# DIAMMONIUM PHOSPHATE IN DAIRY CATTLE RATIONS

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# INTRODUCTION

In animal production, feed costs may constitute as much as half the cost of production; thus, purchase of nutrients is important from the standpoint of controlling production cost and maximizing returns. Protein, a relatively expensive nutrient, must be purchased and fed with scrutiny if optimum production efficiency is to be obtained.

Ruminants have a somewhat unique protein requirement in that non-protein nitrogen (NPN) compounds are utilized by rumen bacteria for microbial synthesis of proteins. Non-protein nitrogen supplements offer, in many situations, a favorable economical advantage over other sources of protein. Urea may be used to lower feed cost without significantly lowering productiveness of the ration; however, returns from urea supplementation depend on a number of factors. Several factors are (a) the level at which urea is substituted in the ration, (b) the cost of urea relative to various feed proteins, and (c) the cost of grain. Other NPN sources which have been investigated are (a) ammoniated industrial products, (b) biuret, (c) diammonium phosphate, (d) creatine and (e) other ammonia-containing compounds.

The potential of diammonium phosphate (DAP) has recently created interest in the feed industry. The present study was prompted by the fact that palatability problems were encountered when lactating cows were fed rations containing 2% DAP.

The objectives of the research reported herein were to study the acceptability and palatability of rations containing various levels and types of DAP. Further study was conducted to measure the relative amounts of ammonia release from several types of DAP under different conditions.

# LITERATURE REVIEW

## Ruminant Utilization of Non-Protein Nitrogen

Ruminant utilization of NPN was studied by Wegner et al. (39) in 1940. They reported an increase in protein precipitate upon incubation of artificially buffered rumen fluid with inorganic nitrogen. Since then, much convincing evidence has been published indicating that protein can be synthesized from NPN by rumen microorganisms (2, 8, 25, 26). Loosli et al. (20) reported that 10 amino acids were synthesized by rumen microorganisms in vitro from a purified diet which contained NPN as the single source of dietary nitrogen. Only minute traces of 9 amino acids were present in the purified diet.

There is evidence that much microbial protein may be synthesized from ammonia (2, 25, 26) and other metabolites such as fatty acids (2, 11). Workers have shown that a portion of ruminal ammonia is derived from dietary protein upon degradation by microorganisms. Bladen et al. (1) found that 28% of 271 strains of bacteria from the rumen produce ammonia from protein. In general, workers have reported that large amounts of ruminal ammonia are produced when some NPN compounds are fed; however, Oltjen et al. (24) and Russell et al. (29) have shown that the amount of ammonia produced in vivo varies greatly for different NPN supplements.

Repp et al. (28) reported that rumen microorganisms do not elaborate sufficient enzymes to degrade adequately all sources of NPN. Their data were supported by other workers who investigated several ammoniated products as NPN supplements (6, 35, 36).

Commercial grade DAP has been considered as an NPN supplement because of its ammonium content and low cost of production.

## Production of Commercial Ammonium Phosphates

Ammonium phosphates, DAP and monoammonium phosphate (MAP), have been produced for many years as sources of phosphate for commercial fertilizers (38). Most commercial processes have depended on industrial sources of ammonia or anhydrous ammonia for reaction with phosphoric acid. In general, MAP or DAP was produced by changing the molar ratio of ammonia to phosphoric acid in a reaction slurry while maintaining other critical factors such as reaction temperature and removal of precipitated ammonium phosphate. Both MAP (5) and DAP (5, 9, 10, 12, 19, 24, 29, 30, 31, 34) have been investigated as a NPN supplement.

## Economics of Feeding DAP in Ruminant Rations

Little work has been reported on the economical aspects of DAP feeding. Work at the Montana station (34) indicated that DAP compares favorably with urea when it replaces the same percentage of crude protein as replaced by urea. The DAP and urea were substituted in supplemental rations which were fed to fattening heifers. No significant difference between feed costs per hundred weight of gain was reported. Cowman et al. (5) indicated that there was no economic difference between urea and DAP when used in a protein supplement for wintering and fattening steers (DAP actual cost = $160/ton). Arizona workers (9) conducted an experiment with fattening steers and found no advantage in replacing 50% of the cottonseed meal-nitrogen in a fattening ration with urea or DAP.

The f.o.b. price of DAP varies considerably ($95 - $110/ton) depending on place of purchase (27). The current price of DAP, shipped to Stillwater, is approximately $120 per ton (23).

Biological Value of DAP

Non-Protein Nitrogen: In general, DAP nitrogen has been shown to be about equal to urea nitrogen for producing weight gain when isonitrogenous amounts were fed. Thomas et al. (34) obtained approximately equal weight gains when DAP replaced urea in a supplemental ration for fattening heifers. Other workers (5) at the Montana station reported similar results when steers were fed a wintering and fattening supplement containing DAP. The steers were wintered for 112 days and fattened for 168 days. Hale et al. (9) reported that neither urea nor DAP was better than cottonseed meal as a protein supplement. Russell et al. (29) showed nitrogen retention from rations which contained either urea or DAP to be 4.4 and 6.8 grams of nitrogen, respectively, for a 5-day collection period. Sheep used in the nitrogen balance study were fed rations which contained approximately 31% of the protein equivalent in the form of NPN. Oltjen et al. (24) fed urea and DAP in separate rations and supplied 62% of the dietary nitrogen as NPN. The rations were fed to 85-pound lambs and nitrogen balance studies were conducted. They found no significant difference in per cent digestibility between the urea and DAP rations. Retention of nitrogen was greater ($P < 0.01$) for the urea ration when compared to the DAP ration. No explanation was given for the greater urinary loss attributed to the DAP ration. Lassiter et al. (19) found no apparent difference in growth rate of dairy heifers when fed either DAP or urea. Approximately 20% of the dietary nitrogen was supplied as DAP, while 33% of dietary nitrogen was supplied as urea. In one trial reported by the Michigan workers (19), DAP appeared to decrease digestibility of dietary nitrogen; however, they stated that results were biased because of palatability problems with the ration containing DAP. In a second trial, a ration containing both urea

and DAP was compared to rations which contained only urea or DAP as an NPN source. In each case NPN furnished 43% of the dietary nitrogen. Nitrogen retention was significantly greater ($P < 0.05$) for the ration which contained both urea and DAP. No significant difference was found between the rations which contained only urea or DAP as the NPN supplement. The results were confounded slightly because of two extremely low retention values for the urea ration.

Phosphorus: The biological value of DAP phosphorus has been found to be high. In six experiments with chicks, DAP had a higher biological value than tricalcium phosphate (standard) or dicalcium phosphate (13, 14, 15, 16, 17, 18).

Cowman et al. (5) reported no significant difference in blood phosphorus levels of beef cattle when supplemental phosphorus was supplied as DAP instead of defluorinated phosphate. Oltjen et al. (24) fed rations containing either urea or DAP and found no significant difference in phosphorus retention, when expressed as per cent of intake. The ration dry matter contained 0.27 and 1.25% phosphorus, respectively. In a later trial, they (24) found no significant difference in phosphorus retention (expressed as per cent of intake) when DAP, in place of a mineral mix, furnished 94% of dietary phosphorus for beef steers.

## Toxicity of DAP

Toxicity has been a factor of importance in feeding NPN. Diammonium phosphate has been found to be much less toxic to ruminant animals than urea when compared on an isonitrogenous basis. Russell et al. (29) found that in order to produce mortality losses in 80-pound lambs, approximately twice as much DAP-nitrogen as urea nitrogen was required. Blood levels of

ammonia were much higher for animals dosed with urea. Oltjen et al. (24) reported that urea was approximately twice as toxic as DAP when an equal amount of nitrogen was supplied by each compound. Blood levels of ammonia in urea-dosed animals were about twice as high as ammonia levels in the DAP-dosed animals. In all cases (24, 29), ruminal pH was higher for the animals which were dosed with urea. Russell et al. (29) suggested that lower pH values observed for DAP-dosed animals might be due, in part, to buffering by the soluble phosphate.

Palatability of Rations Containing DAP

Palatability problems have been encountered when DAP was fed at various levels (9, 19, 24, 31). In 1946, Shaw et al. (31) conducted palatability trials and found that cows would consume rations which contained 1% DAP. Low feed consumption and, in some cases, complete feed refusal were noted at the Oklahoma station when lactating dairy cows were fed rations containing 2% DAP (37). Michigan workers (19) made similar observations in 1962 when lactating dairy cows were fed a ration that contained 2% DAP; however, the cows seemed to adjust after having received a ration containing 1% DAP. When the cows were first offered a 2% DAP ration, four out of nine cows exhibited lower feed consumption. After the cows had consumed rations which contained various amounts of DAP, they were fed a ration containing 4% DAP. Upon receiving the 4% DAP ration, two out of nine cows exhibited palatability problems. Young dairy heifers were fed DAP at a 3% level with no apparent palatability problem (19).

Oltjen et al. (24) reported decreased feed consumption by sheep which were fed DAP at 2 and 5% levels. They also noted that the sheep were able to sort ration components, leaving DAP in the feed boxes. Poor

palatability was attributed to ammonia release from the DAP. Further study indicated that contact of DAP with saliva or water would release 30 and 10 %, respectively, of the ammonia contained therein. The length of time required to release the respective amounts was not reported; however, it was reported that approximately 50% of the DAP ammonia was lost as a result of pelleting.

Publications from the Montana station (5, 34) made no mention of palatability problems with beef cattle when DAP was fed in supplemental rations. In one trial, DAP constituted 5% of the supplement.

## EXPERIMENTAL PROCEDURE

In order to meet the objectives of this study, three trials were conducted. Reagent grade DAP (RGDAP) and several types of commercial grade DAP were used. The types of commercial DAP were (a) regular DAP (RDAP), a product marketed prior to this study; (b) stabilized DAP (SDAP-I), an experimental product at the time of initiation of this study; and (c) a stabilized product (SDAP-II), which was obtained after the initiation of this study.

Two trials with nonlactating cows were conducted to investigate the acceptability of rations containing various types (RDAP, SDAP-I and SDAP-II) and levels of DAP. A third trial with lactating cows was conducted to measure the palatability of rations which contained various levels of SDAP-II. In each trial, laboratory analyses were performed in addition to the feeding experiments.

### TRIAL I

#### Selection of Cows and Assignment of Treatments

Twenty nonlactating dairy cows were obtained from the Oklahoma State University dairy herd as cows in the milking herd were turned dry. The cows were grouped, according to breed, into blocks of four cows each; however, no other consideration was given to breed of animal. There were 12 Ayrshire, 4 Guernsey and 4 Holstein cows used in Trial I.

The cows within each group were assigned randomly to a 4 x 4 balanced Latin square design (Table I) which permitted estimation of carry-over

effects (3). Each cow was fed the various experimental rations in a sequence determined by the Latin square to which that particular cow had been assigned.

TABLE I

TREATMENT SEQUENCES FOR LATIN SQUARE--TRIAL I

| Period | Treatment Sequences | | | |
|---|---|---|---|---|
| 1 | A | B | C | D |
| 2 | B | C | D | A |
| 3 | D | A | B | C |
| 4 | C | D | A | B |

Experimental periods were of 10 days' duration; therefore, the trial required 40 days plus the period allocated for standardization purposes. The primary response criterion for Trial I was pounds of the respective experimental ration consumed.

Composition and Handling of Rations

All rations were mixed by Stillwater Milling Company, Stillwater, Oklahoma. The control ration, which contained no DAP, was mixed first to insure that the ration would contain no contamination of DAP from the other experimental rations. A pair of mixers was available which allowed the ration containing RDAP to be mixed entirely separate from rations containing SDAP-I. When it was necessary for one experimental ration to follow another in the mixing sequence, the ration containing the smaller amount of DAP always preceded the ration containing a greater amount of DAP. Each major individual constituent of the rations was taken from a single

storage bin. The rations (Table II) were fed in loose form. Brightly colored tags were attached to the feed sacks to identify the rations rapidly and accurately.

TABLE II

COMPOSITION OF RATIONS--TRIAL I

| Ingredient | Ration | | | |
|---|---|---|---|---|
| | Control | 1.5% stabilized DAP-I | 3% stabilized DAP-I | 3% regular DAP |
| | (lb/ton) | | | |
| Sorghum grain, finely ground | 1300 | 1300 | 1300 | 1300 |
| Corn, ground | 140 | 190 | 240 | 240 |
| Wheat shorts | 200 | 200 | 200 | 200 |
| Molasses, liquid | 100 | 100 | 100 | 100 |
| Salt | 20 | 20 | 20 | 20 |
| Vitamin-trace mineral mix | 20 | 20 | 20 | 20 |
| Cottonseed meal, 41% | 200 | 120 | 40 | 40 |
| DAP, stabilized-I | -- | 30 | 60 | -- |
| DAP, regular | -- | -- | -- | 60 |
| Defluorinated rock phosphate | 20 | -- | -- | -- |
| Calcium carbonate | -- | 20 | 20 | 20 |
| | 2000 | 2000 | 2000 | 2000 |

Care of Experimental Animals

Standardization Period: The cows to be used in Trial I were placed in an experimental lot one week prior to initiation of the experiment and individually fed ten pounds of grain daily. They had access to a pole barn which was open on the south side. Water was available at all times other than during the feeding and weigh-back period each day. Good quality alfalfa hay was fed free-choice on a group basis in two all-metal,

portable hay bunks approximately 12 feet long and 3 feet wide. Hay was placed in the bunks each morning while the cows were stanchioned or shortly thereafter; thus, hay consumption was nil immediately prior to concentrate feeding. The hay bunks were moved frequently to maximize hay consumption.

All cows were stanchioned at approximately 10:00 A.M. each day and allowed to eat until approximately 11:30 A.M. The amount of feed not consumed in the 1.5-hour period was weighed and recorded.

Observations were made twice daily regarding health and general condition of each cow. Body weights were taken on two consecutive days after initiation of the standardization period.

Comparison Period: Experimental rations were fed in a sequence determined by the Latin square. Twelve pounds of the respective experimental ration were fed daily. All other feeding and management details were conducted as described for the standardization period. Body weights were taken on two consecutive days at the end of each 10-day comparison period.

Laboratory Analysis of Ammonia Release

Collection of Saliva Samples: Saliva was collected from each cow and laboratory analyses were performed to determine the relative extent of ammonia release upon incubation of the sample with a known weight of several types of DAP. Cows were selected for sampling in such a manner that all samples from cows within a given block were taken in the same time period; thus, there was an equal number of saliva samples taken from cows which had received each respective experimental ration eight days prior to sampling. Two saliva samples were taken from each cow. Saliva samples were taken while the cows were restrained in a stanchion located some distance from where the cow was fed.

In order to facilitate sampling, each cow was haltered and a section of regular garden hose, wrapped with masking tape, was placed through her mouth. The cow was allowed to chew on the hose while being sampled.

The sampling apparatus consisted of a small vacuum pump, two 500 ml vacuum flasks and thick-walled tubing. Saliva was drawn from the cheek region of the mouth and caught directly in the screw-cap test tube used for storage of the sample.

Method of Preserving Saliva Samples: The saliva samples were frozen immediately after collection. An insulated ice pack containing dry ice and ethyl alcohol was used. Plastic bags (Nasco Whirl Paks) were placed around the test tubes before they were put into the dry ice-alcohol bath to prevent breakage of the tube. Without the plastic bags, test tubes cracked upon being placed in the alcohol. After the samples were returned to the laboratory, they were stored in the freezing compartment of a refrigerator at approximately -5 C.

Determination of Ammonia Release: The release of ammonia upon incubation of various types of DAP with bovine saliva was measured by a microdiffusion technique described by Conway (4), as modified to meet the conditions of this experiment. All analyses were made in duplicate and the two values were averaged.

Reagents used in the analysis were a dilute standardized HCl solution, approximately 0.01 N, and a 1% boric acid-indicator solution. The HCl served as a titrant and the boric acid-indicator solution was used to absorb a portion of the ammonia released during incubation. Indicator (33 mg. bromocresol green and 66 mg. methyl red dissolved in 100 ml. ethyl alcohol) was added dropwise to a 1% boric acid solution until a

definite, faint, pink color was obtained. This solution served as the boric acid-indicator solution.

Approximately one gram, weighed to the nearest tenth of a milligram, of each type of DAP (RGDAP, RDAP and SDAP-I) was placed into respective Conway units. One milliliter of boric acid-indicator solution was pipetted into the center well. Thin strips of polyethylene, approximately 2.5 inches long and 1 inch wide, were placed next to the center well of the unit, and the DAP sample was located behind the strip to prevent the sample from rolling to the lower side of the unit upon tilting. Each saliva sample was thawed as needed for making an analysis. A pH reading was taken using a Beckman Model N portable pH meter with a single glass electrode. A 1-ml portion of the saliva sample was carefully placed in the lower side of each unit; thus, no mixing between saliva and DAP occurred prior to initiation of the incubation period. Each unit was placed in an oven at 39 C and maintained in a tilted position. The polyethylene strips were removed gently and the lids were affixed with care. The closed units were allowed to heat for 5 minutes. After the equilibration period, the units were quickly tilted in an opposite direction allowing saliva to contact the DAP. The units were maintained in a level position during a 5-minute incubation period, measured from the time that saliva was mixed with the DAP sample until the lids were removed. Rubber stoppers (size 7) were placed over the center wells to prevent further absorption of ammonia while titrations were being made. A 5-ml Exax burette was used for making titrations which were recorded to the nearest hundredth of a milliliter. Calculation of ammonia absorption was made from the number of milliequivalents of HCl required to titrate the boric acid-indicator

solution to a permanent pink end point. The amount of ammonia absorbed was multiplied by the reciprocal of the respective DAP sample weight to express determinations on a weight basis ($\mu$g $NH_3$/g DAP).

A second set of determinations was made by incubating 2-gram DAP samples for two minutes. The absorption values were expressed on a weight basis ($\mu$g $NH_3$/g DAP).

Statistical Analysis of Data

An analysis of variance on the feed consumption data was computed as described by Cochran and Cox (3). The adjusted direct and residual effects were tested for significance.

An analysis of variance was also computed on the data concerning ammonia release by different types of DAP upon contact with saliva. Where the "F" test revealed a significant difference among treatments, Duncan's new multiple-range test (32) was applied to locate the differences.

## TRIAL II

Cows Used and Composition of Rations

During the experiment, 8 Holstein, 8 Ayrshire and 4 Jersey cows were used. Rations fed in Trial II were formulated according to Table III. Other details concerning the assignment of treatments, care of animals, collection of data, and statistical analyses were carried out as described for Trial I.

Laboratory Analysis of Ammonia Release

Collection of Saliva Samples: Each cow was sampled during the last half of the period in which she received the control ration. One cow was not sampled during the time period in which the control ration was fed;

therefore, at the end of the trial, she received control ration for eight days before she was sampled. Sampling was performed by the method described in Trial I; however, the saliva samples were not frozen immediately after collection. Instead, they were placed directly into a freezer which maintained a temperature of approximately -5 C. It was assumed that the samples were frozen within 30 minutes after being placed in the freezer. Several days after sampling, the saliva samples were placed in a precooled, insulated ice pack and transferred to the laboratory. The samples were stored as described in Trial I.

TABLE III

COMPOSITION OF RATIONS--TRIAL II

| Ingredient | Ration | | | |
|---|---|---|---|---|
| | Control | 1.5% stabilized DAP-II | 3% stabilized DAP-II | 3% regular DAP |
| | (lb/ton) | | | |
| Sorghum grain, steam rolled | 600 | 600 | 600 | 600 |
| Corn, ground | 340 | 390 | 440 | 440 |
| Oats, crimped | 300 | 300 | 300 | 300 |
| Wheat bran | 300 | 300 | 300 | 300 |
| Molasses, liquid | 200 | 200 | 200 | 200 |
| Cottonseed meal, 41% | 200 | 120 | 40 | 40 |
| Salt | 20 | 20 | 20 | 20 |
| Vitamin-trace mineral mix | 20 | 20 | 20 | 20 |
| Defluorinated rock phosphate | 20 | -- | -- | -- |
| DAP, stabilized-II | -- | 30 | 60 | -- |
| DAP, regular | -- | -- | -- | 60 |
| Calcium carbonate | -- | 20 | 20 | 20 |

Determination of Ammonia Release: In Trial I, the amount of ammonia produced and the amount absorbed during the incubation period were not the same; however, the ratio of the amount of ammonia absorbed compared to the amount produced was assumed to be closely related. To test this assumption and to more nearly determine the actual amount of ammonia release

from DAP, various types of DAP were reacted with saliva in 50 ml reaction tubes. The ammonia produced therein was swept into absorption test tubes where the ammonia was absorbed by a boric acid-indicator solution. Laboratory grade nitrogen gas was used as a carrier-gas to sweep ammonia from the reaction tubes. The apparatus (Fig. 1) used to make the ammonia determinations consisted primarily of (a) a source of laboratory grade nitrogen, (b) a pressure gauge and regulator, (c) a gas dispersion unit, (d) 8 reaction tubes, (e) 8 injection assemblies, (f) a temperature-controlled water bath and (g) 16 absorption tubes. The nitrogen bottle was connected to a Hoke-Phoenix gas regulator which permitted an adjustment of gas flow in pounds of pressure per square inch. Tygon tubing (3/16 inch ID) was used to make connections in the system which required tubing. An 18-gauge hypodermic needle was inserted inside the outlet nipple on the regulator. The hypodermic needle restricted the flow of gas through the outlet nipple and allowed the rate of gas flow to be regulated by simply adjusting regulator pressure instead of having to establish a particular gas flow rate each time the unit was placed into operation. It was found that 30 pounds of regulator pressure produced the optimum rate of gas flow for the system. The rate of gas flow from the outlet nipple was 16 l per minute. This amount of nitrogen was sufficient to displace the volume of gas in each reaction tube 40 times per minute. The maximum discrepancy in determining the actual amount of ammonia produced during the 1-minute reaction period, due to retention of one reaction tube of $N_2$-$NH_3$ gas mixture at the instant gas flow stopped, was approximately 2%. This discrepancy had no effect on the relative values of ammonia release from the different types of DAP.

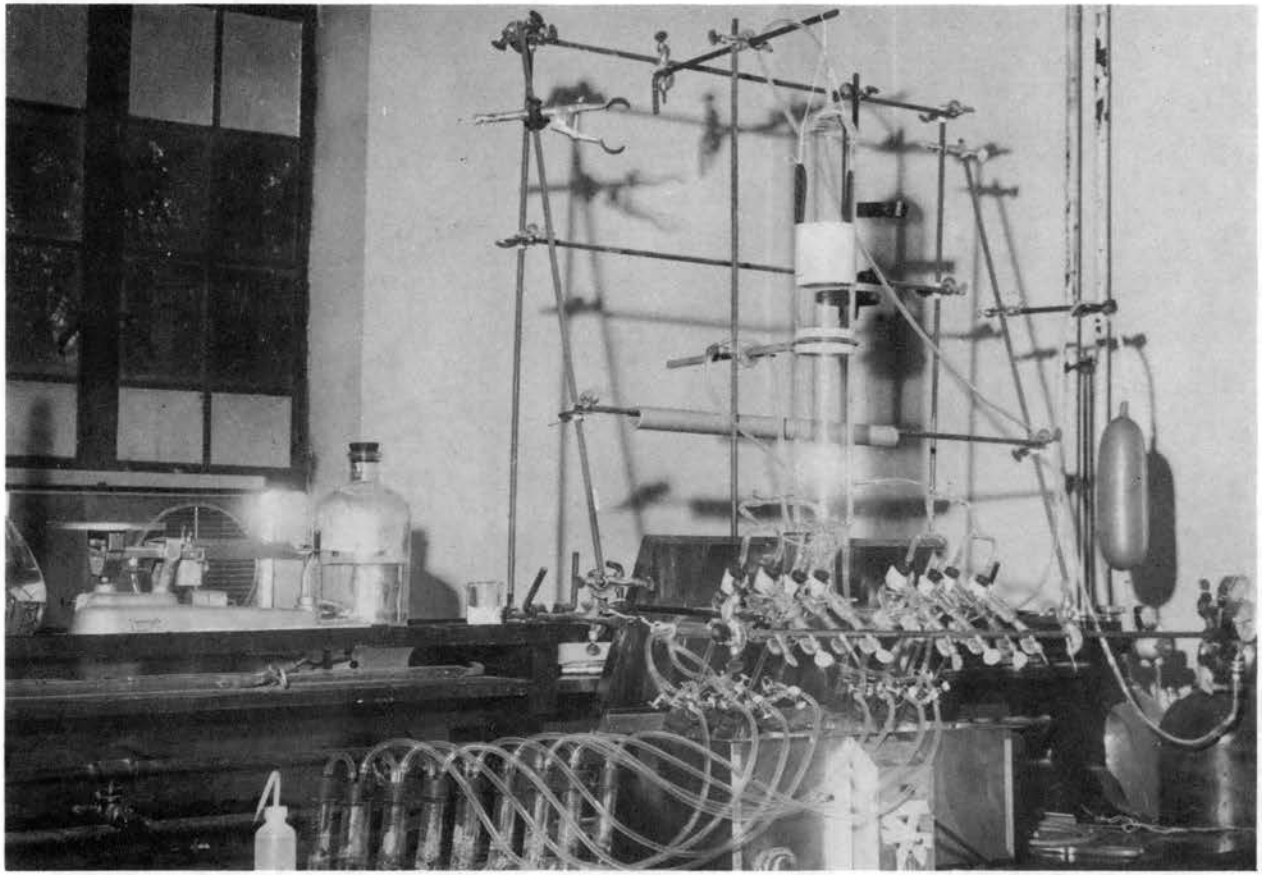

Fig. 1. Apparatus used for making ammonia determinations in Trial II.

Prior to making a given determination, the test tube of saliva was removed from storage and placed in a 500 ml beaker of water for thawing. Absorption test tubes were removed from the oven and 10 ml of the boric acid-indicator solution, described in Trial I, were pipetted into each one. They were then stoppered, but not connected to the reaction tubes. Reaction tubes, each containing a known weight of DAP (approximately one gram) were affixed on the rubber stoppers held by stems of the dropping funnels. When making a determination, saliva was injected into two reaction tubes which contained no DAP and the ml of titrant required to titrate the respective absorption test tubes were averaged. The average value served as a blank to be subtracted from each of the other titrations. Duplicate samples of various types of DAP were placed in the other reaction tubes. After the tubes were affixed to the stoppers, they were lowered into the water bath (39 C) to equilibrate and nitrogen was passed through the system for 30 seconds at 30 pounds of regulator pressure. The nitrogen flow was stopped and the reaction chambers were connected to the absorption units. A very slight flow (less than one pound of regulator pressure) of nitrogen gas was passed through the system to permit adjustment of the flow rate of each reaction tube. The adjustment was made by changing the depth of stoppers in the respective absorption tubes. A 1-ml portion of the saliva sample was pipetted into each dropping funnel. Each funnel was connected to a pressure line which carried nitrogen from a rubber balloon to the mouth of the dropping funnel. The pH of another portion of the saliva sample was taken after the balloon was filled with nitrogen gas. Nitrogen was passed through the system at 30 pounds of regulator pressure and saliva samples were injected into the reaction tubes simultaneously.

Nitrogen flow rate was maintained for one minute and was then stopped instantaneously by opening the side outlet of a glass "T" located in the nitrogen line near the regulator nipple. Reaction tubes were isolated and by-passes were opened by changing positions of the pinch clamps. Nitrogen was passed through the system at 30 pounds of regulator pressure for one minute to sweep residual ammonia from the lines. The absorption units were disconnected from the reaction tubes and glass inlet tubes of the absorption test tubes were rinsed with distilled water which had been boiled immediately prior to rinsing. The boric acid-indicator solution was titrated with standard HCl and calculations of ammonia release were made from the number of milliequivalents of HCl required to titrate the boric acid-indicator solution to a permanent pink end point.

## TRIAL III

### Selection of Cows and Assignment of Treatments

Twenty-four lactating cows were selected from the Oklahoma State University dairy herd and grouped into blocks of 6 cows each. The basis for grouping was (a) breed of animal, (b) stage of lactation and (c) level of production. Cows with a high level of production were selected when possible. Six Holstein and 18 Ayrshire cows were used in the trial.

Three experimental rations were fed cafeteria style with only two rations presented to each animal for selection at a time. The combinations of rations A, B and C were A-B, A-C and B-C. Ration combinations were considered as treatments in the Latin square design (Table IV). Animals within blocks were assigned randomly to treatment sequences. The position of each ration placed before each cow was changed daily to prevent possible bias created by a cow's preference for a particular feed box.

Experimental periods were of two weeks' duration. The response criterion was pounds of the various experimental rations consumed.

TABLE IV

TREATMENT SEQUENCES FOR LATIN SQUARE--TRIAL III

| Period | Block 1 Treatment Sequences | | | Block 2 Treatment Sequences | | |
|---|---|---|---|---|---|---|
| 1 | A-B | A-C | B-C | A-B | A-C | B-C |
| 2 | A-C | B-C | A-B | B-C | A-B | A-C |
| 3 | B-C | A-B | A-C | A-C | B-C | A-B |

Composition and Handling of Rations

Rations were mixed by Stillwater Milling Company, Stillwater, Oklahoma. Mixing, handling and storage was conducted as described for Trial I. All rations were fed in loose form.

Rations fed to the experimental animals were (a) control, (b) 1.25% SDAP-II and (c) 2.5% SDAP-II (Table V).

Care of Experimental Animals

Standardization Period: Animals were brought into the barn and stanchioned seven days prior to the experimental period and adjusted to the control ration. Grain was increased until the cows were being fed ad libitum. Body weights were taken during the standardization period and at the end of each 14-day experimental period. Excellent quality alfalfa hay was fed ad libitum to six blocks of cows; however, due to a hay shortage, medium quality alfalfa hay was fed to the other blocks.

TABLE V

COMPOSITION OF RATIONS--TRIAL III

| Ingredient | Ration | | |
|---|---|---|---|
| | Control | 1.25% stabilized DAP-II | 2.5% stabilized DAP-II |
| | (lb/ton) | | |
| Sorghum grain, steam rolled | 800 | 800 | 800 |
| Corn, ground | 500 | 540 | 580 |
| Oats, crimped | 100 | 100 | 100 |
| Wheat shorts | 200 | 200 | 200 |
| Cottonseed meal, 41% | 200 | 135 | 70 |
| Molasses, blackstrap | 140 | 140 | 140 |
| Vitamin-trace mineral mix | 20 | 20 | 20 |
| Salt | 20 | 20 | 20 |
| Defluorinated rock phosphate | 20 | -- | -- |
| Calcium carbonate | -- | 20 | 20 |
| DAP, stabilized-II | -- | 25 | 50 |

All animals were brought into the barn and stanchioned at 1:00 P.M. and 2:00 A.M. each day. Restraining chains with clips were used to stanchion the cattle. Grain was fed and observations were made regarding health and general condition of each cow. The feed troughs were partitioned, making two feed boxes available for each cow. The partitions were made high enough to prevent feed from being carried between boxes. A predetermined amount of feed was placed in the proper boxes and the cows were allowed to eat for approximately one hour. The cows were permitted to return to the experimental lot as soon as they were milked. The amount of feed not eaten during a 1-hour period was determined and recorded twice daily.

Comparison Period: The cows were managed in the same manner as they were during the standardization period.

Since only two choices were offered each cow at any particular time, the opinion was that enough of each experimental ration should be fed to meet the cow's needs in case one of the rations was refused. This method of feed assignment essentially created ad libitum feeding for all cows consuming rations which were not greatly distinguishable. In order to standardize procedure, the amount needed by each cow was calculated and placed in each feed box regardless of the combination of rations being fed. Feed allowances were made by assuming hay consumption to be approximately two pounds per hundred pounds of body weight. Average butterfat tests for the Holsteins and Ayrshires were estimated to be 3.5 and 4%, respectively. The theoretical amounts of grain to be fed was then obtained from a feeding guide for lactating dairy cows prepared by Stone et al. (33).

Laboratory Analysis of Ammonia Release

Saliva samples were collected by the method described in Trial I and were frozen in the same manner as that described in Trial II. Determinations of ammonia release were made by using the procedure described in Trial II.

Determination of Effect of Freezing Saliva: Four cows were selected randomly from the Oklahoma State University dairy herd for sampling purposes. Each cow was sampled and each sample thus obtained was taken to the Dairy Science laboratory where pH was recorded. Two aliquots were measured. The first aliquot was frozen for later analyses and the second was used immediately for making determinations of ammonia release from several types of DAP (RGDAP, RDAP and SDAP-II). The purpose of this procedure was to investigate the possibility that salivary changes occur upon freezing which alter the amount of ammonia released from DAP.

Determination of Effect of Incubation Time on Release of Ammonia from DAP: Known amounts of RGDAP were placed in the reaction tubes not used in determining the blank. A frozen saliva sample composed of saliva from several cows was thawed and pH was determined. Other basic details of the procedure were performed as described in Trial II. Saliva was injected simultaneously into two reaction tubes containing RGDAP and the two blank tubes. Two injections at a time were made into the other reaction tubes at various time intervals after the initial injections. The time intervals were (a) two minutes after the initial injections and (b) three minutes after the initial injections. The nitrogen flow was stopped instantaneously four minutes after the initial injections and residual ammonia was flushed from the tubing. Calculations of ammonia release were made as described in Trial II.

Determination of Effect of pH on Release of Ammonia from DAP: Buffer solutions of known pH were made from analytical standards (pHydrion buffers) and determinations of ammonia release upon reacting RGDAP with the various buffer solutions were performed. Known amounts of RGDAP were placed in six reaction tubes and one milliliter of a buffer solution was pipetted into each of the eight dropping funnels. Determinations were made, using the basic procedure described in Trial II and the six values ($\mu g$ $NH_3$/g DAP) thus obtained were averaged to facilitate graphing of the data. The pH values of the buffer solutions tested were 4.0, 5.0, 6.0, 7.0, 8.2, 9.0 and 10.0.

## Statistical Analysis of Data

An analysis of variance was computed on the feed consumption data to identify significant sources of variation.

# RESULTS AND DISCUSSION

## TRIAL I

### Voluntary Feed Consumption and Relative Amounts of Ammonia Release

All cows used in the trial remained healthy and in good condition. Two cows calved four days prior to completion of the experiment, and feed consumption for these cows during period four was estimated by multiplying the average daily consumption during the first six days by ten.

Mean daily feed consumption was 9.23, 8.89, 7.86 and 8.26 pounds per day for the control, 1.5% SDAP-I, 3% SDAP-I and 3% RDAP rations, respectively. Direct adjusted treatment effects were not statistically significant ($P > 0.10$). Likewise, adjusted residual effects were not significant. In using the Latin square design, the assumption was made that voluntary feed consumption would test acceptability of the various rations by the cows. It was realized that a precise measure of palatability would not be obtained since cows were not given a choice between rations; however, as a preliminary study, the results were indicative of whether or not dry cows would accept rations containing the various levels of DAP.

At no time was any ration completely refused. Most of the trial was conducted in July and August; therefore, feed consumption was somewhat low for all rations. There was considerable variation among cows in level of concentrate consumption. Some cows were consistently low consumers while others were consistently high consumers. As an extreme example, one cow consumed 135 pounds of concentrate while another cow consumed 311 pounds

of concentrate. The SDAP-I used in the experimental rations appeared to be stabilized to a considerable degree. Relative mean values of ammonia release during five minutes of incubation were 121, 71 and 13 μg $NH_3$/g DAP for RGDAP, RDAP and SDAP-I, respectively. Assuming that absorption in the Conway units was proportional to the amount of ammonia produced, RGDAP and RDAP produced 9.3 and 5.5 times as much ammonia, respectively, as was produced by SDAP-I. The determined value for SDAP-I was suspected of being somewhat high because of the small amount of titrant (0.02N) required for the titration. An error of 1 drop of titrant caused an error in ammonia determination of 7 μg which was half the mean value for SDAP-I. This was of significance since it was necessary to dispense titrant from the burette in quantities of not less than a drop (0.02 ml) in order to make accurate identification of the end point. The most nearly correct value for SDAP-I in this group of determinations was probably less than 13 μg $NH_3$/g DAP.

In a second group of determinations, the amount of DAP placed in each Conway unit was doubled and titrant strength was decreased (0.006 N) in an effort to increase sensitivity of the procedure. This was deemed necessary to obtain an accurate measure of the relative amounts of ammonia released during an incubation period of two minutes. Reliable results were not deemed likely with incubation periods less than two minutes, and some doubt was present concerning accuracy of a 2-minute determination. Relative mean values of ammonia release were 18.7, 10.1 and 2.2 μg $NH_3$/g DAP for RGDAP, RDAP and SDAP-I, respectively. All differences among treatment means were significant ($P < 0.01$). Under these conditions, RGDAP and RDAP were found to release 8.5 and 4.6 times as much

ammonia, respectively, as was released by SDAP-I. The mean value for SDAP-I was probably inflated slightly. No increase in sensitivity resulted from changing the conditions for the determination since the effect of decreasing titrant strength was offset by decrease in ammonia absorption due to shortening of reaction time. In this case, 1 drop of titrant was equivalent to 2 μg of ammonia which was near the mean value for SDAP-I.

TABLE VI

RELATIVE AMOUNTS OF AMMONIA RELEASED FROM DAP UPON SATURATION WITH BOVINE SALIVA AND INCUBATION AT 40 C FOR TWO MINUTES IN CONWAY UNITS

| Ration fed 8 days prior to obtaining saliva sample | No. of saliva samples | Type of DAP | | |
|---|---|---|---|---|
| | | Reagent | Regular | Stabilized-I |
| | | (avg μg $NH_3$/g DAP) | | |
| Control | 5 | 18 | 10 | 2 |
| 1.5% stabilized DAP-I | 5 | 19 | 10 | 3 |
| 3.0% stabilized DAP-I | 5 | 18 | 10 | 2 |
| 3.0% regular DAP | 5 | 18 | 10 | 2 |
| Average | | 18[a] | 10[b] | 2[c] |

[abc]Means with different superscripts differ significantly ($P < 0.01$) according to Duncan's new multiple range test.

The rate of ammonia release was suspected of being greater at short incubation periods; however, differences in the rate of release were not proved or disproved by using the modified Conway procedure. The apparent ammonia release per gram of DAP was approximately 6.5 times as great for 5-minute reaction periods as during 2-minute periods. Ammonia release was shown in Trial III to be a linear function of the reaction time; therefore, 5-minute periods should have produced values approximately 2.5 times as great as values for the 2-minute periods. This discrepancy probably

occurred because of inadequacy of the modified Conway procedure to give correct values with respect to reaction time, especially at the shorter period.

The type of ration fed to the cows had no effect on the amount of ammonia released from DAP upon contact with saliva taken from the cows (Table VI).

## TRIAL II

### Voluntary Feed Consumption and Relative Amounts of Ammonia Release

The cows used in Trial II were in good condition throughout the experiment, although two cows were treated for mastitis. Total feed consumption was 8,775 pounds for Trial II as compared to 6,847 pounds for Trial I. Most of the difference in consumption was attributed to a change in environmental temperature since Trial II was conducted during November and December; however, part of the difference may have been caused by a change in the composition of rations. The rations fed in Trial II may have been more palatable than those fed in Trial I.

Average daily consumption was 11.3, 11.0, 11.1 and 10.5 pounds for the control, 1.5% SDAP-II, 3% SDAP-II and 3% RDAP rations, respectively. Direct adjusted treatment effects were not statistically significant ($P \geq 0.10$). Likewise, adjusted residual effects were not significant. In Trial II, there was much less variation among cows with regard to total feed consumption as compared to Trial I.

The relative amounts of ammonia release were 333, 126 and 16 μg $NH_3$/g DAP for RGDAP, RDAP and SDAP-II, respectively. All differences among treatment means were significant ($P < 0.01$). The ratio of these

TABLE VII

AMMONIA RELEASED FROM DAP UPON SATURATION WITH BOVINE SALIVA AND INCUBATION AT 39 C FOR ONE MINUTE

| Cow No. | Type of DAP | | | Total |
|---|---|---|---|---|
| | Reagent | Regular | Stabilized-II | |
| | μg $NH_3$/g DAP | | | |
| 1 | 296 | 158 | 17 | 471 |
| 2 | 411 | 184 | 37 | 632 |
| 3 | 349 | 134 | 17 | 500 |
| 4 | 401 | 124 | 22 | 547 |
| 5 | 292 | 128 | 15 | 435 |
| 6 | 347 | 122 | 24 | 493 |
| 7 | 265 | 134 | 12 | 411 |
| 8 | 383 | 136 | 22 | 541 |
| 9 | 379 | 128 | 0 | 507 |
| 10 | 323 | 109 | 10 | 442 |
| 11 | 306 | 109 | 10 | 425 |
| 12 | 381 | 139 | 10 | 530 |
| 13 | 403 | 133 | 27 | 563 |
| 14 | 262 | 100 | 9 | 371 |
| 15 | 309 | 111 | 12 | 432 |
| 16 | 306 | 111 | 17 | 434 |
| 17 | 260 | 111 | 12 | 383 |
| 18 | 311 | 107 | 14 | 432 |
| 19 | 389 | 139 | 10 | 538 |
| 20 | 289 | 97 | 14 | 400 |
| Average | 333[a] | 126[b] | 16[c] | 474 |

[abc]Means with different superscripts differ significantly ($P<0.01$) according to Duncan's new multiple range test.

values was 21, 8 and 1, respectively; thus, the data supported the idea that determinations of ammonia release were somewhat high for SDAP-I as measured in Trial I. It was noted that the analytical procedure used in Trial II produced values of ammonia release which were much greater than those produced by the modified Conway procedure (Table VII). This was due to a greater recovery of ammonia obtained with the more direct method used in Trial II.

The amount of ammonia released from DAP varied when DAP was reacted with saliva from different cows (Table VII). There was some indication that ammonia release increased as pH increased (Table VIII); however, the data were difficult to assess because of the small number of samples tested in each pH classification.

TABLE VIII

EFFECT OF pH ON AMMONIA RELEASE BY DAP UPON SATURATION WITH BOVINE SALIVA AND INCUBATION AT 39 C FOR ONE MINUTE

| pH | No. of samples | Type of DAP | | |
|---|---|---|---|---|
| | | Reagent | Regular | Stabilized-II |
| | | (avg µg/g DAP) | | |
| 8.9 | 4 | 338 | 139 | 20 |
| 8.8 | 4 | 355 | 131 | 20 |
| 8.7 | 4 | 306 | 121 | 15 |
| 8.6 | 5 | 355 | 128 | 14 |
| 8.5 | 2 | 284 | 105 | 10 |
| 8.4 | 1 | 328 | 105 | 10 |

TRIAL III

Palatability of DAP Rations

All of the 24 cows used in Trial III remained in relatively good condition and in no instance was loss in body weight noted. One cow was

treated for a foot injury and another for mastitis. One cow was treated for a hormone imbalance and displaced abomasum, and another was diagnosed to have a lung disorder. Average daily consumption of concentrate was 20.2 pounds per cow. Since each cow had simultaneous access to two different rations at all times, the amount of each consumed daily would be expected to be approximately one-half the total amount if the cows did not distinguish between rations. Thus, the average daily consumption was 8.7, 11.7 and 9.8 pounds for the control, 1.25% SDAP-II and 2.5% SDAP-II rations, respectively. Differences among treatment means were not statistically significant ($P > 0.05$). There was only one cow that showed a distinct preference against the 2.5% DAP ration and this was not consistent from one period to another. This cow consumed only 5 pounds of the 2.5% DAP ration and 178 pounds of the 1.25% DAP ration during the third period; however, earlier in the experiment, she had consumed 290 pounds of the 2.5% DAP ration and 60 pounds of the control ration (Table XII).

In other work (21) where palatability was measured cafeteria style, individual calves tended to be quite uniform in their choices over a period of time. Similarly, most of the cows used in Trial III of the present study were highly consistent in their choices over a period of two weeks; however, all cows did not make the same selection when given a choice between two rations. Miller and Clifton (22) have reported such a variability with dairy calves and have suggested that researchers should not use small numbers of animals for cafeteria type palatability experiments.

Effect of Freezing Saliva

Variation in pH was noted among some of the saliva samples used to test for ammonia release in Trials I and II. In further work, a slight

TABLE IX

EFFECT OF FREEZING SALIVA ON SALIVA-DAP REACTION RATE AND pH OF SALIVA SAMPLE[a]

| Sample number[b,c] | Number of days sample was frozen | pH | Type of DAP | | | Total |
|---|---|---|---|---|---|---|
| | | | RGDAP | RDAP | SDAP-II | |
| | | | μg $NH_3$/g DAP | | | |
| 1A | -- | 8.5 | 306 | 116 | 10 | 432 |
| 2A | -- | 8.6 | 223 | 117 | 14 | 354 |
| 3A | -- | 8.6 | 221 | 117 | 27 | 365 |
| 4A | -- | 8.7 | 323 | 134 | 34 | 491 |
| 1B | 26 | 8.8 | 211 | 68 | 10 | 289 |
| 2B | 23 | 8.7 | 257 | 105 | 10 | 372 |
| 3B | 22 | 8.8 | 309 | 105 | 17 | 431 |
| 4B | 20 | 8.7 | 294 | 111 | 9 | 414 |
| Average, fresh samples (A) | -- | 8.6 | 268 | 121 | 21 | 411 |
| Average, frozen samples (B) | 23 | 8.8 | 268 | 97 | 12 | 377 |

[a]Incubation time of one minute at 39 C.

[b]Sample "A" tested within ten minutes after collection.

[c]Sample "B" frozen at -5 C before testing.

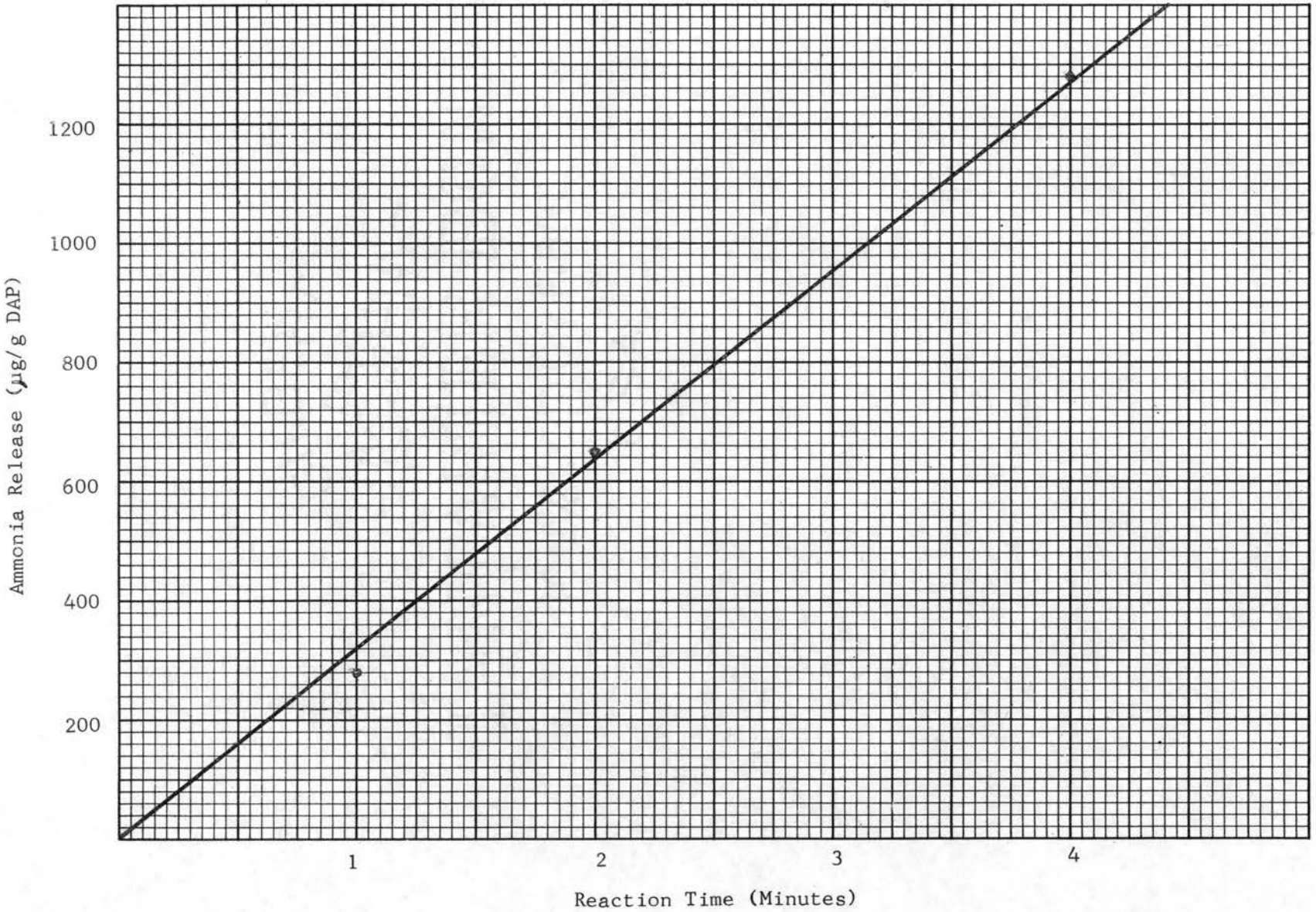


Figure 2. Ammonia Release from RGDAP Upon Saturation with Bovine Saliva and Incubation at 39 C. I. Effect of Reaction Time.

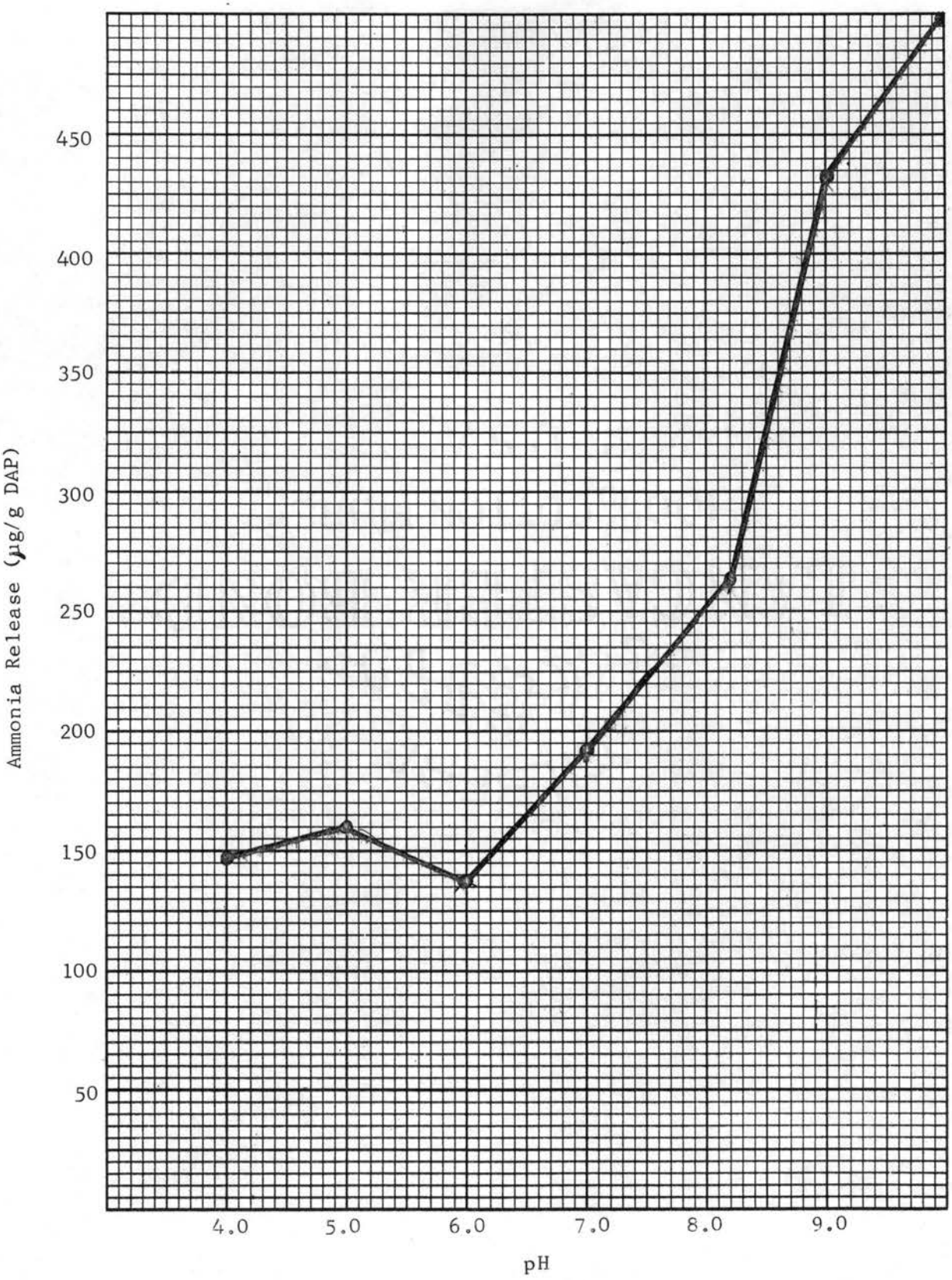


Figure 3. Ammonia Release from RGDAP Upon Saturation with Buffer Solution and Incubation at 39 C for One Minute. II. Effect of pH.

rise in pH was noted upon freezing and storing salive samples; however, the small pH changes did not appear to affect greatly the amount of ammonia release from DAP (Table IX). According to Dukes (7), some of the variation in pH may have been due to loss of $CO_2$ from the saliva samples having higher pH values.

It was concluded that freezing of saliva had little, if any, effect on the rate of ammonia release from DAP upon reaction with saliva that had been frozen.

Effect of Incubation Time on Release of Ammonia from DAP

Ammonia release from DAP upon saturation with bovine saliva was found to be a linear function of the length of reaction time (Fig. 2). Reaction periods greater than four minutes were not studied; however, it seemed reasonable to assume that unstabilized DAP which had been dampened with saliva would liberate a considerable amount of ammonia within a period of several minutes.

Effect of pH on Release of Ammonia from DAP

Ammonia release from DAP was found to be affected greatly by pH (Fig. 3). The greatest rate of change of ammonia release with respect to pH, in the pH range 4.0-10.0, occurred between pH 8.0 and 9.0. Thus, it appears that saliva pH might be critical in determining whether or not cattle find rations containing unstabilized DAP unpalatable. In this study, it was not apparent that cows which produced high-pH saliva upon sampling under the conditions described herein, ate less feed when DAP was included in the ration.

## SUMMARY AND CONCLUSIONS

Two preliminary trials were conducted using nonlactating dairy cows to test the acceptability of rations containing various types and levels of DAP. Another trial was conducted with lactating cows to measure the palatability of rations containing various levels of stabilized DAP (SDAP-II). Laboratory analyses were performed in each trial to determine the extent of ammonia release from different types of DAP under various conditions.

In Trial I, twenty nonlactating cows were grouped according to breed and assigned randomly to a 4 x 4 Latin square which permitted estimation of carry-over effects. Experimental periods were of ten days duration. Alfalfa hay was fed ad libitum on a group basis. Each cow was fed individually twelve pounds of grain daily and the amount not consumed during a 1.5-hour period was determined. The experimental rations were (a) control, (b) 1.5% SDAP-I, (c) 3% SDAP-I and (d) 3% RDAP. Average daily grain consumption was 9.2, 8.9, 7.9 and 8.3 pounds per cow, respectively. Differences among treatment means were not statistically significant. Considerably less ammonia was released from the stabilized DAP upon contact with saliva than from the regular grade of DAP.

In Trial II, the experimental rations were (a) control, (b) 1.5% SDAP-II, (c) 3% SDAP-II and (d) 3% RDAP. Average daily grain consumption was 11.3, 11.0, 11.1 and 10.5 pounds per cow, respectively, for the different rations. Differences among treatment means were not statistically significant.

From Trials I and II, it was concluded that nonlactating cows will accept rations which contain as much as 3% DAP.

In Trial II, ammonia release from RGDAP, RDAP and SDAP-II upon saturation with bovine saliva and incubation at 39 C for one minute was 333, 126 and 16 μg $NH_3$/g DAP, respectively. All differences among treatment means were significant ($P < 0.01$).

In Trial III, twenty-four lactating dairy cows were grouped according to breed, calving date and level of production and fed combinations of three rations, taken two at a time, in a sequence determined by a pair of balanced 3 x 3 Latin squares. The ration combination, with each of the two rations supplying the theoretical grain requirement of each cow, was fed twice daily and the amount not consumed was determined after each feeding. The rations were (a) control, (b) 1.25% SDAP-II and (c) 3% SDAP-II. The combinations were a-b, a-c and b-c. Average daily consumption of grain was 20.2 pounds per cow. Average daily consumption of the experimental rations was 8.7, 11.7 and 9.8 pounds, respectively, for the control, 1.25% SDAP-II and 3% SDAP-II rations. Differences among treatment means were not statistically significant ($P > 0.05$).

The rate of ammonia release from DAP was found to be a linear function of time, in the time interval 0-4 minutes. Ammonia release from DAP increased as pH increased, with the greatest rate of increase occurring in the pH range 8.0-9.0.

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# A P P E N D I X E S

TABLE X

ANALYSIS OF VARIANCE ON RATE OF AMMONIA RELEASE FROM DAP--TRIAL I[a]

| Source of variation | Degrees of freedom | Sums of squares | Mean squares | F Values |
|---|---|---|---|---|
| Total | 59 | 0.1051 | 0.00178 | |
| Cows | 19 | 0.0151 | --- | |
| Blocks (B) | 4 | 0.0033 | 0.0008 | 0.88 |
| Ration treatments (R) | 3 | 0.0006 | 0.0002 | 0.22 |
| B x R | 12 | 0.0112 | 0.0009 | |
| Treatments (T) | 2 | 0.0819 | 0.0409 | 136[b] |
| Treatment x cows | 38 | 0.0081 | --- | |
| T x B | 8 | 0.0015 | 0.0002 | 0.66 |
| T x R | 6 | 0.0001 | 0.0001 | 0.33 |
| T x B x R | 24 | 0.0065 | 0.0003 | |

1% Multiple Range Test for Treatments

| | A | B |
|---|---|---|
| C | 0.09(0.013)[c] | 0.05(0.012) |
| B | 0.04(0.012) | |

[a]Treatments: A - RGDAP
B - RDAP
C - SDAP-I

[b]$P < 0.01$

[c]Numbers in parentheses, least significant range.

TABLE XI

ANALYSIS OF VARIANCE ON RATE OF AMMONIA RELEASE FROM DAP--TRIAL II[a]

| Source of variation | Degrees of freedom | Sums of squares | Mean squares | F Values |
|---|---|---|---|---|
| Total | 59 | 33.9870 | 0.5761 | |
| Treatments (T) | 2 | 32.2059 | 16.1030 | 715.69[b] |
| Cows (C) | 19 | 0.9252 | 0.0487 | 2.16[c] |
| T x C | 38 | 0.8559 | 0.0225 | |

1% Multiple Range Test for Treatments

| | A | B |
|---|---|---|
| C | 1.76(0.134)[d] | 0.60(0.128) |
| B | 1.16(0.128) | |

[a]Treatments: A - RGDAP
B - RDAP
C - SDAP-II

[b]$P < 0.01$

[c]$P < 0.05$, multiple range test not computed.

[d]Numbers in parentheses, least significant range.

TABLE XII

CONSUMPTION OF RATIONS WITH DIFFERENT TYPES OF DAP[a]
BY INDIVIDUAL COWS--TRIAL III

| Block | Cow Number | Period 1 | | Period 2 | | Period 3 | |
|---|---|---|---|---|---|---|---|
| | | lb/period | | | | | |
| 1 | 1 | W 119 | B 127 | W 135 | R 161 | B 181 | R 170 |
| " | 2 | W 114 | R 122 | B 211 | R 149 | W 199 | B 190 |
| " | 3 | B 186 | R 52 | W 143 | B 175 | W 186 | R 105 |
| 2 | 4 | W 109 | B 115 | B 156 | R 117 | W 181 | R 102 |
| " | 5 | W 41 | R 50 | W 9 | B 58 | B 83 | R 57 |
| " | 6 | B 156 | R 98 | W 146 | R 133 | W 134 | B 143 |
| 3 | 7 | W 142 | B 201 | W 142 | R 191 | B 186 | R 159 |
| " | 8 | W 95 | R 379 | B 132 | R 338 | W 160 | B 287 |
| " | 9 | B 173 | R 201 | W 65 | B 314 | W 138 | R 259 |
| 4 | 10 | W 24 | B 192 | B 199 | R 67 | W 246 | R 76 |
| " | 11 | W 22 | R 409 | W 58 | B 404 | B 143 | R 296 |
| " | 12 | B 125 | R 325 | W 283 | R 240 | W 121 | B 315 |
| 5 | 13 | W 115 | B 77 | W 116 | R 54 | B 123 | R 38 |
| " | 14 | W 103 | R 137 | B 96 | R 111 | W 52 | B 115 |
| " | 15 | B 108 | R 70 | W 49 | B 101 | W 72 | R 91 |
| 6 | 16 | W 87 | B 162 | B 117 | R 126 | W 106 | R 138 |
| " | 17 | W 139 | R 134 | W 112 | B 134 | B 116 | R 117 |
| " | 18 | B 117 | R 86 | W 92 | R 75 | W 88 | B 77 |
| 7 | 19 | W 89 | B 275 | W 98 | R 149 | B 158 | R 46 |
| " | 20 | W 179 | R 149 | B 149 | R 140 | W 154 | B 176 |
| " | 21 | B 221 | R 55 | W 185 | B 108 | W 209 | R 76 |
| 8 | 22 | W 158 | B 208 | B 216 | R 94 | W 244 | R 50 |
| " | 23 | W 60 | R 290 | W 53 | B 157 | B 178 | R 5 |
| " | 24 | B 164 | R 91 | W 150 | R 17 | W 133 | B 41 |

[a]W = No DAP, B = 1.25% SDAP-II, R = 2.5% SDAP-II

VITA

James Leonard Reaves

Candidate for the Degree of

Master of Science

Thesis: DIAMMONIUM PHOSPHATE IN DAIRY CATTLE RATIONS

Major Field: Dairy Science

Biographical:

Personal Data: Born in Klamath Falls, Oregon, October 13, 1941.

Education: Graduated from Sheridan High School in 1959; received the B.B.A. degree from Southern State College, Magnolia, Arkansas in 1963 with a major in Agriculture-Business.

Experience: Raised on diversified farm in Arkansas; active in 4-H and F.F.A. work; worked on beef cattle farm and served as assistant manager, 1955-59; worked on diversified farm and in milk plant at Southern State College, 1959-60; served as D.H.I.A. supervisor for Southwest Arkansas Dairy Herd Improvement Association, 1960-63; served as substitute dairy herdsman at Southern State College, summer, 1962; served as Research Assistant at Oklahoma State University, June, 1963 to date; served as correspondence dairy instructor at Oklahoma State University, January, 1964, to date.

Member of: Phi Sigma Society, American Dairy Science Association.