dispersion of granule fragments. The covalent bonds are not severed. At this point, the granule is much less crystalline, but not amorphous (68).

Potato starch exhibits an exceptionally high degree of swelling at low temperatures. This is explained by weak internal bonding and ionizable phosphate groups that display mutual repulsion. This indicates that potato starch should be easily fragmented (44). However, extensive swelling of potato starch is noted before extensive solubilization is observed (45). The effect is a sharper increase in viscosity and the reaching of higher peak viscosities with tuber starch (29,32). Consequently, potato starch develops a long cohesive paste. The cereal grains display two-stage restricted swelling and, when gelatinized, develop a short paste (44).

Retrogradation is the return of swollen, gelatinized or dissolved starch to an aggregated form (42). The alignment of linear molecules allows for the formation of hydrogen bonding. Therefore, amylopectin

shows little tendency to retrograde, mainly as a result of steric effect (76). Factors affecting retrogradation include: 1. pH, 2. presence of non-starch components in the medium, 3. concentration of amylose (76) and 4. origin of amylose (39,64).

Rate of retrogradation can be increased by higher starch concentrations and lowering the temperature. Only small amounts of water are necessary for this phenomenon to occur (28).

Retrograded starch is relatively insoluble in water and is resistant to enzyme hydrolysis. However, retrogradation can be partially prevented by enzymatic hydrolysis before allowing gelatinized starch to cool (76). Damaged starch is digested by amylase at a faster rate than native starch when equal enzyme concentrations are used (65).

Isolated starches are easier to determine the degree of starch gelatinization than with cooked grains. In heterogenous samples, such as cooked grains, total loss of birefringence cannot be considered complete gelatinization (68). Many methods through the years have been developed to measure starch damage. Those include: 1. the loss of birefringence under a polarized microscope (49), 2. staining with congo red (49,65), 3. proton nuclear magnetic resonance which can be used to also follow changes on the molecular level over a wide range of temperatures (42) and 4. numerous modifications of enzyme hydrolysis followed by quantitative measurement of reducing sugars (24,65,70,72).

Liquid Supplements and Their Ingredients

Liquid supplements are produced from a liquid vehicle (molasses, fermentation liquors or distillers solubles) mixed with a source of NPN, water and phosphoric acid. At times minerals, vitamins, antibiotics and other ingredients are included. The liquid product is termed a supplement because

it is not formulated as the sole ration (19,84). The livestock industry's primary purpose for feeding liquid supplements is to supply a source of nitrogen and energy to the animal (13).

A limited number of researchers have compared liquid and dry supplements. They have indicated if both contain the same essential nutrients, blended with urea, animal performance will be similar (41,85). Advantages of liquid supplements are diverse feeding applications, easy handling, superior source of phosphorus, and a palatable method of feeding urea. However, many problems exist in the liquid supplement industry. One major concern is overconsumption when wintering pasture becomes scarce. Also, the closing of liq-tanks is recommended when drinking water is not readily accessible. Many liquid supplements on the market today undergo a layering effect when stored for several days. Other disadvantages include equipment cost, heated storage, and metal corrosion (85).

High producing dairy cattle were fed a ration containing urea and corn silage. There was no advantage to liquid or dry supplements in animal performance. However, lower milk yields were noted when nitrogen was supplied completely from an NPN source. Another investigation showed no difference in milk yields, milk fat content, or feed intake when comparing the two forms of supplement (48).

Fattening cattle studies have not indicated a consistant advantage of dry over liquid protein supplements (5,11). The decision to feed liquid or dry supplements depends on economics and how they are incorporated into the livestock operation (41).

Molasses was first fed to animals over 100 years ago. Advantages of feeding molasses include increased palatibility of complete ration, a source of highly digestible energy, and reduced dustiness. A total ration

containing 10% molasses has been shown to improve animal performance. However, as a by-product, molasses is generally produced with poor quality control and, therefore, subject to batch variation (85).

The net energy value of molasses for milk production is 1.499 M cal/kg when fed at 10%, but only 0.508 M cal/kg when fed at 30% of the total ration. The lower net energy value for high molasses additions was attributed to greater energy requirements for metabolism (48).

The first limiting nutrient for rumen animals grazing forages in dry or winter seasons is protein. With protein being expensive because of competition with monogastric animals, researchers have turned to investigating utilization of NPN sources for providing nitrogen (74).

A diet that supplies all nutrient requirements for rumen microbial activity can utilize urea nitrogen to build microbial protein (56). Urease secreted by rumen microorganisms rapidly hydrolyzes urea to ammonia and carbon dioxide (57). An increase in rumen pH causes the NH_4^+ ion to be converted to NH_3. The ammonia is rapidly absorbed through the rumen wall into the portal vein. The amount of ammonia absorbed depends on the concentration gradient and rumen pH. The liver functions to convert blood ammonia to urea. When the liver can no longer convert all of the blood ammonia to urea, ammonia toxicity may occur. This is due to the concentration of ammonia in peripheral blood (12,18,47,51).

Liquid feeds containing ammoniated sugars have a pH range from 8 to 9. Toxic effects from ammonia were shown to be prevented by lowering the pH with organic and inorganic acids. Examples of acids used were phosphoric, lactic, acetic, propionic, hydrochloric, and sulfuric. The final pH of the liquid supplement was 3 to 6 (81).

Maximum levels of urea that can be safely fed to ruminants is hard to determine because numerous factors are involved in the efficient utilization of urea nitrogen. A few factors are: source of carbohydrate, frequency of feeding, and minerals supplied in the ration (18).

A readily available supply of carbohydrates has been shown to improve urea utilization. Therefore, the degree that energy requirements of microorganisms are met influence their utilization of nitrogen from ammonia-supplying compounds (16). Results indicate that starch is superior to molasses in providing carbon skeletons required for ammonia nitrogen to be converted to microbial protein. Cellulose is degraded too slowly, while mono- and disaccharides are fermented too rapidly to be utilized well (20).

Cane molasses is incorporated into many livestock rations to improve nitrogen utilization from urea. When 10 or 15% molasses was added to a basal ration containing urea, at the expense of corn, nitrogen retention and apparent digestibility of dry matter were improved. This increase in nitrogen retention indicates the microbial environment has been improved for converting NPN to microbial protein (33). This agrees with in vitro studies conducted by Burroughs (15).

Many investigators have reported application of moist heat with cereal grains produces a hydrated starch that can be more rapidly digested by rumen microorganisms than raw starch. This allows the energy from cereal grains to be available at rates that improve urea nitrogen utilization (55, 62,69).

Meyer demonstrated the effect of feed processing on nitrogen utilization. Ground and expanded grain were compared to unprocessed feed. Results showed feed processing lowered rumen ammonia 50%, while increasing bacterial

nitrogen 50%. This indirectly shows that nitrogen utilization improves because of increased carbohydrate utilization (36,53).

The skeleton contains a large percentage of the total body phosphorus. The smaller portion is found in the tissue and makes phosphorus one of the most important minerals required by the animal (54). This phosphorus functions in carbohydrate, fat, protein, and normal muscle metabolism (84).

Many times supplemental phosphorus is needed in rations that already contain sufficient calcium. Phosphoric acid has been shown to be an effective source of phosphorus (59) for Hereford heifer calves being fed dry bluestem grass, soybean meal, and molasses. This acid was highly palatable and produced no ill effects (52). Other studies showed phosphoric acid comparable to dicalcium phosphate and steamed bone meal in supplying the animal's phosphorus requirement (59,73).

Some constituents in molasses cause a gel formation when mixed with phosphoric acid. This normally occurs when pH and temperature of the mixture is increased. The gelling tendencies were prevented by small additions of non-phosphatic acids. Sulfuric acid prevented gelling when added at a rate of 12 parts by weight per 100 parts total weight of 75% phosphoric acid (77).

Phosphoric acid has been utilized in liquid supplements to repel insects, facilitate dissolution of urea in molasses, stabilize the product against fermentation, and reduce viscosity of the supplement. Also claimed was increased vitamin stability because of the lower pH and molasses preventing rapid interreaction with oxygen (3).

Acetic and propionic acids have both been used as grain preservatives. These acids are noted for their antifungal activity (43). Both acids are found in fermented grains and are naturally utilized by ruminants (82).

Feeding Potato Based Material

For many years cull potatoes have been fed to livestock. At times, the potatoes are blended with haylage and stored in silos. This method has been shown to preserve potatoes for at least one year. Potatoes are often dehydrated before feeding. This dried material is comparable to corn in feed value. However, it is expensive to produce dried potato flakes (75).

For animals to efficiently utilize potatoes, the importance of cooking has been clearly demonstrated. Cooked potato flakes showed a greater efficiency of nutrient utilization than raw potatoes. A study with young pigs showed that cooked potato flakes gave a growth response similar to that achieved with corn meal (79,80).

Proximate analyses indicated that dried cooked potato flakes may be utilized in broiler diets. The potato flakes containing 9.5% protein were added in place of corn to starter and finisher diets at levels of 0, 10, 20 and 40%. The birds fed 40% potato flakes grew slower and ate less, while the other three diets all gave similar results in bird performance. The feed efficiency of all diets was equal (78).

Cooked potato flour was used to feed 7-day old calves on an early weaning system. Dried skimmed milk was replaced by 7, 14 and 21% potato flour. Investigations of iso-nitrogenous and iso-energetic diets showed cooked potato flour depressed growth until the calves were 25 days of age. However, calves weaned at 42 days of age showed overall calf performance similar with all diets (38).

Another investigation, using 7-day old calves, studied performance and digestibility of dried cooked potatoes in liquid milk replacers and creep diets. While early depression of growth was noted, performance was not significantly depressed after 35 days on trial. Creep feed intake was not

influenced by incorporating potato starch in milk diets. Bloat was noted in some calves fed 40% potato flakes. The calves on this diet ate less, but grew faster than calves fed corn (37).

CHAPTER III

MATERIALS AND METHODS FOR INVESTIGATIONS OF POTATO STARCH WASTE IN LIQUID SUPPLEMENTS

Potato Starch Waste

Potato starch for these investigations was obtained from a waste stream that occurs from production of potato chips. The waste starch was supplied by the Frito Lay Company[1]. When potato starch is allowed to settle from excess waste water, the sediment is a solid material with the composition of a fine paste. On an "as is" basis, determined by proximate analysis, the starch material contained: .23% protein; 45% dry matter; .05% ash; no measurable level of ether extract or crude fiber; and 54.72% nitrogen-free extract, determined by difference.

Hydrothermal Processing

Experimental liquid supplement samples were processed using a Penick and Ford laboratory[2] continuous jet cooker. The cooker is equipped with a Moyno pump that delivers 1.17 liters per minute of slurry to a hydrothermal heater. Live steam is blended directly with the slurry allowing temperatures of 163 C and 55 psig to be achieved. The jet cooker is equipped with both temperature and pressure gauges. The moisture absorbed from the steam and retention time of the slurry in the cooker are given in Chapter IV. This processing technique causes extensive shear and cooking of starch.

Processing of Liquid Supplement Samples

A starch source was blended with urea (45% N) using a starch source:urea ratio calculated on a dry weight basis. If necessary, water was added to

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[1]Frito Lay Inc., Dallas, Texas.

[2]Penick and Ford, Ltd., Cedar Rapids, Iowa.

form the slurry. The starch source:urea slurry was jet cooked at 141 to 149 C at 40 psig. Cane molasses (3% crude protein and 75% dry matter) was immediately added to adjust the protein equivalent to the desired level. The slurry was cooled to 60 C before adding α-amylase (derived from Bacillus subtilis containing not less than 19,000 Wallerstein Bacterial Amylase Units per gram) to decrease the viscosity of the product. The α-amylase optimum activity for starch liquefacation is at temperatures of 60 to 85 C and is effective at pH's 6.5 to eight. When the desired viscosity was reached, enzyme activity was terminated with 85% food grade phosphoric acid. After the supplement had cooled to 40 C, propionic acid was added to inhibit mold growth.

Analytical Techniques

Dry Matter. Moistures (4) were determined in duplicate using a forced air oven for 1 hr set at 130 C. Percentage dry matter was then calculated.

Crude Protein. Nitrogen (4) content of liquid supplements was determined using the boric acid modification of Macro-Kjeldahl. Analyses were run in duplicate with the nitrogen being multiplied by 6.25 to calculate protein equivalent.

pH. A Beckman pH meter was used to determine pH of liquid supplement samples. Readings were taken to the nearest hundredth of a pH unit.

Viscosity. Viscosity of samples was determined at room temperature (25 to 27 C) using a LVT Brookfield Viscometer[1]. Results are reported in centipoise (cps). Comparison can only be made of cps values determined with the same spindle and rpm. Different spindles and rpm's are used when the range of viscosities being measured has a large variance.

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[1]Brookfield Engineering Laboratories, Inc., Stoughton, Massachusetts.

Free Ammonia. Conway (21) micro-diffusion analysis measures free ammonia (NH_3-N) that is released from the sample when mixed with saturated potassium carbonate. Free ammonia was calculated as milligrams NH_3-N per gram sample. Averages of duplicate samples are reported.

Starch Damage. Sung's (72) method for starch damage is based on Sandstedt's (65) enzyme hydrolysis with β-amylase with a subsequent measurement of reducing sugars using a ferricyanide analysis (1). Sandstedt's (65) method was modified by reducing β-amylase to 60 mg to digest a 1 g sample. Therefore, the results indicate the amount of readily fermentable carbohydrate in the liquid supplement. Results are reported as milligrams of maltose per gram dry matter. A maltose standard was run to adjust for daily variations that occur in the analytical technique.

Protein Synthesis. Barr's (9) in vitro rumen fermentation analysis was used to determine protein synthesized from liquid supplement samples. Rumen fluid was obtained from a rumen fistulated Angus steer fed a urea-containing ration. Fluid was collected from the middle of the ventral sac of the rumen with a syringe and placed in a thermos bottle. Immediately after collection, rumen fluid was strained through four layers of cheesecloth. Ten milliliter aliquots of strained rumen fluid were added to a 1 g sample of liquid supplement in a 50 ml centrifuge tube. A pH buffer, containing electrolytes similar to saliva, was added. The tube was immediately closed with a rubber stopper equipped with a bunsen valve. Following a 4 hr incubation in a water bath at 39 C, samples were centrifuged at 25,400 X G for 15 minutes. The supernatant was discarded and the residue extracted twice with 25 ml of methanol. The precipitate was transferred to Kjeldahl flasks for nitrogen analysis. Crude protein was determined in the following samples: 1. feed samples (liquid supplement + 20 ml buffer + 10 ml rumen

fluid), 2. feed blank (liquid supplement + 20 ml buffer) and 3. rumen fluid blank (10 ml rumen fluid + 20 ml buffer). The feed samples and feed blanks were both incubated before centrifugation, while the rumen fluid blank was not. Milligrams of protein were calculated by:

$$\text{mg protein} = \text{ml acid} \times \text{normality of acid} \times 14 \times 6.25$$

$$\text{mg protein synthesized} = \text{mg protein in feed sample} - (\text{mg protein in feed blank} + \text{mg protein in rumen fluid blank})$$

Protein synthesis results are averages of duplicate determinations reported as milligrams of protein synthesized or as a percentage of the experimental control.

CHAPTER IV

EFFECT OF STORAGE ON POTATO BASED LIQUID SUPPLEMENTS

Many liquid supplements on the market today undergo a layering effect when stored several days. The instability of liquid supplements is the result of ingredients and additives in the formula breaking down during extended storage.

The purpose of these investigations were to determine an acid combination that would stabilize a hydrothermally processed potato based liquid supplement over the 2 to 3 month period that normally would be required for complete consumption. Potato starch used was from the waste stream previously reported.

Materials and Methods

This study was conducted using various levels and combinations of acids to lower the pH and act as preservatives for hydrothermally processed liquid supplement samples. These include: phosphoric acid (85% food grade), propionic acid, sulfuric acid, and methyl-bis propionate[1] (MBP). Researchers have previously reported the first three acids listed as being used for antimicrobial agents or ingredients in liquid supplements (3,43,77,81).

A hydrothermal processing technique was used to produce the potato based liquid supplement. This process was discussed in Chapter III. Table 3 shows formulas for liquid supplements containing potato starch:urea ratios of 1.5:1 and 1.75:1 calculated on a dry weight basis. Water absorbed during processing was due to steam being directly blended with the starch:urea slurry. The

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[1]Methyl-bis propionate was a combination of formaldehyde and propionic acid supplied by Chevron Chemical Company, Norwalk, Iowa.

acid preservative combination added varied from 1.5 to 3.0% by total weight. This range allows a 1.5% difference in the final product weight to occur.

Individual batches of potato based supplement were produced for both 1.5:1 and 1.75:1 potato starch:urea ratios. The total batch weight processed was 27.27 and 11.36 kg, respectively. Potato starch and urea were mixed and then jet cooked at 141 C and 40 psig. This gel was blended with molasses, adjusting the protein equivalent to 30%. After the liquid supplement cooled to 60 C α-amylase was added at the rate of .01% by weight. Enzyme activity was terminated after 4 min with addition of .5% phosphoric acid. The batches were immediately divided into 3.64 kg samples. Table 4 shows acid levels and combinations added to each individual sample, with adjustment for the .5% phosphoric acid already added. Samples were stored in 3.78 l plastic containers at room temperature with lids secured. Samples were prepared twice during week zero of the storage trial to obtain an estimate of reproducibility. Before removing aliquots for analysis during the trial, the liquid supplement in the plastic containers was thoroughly mixed.

The 1.75:1 potato starch:urea ratio formula was evaluated with 1.0% phosphoric acid and various levels of propionic acid as the preservative. This allowed for comparison with the 1.5:1 starch:urea samples treated with these acid combinations.

Factors used for evaluating the effect of storage on the potato liquid supplement samples shown in table 4 were: 1. pH, 2. crude protein, 3. dry matter, 4. free ammonia (NH_3-N), 5. maltose and 6. viscosity. These analyses were run at intervals of 0, 3, 5 and 8 weeks to detect variations within individual samples. Due to the large number of samples in the study, some analyses were not run at the intervals originally specified.

TABLE 3. POTATO BASED LIQUID SUPPLEMENT FORMULAS

	Starch:urea ratios 1.5:1 %	1.75:1 %
Potato starch material (as is) (45% dry matter)	35.83	41.88
Cane molasses	38.50	31.04
H_2O (absorbed in processing)	13.17	14.54
Urea	11.00	11.04
Acid[A]	1.5-3.0	1.5-3.0
Enzyme (α-amylase)	0.01	0.01

[A]Combination of phosphoric, propionic, sulfuric, and MBP varies from 1.5 to 3.0% of total formula.

TABLE 4. ACID[A] COMBINATION AND LEVELS FOR STORAGE EVALUATION

Sample Set 1 & Set 2	Potato starch urea ratio	Phosphoric %	Propionic %	MBP %	Sulfuric %
S1 & S13	1.5:1	1.0	0.5	---	---
S2 & S14	1.5:1	1.0	1.0	---	---
S3 & S15	1.5:1	1.0	2.0	---	---
S4 & S16	1.5:1	1.0	---	0.5	---
S5 & S17	1.5:1	1.0	---	1.0	---
S6 & S18	1.5:1	1.0	---	2.0	---
S7 & S19	1.5:1	0.5	0.5	---	0.5
S8 & S20	1.5:1	0.5	1.0	---	0.5
S9 & S21	1.5:1	0.5	2.0	---	0.5
S10 & S22	1.75:1	1.0	0.5	---	---
S11 & S23	1.75:1	1.0	1.0	---	---
S12 & S24	1.75:1	1.0	2.0	---	---

[A]Percentage acid based on weight of complete formula.

In vitro protein synthesis (87) was determined with potato liquid supplement samples during week zero. At the end of week zero, small portions of each sample were placed in cold storage. Protein synthesis, determined in week eight, used the frozen samples as controls for samples stored in plastic containers.

Aardvark (2) is a computerized statistical analysis of variance that was run on the results from the first five analyses mentioned. To determine real differences among means, all were calculated from sample set 1 and set 2. A least significant difference (LSD) level of .05 was used. Tests for significant differences in weekly means were determined for each acid treatment. Differences occurring between acid treatments were calculated from the sample average mean for the complete trial. The interreaction between weeks and acid treatments were also analyzed. These data are presented in figures to indicate trends from linearity. This method of statistical analysis allowed for larger degrees of freedom to calculate LSD ranges at the .05 level. Protein synthesis was analyzed by studying interreactions between weeks and acid treatments.

Results and Discussions

This investigation was designed to determine combinations and levels of acid that would best stabilize the product from chemical changes as a result of time and microbial activity.

Table 5 gives data on theoretical calculated formula weights and actual weights obtained from the jet cooking process. Set 1 and set 2 were processed on different days. Weighing of formulas was achieved using a Toledo scale with 4 oz divisions. There is less than 2% variation between calculated and final weights. These data indicate that water absorption from steam remains reasonably constant and a product with similar composition

can be reproduced from day to day. There are no indications that percentage water absorbed varies with changes of the potato starch:urea ratio. Calculated water absorbed from table 3 shows that 21 to 22% of the slurry leaving the jet cooker was moisture absorbed from steam. The difference between calculated and final formula weight was due to cooking time and variations in initial weighing.

The potato starch:urea slurry retention time in the jet cooker was determined by adding a red dye at the Moyno pump entrance while processing. Time was measured until the dye initially appeared after the hydrothermal heater. This retention time was 16 to 17 seconds.

pH. There was little effect on sample pH (table 6) when the level of propionic acid or MBP increased from .5 to 2.0%. However, a decrease in pH from 4.05 to 3.80 occurred when changing from a 1.5:1 to a 1.75:1 potato starch:urea ratio treated with .5% propionic acid.

Table 7 shows weekly sample pH averages of set 1 and set 2 of the potato liquid supplement. These data show that little weekly pH variation occurred within a sample over an 8-week period. The samples treated with 1.0 phosphoric and 2.0% propionic acid, with the 1.5:1 starch:urea ratio, were the only acid treatment with a significant difference ($P<.05$). However, raw data in table 1 of appendix A indicates this variation is probably due to error in zeroing the pH meter, rather than to a real change in sample's pH. Figures 1, 2, 3 and 4 show that pH for all acid treatments remained constant throughout the storage trial. The data plotted are averages of corresponding samples from set 1 and set 2 reported in table 7. These figures agree with statistical results in that there was little variation in pH because of time or acid treatment.

TABLE 5. CALCULATED AND FINAL PRODUCT WEIGHT

Set	Potato Starch urea ratio	Calculated formula weight (kg)	Final product scale weight (kg)	Difference between calculated and final (kg)
1	1.5:1	27.27	27.72	.45
1	1.75:1	11.36	11.82	.46
2	1.5:1	27.27	27.05	.22
2	1.75:1	11.36	11.14	.22

TABLE 6. EFFECT OF ACID LEVEL AND COMBINATION ON pH

Sample	Potato starch urea ratio	% Acid 0.5	1.0	2.0
		pH values		
1% Phosphoric + X%[A] propionic	1.5:1	4.05	4.06	4.03
1% Phosphoric + X% MBP	1.5:1	4.02	3.96	3.97
.5% Phosphoric + .5% sulfuric + X% propionic	1.5:1	3.93	3.95	3.96
1% Phosphoric + X% propionic	1.75:1	3.80	3.77	3.74

[A] X = % acid addition.

The average pH over the 8-week trial for each acid treatment is shown in table 7. The samples treated with 1.0 and 2.0% propionic acid at the high starch level were lower ($P<.05$) than samples made with the 1.5:1 starch:urea ratio. The decline in pH is attributed to an increase in potato starch and a decrease in molasses, while holding the percentage of urea in the formula constant. The pH of the other acid treatments were approximately 4.0 and not different ($P<.05$) regardless of acid treatment.

Dry Matter. The average dry matter content over the 9-week trial for samples treated with propionic acid is shown in table 8. These data show the effect starch:urea ratio had on dry matter content of potato liquid supplements. Samples produced with the 1.75:1 starch:urea ratio were significantly ($P<.05$) lower in dry matter than samples made with the 1.5:1 ratio. Samples compared were treated with the same acid combinations. The 1.5:1 ratio contained 6.0% more potato starch than the 1.75:1 ratio. This resulted in approximately a 3.0% difference in dry matter, due to the 55% moisture content of the potato starch waste.

The average weekly dry matter content for each acid treatment is shown in table 9. There were no differences ($P<.05$) within an individual acid treatment for the 9-week period, except for samples S9 and S21 during week six. These samples were treated with .5% phosphoric plus .5% sulfuric plus 2.0% propionic acid using the 1.5:1 starch:urea ratio. However, the dry matter reported for S21 in table 2 of appendix A is exceptionally high. This may indicate an error in the dry matter analysis of this sample. Figures 5, 6, 7 and 8 show average dry matter from set 1 and set 2 plotted against weeks. These data agree with the expected values for samples stored in containers that prevented rapid evaporation. The plots and the statistical analyses in table 9 show dry matter was stable for a 9-week period.

TABLE 7. EFFECT OF STORAGE ON THE pH[A] OF POTATO BASED LIQUID SUPPLEMENTS

Sample Set 1 & Set 2	Week[B] 0	3	5	8	Sample avg[C] for trial
S1 & S13	4.05^{a}	3.91^{a}	4.11^{a}	4.04^{a}	4.03^{h}
S2 & S14	4.06^{a}	3.95^{a}	4.10^{a}	4.08^{a}	4.05^{h}
S3 & S15	4.03^{ab}	3.85^{a}	3.87^{ab}	4.10^{b}	3.97^{h}
S4 & S16	4.02^{a}	3.99^{a}	3.93^{a}	4.10^{a}	4.01^{h}
S5 & S17	3.97^{a}	3.93^{a}	3.95^{a}	4.11^{a}	3.98^{h}
S6 & S18	3.97^{a}	3.89^{a}	3.95^{a}	4.05^{a}	3.97^{h}
S7 & S19	3.93^{a}	3.87^{a}	3.97^{a}	4.06^{a}	3.96^{h}
S8 & S20	3.94^{a}	3.87^{a}	3.95^{a}	4.05^{a}	3.95^{h}
S9 & S21	3.96^{a}	3.84^{a}	3.97^{a}	4.04^{a}	3.95^{h}
S10 & S22	3.80^{a}	3.86^{a}	4.04^{a}	4.03^{a}	3.93^{h}
S11 & S23	3.77^{a}	3.77^{a}	3.83^{a}	3.86^{a}	3.81^{i}
S12 & S24	3.74^{a}	3.76^{a}	3.84^{a}	3.84^{a}	3.80^{i}

[A] pH results are averages of set 1 and set 2.

[B] Row means for weekly averages with the same superscript (a,b) are not significantly different ($P<.05$).

[C] Column means for sample average for the trial with the same superscript (h,i) are not significantly different ($P<.05$).

[D] See table 1 in appendix A for weekly pH values for individual samples.

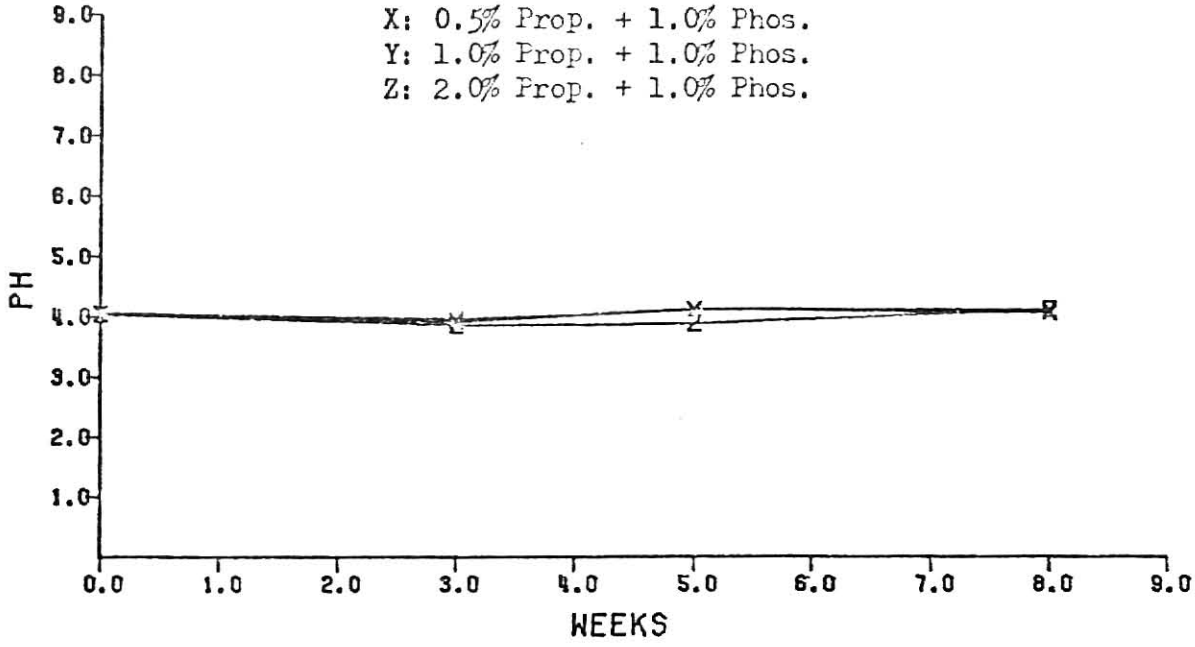

FIGURE 1. PH DURING STORAGE USING PROPIONIC ACID

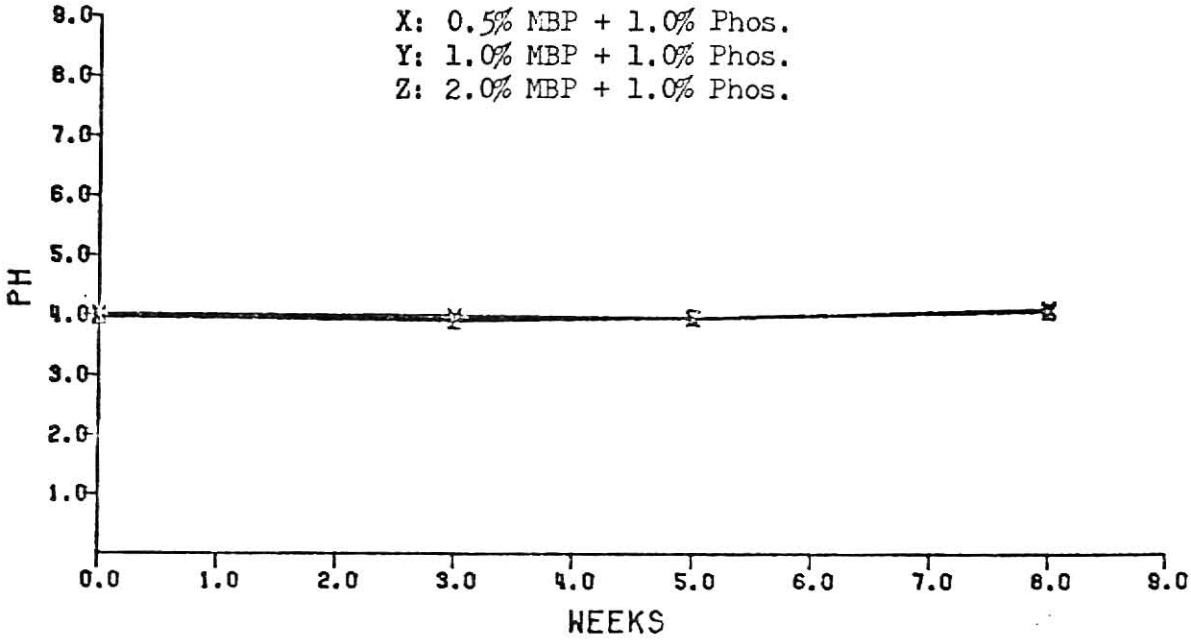

FIGURE 2. PH DURING STORAGE USING MBP

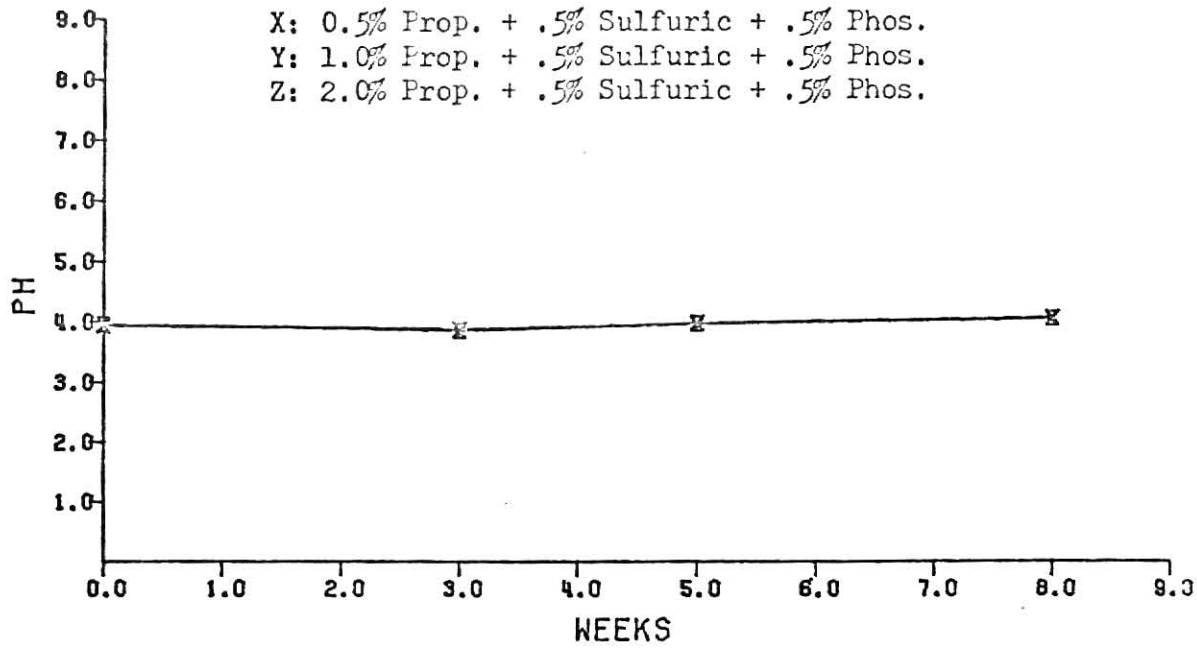

FIGURE 3. PH DURING STORAGE USING SULFURIC AND PROPIONIC ACID

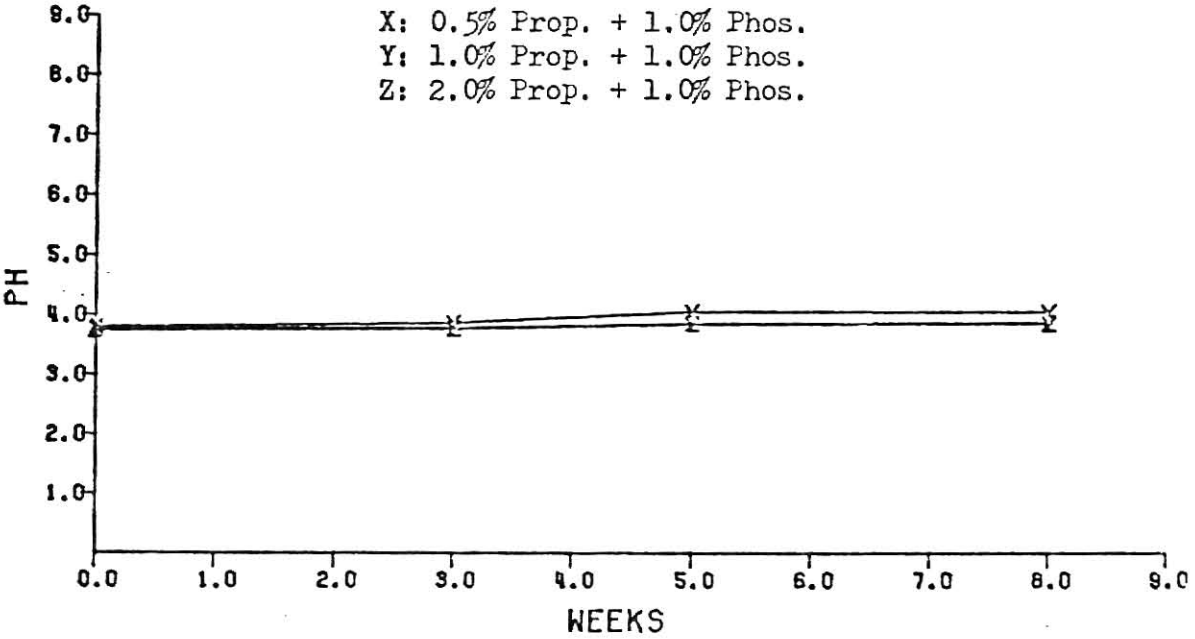

FIGURE 4. PH DURING STORAGE USING PROP. ACID AT HIGH STARCH LEVEL

TABLE 8. EFFECT OF POTATO STARCH:UREA RATIOS ON DRY MATTER CONTENT OF PROCESSED SAMPLES

Sample Set 1 & Set 2	Potato starch urea ratio	Dry matter[AB] %
S1 & S13	1.5:1	53.21[a]
S2 & S14	1.5:1	53.92[a]
S3 & S15	1.5:1	53.74[a]
S10 & S22	1.75:1	51.04[b]
S11 & S23	1.75:1	51.32[b]
S12 & S24	1.75:1	51.18[b]

[A]Dry matter are averages for individual acid treatments from set 1 and set 2 over a 9-week trial.

[B]Column means with the same superscript (a,b) are not significantly different ($P<.05$).

TABLE 9. EFFECT OF STORAGE ON THE DRY MATTER[A] CONTENT OF POTATO BASED LIQUID SUPPLEMENTS

Sample Set 1 & Set 2	Week[B] 0	3	6	9
S1 & S13	53.09[a]	52.58[a]	53.71[a]	53.45[a]
S2 & S14	53.51[a]	53.54[a]	54.75[a]	53.85[a]
S3 & S15	53.31[a]	53.54[a]	54.68[a]	53.44[a]
S4 & S16	53.12[a]	53.85[a]	53.44[a]	52.67[a]
S5 & S17	52.69[a]	53.32[a]	52.84[a]	52.96[a]
S6 & S18	53.51[a]	54.09[a]	53.08[a]	52.97[a]
S7 & S19	54.42[a]	55.70[a]	53.83[a]	53.62[a]
S8 & S20	54.16[a]	51.29[a]	55.06[a]	53.81[a]
S9 & S21	53.90[a]	51.31[a]	56.06[b]	53.99[a]
S10 & S22	50.21[a]	50.77[a]	52.27[a]	50.89[a]
S11 & S23	50.77[a]	50.06[a]	52.45[a]	51.98[a]
S12 & S24	50.84[a]	49.73[a]	52.72[a]	51.42[a]

[A]Dry matter values are averages of set 1 and set 2 reported as a percentage.

[B]Row means with the same superscript (a,b) are not significantly different ($P<.05$).

[C]See table 2, appendix A for weekly dry matter percentages for the individual samples.

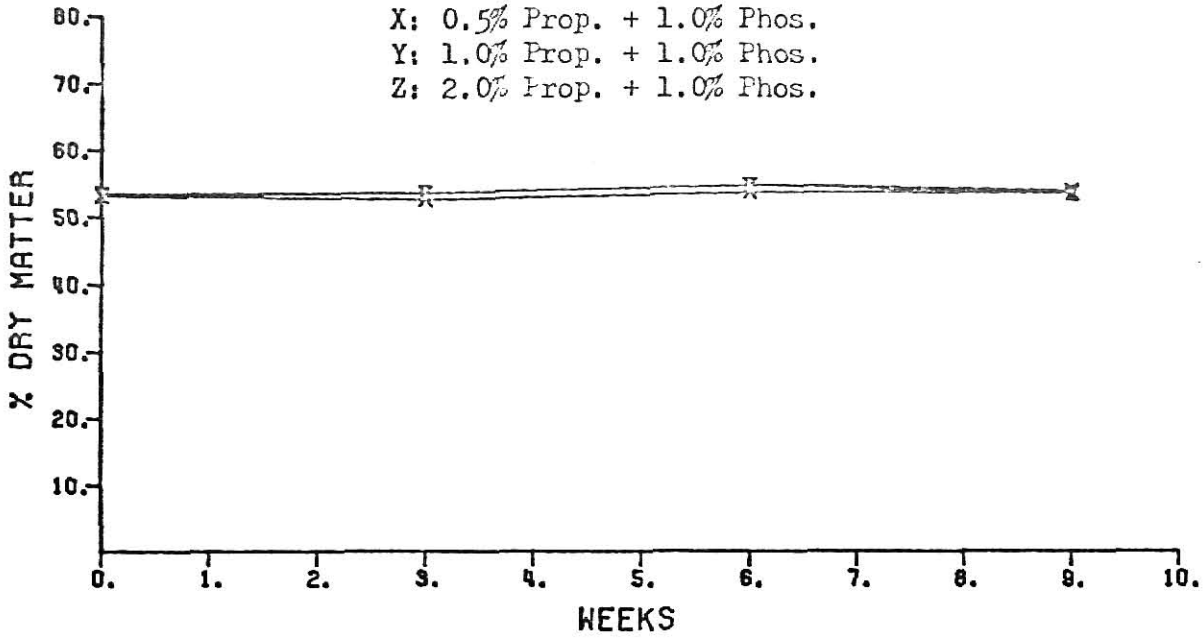

FIGURE 5. DRY MATTER DURING STORAGE USING PROPIONIC ACID

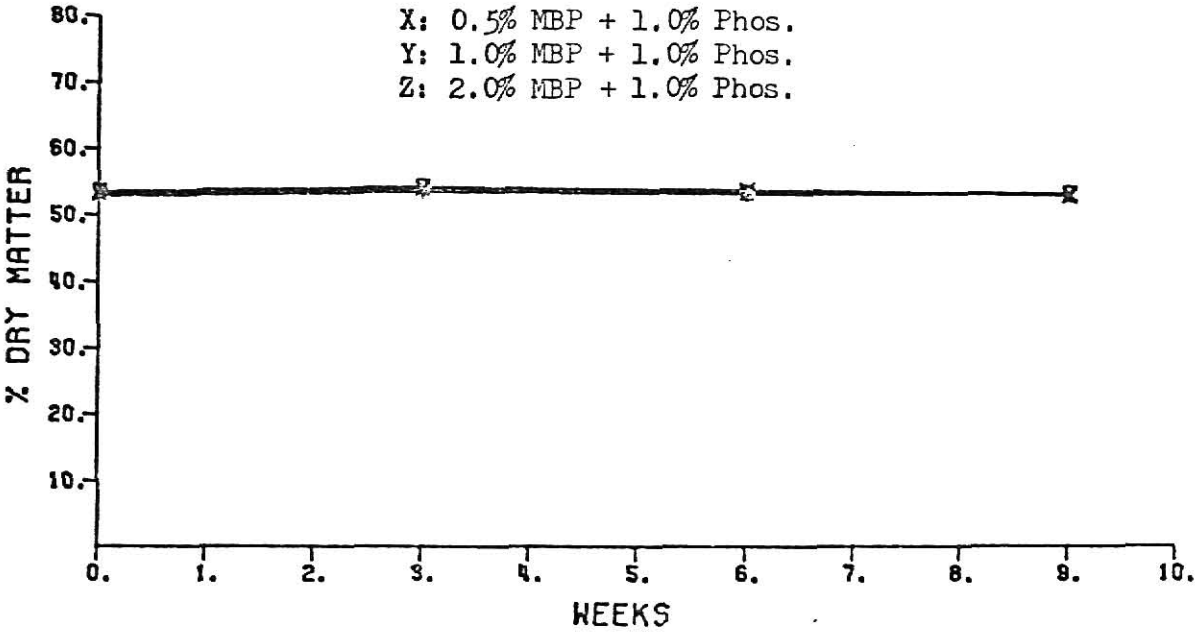

FIGURE 6. DRY MATTER DURING STORAGE USING MBP

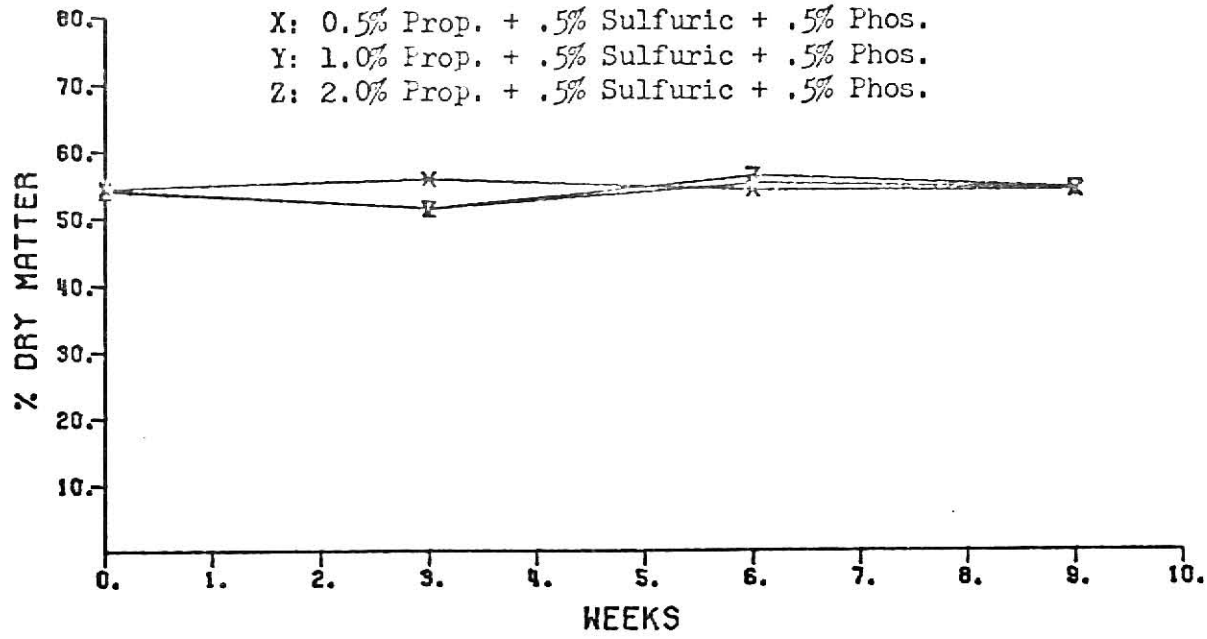

FIGURE 7. DRY MATTER DURING STORAGE USING SULFURIC AND PROPIONIC ACID

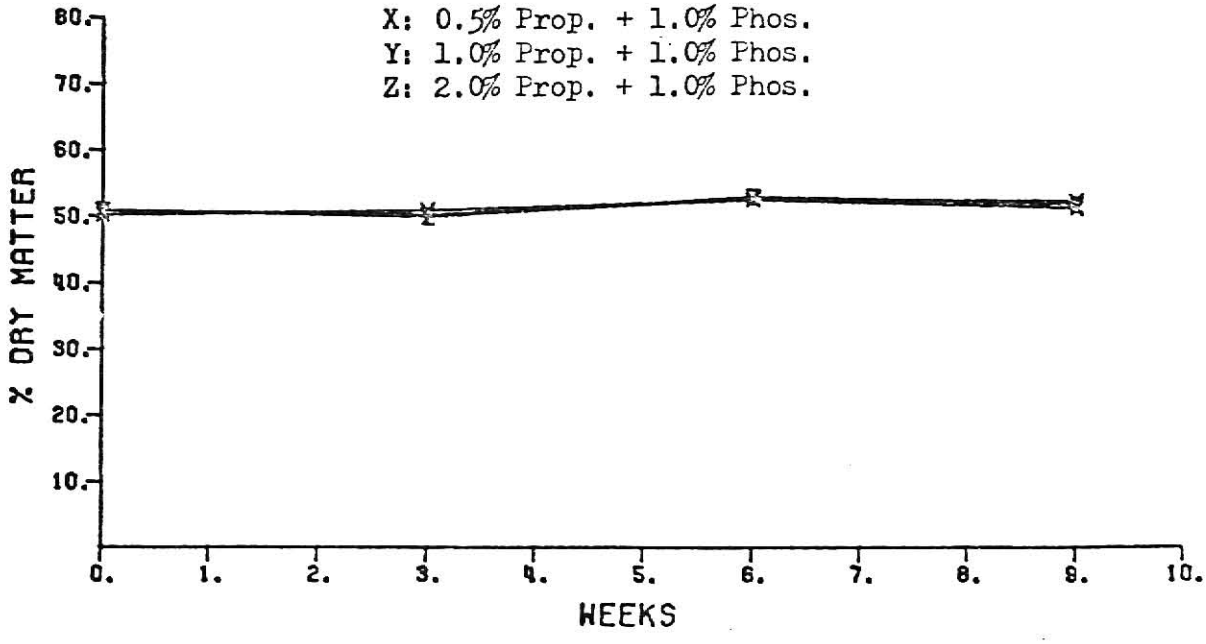

FIGURE 8. DRY MATTER DURING STORAGE USING PROPIONIC AT HIGH STARCH LEVEL

Containers were opened at intervals during the trial to obtain samples for analysis and this allowed interreaction with air.

Crude Protein. Table 10 shows the weekly average crude protein content of samples from set 1 and set 2 on a dry basis. There were no differences ($P<.05$) within any acid treatment during the 9-week period. Figures 9, 10, 11 and 12 illustrate that average protein contents showed no trends from linearity for the storage trial. These data indicate that nitrogen was not lost during storage of the potato liquid supplement.

Table 3 in appendix A shows weekly crude protein of individual samples on "as is" and dry basis. On an "as is" basis, the protein content ranges approximately between 28 and 31%. However, on a dry basis, the protein content of samples made with the 1.75:1 ratio are higher than the 1.5:1 starch:urea ratio. This was because of a lower dry matter content.

Free Ammonia. Table 11 and figures 13, 14, 15 and 16 show results of ammonia released from storage liquid supplement samples when mixed with saturated potassium carbonate (85). Results are weekly averages of two sample sets presented as mg NH_3-N/g dry matter. In all samples, a significant increase ($P<.05$) in free ammonia occurred between week six and nine. Samples treated with .5% phosphoric plus .5% sulfuric plus 2.0% propionic acid showed a significant increase ($P<.05$) as early as week three. Table 4 in appendix A shows free ammonia (NH_3-N) values for individual liquid supplement samples from set 1 and set 2.

The average free ammonia for the first six weeks of the trial was approximately 1.1 mg NH_3-N/g dry matter. Week nine shows average free ammonia levels around 1.35 mg NH_3-N/g dry matter. A 1 g sample of dry matter from a 30% protein equivalent liquid supplement contains 48.00 mg of nitrogen. Therefore, the increase in ammonia from week six to nine was only .52%

TABLE 10. EFFECT OF STORAGE ON THE CRUDE PROTEIN[A] CONTENT OF POTATO BASED LIQUID SUPPLEMENTS

Sample Set 1 & Set 2	Week[B] 0	3	6	9
S1 & S13	57.72[a]	55.43[a]	55.67[a]	56.09[a]
S2 & S14	55.44[a]	55.53[a]	55.27[a]	56.39[a]
S3 & S15	54.94[a]	54.26[a]	54.90[a]	55.49[a]
S4 & S16	55.99[a]	54.64[a]	55.68[a]	57.02[a]
S5 & S17	56.15[a]	54.72[a]	56.40[a]	56.83[a]
S6 & S18	55.09[a]	53.84[a]	56.59[a]	55.13[a]
S7 & S19	56.04[a]	53.54[a]	56.30[a]	56.43[a]
S8 & S20	55.63[a]	57.28[a]	54.08[a]	55.76[a]
S9 & S21	55.48[a]	56.66[a]	54.92[a]	55.56[a]
S10 & S22	60.18[a]	56.84[a]	57.00[a]	59.99[a]
S11 & S23	58.24[a]	57.56[a]	55.26[a]	58.09[a]
S12 & S24	57.70[a]	57.17[a]	54.90[a]	57.17[a]

[A]Crude protein values are averages of set 1 and set 2 on a dry basis and reported as a percentage.

[B]Row means with the same superscript (a) are not significantly different ($P<.05$).

[C]See table 3 in appendix A for weekly crude protein percentage for the individual samples.

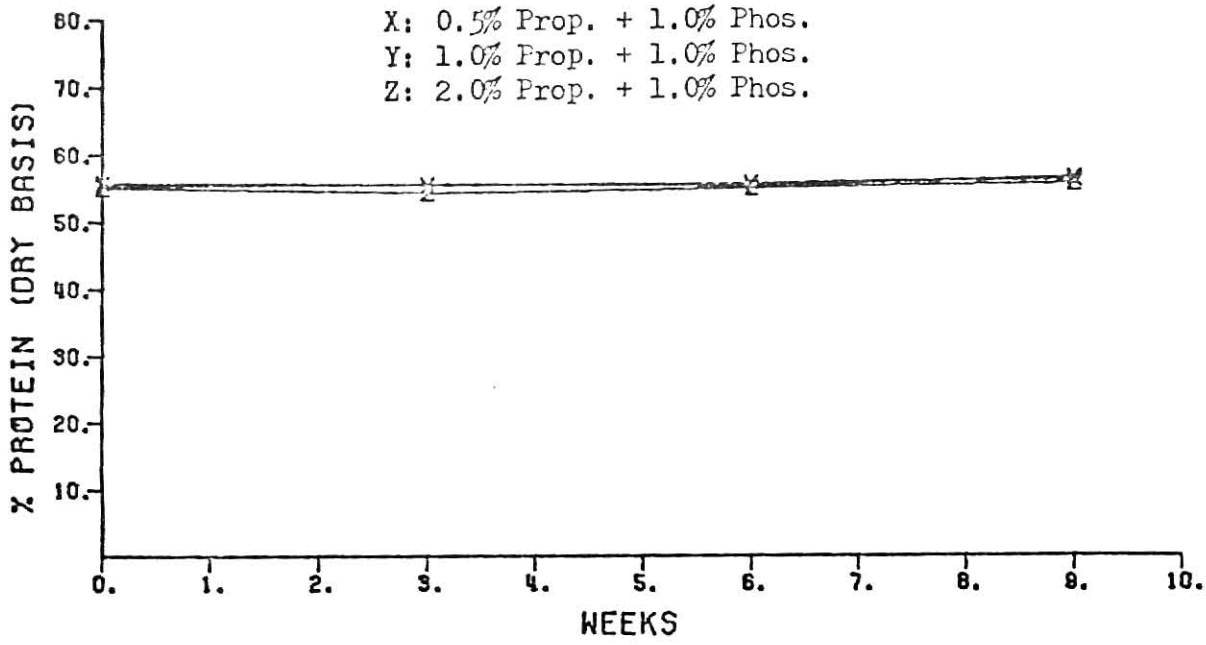

FIGURE 9. PROTEIN DURING STORAGE USING PROPIONIC ACID

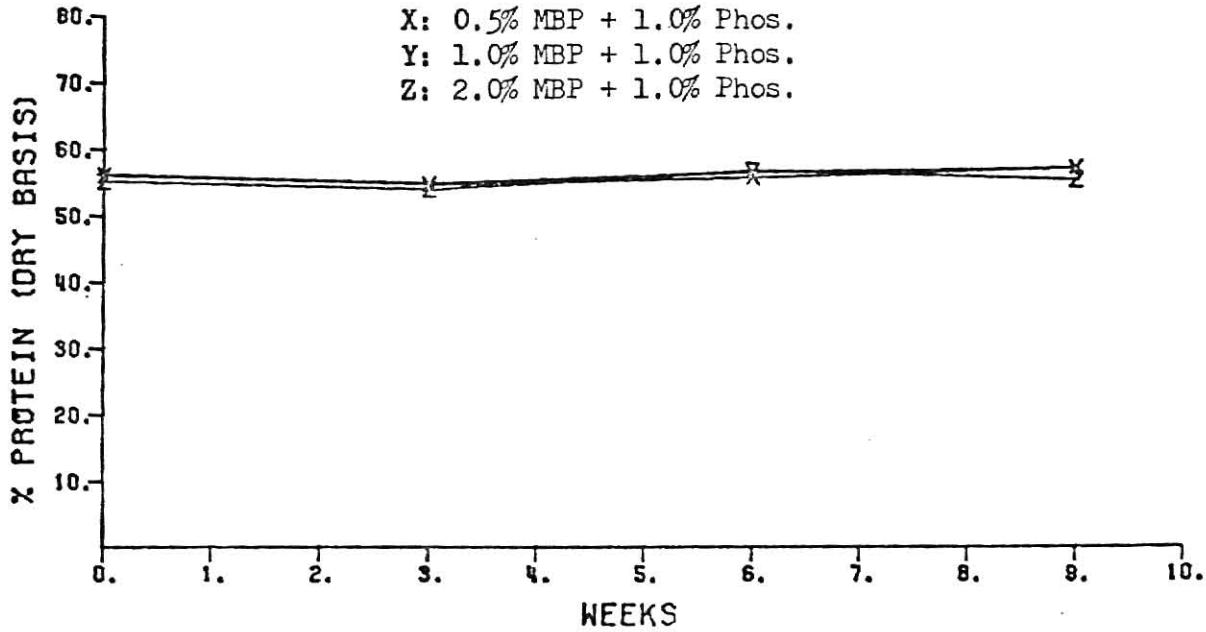

FIGURE 10. PROTEIN DURING STORAGE USING MBP

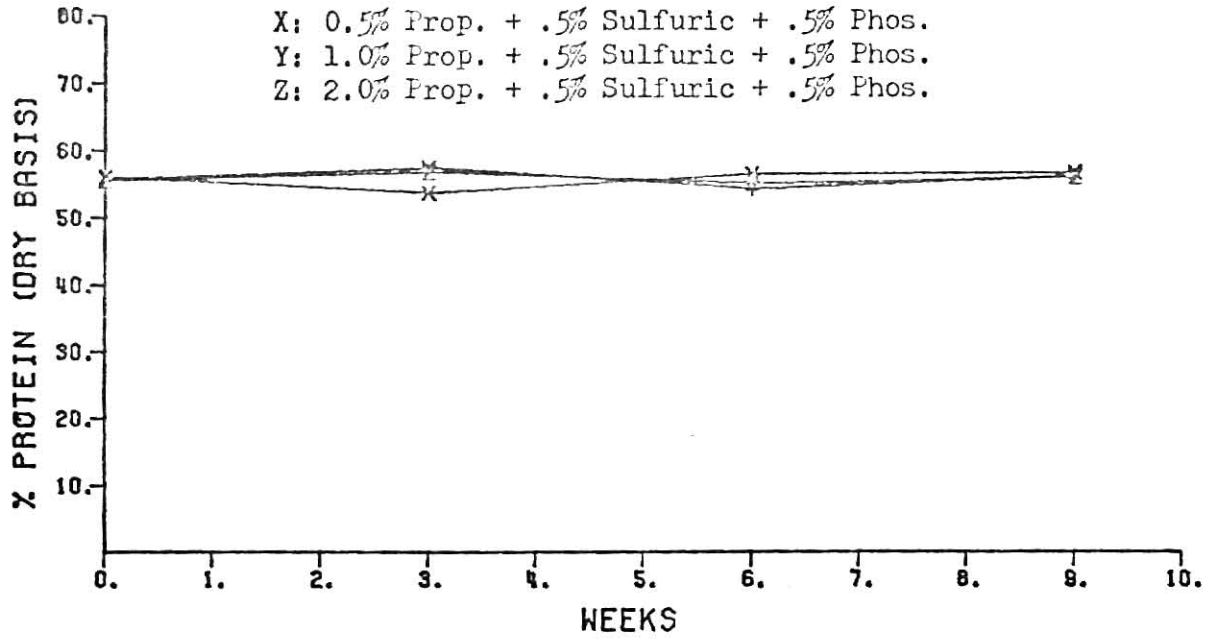

FIGURE 11. PROTEIN DURING STORAGE USING SULFURIC AND PROPIONIC ACID

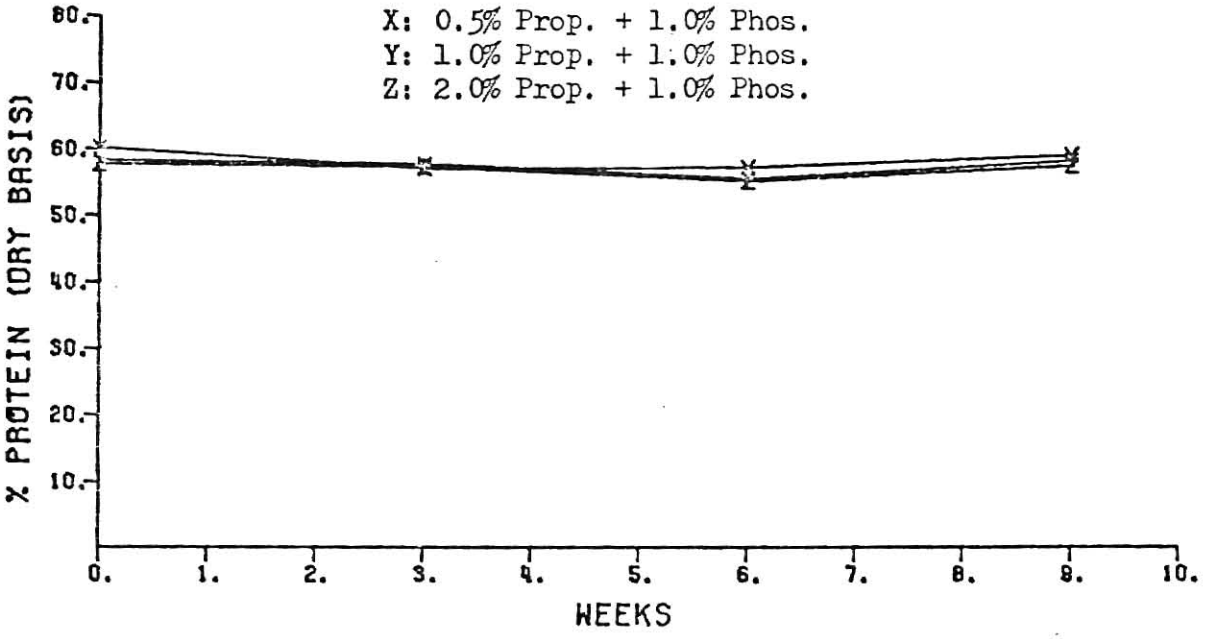

FIGURE 12. PROTEIN DURING STORAGE USING PROP. AT HIGH STARCH LEVEL

of the total nitrogen in the sample. While an increase in free ammonia (NH_3-N) was noted, the pH and protein content of the storage samples remained constant.

Averages for individual acid treatments for the complete trial show several different significant ranges in table 11. Free ammonia data using a starch:urea ratio of 1.5:1 gave no significant differences ($P<.05$) regardless of acid combination. Free ammonia decreased ($P<.05$) when using a starch:urea ratio of 1.75:1. This possibly occurred due to a greater amount of potato starch for the nitrogen to form a complex with.

Maltose. The readily available carbohydrate portion of the stored liquid supplement samples was estimated using β-amylase in the analytical technique followed by the ferricyanide analysis (43). The results of this analysis include the amount of starch susceptible to β-amylase plus the reducing sugars already present in the liquid supplement. Results in table 12 are averages of set 1 and set 2 reported as mg maltose/g dry matter. The only differences ($P<.05$) are from samples treated with 1.0 or 2.0% propionic acid produced using the 1.5:1 potato starch:urea ratio. Table 5 in appendix A shows weekly maltose values on an "as is" and dry basis.

Figures 17, 18 and 19 show that a decrease in maltose produced occurred during the storage trial. S1, a typical example, decreased from 200 to 178 mg maltose over the 9-week period. This indicates some retrogradation of starch occurred in the potato liquid supplement. The trends noted in these figures agree with the analysis of variance run on the average maltose for all samples for each week. Maltose was lower ($P<.05$) in weeks three, six and nine than week zero (table 12). Figure 20 is not as clear for interpreting maltose trends. Note, with lower addition levels of molasses, the

TABLE 11. EFFECT OF STORAGE ON THE FREE AMMONIA[A] (NH_3-N) VALUES OF POTATO BASED LIQUID SUPPLEMENTS

Sample Set 1 & Set 2	Week[B] 0	3	6	9	Sample avg[C] for trial
S1 & S13	1.065^{a}	1.138^{a}	1.171^{a}	1.307^{b}	1.170^{hi}
S2 & S14	1.090^{a}	1.189^{a}	1.093^{a}	1.319^{b}	1.173^{h}
S3 & S15	1.062^{a}	1.146^{a}	1.083^{a}	1.281^{b}	1.143^{hij}
S4 & S16	1.071^{a}	1.131^{a}	1.175^{a}	1.313^{b}	1.173^{h}
S5 & S17	1.066^{a}	1.102^{a}	1.157^{a}	1.332^{b}	1.164^{hij}
S6 & S18	1.078^{a}	1.040^{a}	1.128^{a}	1.349^{b}	1.149^{hij}
S7 & S19	1.099^{a}	1.068^{a}	1.206^{b}	1.362^{c}	1.184^{h}
S8 & S20	1.126^{a}	1.147^{a}	1.119^{a}	1.371^{b}	1.191^{h}
S9 & S21	1.053^{a}	1.207^{b}	1.147^{ab}	1.327^{c}	1.184^{h}
S10 & S22	0.999^{a}	1.082^{a}	1.081^{a}	1.305^{b}	1.117^{ijk}
S11 & S23	1.007^{a}	1.099^{a}	1.069^{a}	1.288^{b}	1.117^{ijk}
S12 & S24	0.947^{a}	1.058^{a}	0.974^{a}	1.288^{b}	1.067^{k}

[A]Free ammonia values are averages of set 1 and set 2 reported as mg NH_3-N/g dry matter.

[B]Row means with the same superscript (a,b,c) are not significantly different ($P<.05$).

[C]Column means for sample average for the trial with the same superscript (h,i,j,k) are not significantly different ($P<.05$).

[D]See table 4 in appendix A for weekly free ammonia values for the individual samples.

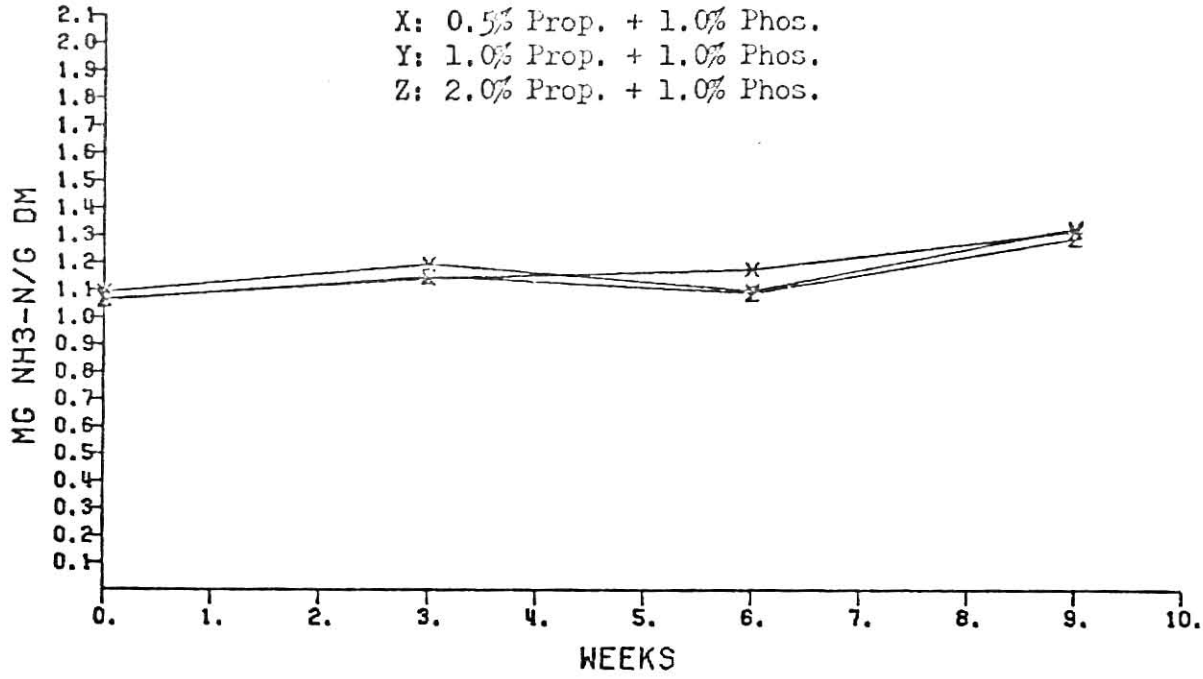

FIGURE 13. FREE AMMONIA DURING STORAGE USING PROPIONIC ACID

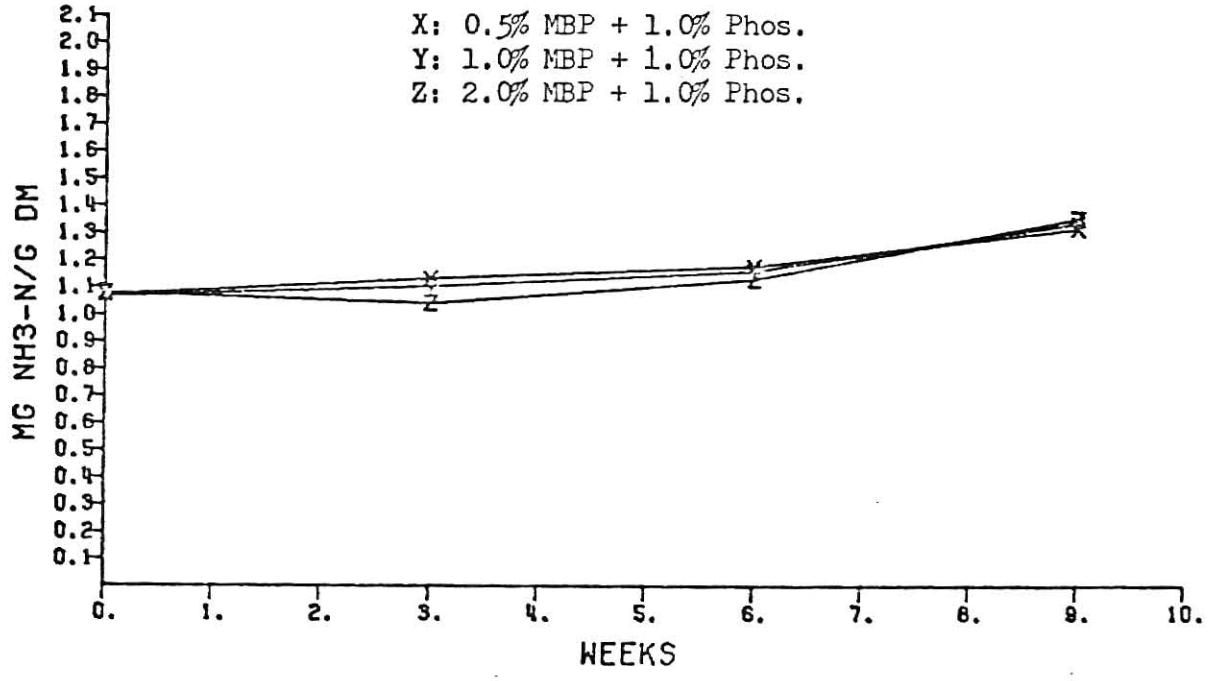

FIGURE 14. FREE AMMONIA DURING STORAGE USING MBP

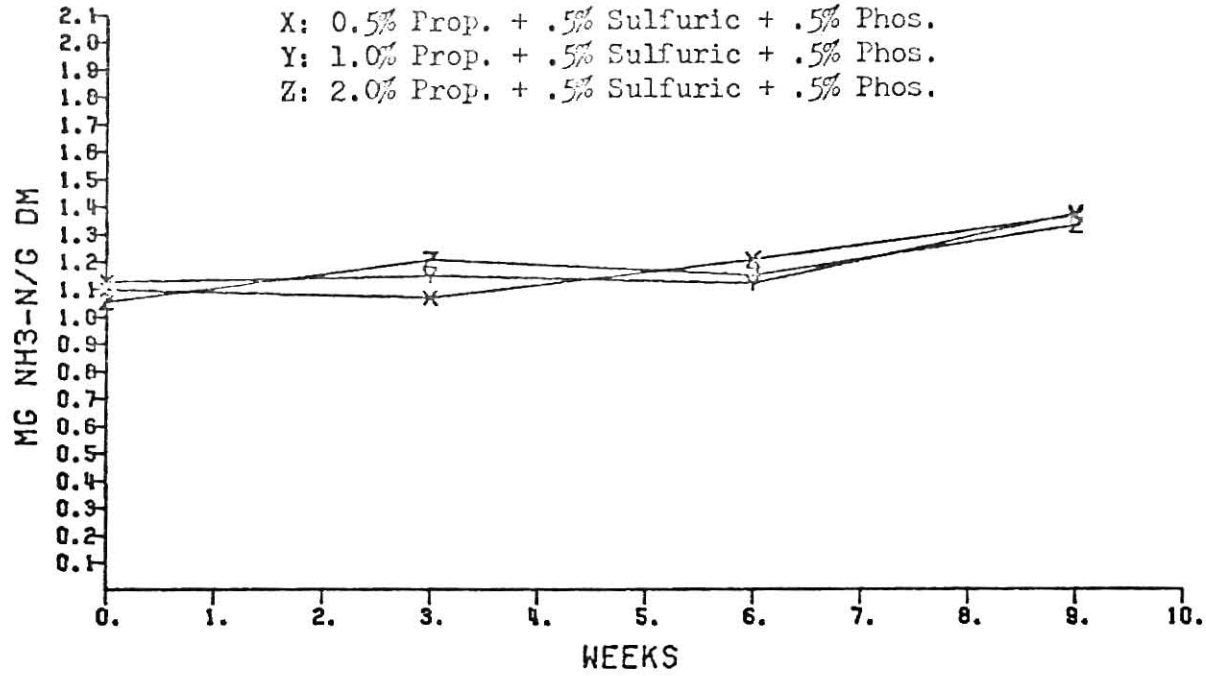

FIGURE 15. FREE AMMONIA DURING STORAGE USING SULFURIC AND PROPIONIC ACID

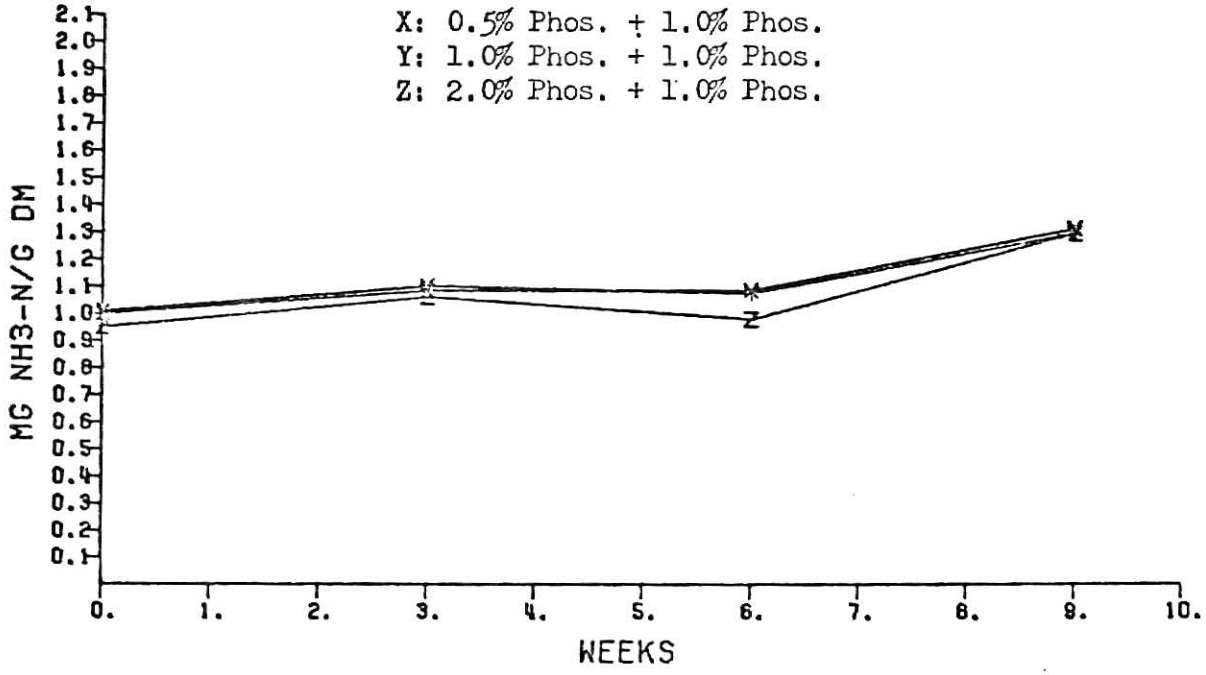

FIGURE 16. FREE AMMONIA DURING STORAGE USING PROP. AT HIGH STARCH LEVEL

TABLE 12. EFFECT OF STORAGE ON THE MALTOSE[A] VALUES OF POTATO BASED LIQUID SUPPLEMENTS

Sample Set 1 & Set 2	Week[B] 0	3	6	9
S1 & S13	379.2[a]	386.9[a]	339.7[a]	334.2[a]
S2 & S14	381.4[a]	351.7[ab]	358.1[ab]	317.9[b]
S3 & S15	368.7[a]	341.0[ab]	300.2[b]	316.0[ab]
S4 & S16	402.9[a]	370.8[a]	360.4[a]	355.0[a]
S5 & S17	407.5[a]	373.9[a]	352.3[a]	347.2[a]
S6 & S18	368.7[a]	335.1[a]	327.1[a]	328.9[a]
S7 & S19	384.2[a]	347.6[a]	356.6[a]	353.3[a]
S8 & S20	370.6[a]	370.0[a]	330.9[a]	333.3[a]
S9 & S21	373.8[a]	368.9[a]	331.2[a]	334.8[a]
S10 & S22	330.3[a]	297.9[a]	315.7[a]	300.7[a]
S11 & S23	324.2[a]	281.1[a]	294.1[a]	330.6[a]
S12 & S24	313.6[a]	281.1[a]	270.8[a]	271.4[a]
Week avg for trial	367.1[a]	342.2[b]	328.1[b]	327.0[b]

[A] Maltose values are averages of set 1 and set 2 reported as mg maltose/g dry matter.

[B] Row means with the same superscript (a,b) are not significantly different ($P<.05$).

[C] See table 5 in appendix A for weekly maltose values for the individual samples.

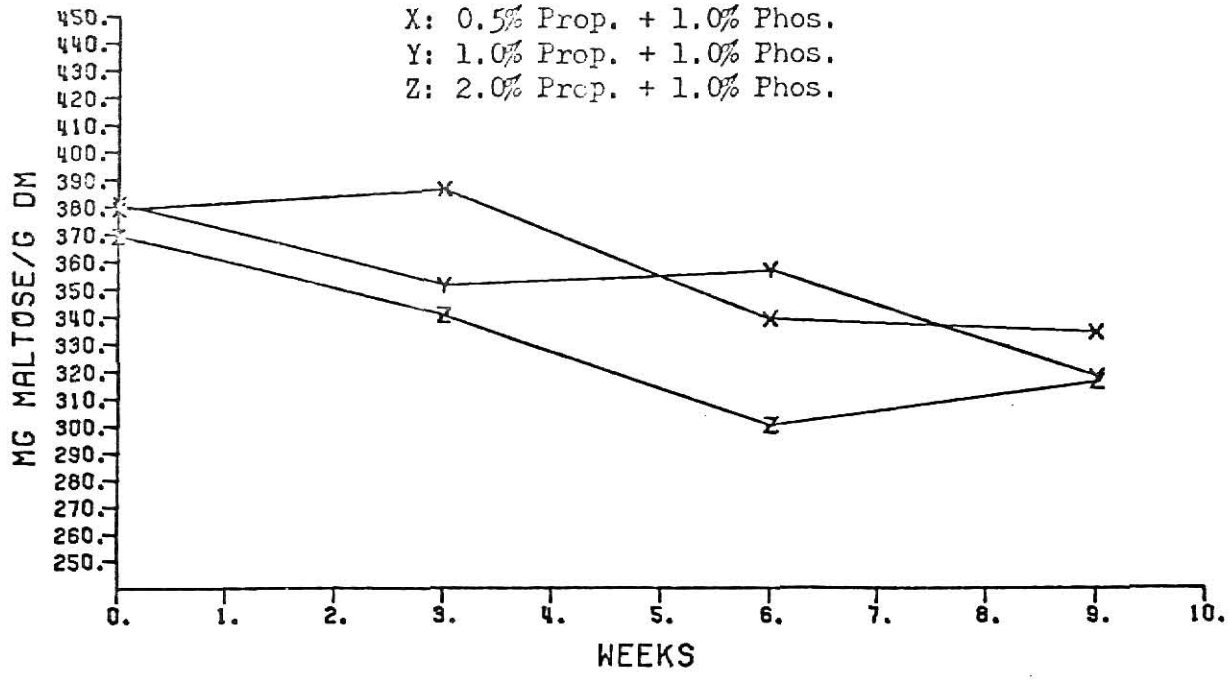

FIGURE 17. MALTOSE DURING STORAGE USING PROPIONIC ACID

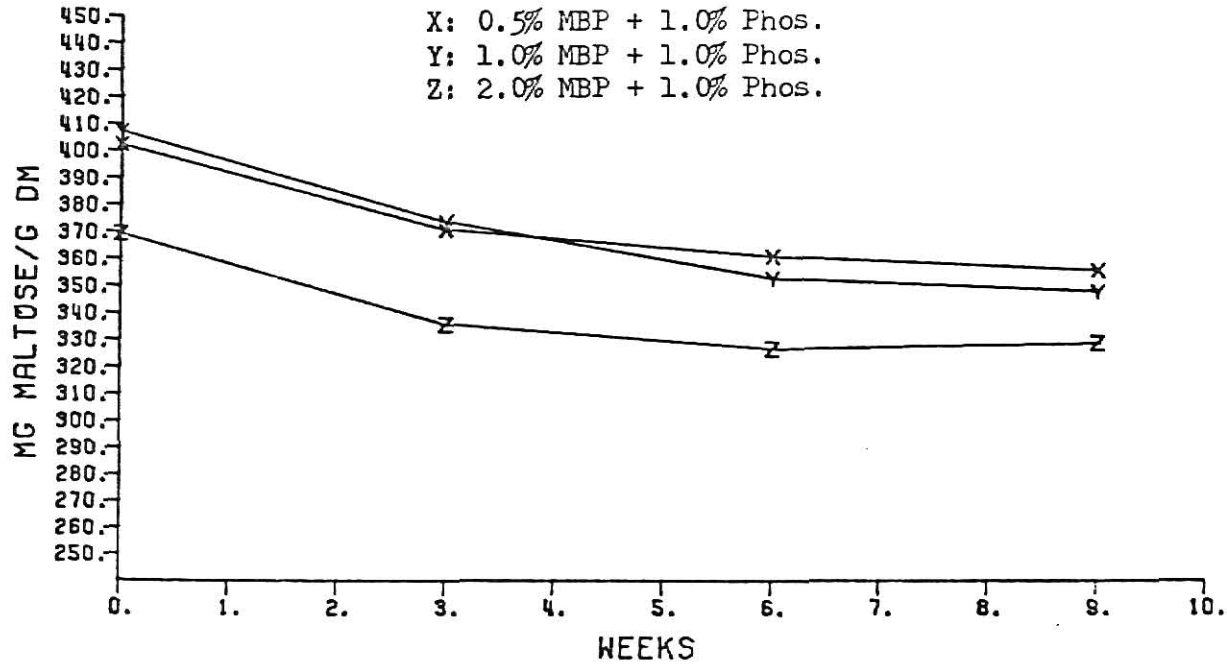

FIGURE 18. MALTOSE DURING STORAGE USING MBP

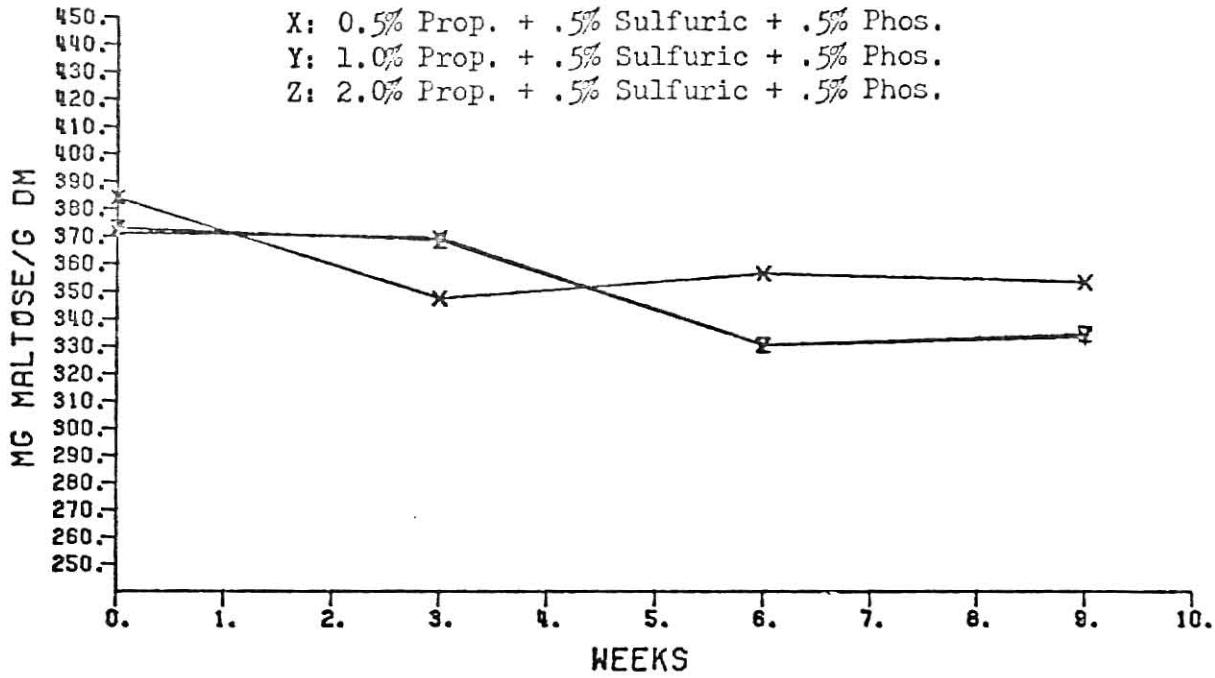

FIGURE 19. MALTOSE DURING STORAGE USING SULFURIC AND PROPIONIC ACID

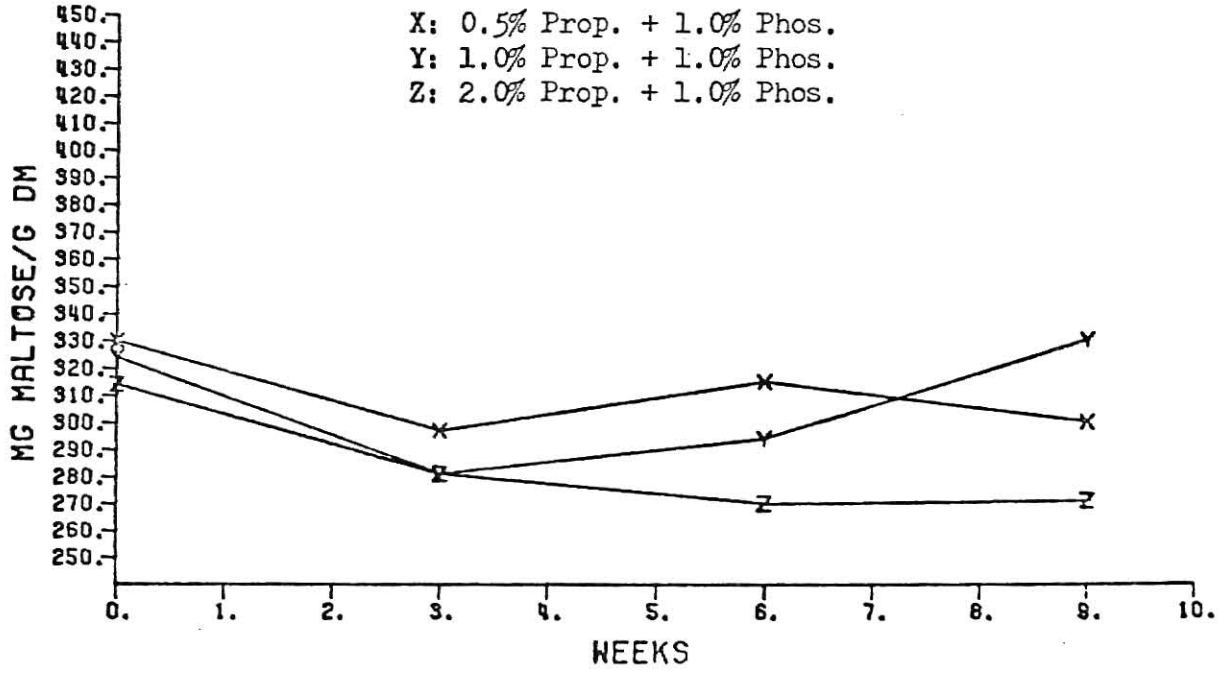

FIGURE 20. MALTOSE DURING STORAGE USING PROP. AT HIGH STARCH LEVEL

1.75:1 starch:urea ratio shows values of maltose lower than with the 1.5:1 ratio by approximately 50 mg/g dry matter. This was due to the amount of reducing sugars present in molasses.

Viscosity. Table 13 shows variations in initial visocity of samples within a sample set ranging from 111 centipoise (cps) to > 200,000 cps. Indications are this may have occurred because of poor mixing of enzyme in batches that were divided into equal parts after enzyme addition. This procedure could be corrected by allowing the supplement to cool, after addition of .5% phosphoric acid, before dividing into separate samples. Protein and dry matter were equivalent between samples, within the sample set, indicating that the dividing procedure used resulted in materials well blended from other aspects.

The large variations in viscosity required use of different spindles and rpm's to obtain cps values. Comparative analysis can only be made between weekly viscosity readings of each sample because the same spindle and rpm were used for each sample, respectively, throughout the trial. Corresponding samples from set 1 and set 2 cannot be compared because rpm's and spindles used were not constant. Table 13 shows increasing viscosity in most of the storage samples over an 8-week period. S1 gradually increased from 7,880 to 12,640 cps during the trial. There was no significant differences in dry matter reported. This upward viscosity trend may be attributed to a gelling of the potato starch in the liquid supplement.

Week Fourteen. The storage trial was extended to 14 weeks using samples with various levels of propionic acid and the 1.5:1 starch:urea ratio (table 14). Values of dry matter, pH and protein were not different from those for week nine. The data for free ammonia indicated an increase of approximately .2 mg of NH_3-N from weeks nine to fourteen. This follows the increase ($P<.05$)

shown from weeks six to nine. However, the protein content remained constant throughout the 14-week trial. Maltose values show approximately a 5 to 10 mg decrease in maltose per gram from weeks nine to fourteen. This indicates that retrogradation of the potato starch is continuing slowly. The 14-week data supports the trends noted in the 9-week trial.

Protein Synthesis. *In vitro* protein synthesis described in Chapter III was run on stored samples to evaluate the effects of the different acid combinations used as preservatives. Daily variations in rumen fluid were adjusted for by converting milligrams protein synthesized to a percentage of the control. Control samples were S1 and S13 for each respective sample set and were assigned values of 100%.

Table 15 shows protein synthesis results for week zero. The data for samples from set 2 are not averaged with set 1. This was because of poor rumen fluid on the day set 2 was run, causing some negative values.

Small portions of each potato liquid supplement samples were placed in cold storage (-4 C) during week zero. This was done to hold samples which would have minimum changes due to microbial activity. These frozen samples were used as experimental controls for liquid supplements stored at room temperature for 8 weeks. Protein synthesis feed blanks (described in protein synthesis of Chapter III) for frozen samples gave identical results to those of week zero. The feed blanks are not run with rumen fluid. Therefore, these results indicate the frozen samples did not change with time. Protein synthesis for frozen samples and samples stored at room temperature for eight weeks are shown in table 15. Also shown, are the averages of the three *in vitro* runs during the storage trial. Analysis of variance was run as previously described.

The results in table 15 show that average protein synthesized from samples treated with 1.0 or 2.0% MBP were lower ($P<.05$) than any other acid

treatment. At the 2.0% addition level, protein synthesis was decreased by approximately 80%. This indicates MBP inhibited microbial activity in the in vitro system. Therefore, MBP is not a good preservative. There were no other significant differences between acid treatments. The highest average protein synthesized was obtained with .5% propionic plus 1.0% phosphoric acid using the 1.5:1 potato starch:urea ratio. All other variations in the protein synthesis averages were considered trends. Table 6 in appendix A shows results for protein synthesis for individual samples. These data are presented as milligrams protein synthesized per gram liquid supplement and as a percentage of the control.

Table 15 also gives a comparison of protein synthesis for weeks zero, eight and frozen samples. A 11.30% difference in percentage protein synthesized within an acid treatment was required for a significant difference ($P<.05$). These data indicate that most acid combinations preserved the liquid supplement for an 8-week period. Samples treated with MBP showed the widest variation. Significant differences ($P<.05$) shown in samples S9, S21 and S11, S23 were marginal cases. This author believes that frozen samples performed similar to samples stored for 8-weeks at room temperature.

These results (table 15) indicate that differences ($P<.05$) in maltose, free ammonia and changes in viscosity had little or no effect on protein synthesis. Protein, pH and dry matter remained constant throughout the investigation, regardless of acid combination utilized. These studies show that a hydrothermally processed potato based liquid supplement can be stored for a 2-month period when preserved with .5% propionic plus 1.0% phosphoric acid. This was also the best treatment when considering the economics of the other acid combinations. The phosphoric acid will be utilized by the animals as a source of phosphorus, while propionic acid can be used as a source of energy and acts as an antifungal agent.

TABLE 13. EFFECT OF STORAGE ON THE VISCOSITY[A] OF POTATO BASED LIQUID SUPPLEMENTS

Sample Set 1	Week 0	3	5	8
S1	7,880	10,180	10,400	12,640
S2	118,000	> 200,000	> 200,000	> 200,000
S3	86,200	125,800	157,000	163,000
S4	107,200	150,600	192,000	> 200,000
S5	120,200	167,200	158,600	> 200,000
S6	103,400	176,000	154,000	155,000
S7	130,000	188,600	196,000	> 200,000
S8	130,600	196,000	334,400	185,200
S9	115,000	169,000	167,000	179,800
S10	520	372	554	346
S11	60,100	81,600	83,600	17,800
S12	50,200	46,300	67,800	12,700
Set 2				
S13	119,400	173,400	196,800	> 200,000
S14	105,200	106,600	188,600	195,000
S15	22,000	> 50,000	> 50,000	> 50,000
S16	157	121	110	107
S17	163	111	96	91
S18	14,680	6,680	5,400	49,000
S19	78,200	152,800	177,600	168,800
S20	94,000	172,800	188,600	186,600
S21	85,600	169,000	168,000	198,400
S22	> 200,000	> 200,000	> 200,000	> 200,000
S23	> 200,000	> 200,000	> 200,000	> 200,000
S24	> 200,000	> 200,000	> 200,000	> 200,000

[A]Viscosity reported as centipoise (cps).

TABLE 14. EFFECTS OF 14-WEEKS OF STORAGE ON POTATO BASED LIQUID SUPPLEMENTS

Sample Set 1	pH	Dry matter %	Protein %[C] as is	db	Free ammonia[A] as is	db	Maltose[B] as is	db
S1	4.15	52.43	29.37	56.01	0.910	1.735	169.4	323.1
S2	4.02	53.52	29.53	55.17	0.871	1.627	161.8	302.3
S3	3.90	53.22	29.63	55.67	0.900	1.691	155.2	291.6
Set 2								
S13	4.00	54.05	29.98	55.47	1.025	1.896	171.6	317.5
S14	4.05	54.04	29.95	55.42	0.863	1.597	163.6	302.7
S15	4.00	52.90	29.67	56.08	0.913	1.726	153.1	289.4

[A]Free ammonia reported as mg NH_3-N/g on "as is" and dry basis.

[B]Maltose reported as mg maltose/g on "as is" and dry basis.

[C]Protein reported on an "as is" and dry basis as a percentage.

TABLE 15. EFFECT OF STORAGE ON IN VITRO PROTEIN SYNTHESIS (REPORTED AS A PERCENTAGE OF THE CONTROL[A])

Sample Set 1 & Set 2	Week[BD] 0	Week[CD] 8	Frozen Samples[CD]	Average[E]
S1 & S13	100[a]	100[a]	100[a]	100[h]
S2 & S14	95[a]	90[a]	93[a]	93[h]
S3 & S15	84[a]	80[a]	82[a]	82[h]
S4 & S16	86[a]	93[a]	90[a]	89[h]
S5 & S17	52[a]	80[b]	91[b]	74[i]
S6 & S18	28[ab]	20[a]	36[b]	28[i]
S7 & S19	97[a]	91[a]	98[a]	95[h]
S8 & S20	94[a]	89[a]	89[a]	90[h]
S9 & S21	79[a]	81[a]	93[b]	84[h]
S10 & S22	85[a]	85[a]	94[a]	88[h]
S11 & S23	80[a]	78[a]	92[b]	83[h]
S12 & S24	79[a]	84[a]	87[a]	83[h]

[A]Control is sample S1 or S13 and assigned a value of 100%.

[B]Protein synthesis on set 2 was not run in week zero. Data reported is only from sample set 1.

[C]Values are reported as averages of the percentage of control from set 1 and set 2.

[D]Row means with the same superscript (a,b) are not significantly different ($P<.05$).

[E]Column means with the same superscript (h,i) are not significantly different ($P<.05$).

[F]See table 6 in appendix A for protein synthesis data of individual samples.

CHAPTER V

COMPARISONS OF POTATO AND CORN BASED LIQUID SUPPLEMENTS

To alleviate the problem of disposing of the waste material, interest developed in channeling potato starch waste to the animal feed industry by incorporating it into a hydrothermally processed liquid supplement.

A liquid supplement was produced to improve nitrogen and carbohydrate utilization by ruminants. The product was produced by blending molasses with a hydrothermally processed starch:urea slurry. Three studies were conducted to determine the effect of using corn or potato starch waste as the carbohydrate source.

Materials and Methods

Study one was a comparison of corn and potato based liquid supplements with different starch:urea ratios. The potato supplement was produced with 1.5:1 and 1.75:1 starch:urea ratios, while the corn supplement was produced with a 2:1 ratio.

Corn was micropulverized before blending with urea using a 2:1 ratio calculated on an "as is" basis. Table 16 shows the corn based liquid supplement formula. Water was added to corn:urea blends to obtain a slurry containing 41% dry matter. The slurry was hydrothermally processed at 149 C and 40 psig. Molasses was added to adjust the protein equivalent to 30%. Four minutes after α-amylase was added, activity was stopped with 1.0% phosphoric acid. Propionic acid was added as a preservative. The enzyme was added at 60 C, while propionic acid was incorporated at 40 C. Samples produced on different days were designated at C1 and C2 (table 17).

The potato based liquid supplement used for comparison with the corn based product were samples S1, S2, S10, S11, S13, S14, S22 and S23 from the

storage trial. These samples were treated with .5% or 1.0% propionic acid. Comparisons were made with both 1.5:1 and 1.75:1 potato starch:urea ratios. The potato based liquid supplement samples in table 17 are designated as they were in the storage trial (Chapter IV).

Phase two of this study compared corn and potato based liquid supplements formulated with a 2:1 starch source:urea ratio. This ratio was calculated on a 90% dry matter basis for both supplements. Table 16 shows these two formulas, with the only variation being starch sources. Processing techniques were used as previously described for the corn based liquid supplements. The only exception was addition of phosphoric and propionic acids 72 hr after addition of α-amylase. Table 19 shows potato samples labeled P1 and P2, while corn is designated as C3 and C4. Sample set 1 and set 2 were processed on different days.

The third phase of this study was designed to determine animal acceptance of corn or potato based liquid supplements. Thirty-three head of crossbred heifers were used for this trial. Their ration consisted of: 6.2 kg alfalfa hay and M-60[1] fed ad libitum. Water was also supplied ad libitum. A two-wheel liq-tank was divided in half, with the corn and potato supplements placed on either side. The ends were reversed every 3 days during the 3-week trial. Average daily consumption for each supplement was calculated from the total consumption for each week. The potato 1.5:1 and corn 2:1 liquid supplement formulas are shown in table 3 of Chapter IV and table 16 of Chapter V, respectively.

Results and Discussion

The potato starch waste offers unique processing characteristics. With a dry matter content of 45%, waste potato starch is a solid material. French

[1]M-60 is a pelleted mill run product produced at the KSU Feed Mill.

TABLE 16. CORN AND POTATO BASED LIQUID SUPPLEMENTS USING A 2:1 STARCH:UREA RATIO

Ingredient	Corn based %	Potato Based %
Corn	18.12	.00
Potato starch (45% dry matter)	.00	36.24
Urea	9.90	9.90
Molasses	12.94	12.94
H_2O (added prior to processing)	35.37	17.25
H_2O (absorbed during processing)	22.17	22.17
Phosphoric acid	1.00	1.00
Propionic acid	.50	.50
Enzyme (α-amylase)	.01	.01

(28) reports when water is used as the solvent, urea can break hydrogen bonding within some origins of starch. When dry urea was mixed with potato starch waste, the viscosity decreased extensively. This allowed for a higher level of solids to be jet cooked than could be achieved with corn as the starch source. Table 17 shows the effect of carbohydrate source on dry matter content of hydrothermally processed liquid supplements. The potato based liquid supplements were produced containing greater than 50% dry matter when crude protein was adjusted to 30% with molasses. The corn based supplement contained 34% dry matter. These data show that dry matter varied with carbohydrate source because of processing limitations.

Table 17 shows pH, dry matter, protein and maltose values for the first phase of this investigation. The protein between samples ranged from 29 to 30%. On a dry basis, protein was 55% for potato based, compared to 86% for corn based supplements. Large variations in maltose produced occurred between samples containing different starch sources and starch:urea ratios. The 1.5:1 ratio for potato based supplement had 380 mg maltose/g dry matter, while the 1.75:1 ratio had 341 mg maltose. The corn based supplement had 185 mg maltose/g dry matter. Variations in maltose were largely due to the amount of molasses required to adjust the protein equivalent of the cooked starch:urea gel to 30%. Therefore, the maltose produced did not indicate starch damage, but rather the amount of readily available carbohydrate per gram of liquid supplement. The pH, regardless of starch origin or starch:urea ratio, ranged from 3.8 to 4.1. This was considered a small variation.

Table 18 shows <u>in vitro</u> protein synthesis for liquid supplements produced with different starch sources. A 1 g liquid supplement sample was used in all fermentations. Results are shown for two <u>in vitro</u> runs, with the corn sample

of each respective set designated as the control and given a value of 100%. Data are presented on an "as is" and dry basis. Note, there is a difference in the amount of cooked starch, dry matter and molasses between supplements produced from different starch sources and/or starch:urea ratios. The statistical analysis of variance used a nested model where effects of samples within treatments were removed from the error. The 1.5:1 potato based supplement on an "as is" basis was significantly ($P<.05$) higher in protein synthesized than 1.75:1 potato or corn supplements. This difference was approximately a 15% improvement. These data indicate on an "as is" basis, the 1.5:1 potato based supplement will be better utilized by the ruminant. When data were corrected for dry matter variations, the results were reversed. The corn based supplement was higher ($P<.05$) in protein synthesized than 1.5:1 or 1.75:1 potato supplements. This difference was approximately 25%. On a dry basis, there was no difference ($P<.05$) between the two potato supplements. However, the 1.5:1 ratio was approximately 5% higher in protein synthesized than 1.75:1. The differences noted between results of protein synthesis reported on an "as is" and dry basis were attributed to the variations in the formulas previously mentioned.

Phase two of this study was designed for a direct comparison of liquid supplements produced from the two starch sources. Formulas from table 16 were used to hydrothermally produce both corn and potato based liquid supplements. Samples in set 1 and set 2 in table 19 were processed on different days. The analytical results are presented in this table. The average dry matter content of potato based samples was 39.4%, while the corn average was 36.5%. There was no difference in pH, regardless of starch source. The crude protein content of samples ranged from 26.5 to 28%.

Maltose data in table 19 show potato based samples produced approximately 65 more milligrams of maltose per gram than the corn supplement. The variance in maltose could be due to corn being approximately 70% starch, while the potato starch was higher in carbohydrate material. A second factor could be that potato granules may have been damaged to a greater degree than starch in the corn, causing it to be more susceptible to β-amylase.

Table 19 shows the in vitro protein synthesis results on an "as is" and dry basis. Results are averages of two runs for protein synthesis data. One-gram samples were used for in vitro fermentations. The potato based supplement averaged 11% more protein synthesized than the corn supplement. Although this variation was not greater ($P<.05$) it indicated that a hydrothermally processed potato supplement can perform equally with the corn supplement.

The third phase of this study was conducted to determine feed acceptance of the two liquid supplements. A 1.5:1 potato supplement was compared to a 2:1 corn supplement. The protein equivalent of the two supplements was 29 to 30%. However, the dry matter of the potato supplement was 17.5% higher than the corn supplement. Results of the trial are shown in table 20. During week one, similar amounts of each supplement were consumed (.295 kg/day potatoes and .275 kg/day corn). Consumption rate declined throughout the 3-week trial. The trial average intake per head of potato liquid supplement was .213 kg on an "as is" basis and .110 kg on a dry basis. The corn intake was lower at .141 kg on an "as is" basis and .048 kg on a dry basis. This indicates that potato liquid supplement was acceptable to the animal. The difference in consumption rates could possibly be due to the potato liquid supplement containing 25% more molasses than the corn supplement.

TABLE 17. ANALYTICAL DATA FOR CORN AND POTATO BASED LIQUID SUPPLEMENTS

Sample Set 1	Starch: urea ratio	pH	Dry matter %	Protein % as is	Protein % db	Maltose mg maltose/ g sample	Maltose mg maltose/ g dm
S[A]1	1.5:1	4.20	52.40	29.21	55.74	200.4	382.4
S2	1.5:1	4.08	53.31	29.31	54.98	201.3	377.6
S10	1.75:1	3.86	48.72	29.02	59.56	169.6	348.1
S11	1.75:1	3.78	49.33	28.58	57.94	164.8	334.0
C[A]1	2:1	3.80	34.10	29.85	84.60	62.9	184.5
Set 2							
S13	1.5:1	3.91	53.78	29.96	55.71	202.2	376.0
S14	1.5:1	4.05	53.72	30.06	55.91	207.0	385.3
S22	1.75:1	3.75	51.70	31.44	60.81	161.6	312.6
S23	1.75:1	3.76	52.20	30.56	58.54	164.2	314.4
C2	2:1	3.89	33.54	29.64	88.37	62.4	186.0

[A]S and C designate potato or corn starch source, respectively.

TABLE 18. IN VITRO PROTEIN SYNTHESIS[A] COMPARISON OF CORN AND POTATO BASED LIQUID SUPPLEMENTS

Sample Set 1	% of corn run 1 as is	run 1 db	run 2 as is	run 2 db	Average of run 1 & run 2[BC] as is	db
S1	108	70	112	73	110	72
S2	96	61	100	64	98	62
S10	94	66	98	68	96	67
S11	95	65	92	64	94	64
C1	100	100	100	100	100	100
Set 2						
S13	132	82	130	81	131	81
S14	121	76	125	78	123	77
S22	89	70	130	84	110	77
S23	80	57	132	85	106	71
C2	100	100	100	100	100	100

[A]Potato values are reported as a percentage of the corn from respective sample sets on "as is" and dry basis.

[B]The mean of S1, S2, S13, S14 is significantly ($P<.05$) higher than means S10, S11, S22, S23 and C1, C2 on an "as is" basis.

[C]The mean of C1, C2 is significantly ($P<.05$) higher than means S1, S2, S13, S14 and S10, S11, S22, S23 on a dry basis.

TABLE 19. CORN AND POTATO BASED LIQUID SUPPLEMENTS WITH EQUAL STARCH SOURCE:UREA RATIOS

Sample Set 1	pH	Dry matter %	Protein %[A] as is	db	Maltose[B] as is	db	Protein synthesis[CD] as is	db
P1	3.4	39.48	26.49	67.10	238.7	604.6	121	112
C3	3.6	37.38	27.67	74.02	174.6	467.1	100	100
Set 2								
P2	3.3	39.27	28.07	71.48	231.3	589.0	112	101
C4	3.4	35.54	28.35	79.77	167.2	470.5	100	100

[A]Protein reported as a percentage on "as is" and dry basis.

[B]Maltose reported as mg maltose/g sample on "as is" and dry basis.

[C]Potato supplement protein synthesis values reported as a percentage of corn from respective sample sets on an "as is" and dry basis.

[D]The protein synthesis mean of C3, C4 were not significantly different ($P<.05$) from the mean of P1, P2 on either an "as is" or dry basis.

TABLE 20. LIQUID SUPPLEMENT CONSUMPTION OF 33 CROSSBRED HEIFERS

Week	Potato liquid supplement[A] kg/week as is	kg/day/head as is	db	Corn liquid supplement[B] kg/week as is	kg/day/head as is	db
1	68.18	.295	.153	63.64	.275	.094
2	54.54	.236	.122	22.73	.098	.034
3	25.00	.108	.056	11.36	.049	.017
Average	49.24	.213	.110	32.66	.141	.048

[A]Potato liquid supplement was a 1.5:1 starch:urea ratio with 29.35% protein and 51.81% dry matter.

[B]Corn liquid supplement was a 2:1 starch:urea ratio with 29.82% protein and 34.32% dry matter.

CHAPTER VI

EFFECTS OF HYDROTHERMAL PROCESSING

A liquid supplement was produced to improve the nitrogen and carbohydrate utilization in ruminants by blending a hydrothermally processed starch source:urea slurry with molasses. This investigation studied the effects of hydrothermal processing and the effect of the addition of α-amylase on a potato starch based urea liquid supplement. A hydrothermally processed potato starch:urea slurry was compared to a raw potato starch:urea slurry when incorporated into a liquid supplement.

Materials and Methods

Potato starch:urea slurries were hydrothermally processed at 149 C and 40 psig. The potato starch:urea ratios of 1.5:1 and 1.75:1 were studied. These formulas are shown in table 3. Molasses was added to the hydrothermally processed slurry to adjust crude protein to 30%. Alpha-amylase was added at a rate of .01% by weight when samples were 60 C. Enzymatic activity was terminated after $5\frac{1}{2}$ min for the 1.5:1 ratio and 9 min for the 1.75:1 ratio with 1.0% phosphoric acid. Propionic acid (.5%) was added, to inhibit mold growth, when liquid supplement samples were 40 C. Table 21 identifies these samples as C+E (hydrothermally processed starch:urea plus enzyme).

Samples in table 21, identified as UC+E (raw potato starch:urea plus enzyme), were processed by mixing raw potato starch with urea and molasses. Tap water was added to the unprocessed samples to adjust for moisture absorption that occurred during the cooking process. Samples were heated in a water bath at 60 C. Enzyme and acids were immediately added as described for the C+E potato liquid supplement samples.

The UC-E (raw potato starch:urea prepared without enzyme) samples in table 21 were prepared by blending raw potato starch with urea, molasses, water (to adjust for moisture absorbed from steam) and the acids. These samples were not heated nor treated with α-amylase.

Data from the liquid supplement samples from the three processing techniques described above are shown in table 21. Samples from set 1 and set 2 were processed on different days. These processing techniques were used to process 1.5:1 and 1.75:1 starch:urea ratios. The effect of hydrothermal processing and enzyme treatment was evaluated mainly with in vitro protein synthesis.

Results and Discussions

Table 21 shows analytical results of potato liquid supplement samples produced with the three different processing techniques. The protein equivalent of the hydrothermally processed samples is consistently 2 to 3% lower than raw potato supplements. Dry matter also varied between cooked and uncooked samples within each starch:urea ratio. These variations in protein and dry matter were due to inaccurate adjustments for moisture absorbed from steam in the jet cooking process. More tap water should have been added to the uncooked samples.

Table 22 shows two runs of in vitro protein synthesis for this study. The C+E sample produced with the 1.5:1 starch:urea ratio, in each respective set, was designated as the control and given a value of 100%. One gram samples were used for all fermentations. Data are presented on a dry basis to adjust for the dry matter variations previously mentioned. This method gave an advantage to the raw samples because of their higher protein content.

The statistical analysis of these data was designed as described in Chapter V. There were no differences ($P<.05$) between C+E samples produced

with the 1.5:1 and 1.75:1 starch:urea ratio. This agrees with results in Chapter V. The samples that were not hydrothermally cooked (with or without α-amylase) gave 25% lower protein synthesis. This decrease is more than two significant ($P<.05$) ranges lower than the jet cooked samples. These results were the same, regardless of potato starch:urea ratio. There were no differences ($P<.05$) between UC+E or UC-E samples. This study shows the hydrothermal processing technique significantly ($P<.05$) improved nitrogen and carbohydrate utilization.

TABLE 21. EFFECT OF HYDROTHERMAL PROCESSING ON POTATO LIQUID SUPPLEMENTS

Sample Set 1	Sample description	Starch:urea ratio	pH	Dry matter %	Protein % as is	Protein % db
Y1	C+E[A]	1.5:1	3.90	52.43	29.37	56.01
Y2	C+E	1.75:1	4.00	48.82	29.88	61.20
Y3	UC+E[B]	1.5:1	3.85	54.20	32.00	59.04
Y4	UC-E[C]	1.5:1	3.90	54.09	31.87	58.92
Y5	UC+E	1.75:1	3.85	51.65	31.94	61.83
Y6	UC-E	1.75:1	3.80	51.05	32.17	63.01
Set 2						
Y7	C+E	1.5:1	3.95	54.05	29.98	55.47
Y8	C+E	1.75:1	3.90	49.27	28.77	58.39
Y9	UC+E	1.5:1	3.85	53.97	31.87	57.05
Y10	UC-E	1.5:1	3.80	53.84	32.23	59.86
Y11	UC+E	1.75:1	3.90	50.21	31.76	63.25
Y12	UC-E	1.75:1	3.95	49.79	31.82	63.91

[A] C+E: jet cooked with enzyme added after cooking.

[B] UC+E: raw potato starch:urea slurry with enzyme added at 60 C.

[C] UC-E: raw potato starch:urea slurry with no enzyme addition.

TABLE 22. IN VITRO PROTEIN SYNTHESIS: EFFECTS OF HYDROTHERMAL PROCESSING ON POTATO LIQUID SUPPLEMENTS

Sample Set 1	% of control[A] Run 1	Run 2	Average[BC]
Y1	100	100	100
Y2	98	112	105
Y3	78	77	77
Y4	73	70	71
Y5	64	85	74
Y6	62	70	66
Set 2			
Y7	100	100	100
Y8	112	87	99
Y9	72	69	70
Y10	75	73	74
Y11	73	63	68
Y12	75	61	68

[A]Percentage of control calculated by giving mg prot syn/g dry matter from sample Y1 and Y7 values of 100%.

[B]Mean of Y1, Y7 are significantly ($P<.05$) higher than means Y3, Y9 and Y4, Y10.

[C]Mean of Y2, Y8 are significantly ($P<.05$) higher than means Y5, Y11 and Y6, Y12.

CHAPTER VII

SUMMARY AND CONCLUSIONS

From the data presented in the previous chapters, several conclusions can be drawn concerning the effects of incorporating potato starch waste into a hydrothermally processed liquid supplement. The hydrothermally processed liquid product contains a starch source, molasses, nonprotein nitrogen (urea), phosphoric acid, propionic acid and water. The potato starch waste was a food by-product.

There are no indications that percentage water absorbed from steam addition during the jet cooking process varied with potato starch:urea ratio. The 1.5:1 and 1.75:1 potato starch:urea ratios showed 21 to 22% of the gel leaving the cooker was moisture absorbed from steam. The retention time in the jet cooker was 16 to 17 seconds.

Storage trial results showed the potato based liquid supplement decreased in maltose produced, while viscosity increased, indicating a retrogradation of the cooked potato starch. During the last three weeks of the trial, free ammonia (NH_3-N) significantly ($P<.05$) increased. Protein, pH and dry matter remained constant throughout the trial, regardless of acid combinations utilized. Methyl-bis propionate inhibited microbial activity in the in vitro system. The highest average quanity of protein synthesized was obtained with .5% propionic plus 1.0% phosphoric acid using a 1.5:1 potato starch:urea ratio. Protein synthesis results showed that this potato liquid supplement could be stored safely for at least 2 months.

The viscosity of the potato starch waste decreased extensively when blended with urea. This allowed a higher level of solids to be processed

than could be achieved with the corn based supplements that were processed similarly. This characteristic allowed a potato liquid supplement to be produced that was greater than 50% dry matter.

The 1.5:1 potato based supplement on an "as is" basis was higher ($P<.05$) in protein synthesized than either the 1.75:1 potato or 2:1 corn liquid supplements. On a dry basis the corn supplement was greater ($P<.05$) in protein synthesized than either potato based supplements. Comparison of a 2:1 corn supplement with a 2:1 potato supplement showed the potato product performed at least equal to, if not better than, the corn supplement in amount of protein synthesized. Maltose values were higher with potato based liquid supplements.

An animal acceptability study showed a preference for the 1.5:1 potato based supplement over the 2:1 corn based supplement. On a dry basis the average daily intake per head was .110 kg for the potato and .048 kg for the corn supplement. The consumption rate of both supplements declined throughout the 3-week trial.

The hydrothermal processing technique used for cooking the potato starch:urea slurry improved ($P<.05$) protein synthesis compared to an unprocessed potato starch:urea slurry. These results were achieved at both the 1.5:1 and 1.75:1 potato starch:urea ratios. The improvement was approximately 25%. This study showed that hydrothermal processing significantly improved urea nitrogen and potato starch utilization.

These data should be of value to both the livestock and potato industry. This product provides a stable liquid supplement that incorporates an inexpensive energy source and helps relieve a waste disposal problem.

LITERATURE CITED

1. A.A.C.C. 1962. Cereal Laboratory Methods (7th Ed.). American Association of Cereal Chemists. St. Paul, Minnesota.

2. Aardvark. Computerized statistical program. Dept. of Statistics, Cont. 209. Kansas State University.

3. Anderson, P. C. 1956. Feed supplements for ruminants. U.S. Patent No. 2,748,001.

4. A.O.C.A. 1975. Official Methods of Analysis (12th Ed.) Association of Official Analytical Chemists. Washington, D.C.

5. Arias, C., W. Burroughs, P. Gerlaugh and R. M. Bethke. 1951. The influence of different amounts and sources of energy upon in vitro urea utilization by rumen microorganisms. J. Anim. Sci. 10:683.

6. Badenhuizen, N. P. 1969. Address presented 54th National Meeting of A.A.C.C. Cer. Sci. Today. Vol. 14.

7. Badenhuizen, N. P. 1965. Occurrence and development of starch in plants. In R. L. Whistler and E. F. Paschall (Ed.). Starch: Chemistry and Technology. Vol. I. Acad. Press. New York, New York.

8. Barker, C. C., E. L. Hirst and G. T. Young. 1940. Linkage between repeating units in the starch molecule. Nature 147:296.

9. Barr, G. W. 1974. In vitro rumen fermentation techniques for studying urea utilization in the bovine rumen. PhD. Thesis. Kansas State University.

10. Bates, F. L., D. French and R. E. Rundle. 1943. Amylose and amylopectin content of starches determined by their iodine complex formation. J. Amer. Chem. Soc. 65:142.

11. Bell, M., C. Willis and C. K. Whitehair. 1953. Value of urea nitrogen in rations containing different carbohydrate feeds. J. Anim. Sci. 12:787.

12. Bloomfield, R. A., E. O. Keasley, D. O. Creach and M. E. Muhrer. 1963. Ruminal pH and absorption of ammonia and UFA. J. Anim. Sci. 22:833. (Abstr.).

13. Bond, J. and T. S. Rumsey. 1973. Liquid molasses-urea or biuret (NPN) feed supplements for beef cattle: Wintering performance, ruminal differences, and feeding patterns. J. Anim. Sci. 37:537.

14. Brautlecht, C. A. 1953. In Starch: Its Sources, Productions and Uses. Reinhold. New York, New York.

15. Burroughs, W., J. Long, P. Gerlaugh and R. M. Bethke. 1950. Cellulose digestion by rumen microorganisms as influenced by cereal grains and protein rich feeds commonly fed to cattle using an artifical rumen. J. Anim. Sci. 9:523.

16. Burroughs, W., F. H. McGuire and C. Cooper. 1968. Attempts to improve liquid and dry all-urea type cattle supplements with fish solubles. Iowa Cattle Feeding Exp. Rep. A. S. Leaflet R116.

17. Carrol, B. and H. C. Cheung. 1964. Determination of amylose sorbition of congo red. In R. L. Whistler and E. F. Paschall (Ed.). Starch: Chemistry and Technology. Vol. IV. Acad. Press. New York, New York.

18. Chalupa, W. 1968. Problems in feeding urea to ruminants. J. Anim. Sci. 27:207.

19. Church, D. C. 1971. Digestive Physiology and Nutrition of Ruminants. D. C. Church publisher, distributed by O and B Books, Corvallis, Oregon 1:30.

20. Church, D. C. 1972. Digestive Physiology and Nutrition of Ruminants. D. C. Church publisher, distributed by O and B Books, Corvallis, Oregon 3:147.

21. Conway, E. J. 1957. Microdiffusion Analysis and Volumetric Error (4th Ed.). Crosby, Lockwood and Sons, LTD. London, England.

22. deHoan, F. A. M. and Zwerman. 1973. Land disposal of potato starch processing waste water in the Netherlands. In Food Processing and Waste Management. Cornell Agricultural Waste Management Conference.

23. Deatherage, W. L., M. M. MacMasters and C. E. Rist. 1955. A partial survey of the amylose content in starch from domestic and foreign varieties of corn, wheat and sorghum, and some other starch bearing plants. Trans. of A.A.C.C. 13:31.

24. Donelson, J. R. and W. T. Yamazalsi. 1962. Note on a rapid method for the estimation of damaged starch in soft wheat flours. Cer. Chem. 39:460.

25. Dronzek, B. L., H. Paulina and W. Bashuls. 1972. Scanning electron microscope of starch from sprouted wheat. Cer. Chem. 49:232.

26. Eipeson, W. E. and K. Paulus. 1973. Investigations on some chemical constituents of potatoes and their influence on the behavior during canning. Potato Research 16:270.

27. Fossum, G. O. 1973. Pretreatment of combined municipal and potato processing wastes. In Food Processing and Waste Management. Cornell Agricultural Waste Management Conference.

28. French, D. 1973. Chemical and physical properties of starch. J. Anim. Sci. 37:1048.

29. Furia, T. E. 1968. Starch in the food industry. In T. E. Furia (Ed.). C.R.C. Handbook of Food Additives. Cleveland, Ohio.

30. Gallant, D., C. Mercier and A. Guilbot. 1972. Electron microscopy of starch granules modified by bacterial α-amylase. Cer. Chem. 49:354.

31. Gracza, R. 1965. Minor constituents of starch. In R. L. Whistler and E. F. Paschall (Ed.). Starch: Chemistry and Technology. Vol. I. Acad. Press. New York, New York.

32. Greenwood, C. T. 1964. Structure, properties and amylolytic degradation of starch. Food Tech. 138:732.

33. Hatch, C. F. and W. M. Beeson. 1972. Effect of different levels of cane molasses on nitrogen and energy utilization in urea rations for steers. J. Anim. Sci. 35:854.

34. Hawkins, E. O. E., J. K. N. Jones and G. T. Young. 1940. The constitution of banana starch. J. Chem. Soc. p. 390.

35. Haworth, W. N., E. L. Hirst and F. A. Isherwood. 1937. Polysaccharides part XXIII. Determination of the chain length of glycogen. J. Chem. Soc. p. 577.

36. Helmer, L. E., E. E. Bartley, C. W. Deyoe, R. M. Myer and H. B. Pfost. 1970. Feed processing, V. Effect of an expansion processed mixture of grain and urea (Starea) on nitrogen utilization in vitro. J. Dairy Sci. 53:330.

37. Hinks, C. E. and A. M. Armishaw. 1975. The nutritive value of cooked potato in liquid and creep diets for early-weaned calves. Anim. Prod. 21:31.

38. Hinks, C. E., D. G. Peers and A. M. Armishaw. 1974. The replacement of skimmed milk by cooked potato flour in milk diets for calves. Anim. Prod. 19:351.

39. Hjermstad, E. T. 1964. Starch gels. In R. L. Whistler (Ed.). Methods in Carbohydrate Chemistry. Acad. Press. New York, New York.

40. Hough, L. and J. N. L. Jones. 1954. The chemical evidence for the structure of starch. In J. A. Radley (Ed.). Starch and Its Derivatives. Chapman and Hall, LTD. London, England.

41. Huber, J. T. 1972. Research on liquid nitrogen supplements for dairy cattle. J. Anim. Sci. 33:166.

42. Jaska, E. 1971. Starch gelatinization as detected by proton magnetic resonance. Cer. Chem. 48:438.

43. Jorgensen, N. A. and H. J. Larsen. 1974. Use of high moisture grains prepared with acids for livestock feeding. University of Wisconsin.

44. Leach, H. W. 1965. Gelatinization of starch. In R. L. Whistler and E. F. Paschall (Ed.). Starch: Chemistry and Technology. Vol. I. Acad. Press. New York, New York.

45. Leach, H. W., L. D. McCowen and T. J. Schoch. 1959. Structure of the starch granule. I. Swelling and solubility patterns of various starches. Cer. Chem. 36:534.

46. Leach, H. W. and T. J. Schoch. 1961. Structure of the starch granule. II. Action of various amylases on granular starches. Cer. Chem. 38:34.

47. Lewis, D., K. J. Hill and E. F. Annison. 1957. Studies on the portal blood of sheep. I. Absorption of ammonia from the rumen of the sheep. Biochem. J. 66:587.

48. Lofgreen, G. P. and K. K. Otagaki. 1960. The net energy of blackstrap molasses for lactating dairy cows. J. Dairy Sci. 43:220.

49. MacMasters, M. M. 1964. Microscopic techniques for determining starch granule properties. In R. L. Whistler and E. F. Paschall (Ed.). Starch: Chemistry and Technology. Vol. IV. Acad. Press. New York, New York.

50. MacMasters, M. M. 1953. The return of birefringence to gelatinized starch granules. Cer. Chem. 30:60.

51. McDonald, I. W. 1948. The absorption of ammonia from the rumen of sheep. Biochem. J. 42:584.

52. Menzies, C. S., D. Richardson, F. H. Baker and R. F. Cox. 1955. Phosphoric acid as a source of phosphorus for ruminants. J. Anim. Sci. 14:1217.

53. Meyer, R. M., E. E. Bartley, C. W. Deyoe and V. F. Colenbrander. 1967. Feed processing. I. Rations effect on rumen microbial protein synthesis and amino acid composition. J. Dairy Sci. 50:1327.

54. Mitchell, H. H. 1947. The mineral requirements of farm animals. J. Anim. Sci. 6:365.

55. Newland, H. W., W. T. Mcgee, G. A. Branaman and L. H. Blakeslee. 1962. Effect of heat-processing and pelleting corn for steers and lambs. J. Anim. Sci. 22:711.

56. Oltjen, R. R., R. J. Sirny and A. D. Tillman. 1962. Purified diets studies with sheep. J. Anim. Sci. 21:277.

57. Pearson, R. M. and J. A. B. Smith. 1943. The utilization of urea in the bovine rumen. 2. The conversion of urea to ammonia. Biochem. J. 37:148.

58. Potter, A. L. and W. Z. Hassid. 1948. Starch I. End groups determination of amylose and amylopectin by periodate oxidation. J. Amer. Chem. Soc. 70:3488.

59. Richardson, D., F. H. Baker, E. F. Smith and R. F. Cox. 1961. Phosphoric acid as a phosphorus source for beef cattle. J. Anim. Sci. 20:522.

60. Robyt, J. F. and F. Dexter. 1967. Multiple attack hypothesis of α-amylase action: Action of porcine pancreatic, human saliva, and aspergillus oryzae α-amylase. Arch. Biochem. Biophys. 122:8.

61. Rundle, R. E. and D. French. 1943. The configuration of starch and the starch-iodine complex. II. Optical properties of the crystalline starch fraction. J. Amer. Chem. Soc. 65:558.

62. Salsbury, R. L., J. A. Hoefen and R. W. Luecke. 1961. Effect of heating starch on its digestion by microorganisms. J. Anim. Sci. 20:569.

63. Samec, M. 1957. Some recent physiochemical studies on starch. J. Polymer Sci. 23:801.

64. Sandstedt, R. M. 1965. Fifty years of progress in starch chemistry. Cer. Sci. Today 10:305.

65. Sandstedt, R. M. and P. J. Mattern. 1960. Damaged starch. Quantitative determination in flour. Cer. Chem. 37:379.

66. Schoch, T. J. 1945. The fractionation of starch. In W. W. Pigman and M. L. Wolfrom (Ed.). Advances in Carbohydrate Chemistry. Vol. I. Acad. Press. New York, New York.

67. Schwimmer, S. and H. K. Burr. 1959. Structure and chemical composition of the potato tuber. In W. Talburt and O. Smith (Ed.). Potato Processing. Avi. Pub. Co. Westport, Connecticut.

68. Seib, P. 1971. Starch gelatinization: Chemical and physical effects. Feedstuffs 42:44.

69. Shaw, J. C., R. R. Robinson, M. E. Senger, S. Lakshmanan and T. K. Lewis. 1959. Production of low fat milk. I. Effect of quality and quantity of concentrate on volatile fatty acid of rumen and on its composition of the milk. J. Nutr. 69:235.

70. Shetty, R. M., D. R. Lineback and P. Seib. 1974. Determining the degree of starch gelatinization. Cer. Chem. 51:364.

71. Smith, O. and C. O. Davis. 1968. Potato processing. In O. Smith (Ed.). Potatoes: Production, Storing, Processing. Avi. Pub. Co. Westport, Connecticut.

72. Sung, A. 1968. Enzymatic evaluation of changes in processed grain. Master's Thesis. Kansas State University.

73. Tillman, A. D. and J. R. Brethour. 1958. Dicalcium phosphate and phosphoric acid as phosphorus sources for beef cattle. J. Anim. Sci. 17:100.

74. Tillman, A. D. and K. S. Sidhu. 1969. Nitrogen metabolism in ruminants: Rates of ruminal ammonia production and nitrogen utilization by ruminants - A review. J. Anim. Sci. 28:689.

75. Treadway, R. H. 1947. Industrial utilization of cull and surplus potatoes. Amer. Potato J. 24:361.

76. Watson, S. A. 1964. Determination of the rate of starch retrogradation. In R. L. Whistler (Ed.). Methods of Carbohydrate Chemistry. Acad. Press. New York, New York.

77. Weber, G. R. and F. D. Miller. 1965. Prevention of gel formation. U.S. Patent No. 3,165,143.

78. Whittemore, C. T., I. W. Maffat and J. Mitchell-Manson. 1974. Performance of broilers fed on diets containing cooked potato flakes. Br. Poultry Sci. 15:225.

79. Whittemore, C. T., A. G. Taylor and P. Crooks. 1974. The nutritive value for young pigs of cooked potato flakes in comparison with maize meal. J. Agric. Sci., Camb. 83:1.

80. Whittemore, C. T., A. G. Taylor, F. W. H. Ensley. 1973. The influence of processing upon the nutritive value of the potato: Digestibility studies with pigs. J. of the Sci. of Food and Agric. 24:539.

81. Wiggins, L. F., and S. Augustine. 1958. Feed supplements for ruminants. U.S. Patent No. 2,853,385.

82. Wilcox, R. A., C. W. Deyoe, D. B. Sauer and K. K. Bolsen. 1972. Grain preservatives. Cooperative Extension Service. Kansas State University.

83. Wills, C. G. 1973. Government policies relating to water pollution control. In Food Processing and Waste Management. Cornell Agricultural Waste Management Conference.

84. Woods, R. 1947. Mineral review 1. Borden's Rev. Nutr. Research 8:4.

85. _____. 1969. Liquid supplements for livestock feeding. Chas. Pfizer and Co.

86. _____. 1948. Summary of papers presented at the potato conference held at the eastern regional research laboratory. Potato Research 26:240.

APPENDIX A

TABLE 1. STORAGE TRIAL: pH

Sample Set 1	Week 0	3	5	8
S1	4.20	4.09	4.21	4.10
S2	4.08	3.90	4.19	4.08
S3	4.10	3.90	3.70	4.10
S4	4.06	3.98	3.85	4.09
S5	4.00	4.04	3.83	4.06
S6	3.96	4.00	3.85	4.01
S7	3.90	3.90	3.86	4.08
S8	3.92	3.95	3.86	4.00
S9	3.90	3.91	3.90	4.00
S10	3.86	3.89	4.20	4.15
S11	3.78	3.70	3.88	4.00
S12	3.72	3.78	3.88	4.00
Set 2				
S13	3.91	3.74	4.02	3.99
S14	4.05	4.00	4.01	4.08
S15	3.97	3.80	4.05	4.11
S16	3.98	4.00	4.01	4.11
S17	3.94	3.82	4.06	4.15
S18	3.98	3.79	4.06	4.10
S19	3.96	3.84	4.09	4.04
S20	3.98	3.79	4.04	4.10
S21	4.02	3.77	4.03	4.08
S22	3.75	3.83	3.88	3.91
S23	3.76	3.85	3.78	3.72
S24	3.76	3.75	3.80	3.69

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TABLE 2. STORAGE TRIAL: PERCENTAGE DRY MATTER[A]

Sample Set 1	Week 0	3	6	9
S1	52.40	53.31	51.66	52.75
S2	53.31	55.02	53.18	53.36
S3	53.43	55.82	53.36	53.12
S4	54.13	55.78	53.62	53.73
S5	53.85	55.09	53.38	53.98
S6	53.76	55.80	52.65	53.14
S7	54.90	56.51	53.27	53.85
S8	54.11	50.46	53.34	53.60
S9	53.90	50.23	53.19	53.88
S10	48.72	50.21	48.19	48.82
S11	49.33	49.46	50.06	50.05
S12	49.68	49.15	49.36	51.04
Set 2				
S13	53.78	51.85	55.77	54.15
S14	53.72	52.07	56.32	54.35
S15	53.19	51.26	56.01	53.76
S16	52.11	51.93	53.28	51.61
S17	51.54	51.56	52.31	51.95
S18	53.26	52.39	53.52	52.80
S19	53.94	54.89	54.40	53.39
S20	54.22	52.13	56.79	54.03
S21	53.91	52.39	58.94	54.11
S22	51.70	51.34	56.36	52.96
S23	52.20	50.67	54.84	53.92
S24	52.00	50.32	56.09	51.60

[A]Dry matter is average of duplicate determinations.

TABLE 3. STORAGE TRIAL: PERCENTAGE CRUDE PROTEIN[A]

Sample Set 1	Week 0		3		6		9	
	as is	db	as is	db	as is	db	as is	db
S1	29.21	55.74	29.16	54.70	28.81	55.77	29.09	55.15
S2	29.31	54.98	29.34	53.33	29.17	54.85	29.60	55.47
S3	28.96	54.20	28.91	51.79	28.89	54.14	29.42	55.38
S4	29.69	54.85	29.27	52.47	29.52	55.05	29.76	55.38
S5	29.26	54.34	29.07	52.77	29.10	54.51	29.74	55.09
S6	28.96	53.87	28.84	51.68	28.89	54.87	28.57	53.76
S7	29.92	54.50	29.44	52.10	29.92	56.17	30.42	56.49
S8	29.13	53.83	29.14	57.75	29.40	55.12	29.83	55.65
S9	29.40	54.55	28.77	57.28	28.95	54.43	29.84	55.38
S10	29.02	59.56	28.86	57.49	28.97	60.12	29.88	61.20
S11	28.58	57.94	28.87	58.37	28.57	57.07	30.13	60.20
S12	28.73	57.83	28.33	57.64	28.39	57.52	29.14	57.09
Set 2								
S13	29.96	55.71	29.12	56.16	30.99	55.57	30.88	57.03
S14	30.05	55.91	30.06	57.73	31.37	55.70	31.15	57.31
S15	29.62	55.69	29.08	56.73	31.18	55.67	29.89	55.60
S16	29.77	57.13	29.50	56.81	30.88	56.32	30.28	58.67
S17	29.88	57.97	29.22	56.67	30.49	58.29	30.43	58.57
S18	29.99	56.31	29.34	56.00	31.21	58.31	29.83	56.50
S19	31.06	57.58	30.18	54.98	30.70	56.43	30.10	56.38
S20	31.14	57.43	29.62	56.82	30.13	53.05	30.32	56.12
S21	30.41	56.41	29.36	56.04	32.66	55.41	30.37	56.13
S22	31.44	60.81	28.85	56.19	30.37	53.89	29.86	56.38
S23	30.56	58.54	28.76	56.75	29.32	53.46	30.08	55.79
S24	29.94	57.58	28.54	56.71	29.33	52.29	29.74	57.41

[A]Crude protein are averages of duplicate determinations reported on "as is" and dry basis.

TABLE 4. STORAGE TRIAL: FREE AMMONIA[A]

Sample Set 1	Week 0		3		6		9	
	as is	db	as is	db	as is	db	as is	db
S1	0.540	1.029	0.627	1.175	0.606	1.173	0.720	1.365
S2	0.589	1.104	0.667	1.213	0.569	1.070	0.718	1.345
S3	0.567	1.060	0.643	1.151	0.580	1.086	0.695	1.308
S4	0.567	1.047	0.605	1.084	0.599	1.116	0.708	1.317
S5	0.553	1.028	0.601	1.091	0.593	1.111	0.726	1.344
S6	0.578	1.076	0.579	1.035	0.603	1.143	0.696	1.310
S7	0.595	1.083	0.589	1.043	0.653	1.226	0.744	1.381
S8	0.556	1.008	0.617	1.222	0.600	1.124	0.724	1.351
S9	0.544	1.008	0.613	1.220	0.632	1.189	0.727	1.350
S10	0.485	0.983	0.553	1.101	0.555	1.152	0.658	1.348
S11	0.489	0.991	0.536	1.083	0.557	1.112	0.656	1.310
S12	0.417	0.839	0.511	1.040	0.503	1.018	0.634	1.242
Set 2								
S13	0.593	1.102	0.571	1.101	0.652	1.170	0.677	1.249
S14	0.579	1.077	0.607	1.166	0.628	1.116	0.703	1.293
S15	0.566	1.064	0.585	1.142	0.605	1.080	0.674	1.254
S16	0.571	1.095	0.612	1.179	0.657	1.234	0.676	1.310
S17	0.570	1.105	0.574	1.113	0.629	1.203	0.686	1.321
S18	0.575	1.080	0.547	1.045	0.596	1.114	0.734	1.389
S19	0.627	1.116	0.560	1.093	0.646	1.187	0.717	1.343
S20	0.636	1.224	0.559	1.072	0.630	1.115	0.752	1.392
S21	0.593	1.099	0.626	1.195	0.621	1.105	0.706	1.304
S22	0.525	1.016	0.546	1.063	0.569	1.010	0.668	1.262
S23	0.535	1.024	0.565	1.116	0.563	1.026	0.683	1.266
S24	0.549	1.056	0.548	1.077	0.522	0.930	0.691	1.334

[A]Free ammonia reported as mg NH_3-N/g sample reported on "as is" and dry basis.

TABLE 5. STORAGE TRIAL: MALTOSE[A] PRODUCED

Sample Set 1	Week 0		3		6		9	
	as is	db	as is	db	as is	db	as is	db
S1	200.4	382.4	187.4	351.5	177.0	342.6	177.7	336.8
S2	201.3	377.6	192.7	350.2	172.0	323.4	172.7	323.6
S3	194.7	364.4	185.8	332.8	165.0	309.2	170.0	320.0
S4	199.6	368.7	189.2	339.2	172.0	320.8	176.7	328.9
S5	199.4	370.3	183.4	332.9	171.0	320.3	170.3	315.5
S6	199.6	371.3	178.0	319.0	161.0	305.8	171.9	323.5
S7	205.6	374.5	192.1	340.0	180.0	338.0	187.6	348.4
S8	204.7	378.3	191.4	379.3	176.0	329.9	173.3	323.3
S9	201.9	374.6	186.4	371.1	177.6	333.9	180.1	334.3
S10	169.6	348.1	154.0	306.7	177.6	368.2	160.3	328.3
S11	164.8	334.1	140.0	283.0	158.8	317.2	143.4	286.5
S12	160.2	322.5	136.0	277.0	142.0	287.7	143.5	281.1
Set 2								
S13	202.2	376.0	219.0	422.4	187.9	336.9	179.6	331.7
S14	207.0	385.3	184.0	353.3	221.2	392.8	169.7	312.7
S15	198.4	373.0	179.0	349.2	163.1	291.2	167.8	312.1
S16	227.8	437.1	209.0	402.5	213.1	400.0	196.7	381.1
S17	228.7	444.7	214.0	415.0	201.1	384.4	196.9	379.0
S18	195.0	366.1	184.0	351.2	186.5	348.5	176.5	334.3
S19	212.5	394.0	195.0	355.3	204.1	375.2	191.3	358.3
S20	196.6	363.0	188.1	360.8	187.5	331.9	185.5	343.3
S21	201.1	373.0	192.1	366.7	193.6	328.5	181.5	335.4
S22	161.6	312.6	148.5	289.2	148.4	363.3	144.7	273.2
S23	164.2	314.4	141.6	279.4	148.7	271.1	142.3	274.7
S24	158.5	304.8	143.5	285.2	142.4	253.9	135.5	261.6

[A]Maltose reported as mg maltose/g sample on "as is" and dry basis.

TABLE 6. STORAGE TRIAL: IN VITRO PROTEIN SYNTHESIS

Sample Set 1	Mg prot syn/g (week 8)	% of S1	Mg prot syn/g (frozen samples)	% of S1
S1	15.34	100	15.34	100
S2	13.70	89	13.70	89
S3	12.60	82	14.25	93
S4	14.25	93	15.89	103
S5	14.80	96	16.44	107
S6	4.93	32	7.12	46
S7	18.85	123	16.44	107
S8	16.44	107	15.34	100
S9	14.25	93	14.80	96
S10	13.40	87	13.40	87
S11	13.48	88	12.60	82
S12	16.44	107	13.70	89
Set 2		% of S13		% of S13
S13	20.27	100	19.94	100
S14	18.63	92	19.17	96
S15	15.89	78	14.25	71
S16	18.63	92	15.34	77
S17	12.69	63	15.01	75
S18	1.86	9	4.93	25
S19	12.05	59	17.75	89
S20	14.25	70	15.34	77
S21	13.69	68	17.75	89
S22	16.53	82	19.94	100
S23	13.69	68	20.27	102
S24	12.27	61	16.98	85

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UTILIZATION OF WASTE POTATO STARCH IN UREA CONTAINING LIQUID SUPPLEMENTS

by

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Hydrothermally processed starch source:urea mixtures were used in liquid supplements to improve nitrogen utilization by ruminants. Starch sources used were corn and potato starch obtained from a potato chip waste stream. The starch source was blended with urea and water using a specific starch source:urea ratio. This slurry was hydrothermally processed at 141 C and 40 psig. Phosphoric and propionic acids were added to lower pH and act as an antifungal agent, respectively. Investigations were evaluated by crude protein, dry matter, pH, viscosity, NH_3-N, maltose produced with β-amylase and in vitro protein synthesis.

Effect of storage on potato based liquid supplements was studied using 1.5:1 and 1.75:1 potato starch:urea ratios. Various blends of propionic acid, phosphoric acid, methyl-bis propionate and sulfuric acid were used to stabilize the supplement. Protein, pH and dry matter remained constant ($P<.05$) for eight weeks, regardless of the acid combination used. Maltose produced by β-amylase decreased during the trial indicating retrogradation of the cooked potato starch may have occurred. Between weeks 6 and 9, NH_3-N increased ($P<.05$). In vitro protein synthesis results indicated the liquid supplement could be stored for at least 2 months. The highest average protein synthesized was obtained with .5% propionic plus 1.0% phosphoric acid and a 1.5:1 potato starch:urea ratio. Methyl-bis propionate at levels of 1.0 and 2.0% inhibited microbial activity. The storage trial was extended to 14 weeks using potato liquid supplement samples treated with .5% propionic plus 1.0% phosphoric and the 1.5:1 starch:urea ratio. Crude protein, dry matter and pH were not different after week nine. Ammonia-nitrogen and maltose followed trends from linearity noted in the first 9 weeks.

The hydrothermal process injects live steam with the starch:urea slurry. Both 1.5:1 and 1.75:1 potato starch:urea ratios had 21 to 22% moisture leaving

the cooker as moisture absorbed from steam. Retention time in the jet cooker, measured with dye, was 16 to 17 seconds.

The potato starch waste is a solid material containing 45% dry matter. When blended with urea, the viscosity of the potato starch was extensively decreased. This allowed a potato based liquid supplement to be produced that exceeded 50% dry matter using the 1.5:1 ratio.

Hydrothermal processed liquid supplements, using potato starch and corn as the carbohydrate sources, were compared using in vitro protein synthesis. The 1.5:1 potato based supplement on an "as is" basis was higher ($P<.05$) in protein synthesized than either the 1.75:1 potato or 2:1 corn liquid supplements. On a dry basis the corn supplement was greater ($P<.05$) in protein synthesized than either potato based supplements due to differences in dry matter content. Comparison of a 2:1 corn supplement with a 2:1 potato supplement showed the potato product performed at least equal to, if not better than, the corn liquid supplement in amount of protein synthesized.

An animal acceptability study, using 33 head of crossbred heifers, indicated a preference for the 1.5:1 potato based supplement over the 2:1 corn liquid supplement. On a dry basis, average daily intake per head, from a two-wheel liq-tank, was .110 kg for the potato and .048 kg for the corn supplement. The consumption rate of both supplements declined throughout the 3-week trial.

The effect of the hydrothermal processing technique was determined using in vitro protein synthesis. Comparisons of raw potato based supplement with the hydrothermally processed potato based supplements were made with both 1.5:1 and 1.75:1 starch:urea ratios. The hydrothermal processing technique improved ($P<.05$) protein synthesis compared to the unprocessed supplement by approximately 25%. This study showed the hydrothermal processing

technique significantly improved urea nitrogen and potato starch utilization by rumen microorganisms.