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THE STABILITY OF VITAMIN A PRODUCTS IN  
CATTLE SUPPLEMENTS

by

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## INTRODUCTION

Vitamin A is an essential nutrient to all animals and its unstable characteristic has been known for many years. The poor stability of natural vitamin A sources such as carotenoid compounds and vitamin A from fish liver oils has resulted in the development and production of stabilized vitamin A sources.

There are a number of studies dealing with processing of stabilized vitamin A (4,8,28,38,43,59,65). One product is produced by the flowing procedure (17): vitamin A palmitate is melted under a protective nitrogen blanket. Antioxidants are added at this point to provide optimum stability to the finished product. Concurrently, a gelatin sugar syrup is prepared of appropriate viscosity and temperature. The liquid vitamin A palmitate with antioxidant is then homogenized under pressure into the gelatin-sugar syrup. The result is a very fine dispersion of vitamin A palmitate droplets in the syrup. The droplets average 2-5 microns in diameter. The resulting dispersion is spun into an oil bath, forming solid spherical droplets of the gelatin dispersion. Upon chilling the oil bath, these gelatin droplets solidify and settle from the oil and are separated. The solid gelatin beadlets are then solvent washed. The washed beadlets are dried, screened and packaged.

Stability of these sources is much improved over natural sources but problems may still occur.

The objective of this study was to evaluate the stability of two



sources (Rovimix A-650 and Rovimix A-325)\* of stabilized vitamin A when subjected to different pelleting conditions in cattle supplements.

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\*Registered trademark by Hoffman-La Roche Company in Nutley, New Jersey.

## REVIEW OF LITERATURE

There are a number of published reports dealing with the stability of vitamin A in feeds during storage, and the effect on the rate of this loss of factors such as moisture, temperature, pH, minerals, aqueous solubility, melting point, oxidizing and reducing compounds, particle size, storage time and pelleting operations.

### The Stability of Vitamin A in Natural Sources

The losses of vitamin A in feeds containing cod liver oil or other natural sources of the vitamin have been the subject of many investigations. Dunn (20) found vitamin A was destroyed when cod liver oil was mixed with starch and the mixture granulated and stored in corked bottles in the dark for six months. Marcus (50) showed that vitamin A from cod liver oil was unstable when added to a finely divided solid. Approximately 85 percent of the vitamin, as measured by the Carr-Price reaction, was lost in 10 days when the unsaponifiable fraction was added to a vitamin A free rat ration (U.S.P. basal diet) or to granulated lactose. No difference in vitamin A destruction was noted between storage under air or carbon dioxide. Holmes et al. (36) reported that vitamin A in cod liver and in halibut liver oil was destroyed in 6 and 21 weeks, respectively. When the oils were stored in partially filled bottles at room temperature in diffuse light, stability was improved to some degree by the addition of antioxidants such as Hydroquinone and lecithin.

Holder and Ford (40) and Bethke (10) measured vitamin A losses in a mixed ration containing added cod liver oil using a chick assay method. The ration was stored in burlap sacks at room temperature (21 to 27°C). Holder

and Ford (40) found no loss of vitamin A during storage periods of 8 weeks or less but were able to demonstrate a slight loss in a sample which was stored for 10 weeks. Baird et al. (6) conducted seven experiments covering respectively 0, 4, 8, 12, 16, 21, and 25 weeks of storage of a ration to determine the stability of vitamin A in mixed feed ingredients. The vitamin A from fortified cod liver oil, mixed in a ration, was not completely destroyed, even when stored in burlap bags at summer temperature for 25 weeks. It underwent progressive destruction during storage however.

Thus, the data in the literature gives ample proof of the instability of the vitamin A of cod liver oil in feeds and rations. The factors which retard the destruction noted have not been adequately investigated, and no explanations for the observed discrepancies are evident.

Carotene also has poor stability. Fraps and Treichler (23) reported losses in vitamin A potency of 30-80 percent in 6 to 19 months in alfalfa leaf meal, dried black-eyed peas, dried green sweet peppers, yellow corn (whole or ground) and powdered whole milk. Thompson (70) found that alfalfa (lucerne) meal retained only 54 percent of its original carotene content after 16 weeks at 25°C. Wall and Kelley (71) showed that carotene, when added to chicken mash at 3,000 international units (I.U.) per pound, lost 50 percent of its potency in less than three months. Kamstra et al. (45) using alfalfa meal and carrot oil as carotene sources studied the effect of trace minerals and several other feed ingredients on the stability of carotene in stored poultry feeds. These workers found alfalfa to have greater stability as a carotene source than carrot oil under the conditions of the study.

Fraps and Kemmerer (22) reported a 79 to 100 percent loss of the vitamin A added to different feed mixtures, in the form of fish oils or

concentrates, after four weeks storage at either 7° or 28°C. The loss of carotene in oil, or from alfalfa meal, under the same condition was not as rapid and the alfalfa leaf meal was a slightly more stable source of vitamin A activity in stored feed than cod liver oil.

Halpern and Biely (31) compared the utilization of vitamin A, in various carriers, and concluded that vitamin A oil had greater value in water emulsion than in vegetable oil solution. Johnson et al. (44) found that carotene in dehydrated alfalfa meal gave better chick growth and liver storage than vitamin A from fish liver oil. Reynolds et al. (62) obtained better chick growth with a ration containing stabilized carotene, stabilized vitamin A or vitamin A ester. Halpern et al. (32) found no difference in the biological activity or stability of vitamin A, supplied in a feeding oil or an emulsion, when mixed and stored in a chick ration.

#### Factors Affecting the Stability of Vitamin A from Natural Sources

Stability of vitamin A from cod liver oil or carotene among feed ingredients and supplements of various kinds vary considerably. Stabilizing or destructive properties are apparent between different ingredients. Therefore, the evaluation of cod liver oil or carotene stability in mixed feeds is a complex phenomenon.

Bethke et al. (10) reported: a significantly greater loss of both vitamin A and carotene occurred in a basal ration containing meat scraps and dried skim milk than in a comparable ration where casein replaced the above ingredients. Only about 50 percent of the added vitamin A from cod liver oil or carotene from alfalfa leaf meal was lost from the casein ration after six months storage. In contrast approximately 75 percent losses of both factors

occurred in meat scraps and dried skim milk ration. The addition of 0.1 percent of hydroquinone as an antioxidant to the diluted cod liver oil solution before incorporation in the meat scrap-dried skim milk ration did not affect the loss of vitamin A during storage.

It was reported that greater losses of carotene and of vitamin A occurred in the meat scraps-dried skim milk rations. The fat or free fatty acid content of the ration may have influenced the rate of carotene and vitamin A destruction. Meat scraps, fish meal, and other sources of animal protein concentrates may be quite high in free fatty acid content. Powick (60) reported that rancid lard, mixed into rations, destroyed the vitamin A present. Schroeder et al. (67) reported that meat scraps, high in free fatty acids, brought about inactivation of vitamin A, presumably due to oxidation. Holder and Ford (40) and Bethke et al. (10) found a destructive effect of meat scraps upon the vitamin A of cod liver oil in mixed feeds. Gray and Robinson (29) conducted experiments to study the significance of the free fatty acid content and peroxide value of the fat present in animal protein concentrates. The effect of rancidity in fats on the stability of vitamin A in a poultry ration was also observed. A marked loss of vitamin A was found in halibut liver and in salmon oil on exposure to air in diffuse light in partially filled flasks (Lowen et al., 48).

Anderegg and Nelson (3) noted a characteristic odor of decomposition when cod liver oil was added to dry powders such as skim milk, whole milk, starch or dextrin. Addition of ether, chloroform or benzene did not prevent the decomposition, but alcohol, wheat germ oil or water had a protective action. Buxton (14) found that an antioxidant retarded the destruction of vitamin A in oils. Siedler et al. (68) investigated the value of various

antioxidants as stabilizers for fats added to feed and the effect upon vitamin A and carotene stability. Siedler and Schweigert (69) reported that addition of stabilized animal fat reduced the loss of vitamin A from fish liver oils when mixed and stored as feed.

Burns and Quackenbush (13) studied the effect of several feed ingredients on the stability of vitamin A supplied by several types of commercial concentrates, and reported that stability was enhanced by mixing with soybean oil meal and to a lesser degree corn meal. The addition of the concentrate to glucose (cerelose) diminished the stability of vitamin A.

Wall and Kelley (71), studying factors affecting the stability of vitamin A and carotene in dry mixtures, reported that temperature, concentration, type of carrier and the source of vitamin A had a marked effect on the stability of a number of carotene and vitamin A concentrates. Temperature was the most important variable and in most cases it outweighed all other factors combined. Stability was stated to be inversely proportional to temperature. A comparison of the rate of breakdown of vitamin A added as fish liver oil to feed mixtures stored at 42°F and at room temperatures (66-96°F) showed the rate was twice as high at the higher temperature (Reid, Daugherty and Couch, 61).

Many mineral salts are known to catalyze decomposition of micro-ingredients. Salts of copper, iron, zinc, manganese, and iodine, have been widely incriminated.

Miller and associates (52) demonstrated that a drying agent such as manganese sulphate (0.5 percent in the diet) would destroy vitamin A of cod liver oil in a mixture in which the cod liver oil and manganese was allowed to come in intimate contact. Halverson and Hart (33) showed the extremely

rapid destruction of vitamin A induced by adding Fe, Cu, Co, and Mn to an unsealed container of white corn meal and to cod liver oil was prevented to a large extent when the minerals were added in a dry gelatin-mineral mixture rather than in free form. Effective prevention of vitamin A destruction in practical poultry rations and in certain livestock feeds commonly containing small amounts of added trace minerals is an important problem. Halverson and Hart indicated that a practical solution to the problem depends upon the addition of the trace minerals in a form which limits their ability to come in contact and react with other constituents of the ration.

But, Creek et al. (18) found the addition of 200 P.P.M. of manganese as manganese sulfate to a complete feed or 746 P.P.M. to a concentrate did not affect the stability of thiamine, choline, niacin, calcium pantothenate, riboflavin or carotene in feeds stored 4 months.

Almquist et al. (1) reported evidence that activated charcoal could exert a destructive or inactivating effect on certain vitamins in the ration. This effect could have a serious effect if the ration did not have a sufficient excess of these vitamins to compensate for losses caused by absorption on charcoal. Almquist and Zanden (2) in other studies concluded that a chick diet adequate for normal growth and health was rendered, in effect, deficient in vitamin A, K, B<sub>2</sub> and the gizzard factor by the addition of an activated charcoal.

It has occasionally been reported that pelleting may improve vitamin stability. The fat-soluble vitamins and xanthophylls were originally and still are widely believed to be more stable in alfalfa pellets than in meal form. According to other reports (73,66) this may not be true.

Actually, some carotene is destroyed by the pelleting operation.

Mitchell (53) reported that pellets may contain slightly less carotene than the corresponding meal after several months storage. Mitchell (54) also showed that the carotene of oiled and pelleted dehydrated alfalfa meal was more stable than that of oiled but unpelleted meal and Wornick (74) reported occasional feed formulas in which vitamin A oils were more stable in pellets than in mash.

#### The Stability of Dry Preparations of Vitamin A

Dry preparations of vitamin A have appeared that are stabilized by the addition of an antioxidant and/or by encapsulation in an aerophobic matrix, such as gelatin, ethyl cellulose, gum acacia, wax or hard fat. In some products, the vitamin was adsorbed to particles of carriers usually plant products, or to wax or fat. Synthetic vitamin A palmitate or acetate, and fish oil, are the main sources of the vitamin in such preparations.

Hellstrom et al. (37) studied a basal pig ration that was used to add 13 separate powder forms of vitamin A concentrates to give mixtures containing about 25 I.U. vitamin A per gram. Sample of 200 g. were stored in paper bags for 1, 3, 6, and 12 months at 15°C. After 1 month, 3 vitamin A sources lost 66-92 percent of their activity, 3 others lost 23-31 percent, and in 7 there was no significant loss. In a mixture containing cod liver oil vitamin A was almost absent after 1 month. Benterud (9) reported data on stability of vitamin A concentrates diluted with vegetable oil to 100,000 I.U. per gram and exposed to air in the dark at 37°C. He found the order of stability was from vitamin A acetates with stabilizers added, to natural acetate, to synthetic acetate, to vitamin A alcohol.

Ringnes (63) found a synthetic vitamin A premix powder showed only



insignificant losses after 6 months storage at room temperature in open containers. There was a loss of about 30 percent after 3 months storage when mixed in a chick ration, and much higher losses when intimately mixed with trace minerals. The powder showed lower losses when the potency was greater, and its vitamin stability was much greater than that of cod liver oil.

Halverson and Hendrick (34) determined losses of vitamin A from cod liver oil and two dry preparations when mixed and stored in three vegetable type poultry diets. Studies on effects of diet modifications included addition of meat scraps, limestone, and salts of Mn, Fe, Cu, and Co. Vitamin A stability was fairly good with each diet and was reasonably uniform, except that trace minerals caused greater losses. A wax-coated supplement had greater stability than an oil or fat type supplement.

Reid et al. (61) found there was a rapid loss of vitamin A activity from fish liver oil mixed in several types of rations. A stabilized dry concentrate in which the vitamin was in a micro-crystalline wax carrier usually retained more than 85 percent of its potency after a 4 month storage period.

Davies and Worden (19) concluded, from studies under a variety of storage conditions, that loss of vitamin A was greater in a ration of coarse particulate sizes but pelleting protected the vitamin. The adverse effect of incorporating mineral salts intimately with the vitamin in a ration was confirmed. Some feedstuffs exerted a protective action, the loss being slower in the presence of fish meal or liver meal than various cereal products. Losses were greater in the presence of dried brewer's yeast. A protective value of gelatin was demonstrated.

Geczy (26) studied the stability of a powdered dried vitamin A preparation and a gelatin coated pearl preparation in mixtures with mineral salts at 7.5, 13.0, and 17.7 percent moisture content at normal temperatures and at 65°C. The vitamin A content of powdered dry vitamin A preparations in mixtures decreased to 60, 50, and 40 percent of the starting value during 4 months of storage at normal temperatures and at 7.5, 13.0, and 17.7 percent moisture, respectively. The gelatin coated pearl preparation was stable under these conditions. A powdered dry vitamin A preparation stored 3 days in mixtures at 65°C lost 30-65 percent vitamin A, while a gelatin coated pearl preparation lost only 5-25 percent, respectively.

#### The Availability of Dry Preparations of Vitamin A for Animals

While these products (dry preparations of vitamin A) represent considerable progress in the conservation of vitamin A, they raise the question of relative stability, availability and utilization. The possibility exists that stabilization processes could be carried so far that the vitamin was unavailable to the animal organism.

Nestler et al. (56) reported that vitamin A acetate was utilized more efficiently by quail than either the alcohol or the natural ester. Carotene in cottonseed oil or in alfalfa meal were equally effective, but much less so than stabilized vitamin A sources. Gurcay et al. (30), on the basis of growth, liver storage and blood plasma levels of poults, concluded that crystalline vitamin A acetate was used most effectively, while carotene least effectively and cod liver oil intermediate. Castano et al. (16) found crystalline vitamin A acetate to be more effective than "black cod" liver oil or crystalline carotene, as judged by blood plasma concentration and liver

storage. Kramke et al. (47) using growth of chicks and poults to measure the utilization of vitamin A reported that a dry stabilized vitamin supplement gave better results than the U.S.P. reference standard oil solution.

Gledhill and Smith (27) reported that chicks receiving vitamin A from a dry carrier made significantly greater gains to 10 weeks of age, and had a better feed efficiency, higher liver storage of the vitamin, and lower mortality, than chicks receiving the vitamin from fish oil or alfalfa. The dry vitamin A oil used was encapsulated in hydrogenated fat of a critical melting point and dispersed throughout a finely divided base of defatted, enzyme inactivated soybean flour. The process also involved a spray chilling operation with antioxidants and an emulsifying agent included in the fatty vehicular material. The authors concluded the dry vitamin A carrier presented the vitamin in a more efficient form. Harms et al. (35) compared fish oil and a stabilized vitamin A concentrate (made by the same process as that used by Gledhill and Smith (27)), using chick growth and liver storage as criteria in experiments of 4 or 10 weeks duration. They showed that growth and liver storage of chicks fed the stabilized vitamin A, was increased over that of chicks fed fish oil.

Camp et al. (15) reported that substitution of 2,000 I.U. of dry, stabilized vitamin A per pound of feed for 2,300 I.U. as dehydrated alfalfa meal, in a high energy broiler ration, gave a significant increase in growth and an improvement in feed efficiency. Høie and Sandvik (39), using dry or stabilized vitamin A products, found no differences in growth rate when the feeds were used at one week or stored for a longer period. Matterson et al. (51) found that liver storage of vitamin A in chicks at 8 weeks of age, fed various levels of fish oil, were several times greater when diphenyl-

phenylenediamine (DPPD) was included in the ration. Plasma levels of vitamin A were increased when DPPD was included in the ration and maximum growth was obtained with one-third less of the vitamin. Ascarelli (5) found that vitamin A, supplied as a stabilized concentrate was utilized better by chicks than vitamin A supplied as fish oil. Fritz et al. (24) observed that fat-soluble coating agents were effective in protecting vitamin A, but care in selection of the coating agent was necessary to insure that the coated vitamin was available to the chick. High-melting fats, used as coating agents, impaired the utilization of the vitamin. Water-soluble coating agents were satisfactory only when the feed was stored at low humidities. The addition of Butylated Hydroxyanisole (BHA) or DPPD improved the apparent utilization of vitamin A or carotene.

Particle size was also reported a significant factor in biological availability of vitamin A. Luther (49) using rat liver storage and vitamin A stabilized in aqueous emulsion vehicles established that biological availability varied with particle size and that for best availability, particles should be in a range below 15 microns in diameter and preferably from 1 to 6 microns. Luther (49) also found similar effects using a gelatin-stabilized vitamin A product with controlled particle size in tests comparing oil-carried vitamin A with rats, chickens, swine, calves, steers, and infants. He concluded the gelatin-stabilized vitamin form had considerably greater biological availability over the oil-carried form. Brubacher et al. (12) using a tracer technique to determine absorption efficiency of gelatin-stabilized vitamin A in chickens, showed that at least 97 percent of the vitamin A intake was absorbed, thus providing evidence the stabilized form was absorbed even more completely than the unstabilized oil preparations.

Roche (64) showed the results of a biological availability study with chicks under both laboratory and commercial conditions. Using liver storage of vitamin A as criterion their data showed Rovimix A-650 (acetate) had equal bioavailability to Rovimix A-325 (palmitate). There was evidence that biological availability of vitamin A was not affected by its chemical form, either as an alcohol, acetate or palmitate.

#### The Stability of Dry Stabilized Vitamin A

Many stabilized dry vitamin A preparations now consist of antioxidant-protected vitamin A finely dispersed throughout a gelatin-type base. Olsen et al. (57) studied the availability and stability during storage in a mixed ration, of vitamin A in several commercial dry-form preparations, and found the liver storage of a vitamin A gelatin coated preparation was superior to preparations in which the vitamin was coated with wax or fat, or was absorbed to vegetable protein. Feeding cod liver oil and dehydrated cereal grass were the poorest sources. The superiority of gelatin preparations was indicated. Wornick (74) showed that micro-ingredient stability was strongly affected by feed composition. Tests in Pfizer laboratories on eight different commercial pelleted feed formulas showed the two-month retention of the same lot of vitamin A in gelatin varied from 54-98 percent.

Luther (49) studied the stability of various vitamin A products with different mixtures over five years and brought together stability test results that follow:

# FIVE YEAR STABILITY SUMMARY OF VITAMIN PRODUCTS<sup>1</sup>

Numbers represent percent potency retention after 12 weeks storage, unless otherwise noted. Figures in parenthesis are the number of lots averaged (8 determinations per sample).

Type Product	STABILITY - PERCENT RETENTION			
	High-Moisture Feed 70% R.H., 88°F	Mineral Mix 115°F	Broiler Mash 70°F	Pelleted Broil. Mash 70°F
Gel. Stab.	<u>85%</u> (45) Range 73-97%	<u>72%</u> (20) 51-93%	<u>85%</u> (19) 77-92%	<u>68%</u> (18) 52-88%
Fish Liver Oil	<u>17%</u> (13) Range 12-23%	<u>0%</u> <sup>†</sup> (13) 0-0%	<u>26%</u> (8) 11-45%	<u>27%</u> (8) 15-36%
Wax Stab.	<u>62%</u> <sup>*</sup> (17) Range 22-87%	<u>25%</u> <sup>*</sup> (20) 0-69%	<u>53%</u> (4) 30-70%	<u>33%</u> (4) 29-39%
Fat Stab.	<u>27%</u> (6) Range 19-56%	<u>10%</u> <sup>φ</sup> (6) 0-14%	<u>37%</u> (2) 33-38%	<u>35%</u> (2) 33-37%
<div> <div>† 2-week data</div> <div>φ 6-week data</div> <div>* 8-week data</div> </div>				

<sup>1</sup>From Chas. Pfizer Company, Sixth Annual Research Conference Proceedings, p. 78.

This compilation shows the poor stability of fish liver oil in feeds and its rapid destruction in mineral preparations. The wax-stabilized products have exhibited fair stability in most instances and poor stability in mineral mixtures. There has been a fairly wide range of values found among products of the type studied. The fat-stabilized products did not exhibit good stability in any of the test systems. Gelatin-stabilized products as a group showed good stability under all conditions. There was a fairly wide range in stability in mineral mixtures, this was caused by the poor stability of one of the products included.

In view of the severe conditions imposed during feed pelleting, it is not surprising that stability problems were found with various vitamin A sources. Bierer (11) reported a problem in turkey breeder "fresh" pellets which had suffered a 32 percent loss in vitamin A potency. Wornick (74) showed the results of commercial pelleting tests on various vitamin A sources. The potency losses of stabilized vitamin A as a result of the pelleting operation itself were seldom significant, but losses in pelleted feeds during storage usually were greater than in mash and he concluded that pelleted feeds could be stored from 8 to 12 weeks before being consumed.

Bauernfeind (7) showed that a concentrate of synthetic stabilized vitamin A was two to eight times as stable as fish oil stored under similar conditions. In five comparative tests, in which the stabilized supplement and a fish oil were individually mixed into complete feeds and stored for 16 weeks at 75°F, the average retention of the stabilized supplement was reported to be 90 percent, whereas the feed containing fish oil retained only 28 percent of the original vitamin A content. Klaui et al. (46) showed the losses of vitamin A can be minimized by using it in gelatin encapsulated form. Vitamin A retention in mixes of feeds were: after 1 month storage at 45°C and humid conditions 4.85-54.1 percent, 1 month at 45°C and dry 5.3-97.0 percent. In pelleted feeds after 1 month at 45°C and humid conditions, 13.2-38.6 percent and 1 month at 45°C and dry 28.9-57.8 percent. Humphris (42) reported studies on six commercial poultry mashes, containing vitamin A stabilized in a gelatin base that were stored for 6 months under normal summer conditions. Vitamin A and carotene was determined monthly. They retained 74-90 percent of their initial vitamin A content and 16-40 percent of their initial carotene. Geczy (25) showed that at 65°F and 7.5 percent

moisture, 6-7 percent of the vitamin A activity was lost after 72 hours in the presence of strong oxidizing mineral mixtures containing stable pearl-shaped granules of vitamin A.

Florenskaya et al. (21) showed decreases of vitamins occurred in feed enriched with protein preparations containing either vitamins alone or vitamins and salts of Co, Cu, Mn, Fe, and I. Before addition to the feed the preparations were mixed with soybean groats. The vitamin content of the feed was estimated after 2, 4, and 8 weeks in the enriched feed. The content of cyanocobalamin, riboflavin, vitamin A, vitamin PP, and choline chloride decreased by 69, 26, 10-25, 13-20, and 13-20 percent, respectively. In feed enriched with the preparation containing trace elements, cyanocobalamin and riboflavin decreased by 87 and 30-33 percent, respectively. The decomposition of vitamins caused by trace elements was reduced by higher levels of soybean groats. Pararskyte et al. (58) studied the effect of a few trace elements added to feed mixtures enriched with a number of vitamins on their preservation. The level of vitamins was tested before the experiment and then monthly for 6 months. The addition of trace elements in solution as well as in dry form had no practical effect on the preservation of vitamins during storage. The concentration of riboflavin decreased during this period by 45-53 percent, and the amount of nicotinic acid by 63-66 percent. The analogous figures for the decrease of vitamin A and D<sub>2</sub> were 17-25 percent and 10-36 percent, respectively, after the first month of storage, and 22-34 percent and 16-30 percent, respectively, after 6 months. The amounts of choline, vitamin B<sub>12</sub> and biomyacin were not altered.



## MATERIALS AND METHODS

### Materials Preparation

Three 32 percent crude protein cattle supplements using different vitamin A sources were prepared. The composition of the diets is shown in Table 1. Diet 1 was a soybean meal based cattle supplement. Diet 2 contained 4 percent urea as partial replacement of the soybean meal protein. Diet 3 contained 4 percent urea and 4 percent fat. The three experiential formulas were adjusted to the same nutrient composition being formulated to crude protein, energy, calcium and phosphorous levels based on National Research Council recommendation (55).

The vitamin A sources were added to each diet at a level of 50,000 I.U. per pound of feed based on the potency guaranteed by the manufacturer.

The vitamin A products used in these studies consisted of Rovimix A-650 and Rovimix A-325 and which were stabilized gelatin-sugar starch beadlets containing vitamin A as the acetate and palmitate esters. These products contained a minimum of 650,000 I.U. and 325,000 I.U. vitamin A per gram.

The vitamin A used in producing the formulas was kept in a freezer (-20°C) till the time of production of the formulas. Material for producing the formulas was then removed and the remainder was returned to the freezer.

Ten pounds of premix containing sufficient quantities of minerals and of each test vitamin A product to fortify 1,000 pounds of ration were prepared on the day the feed was to be pelleted.

TABLE 1. COMPOSITION OF THE CATTLE SUPPLEMENTS USED FOR VITAMIN A STABILITY STUDIES

Ingredient Bulk (lbs)	Diet 1			Diet 2			Diet 3		
	A	B	C	A	B	C	A	B	C
Soybean Oil	647	647	647	325	325	325	336	336	336
Meal (44%)									
Ground Sorghum	93	93	93	190	190	190	219	219	219
Grain									
Dehydrated Alfalfa	50	50	50	50	50	50	50	50	50
Meal									
Ground Wheat	100	100	100	180	180	180	100	100	100
Wheat Midds	-	-	-	100	100	100	100	100	100
Molasses	50	50	50	50	50	50	50	50	50
Fat	-	-	-	-	-	-	40	40	40
Dicalcium Phosphate	30	30	30	35	35	35	35	35	35
Salt	20	20	20	20	20	20	20	20	20
Urea (45% N.)	-	-	-	40	40	40	40	40	40
<u>Sub-Total 1</u>	990	990	990	990	990	990	990	990	990
Premix (grams)									
Chelated Trace Minerals <sup>a</sup>	454	454	454	454	454	454	454	454	454
Rovimix <sup>®</sup> A-325 <sup>b</sup>	-	154	-	-	154	-	-	154	-
Rovimix <sup>®</sup> A-650 <sup>b</sup>	-	-	77	-	-	77	-	-	77
Ground Sorghum	4086	3932	4009	4086	3932	4009	4086	3932	4009
Grain									
<u>Sub-Total 2</u>	4540	4540	4540	4540	4540	4540	4540	4540	4540
Total (lbs.)	1000	1000	1000	1000	1000	1000	1000	1000	1000

<sup>a</sup>Manufactured by Erly-Fat Livestock Feed Company, Guaranteed Analysis:  
Iodine 0.085%; Iron (Fe) 1.60%; Manganese (Mn) 1.65%; Copper (Cu) 0.49%;  
Cobalt (Co) 0.41%; and Zinc (Zn) 2.15%.

<sup>b</sup>Trademark registered, produced by Hoffman-La Roche Company.

### Manufacturing Conditions and Procedure

The manufacturing conditions used in these studies were: three replicated pelleting trials under each of the following conditions:

(1) Conditioning temperature during pelleting was adjusted as nearly as possible to 70°C, and 50°C. It was not possible to pellet the 4 percent urea formulas at a temperature higher than 50°C. Therefore, the 70°C study was not conducted.

(2) Pelleting studies were conducted using a 3/16 inch diameter round hole die with a 1 3/4 inch thickness.

The grain used in the formulas was ground through a 1/8 inch hammermill screen. Premixes were added in the ration when mixing was started. Fat and molasses in each trial was added at the horizontal batch mixer and mixing continued an additional four minutes before being discharged. Total mixing time for these formulas was seven minutes.

The feed material for each test consisted of one 1,000 pound batch. Following mixing, ingredients were transferred to a bin above the pellet mill. The pellet mill used was equipped with a conditioning chamber and steam was available to condition the feed.

In each test the ration containing 0.0 percent vitamin A (control) was pelleted first to establish its optimum feed rate. The rations with Rovimix A-325, and Rovimix A-650 were pelleted at the same rates of production.

Pelleting was with a 25 hp California Master Model Pellet Mill. Pellets were cooled in a California Pellet Mill vertical pellet cooler, size 2B.

Pellets dropped into the vertical pellet cooler with the fan on during pelleting. The desired pellet mill operating conditions were reached before

pellets were discharged into the cooler. The pellets in the cooler were cooled an additional ten minutes after pelleting of the batch was complete.

After cooling, the pellets were cycled from the cooler to and over a scalper and then to the sack-off bin to be packed and weighed.

The temperatures of the mash before conditioning (in mixer) and after going through the conditioning chamber were recorded. The temperatures of the pellets before and after cooling were recorded.

The fines from the scalper were collected for each ration on all pellets given the ten minute cooling treatment and weight of fines was recorded.

#### Sampling and Storage

Samples of the material were collected and moisture determinations were made on the mash (in mixer), on the conditioned mash, on hot pellets and on cooled pellets (at sack-off). Samples of the feed were collected on the mash (in mixer) for particle size determination using the ASAE official method (73).

Samples of the test mash were collected in the mixer at different positions and samples of the test pellets were collected periodically at sack-off for vitamin A tests of all rations. Three 50 pound bags of each material produced (mash or pelleted) were stored for sampling at various storage intervals.

The sacks used permitted free moisture passage during storage. Bags were stacked flat in rows with an inch space between adjacent stacks. Temperatures in the area where the samples were stored were monitored to obtain information on normal temperature variation during the storage period (Tables 8, 9, and 10).

Vitamin A analyses were done on the initial mash material prior to pelleting and on the cooled pellets at 0, 2, 4, 8, and 16 weeks under room storage conditions.

Sampling was done using a probe sampler. Samples were collected by inserting the probe diagonally starting at one corner and inserting it to the opposite corner at the other end of the bag.

Both mash and pellet samples were resampled for assay initially and at each interim period. Approximately 5 pounds of sample of each diet were taken at each sampling. Following sampling, all samples were placed in a freezer until prepared for shipment.

The preparation for shipment was as follows: the samples were taken from the deep freeze and reduced to approximately a one pound sample using a sample splitter. This sample was placed in a plastic bag that was then flushed with nitrogen, closed, and then placed inside a manila envelope. These were placed in a second plastic bag which was also flushed with nitrogen and placed inside a larger container. After all samples were placed in the container it was flushed with nitrogen prior to closing. The container was shipped by air freight that afternoon to Hoffman-La Roche Laboratory in Nutley, New Jersey for vitamin A analysis.

## RESULTS AND DISCUSSION

Table 2 shows results of particle size tests with Rovimix A-325, Rovimix A-650 and the meal mix for three cattle supplements. Information obtained on geometric mean diameter (DGW) by weight distribution of sample, geometric log-normal standard deviation of sample estimate by weight distribution, the sample particle range from 16-84%, the total area per gram and the number of particles per one gram sample are given in Table 2.

TABLE 2. PARTICLE SIZE ANALYSIS<sup>a</sup>

	DGW <sup>b</sup> (microns)	SGW <sup>c</sup>	68% Particle Size Range (microns)	Surface Area sq. cms. per gram	No. of Particles per gram
Rovimix A-325	335	1.478	226-495	155	42433
Rovimix A-650	274	1.380	198-338	185	62263
Cattle Supplements:					
1. Based Soybean Oil Meal	635	2.076	306-1317	94	32888
2. Containing 4% Urea	645	2.157	299-1391	95	37470
3. Containing 4% Urea and 4% Fat	661	1.972	335-1303	86	20795

<sup>a</sup>Each value is the average of three replications.

<sup>b</sup>DGW = Geometric Mean Diameter by Weight Distribution of sample.

<sup>c</sup>SGW = Geometric Log-Normal standard deviation of sample estimate by weight distribution.

The number of particles per one gram sample of Rovimix A-650 was much higher than Rovimix A-325. The particle size of the three cattle supplements

were similar except that the diet containing 4% urea and 4% fat was somewhat higher. The range in particle size of the two vitamin A products (68% of the particle) was similar to the smaller particles found for supplement ingredients.

Data was also obtained on the higher potency vitamin A to determine if the smaller quantity of Rovimix A-650 required to fortify feeds would provide for a uniform dispersion. The comparisons to Rovimix A-650 and Rovimix A-325 are presented in Table 3.

TABLE 3. COMPARISONS OF BEADLET SIZE, BEADLET COUNT/GRAM, POTENCY AND DISTRIBUTION IN FEED WITH ROVIMIX A-650 VS. ROVIMIX A-325

	Rovimix A-650	Rovimix A-325
Beadlet size range, microns (includes 68% of particle)	198-338	226-495
Number of beadlets/gm., av.	62,263	42,433
Units of activity per gm.	650,000	325,000
I.U. per beadlet, av.	10.4	7.7
Distribution in feed at 50,000 I.U./lb.:		
Beadlets/454 grams of finished feed	4,808	6,493
Estimated coefficient of variation based on number of particles, %	1.44	1.24

Upon consideration of this data, it appears that the greater number of beadlets in a gram of Rovimix A-650 should assume a uniform dispersion in finished feed.

Data presented in Table 4 gives the moisture content of the samples from different locations. The survey shows that added moisture during conditioning

was 0.5-1.0% at 50°C conditioning temperature, and added moisture levels were 1.8-2.4% at 70°C conditioning temperature. Pelletting with a high conditioning temperature (70°C) would be expected to add more moisture to the mash than a low conditioning temperature (50°C). The moisture contents were nearly the same in the mash before conditioning and for the cooled pellets after cooling in all three cattle supplements. Beginning mash and final pellet moisture levels were similar even though conditioning temperatures were different.

Data presented in Table 5 shows the fines during pelletting and production rates for the three cattle supplements under different conditioning temperatures. Scalper fines were low for all three cattle supplements. All contained 5% molasses which would be expected to reduce fines. Differences between the three cattle supplements were small but the data on scalper fines indicates a trend in favor of the cattle supplements containing 4 percent urea and no fat. It should be noted, however, that the time required to pellet the formulas varied with the 4 percent urea cattle supplement requiring more time to pellet than other cattle supplements. Therefore, the improvement in amount of scalper fines was a result of slower pelletting. Adding 4 percent fat decreased time needed for pelletting. This was also reported by Headley (36). Increasing conditioning temperatures decreased the time to pellet cattle supplements based on soybean oil meal, but this trend did not occur with cattle supplements containing 4% urea and 4% fat. The cattle supplement containing 4% urea, alone, could only be pelleted at 50°C.

The proximate composition of the three cattle supplements determined using AOAC methods is shown for 0 and 24 weeks storage (Tables 6 and 7). These values show equivalent protein, fat, ash and fiber content for the



supplements at 0 weeks and 24 weeks. Moisture content decreased about 4 to 6 percent during 24 weeks of storage and protein and fat were proportionately increased.

The levels of Rovimix A-650 and Rovimix A-325 in three cattle supplements stored at room temperature for four months appear in Tables 8, 9, and 10. The average values of vitamin A given in each table includes data from three replicated tests. A minimum of two assays was performed on each sample analyzed.

The vitamin A levels with the mixed mash diets containing 4% urea or 4% urea and 4% fat (Diet 2 or Diet 3) were lower ( $P \leq 0.05$ ) than those observed with the mash diet based on soybean oil meal (Diet 1). Diets 2 and 3 were equal. The analysis of variance (Tables 11 and 12) indicate the differences were statistically significant at  $P \leq 0.05$ . This difference may have resulted from segregation and sampling errors which are indicated by the increased amount of variation in this diet when compared to the other two diets.

The influence of feed processing on vitamin A levels in mash and pelleted diets (50°C and/or 70°C) containing Rovimix A-325 and Rovimix A-650 is also given in Tables 8, 9, and 10. Vitamin A levels for pellets were lower than mash and were equal at 50°C and 70°C. Levels were approximately 15% lower in pellets than in mash.

Losses of vitamin A during storage in the diet mixtures tested were not significant. The extent of the vitamin A losses during storage were not dependent upon the length of storage, pelleting temperature, diet composition, or diet forms (mash, pellet). These results are not in agreement with those reported by Wornick (74).

Vitamin A levels were significantly greater in the Rovimix A-650 than

Rovimix A-325 ( $P \leq 0.05$ ). These values represent averages of three tests and also represent the total vitamin A retention from the original mash to the final stored pellets.

It should also be noted that initial vitamin A activity, not including carotene for the three cattle supplements, was generally higher than expected at 0 weeks of storage, probably because the vitamin A contained a higher level than guaranteed. All diets had vitamin A contents above calculated vitamin A levels, even after four months storage except for the pelleted diet (50°C or 70°C) containing 4% urea and 4% fat.

TABLE 4. MOISTURE CONTENT OF SAMPLES FROM DIFFERENT LOCATIONS

Diet	Vitamin A Sources	Form of Diets	Moisture %			
			Mash		Pellets	
			Conditioned Before	After	Cooling Before	After
Diet 1 <sup>a</sup>	No Vitamin A	Pellet (50°C)	11.0	11.5	11.6	10.5
		Pellet (70°C)	10.8	12.7	12.2	10.3
	Rovimix A-325	Pellet (50°C)	10.6	11.2	11.2	9.9
		Pellet (70°C)	10.7	12.5	11.6	10.0
	Rovimix A-650	Pellet (50°C)	10.4	11.1	11.2	9.8
		Pellet (70°C)	10.5	12.3	11.8	10.0
Diet 2 <sup>b</sup>	No Vitamin A	Pellet (50°C)	11.1	11.5	10.9	9.4
	Rovimix A-325	Pellet (50°C)	11.1	11.4	10.7	10.0
	Rovimix A-650	Pellet (50°C)	10.8	11.3	10.8	9.5
Diet 3 <sup>c</sup>	No Vitamin A	Pellet (50°C)	10.4	11.2	11.7	10.5
		Pellet (70°C)	10.5	12.7	12.9	10.7
	Rovimix A-325	Pellet (50°C)	10.5	11.2	11.3	10.4
		Pellet (70°C)	10.6	12.5	12.6	10.9
	Rovimix A-650	Pellet (50°C)	10.6	11.2	11.5	10.4
		Pellet (70°C)	10.8	12.6	12.7	11.0

<sup>a</sup>Cattle Supplement based on soybean oil meal.

<sup>b</sup>Cattle Supplement containing 4 percent urea.

<sup>c</sup>Cattle Supplement containing 4 percent urea and 4 percent fat.

TABLE 5. THE AMOUNT OF FINES AND TIME IN THE THREE CATTLE SUPPLEMENTS WITH DIFFERENT CONDITIONING TEMPERATURES

Diet	Vitamin A Sources	Form of Diets	Scalper Fines (%)	Minutes to Pellet (1,000 lb.)
Diet 1 <sup>a</sup>	No Vitamin A	Pellet (50°C)	1.85	28.8
		Pellet (70°C)	1.20	18.8
	Rovimix A-325	Pellet (50°C)	1.02	27.5
		Pellet (70°C)	1.58	20.3
	Rovimix A-650	Pellet (50°C)	1.03	26.1
		Pellet (70°C)	1.24	21.7
Diet 2 <sup>b</sup>	No Vitamin A	Pellet (50°C)	0.72	36.1
	Rovimix A-325	Pellet (50°C)	0.73	39.0
	Rovimix A-650	Pellet (50°C)	0.85	41.0
Diet 3 <sup>c</sup>	No Vitamin A	Pellet (50°C)	1.51	27.9
		Pellet (70°C)	2.51	27.2
	Rovimix A-325	Pellet (50°C)	2.79	23.7
		Pellet (70°C)	3.03	25.7
	Rovimix A-650	Pellet (50°C)	2.35	26.0
		Pellet (70°C)	2.56	27.9

<sup>a</sup>Cattle Supplement based on soybean oil meal.

<sup>b</sup>Cattle Supplement containing 4 percent urea.

<sup>c</sup>Cattle Supplement containing 4 percent urea and 4 percent fat.

TABLE 6. COMPOSITION OF THREE CATTLE SUPPLEMENTS AT 0 WEEK STORAGE

Diet	Vitamin A Sources	Form of Diets	Moisture %	Protein %	Fat %	Ash %	Fiber %
Diet 1 <sup>a</sup>	No Vitamin A	Mash	10.9	31.0	2.6	9.2	5.7
		Pellet (50°C)	10.5	31.1	2.4	9.2	5.8
		Pellet (70°C)	10.3	31.3	2.5	9.2	5.8
	Rovimix A-325	Mash	10.7	31.7	2.5	9.4	5.5
		Pellet (50°C)	9.9	31.4	2.6	9.3	5.9
		Pellet (70°C)	10.0	31.5	2.6	9.4	5.7
	Rovimix A-650	Mash	10.4	32.2	2.7	9.0	6.0
		Pellet (50°C)	9.8	32.0	2.6	9.0	5.8
		Pellet (70°C)	10.0	31.1	2.6	9.1	6.0
	No Vitamin A	Mash	11.1	32.2	2.0	8.0	5.3
		Pellet (50°C)	9.4	32.5	2.0	8.2	4.6
Diet 2 <sup>b</sup>	Rovimix A-325	Mash	11.1	32.0	2.2	8.4	5.0
		Pellet (50°C)	10.1	32.9	2.3	8.4	4.9
	Rovimix A-650	Mash	10.8	30.3	2.0	8.0	5.0
		Pellet (50°C)	9.5	31.0	2.5	8.3	4.7
	No Vitamin A	Mash	10.5	33.1	5.6	8.0	4.8
		Pellet (50°C)	10.5	33.0	5.7	8.1	4.9
		Pellet (70°C)	10.7	32.8	5.9	8.4	4.8
	Rovimix A-325	Mash	10.6	32.9	6.3	8.5	5.0
		Pellet (50°C)	10.4	33.0	6.4	8.4	4.7
		Pellet (70°C)	10.9	33.0	6.2	8.4	2.2
Diet 3 <sup>c</sup>	Rovimix A-650	Mash	10.7	32.3	6.3	8.2	4.8
		Pellet (50°C)	10.4	32.8	6.2	8.2	4.5
		Pellet (70°C)	11.0	33.1	5.9	8.4	4.3
	No Vitamin A	Mash	10.5	33.1	5.6	8.0	4.8
		Pellet (50°C)	10.5	33.0	5.7	8.1	4.9
		Pellet (70°C)	10.7	32.8	5.9	8.4	4.8

<sup>a</sup>Cattle Supplement based on soybean oil meal.<sup>b</sup>Cattle Supplement containing 4 percent urea.<sup>c</sup>Cattle Supplement containing 4 percent urea and 4 percent fat.

TABLE 7. COMPOSITION OF THREE CATTLE SUPPLEMENTS AT 24 WEEKS STORAGE

Diet	Vitamin A Sources	Form of Diets	Moisture %	Protein %	Fat %
Diet 1 <sup>a</sup>	No Vitamin A	Mash	6.7	31.2	2.8
		Pellet (50°C)	6.6	31.7	2.5
		Pellet (70°C)	6.8	32.1	2.8
	Rovimix A-325	Mash	6.1	31.8	2.8
		Pellet (50°C)	6.7	32.6	2.9
		Pellet (70°C)	7.0	32.8	2.9
	Rovimix A-650	Mash	6.6	31.8	2.7
		Pellet (50°C)	6.2	32.2	2.8
		Pellet (70°C)	6.3	31.9	2.9
Diet 2 <sup>b</sup>	No Vitamin A	Mash	5.7	33.2	2.5
		Pellet (50°C)	5.3	33.9	2.3
	Rovimix A-325	Mash	5.5	33.6	2.6
		Pellet (50°C)	5.2	34.2	2.4
	Rovimix A-650	Mash	5.9	31.8	2.4
		Pellet (50°C)	5.4	32.3	2.2
Diet 3 <sup>c</sup>	No Vitamin A	Mash	5.2	34.2	5.7
		Pellet (50°C)	5.4	34.4	5.6
		Pellet (70°C)	6.1	34.1	5.7
	Rovimix A-325	Mash	5.1	34.0	6.6
		Pellet (50°C)	4.7	34.7	6.4
		Pellet (70°C)	4.6	34.8	6.3
	Rovimix A-650	Mash	5.3	33.9	6.4
		Pellet (50°C)	4.7	34.0	6.4
		Pellet (70°C)	4.8	34.4	6.2

<sup>a</sup>Cattle Supplement based on soybean oil meal.

<sup>b</sup>Cattle Supplement containing 4 percent urea.

<sup>c</sup>Cattle Supplement containing 4 percent urea and 4 percent fat.

TABLE 8. VITAMIN A CONTENT OF A CATTLE SUPPLEMENT (SOYBEAN MEAL BASE)  
AFTER PERIODS OF STORAGE<sup>a</sup>

Vitamin A Sources	Forms of Diet	Nominal Vitamin A Level	Vitamin A Remaining (International Units per lb.)											
			Weeks of Storage, and Temperature Ranges											
			0	2 (33-27°C)	4 (33-23°C)	8 (31-21°C)	16 (29-18°C)							
		%	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	
Rovimix A-325	Mash	50,000	100	65,567	131	61,667	123	60,167	120	59,000	118	70,100	140	
	Pellet (50°C)	50,000	100	51,900	104	49,900	100	54,216	108	51,607	103	49,550	99	
	Pellet (70°C)	50,000	100	49,200	99	48,833	98	50,133	100	50,278	101	49,626	99	
Rovimix A-650	Mash	50,000	100	61,267	123	73,733	147	66,333	133	59,500	119	78,800	157	
	Pellet (50°C)	50,000	100	60,333	121	57,233	115	50,383	101	51,000	102	51,355	103	
	Pellet (70°C)	50,000	100	55,600	111	54,000	108	52,666	105	51,107	102	53,766	108	

<sup>a</sup>Storage was in a warehouse under normal environment conditions. Products were prepared in June and July.

<sup>b</sup>Values are given as percent remaining from nominal vitamin A level.

TABLE 9. VITAMIN A CONTENT OF A CATTLE SUPPLEMENT (CONTAINING 4% UREA)  
AFTER PERIODS OF STORAGE<sup>a</sup>

Vitamin A Sources	Forms of Diet	Nominal Vitamin A Level	Vitamin A Remaining (International Units per lb.)										
			Weeks of Storage, and Temperature Ranges										
			0	2 (33-27°C)	4 (33-23°C)	8 (31-21°C)	16 (29-18°C)						
		%	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>		
Rovimix A-325	Mash	50,000	100	57,133	114	54,233	109	51,500	103	58,117	116	51,153	102
	Pellet (50°C)	50,000	100	52,533	105	51,700	103	51,350	103	51,533	103	50,383	101
Rovimix A-650	Mash	50,000	100	55,033	110	67,167	134	54,833	110	56,667	113	60,850	122
	Pellet (50°C)	50,000	100	53,700	107	48,867	98	51,333	103	50,000	100	47,650	95

<sup>a</sup>Storage was in a warehouse under normal environment conditions. Products were prepared in June and July.

<sup>b</sup>Values are given as percent remaining from nominal vitamin A level.



TABLE 10. VITAMIN A CONTENT OF A CATTLE SUPPLEMENT (CONTAINING 4% UREA AND 4% FAT)  
AFTER PERIODS OF STORAGE<sup>a</sup>

Vitamin A Sources	Forms of Diet	Nominal Vitamin A Level	Vitamin A Remaining (International Units per lb.)										
			Weeks of Storage, and Temperature Ranges										
			0	2 (33-27°C)	4 (33-23°C)	8 (31-21°C)	16 (29-18°C)	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	% <sup>b</sup>	
Rovimix A-325	Mash	50,000	100	58,633	117	55,400	111	52,883	106	59,335	119	52,400	105
	Pellet (50°C)	50,000	100	47,367	95	44,467	89	46,683	93	44,333	89	43,850	88
	Pellet (70°C)	50,000	100	50,367	101	49,733	100	53,017	106	53,333	107	47,917	96
Rovimix A-650	Mash	50,000	100	62,933	126	59,767	120	60,683	121	56,916	114	55,558	111
	Pellet (50°C)	50,000	100	55,400	111	54,700	109	52,667	105	49,000	98	46,792	94
	Pellet (70°C)	50,000	100	51,867	104	55,400	111	52,833	106	45,667	91	48,800	98

<sup>a</sup> Storage was in a warehouse under normal environment conditions. Products were prepared in June and July.

<sup>b</sup> Values are given as percent remaining from nominal vitamin A level.

TABLE 11. ANALYSIS OF VARIANCE FOR VITAMIN STABILITY FOR DIET 1,  
DIET 2, AND DIET 3 COMPARING MASH AND PELLETS AT 50°C

Source of Variation	Degrees of Freedom	Mean Square	F Value
Main Plots:			
Replication	2	257.4102	0.764
Treatment	1	15124.9883	44.899**
Diet	2	2691.0625	7.988**
Treatment X Diet	2	761.8225	2.262
Vitamin	1	2053.6782	6.096*
Treatment X Vitamin	1	115.1909	0.342
Diet X Vitamin	2	165.5681	0.492
Treatment X Diet X Vitamin	2	274.3184	0.814
Main plot error	22	336.8711	2.949
Sub-plots:			
Storage Time	4	242.2705	2.121
Treatment X Time	4	297.9685	2.608
Diet X Time	8	231.8427	2.029
Treatment X Diet X Time	8	219.1423	1.918
Vitamin X Time	4	258.6614	2.264
Treatment X Vitamin X Time	4	280.3943	2.454
Diet X Vitamin X Time	8	48.4818	0.424
Treatment X Diet X Vitamin X Time	8	94.0318	0.823
Sub-plot error	96	114.2472	
Total	179		

\*\* Significant at  $P \leq 0.01$  level.

\* Significant at  $P \leq 0.05$  level.

TABLE 12. ANALYSIS OF VARIANCE FOR VITAMIN STABILITY FOR DIET 1  
AND DIET 3 COMPARING MASH, PELLETS AT 50°C  
AND PELLETS AT 70°C

Source of Variation	Degrees of Freedom	Mean Square	F Value
Main Plots:			
Replication	2	336.0068	1.110
Treatment	2	8983.3203	29.676**
Diet	1	3380.0027	11.166**
Treatment X Diet	2	825.7068	2.728
Vitamin	1	2177.0947	7.192*
Treatment X Vitamin	2	112.7048	0.372
Diet X Vitamin	1	6.4206	0.021
Treatment X Diet X Vitamin	2	214.4031	0.708
Main plot error	22	302.7136	3.010
Sub-plots:			
Storage time	4	235.3444	2.340
Treatment X Time	8	149.3376	1.485
Diet X Time	4	334.1252	3.322*
Treatment X Diet X Time	8	218.1533	2.169
Vitamin X Time	4	308.8762	3.071*
Treatment X Vitamin X Time	8	98.9345	0.984
Diet X Vitamin X Time	4	49.3528	0.491
Treatment X Diet X Vitamin X Time	8	77.6902	0.772
Sub-plot error	96	100.5838	
Total	179		

\*\* Significant at  $P \leq 0.01$  level.

\* Significant at  $P \leq 0.05$  level.

## SUMMARY

Data have been presented showing the stability of two vitamin A products. These were compared under different pelleting conditions using three cattle supplements.

Analysis of vitamin A retentions indicated some loss of vitamin A during pelleting. Conditioning temperatures during pelleting did not affect vitamin A retentions. Values for vitamin A levels during 4 months of storage did not show significant changes in vitamin A levels in either mash or pellets. Differences in vitamin losses were not found to be affected by diet formulation.

The vitamin A values found in the diets at all sampling periods were higher when Rovimix A-650 (acetate) was the source. Rovimix A-325 (palmitate) values were lower. This probably indicates higher overformulation in the original material.

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THE STABILITY OF VITAMIN A PRODUCTS IN  
CATTLE SUPPLEMENTS

by

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AN ABSTRACT OF A MASTER'S THESIS

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A study was conducted to evaluate the stability of two sources, Rovimix A-650\* and Rovimix A-325,\* of stabilized vitamin A when subjected to different pelleting conditions in cattle supplements.

Three 32 percent crude protein cattle supplements were based on either soybean oil meal, with 4% added urea, or with 4% urea and 4% fat. Conditioning temperature during pelleting was approximately 50°C and 70°C. Vitamin A analyses were performed on the initial mash material prior to pelleting and on the cooled pellets at 0, 2, 4, 8, and 16 weeks. Materials were at warehouse storage conditions.

The data obtained permit the following conclusions:

Formulations of the cattle supplements used had no effect upon vitamin A retention over 4 months storage.

Vitamin A levels were significantly affected by the pelleting operation. Conditioning temperature had no effect on vitamin A levels during the 16 week period covered by the study. The data collected indicate the vitamin A sources were stable during storage and processing.

Initial vitamin A values in the feed from use of Rovimix A-325 and Rovimix A-650 were higher than calculated and indicate vitamin A potency of the materials used was higher than the guarantee.

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\* Registered trademark by Hoffman-La Roche Company in Nutley, New Jersey.