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THE STABILITY OF VITAMIN A IN SYNTHETIC VITAMIN A CONCENTRATES (ACETATE OR PALMITATE). I.— IN PHOSPHATIC STOCK LICKS WITH AND WITHOUT TRACE ELEMENTS

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INTRODUCTION

The stability of Vitamin A has been the subject of extensive investigation. It has been shown to be unstable in the natural form as fish liver oil and its loss of potency has been studied by Dunn (1924), Anderegg & Nelson (1926), Powick (1925), Marcus (1931), Holmes, Pigott & Menard (1930), Schroeder, Redding & Huber (1936), Lowen, Anderson & Harrison (1937), Bethke, Record & Wilder (1939), Baird, Ringrose & MacMillan (1939), Miller, Joukovsky & Hokenstad (1942), and Halverson & Hart (1950).

This instability of the natural fish oil was ascribed to the instability of the fat or fatty acids which on oxidation hastened the destruction of the Vitamin A (Powick, 1925; Schroeder *et al.*, 1936; and Bethke *et al.*, 1939).

The loss of potency of fish oils could be slowed down by the presence of antioxidants (Buxton, 1947). Furthermore, Halverson & Hendrick (1955) showed that the presence of certain minerals was instrumental in catalysing the oxidative processes whereby the Vitamin A is destroyed. This destruction was notably less in a mixture of a finely divided texture containing gelatin. Manufacturers of commercial feeds have long been troubled with this problem of vitamin instability in feeds. In order to overcome this problem dry preparations, which are claimed to be stable, have been on the market in recent years. These preparations were stabilized by the addition of antioxidants, for example diphenyl p-phenylene diamine (DPPD), butylated hydroxy anisole (BHA), Vitamin E, etc.; also in some instances by encapsulation in an aerophobic matrix, such as gelatin, gum acacia, or wax. Synthetic palmitate or acetate is the main source of Vitamin A in these preparations. These preparations were shown to be suitable when used for the fortification of most feeds as demonstrated by results on growth studies and liver Vitamin A assays (Siedler & Schweigert, 1954; Halverson & Hendrick, 1955; Reid, Daugherty & Couch, 1955; Harms, Reid & Couch, 1955; Camp, Cartrite, Quissenberry & Couch, 1955; Matterson, Bunnel, Stinson, Singsen & Potter, 1955; Siedler, Enzer & Schweigert, 1956; and Fritz, Wharton, Henley & Schoene, 1956).

Fritz *et al.* (1956) concluded from their growth studies with chicks that losses of Vitamin A from feeds were reduced by—

(*a*) a suitable coating to minimise air contact, and by

(*b*) the use of antioxidants.

Water soluble coating agents (gelatin) were satisfactory when feeds were stored at low humidity. However, they did not give adequate protection when feed was stored under high humidity conditions. Fat soluble coating agents were also effective, provided the agents were biologically active, for example high melting waxes served the purpose well.

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It was therefore established that dry products represent an improvement over Vitamin A oils. It cannot be claimed that the loss of potency was altogether eliminated, since all products lost some Vitamin A during storage at room temperature. Most feeds fortified with stabilized Vitamin A, however, retained potencies up to 50 per cent during six months storage (Fritz *et al.*, 1956). In the present study gelatin-coated synthetic Vitamin A esters (acetate or palmitate) were mixed with phosphatic supplements commonly used as stock licks.

Experimental

Nine brands of commercial products submitted for chemical analysis, were chosen. Certain commercial firms and farmers suggested that these synthetic preparations, if they were stable, could be given to range animals along with their phosphatic licks. In order to test the feasibility of this practice, quantities of the stabilized products were mixed into the licks. The licks were exposed to the atmosphere and their Vitamin A potencies tested periodically up to one week.

The chemical composition of these licks is shown in Table 1.

Table 1.—*Composition of licks*

Sample No.	Description	Calcium (Ca)	Phosphorus (P)	NaCl	Iron (Fe)	Copper (Cu)	Fluorine (F)	Moisture
		%	%	%	%	ppm	ppm	%
526	Bone meal.....	21·3	9·5	0	—	—	—	5·3
530	Bone meal, mineralised (salt + trace elements)	20·3	7·77	24·5	0·29	0·06	290	6·4
708	Bone meal, molassed (+ trace elements)	17·9	10·0	0	0·13	0·047	—	3·5
634	Bone meal, mineralised (+ trace elements)	26·0	11·6	0	0·16	0·024	—	4·6
635	Bone meal, mineralised (salt + trace elements)	16·5	6·8	25·5	0·40	0·032	—	4·2
527	Phosphate, mineralised (salt + trace elements)	15·0	5·52	31·9	0·28	0·02	450	5·0
713	Mineral supplement (trace elements)	20·4	7·8	0	0·42	0·024	—	3·8
714	Mineral supplement (trace elements)	22·4	8·8	0	0·25	0·027	—	3·5
233	Dicalcium phosphate ($CaHPO_4$)	23·3	18·2	0	—	—	110	5·7

Experiment 1 (Table 2)

The samples were left exposed to the atmosphere during a period of hot summer days (temperatures varied from a minimum of 51°F to maximum of 82°F). The weather was rainy and as a result the humidity high (percentage relative humidity 60–70). The tests were, however, carried out indoors in diffused sunlight. Analyses for Vitamin A were carried out on samples at varying intervals up to one week.

TABLE 2.—*Stability of a synthetic Vitamin A-concentrate (palmitate-gelatin coated) mixed in phosphate licks*

Description of lick	Period of exposure				Conditions
	0 Hours	24 Hours	48 Hours	168 Hours	
No. 526 Bone meal salt (NaCl) (2: 1).....	Aver.: 183,100	135,000	78,000	58,750	Exposed to air: diffused sunlight
	(174,000–187,000)	(106,000–154,000)	(76,000–80,000)	(48,000–68,000)	Temp. Min. 51 °F, Max. 82 °F
	Control: Vit. A Synthetic conc. 241,400 i.u.	Control: 214,400	Control: 200,000	Control: 200,000	Humidity (Percentage Rel. Humidity 60–70)
Percentage loss of potency.....	14·5	36·8	61·0	70·6	
No. 233 Dicalcium phosphate.....	162,500	132,916	134,680	64,800	Exposed to air: diffused sunlight
	(140,000–184,000)	(110,000–159,750)	(125,400–140,000)	(41,000–97,600)	Temp. Min. 51 °F, Max. 82 °F
	Control: 200,000	Control: 200,000	Control: 200,000	Control: 200,000	Humidity (Percentage Rel. Humidity 60–70)
Percentage loss of potency.....	18·75	33·5	32·6	67·6	
No. 530 "Mineralised" bone meal salt (1: 1) plus trace elements	195,688	148,000	130,760	105,250	Exposed to air: diffused sunlight
	(184,000–208,000)	(147,000–150,000)	(130,000–132,000)	(96,000–106,000)	Temp. Min. 51 °F, Max. 82 °F
	Control: 200,000	Control: 200,000	Control: 200,000	Control: 200,000	Humidity (Percentage Rel. Humidity 60–70)
Percentage loss of potency.....	8·75	26·0	34·6	47·3	
No. 708 "Molassed" bone meal plus trace elements	161,625	134,043	81,666	29,564	Exposed to air: diffused sunlight
	(150,000–176,000)	(129,600–144,000)	(80,000–85,500)	(17,040–55,000)	Temp. Min. 51 °F, Max. 82 F
	Control: 214,400	Control: 214,400	Control: 214,400	Control: 214,400	Humidity (Percentage Rel. Humidity 60–70)
Percentage loss of potency.....	24·7	35·8	62·0	86·2	
No. 634 Bone meal mineralised with trace elements	164,103	93,680	107,225	63,620	Exposed to air: difiused sunlight
	(155,210–169,600)	(86,400–104,000)	(86,000–124,000)	(62,000–68,000)	Temp. Min. 51 °F, Max. 82 °F
	Control: 214,400	Control: 214,400	Control: 214,400	Control: 200,000	Humidity (Percentage Rel. Humidity 60–70)
Percentage loss of potency.....	23·5	56·5	50·0	68·2	

TABLE 2.—*Stability of a synthetic Vitamin A-concentrate (palmitate-gelatin coated) mixed in phosphate licks* (continued)

Description of lick	Period of exposure				Conditions
	0 Hours	24 Hours	48 Hours	168 Hours	
No. 635 Bone meal mineralised with salt and trace elements	185,750 (184,000–189,000)	110,666 (10,600–120,000)	68,333 (62,000–75,000)	23,650	Exposed to air: diffused sunlight
	Control: 214,400	Control: 214,400	Control: 214,400	Control: 214,400	Temp. Min. 51 °F, Max. 82 °F
Percentage loss of potency.....	13·3	48·5	68·8	89·0	Humidity (Percentage Rel. Humidity 60–70)
No. 527 "Mineralised phosphate" salt plus trace elements	150,066 (148,000–154,000)	165,400 (128,500–152,500)	120,000 (112,000–128,000)	70,750 (66,000–75,000)	Exposed to air: diffused sunlight
	Control: 214,400	Control: 214,400	Control: 214,400	Control: 200,000	Temp. Min. 51 °F, Max. 82 °F
Percentage loss of potency.....	30·0	23·2	44 0	65·0	Humidity (Percentage Rel. Humidity 60–70)
No. 713 "Mineral Supplement" plus trace elements	117,166 (107,500–137,500)	98,400 (96,000–100,000)	74,306 (73,333–76,252)	77,083 (75,000–80,000)	Exposed to air: diffused sunlight
	Control: 214,400	Control: 214,400	Control: 214,400	Control: 214,400	Temp. Min. 51 °F, Max. 82 °F
Percentage loss of potency.....	45·2	56·0	65·5	64·0	Humidity (Percentage Rel. Humidity 60–70)
No. 714 "Mineral Supplement" plus trace elements	153,600 (142,000–164,000)	130,250 (120,000–136,500)	57,200 (48,000–66,000)	16,835 (15,120–21,500)	Exposed to air: diffused sunlight
	Control: 200,000	Control: 200,000	Control: 200,000	Control: 200,000	Temp. Min. 51 °F, Max. 82 °F
Percentage loss of potency.....	23·2	34·8	75·5	91·5	Humidity (Percentage Rel. Humidity 60–70)

Vitamin A assays were done with due regard to the possibility of the extraction of minerals which may accelerate the Vitamin A destruction during the procedure. The tests were done according to the Carr-Price colour procedure and evaluation of Vitamin A in a spectro-photometer (Zeiss-opton) read at 620 millimicrons with a colour development of 15 seconds. For this purpose the galvanometer unit of the colorimeter was set to provide quick, direct readings so as to eliminate the time usually required for manual adjustments of balance circuit resistances, etc., with each test. In order to facilitate quick delivery of the reagent to the sample an automatic pipette was employed.

The reagents were of special purity generally prescribed for Vitamin A assays. The vitamin was extracted by means of hot alcohol, the alcohol extract saponified and finally the Vitamin A extracted by means of ethyl ether. The ethereal phase was then washed free from alcohol, alkali and soaps in the conventional way, and ultimately dried with anhydrous sodium sulphate. Finally the ether containing the Vitamin A was cautiously evaporated to dryness in a water bath under an atmosphere of nitrogen or carbon dioxide. After all the ether had evaporated the residue was taken up in chloroform in appropriate volumes to facilitate reading in the spectro-photometer. The tests were repeated to obtain average values.

Experiment 2 (Table 3)

Similar tests, as described above, were carried out to ascertain the influence of exposure to direct sunlight outdoors in comparison with the influence of diffused sunlight indoors. The assays of the samples of fortified phosphate licks were done at intervals up to one week. An acetate synthetic vitamin ester was put to the test in this experiment.

Experiment 3 (Table 4)

Two licks fortified with a mixed vitamin product and encapsulated with gelatin were similarly exposed to diffused and bright sunlight. The vitamins were a mixture of Vitamin A synthetic concentrate (acetate), riboflavin (Vitamin B_2), and Vitamin D_3 compounded in appropriate ratios and quantities for the fortification of chick rations. The assay of the samples was done at intervals for a period of one week.

Experiment 4 (Table 5)

Two licks enriched with Vitamin A palmitate encapsulated in gelatin were tested under conditions of diffused and direct sunlight, as well as to test the effect of leaving the fortified lick in the feeding trough of sheep for one week, when the animals had free access to the lick.

TABLE 3.—*Influence of sunlight on the stability of a synthetic Vitamin A concentrate in phosphatic licks.* (a) *Diffused sunlight* (b) *Direct sunlight. Control: Vit. A. conc. (acetate)*

Type of lick	Period of exposure	24 Hours		48 Hours		168 Hours		Control
1. Bone meal	Potency Vit. A (i.u.)	(*a*) 45,500	(*b*) 35,500	(*a*) 30,000	(*b*) 24,200	(*a*) 20,850	(*b*) 13,300	50,000 (i.u.)
	Percentage loss	(*a*) 9·0	(*b*) 29·0	(*a*) 40·0	(*b*) 51·6	(*a*) 58·3	(*b*) 73·4	
2. Bone meal salt (NaCl) (2: 1) No. 526	Potency (i.u.)	(*a*) 39,000	(*b*) 24,200	(*a*) 24,200	(*b*) 13,000	(*a*) 20,000	(*b*) 17,500	48,000
	Percentage loss	(*a*) 18·7	(*b*) 49·5	(*a*) 49·5	(*b*) 75·2	(*a*) 58·5	(*b*) 63·5	
3. Mineralised phosphate No. 527 salt plus trace element	Potency (i.u.)	(*a*) 30,700	(*b*) 26,400	(*a*) 16,400	(*b*) 13,000	(*a*) 10,300	(*b*) 8,400	48,000
	Percentage loss	(*a*) 36·0	(*b*) 45·0	(*a*) 66·0	(*b*) 73·0	(*a*) 78·8	(*b*) 82·5	
4. "Mineral supplement" plus trace elements No. 714	Potency (i.u.)	(*a*) 35,500	(*b*) 23,500	(*a*) 31,500	(*b*) 15,200	(*a*) 21,250	(*b*) 15,500	48,000
	Percentage loss	(*a*) 26·0	(*b*) 51·0	(*a*) 34·5	(*b*) 68·5	(*a*) 55·5	(*b*) 68·0	
5. Mineral supplement plus trace elements No. 713	Potency (i.u.)	(*a*) 33,200	(*b*) 27,500	(*a*) 30,000	(*b*) 20,500	(*a*) 25,200	(*b*) 10,250	48,000
	Percentage loss	(*a*) 30·8	(*b*) 42·5	(*a*) 37·6	(*b*) 57·5	(*a*) 47·5	(*b*) 79·0	
6. "Mineralised" bone meal plus trace elements No. 634	Potency (i.u.)	(*a*) 27,000	(*b*) 26,000	(*a*) 30,000	(*b*) 22,000	(*a*) 18,500	(*b*) 12,500	48,000
	Percentage loss	(*a*) 43·8	(*b*) 46·0	(*a*) 37·7	(*b*) 54·0	(*a*) 61·5	(*b*) 74·0	
7. Mineralised bone meal plus trace elements No. 635	Potency (i.u.)	(*a*) 32,000	(*b*) 32,000	(*a*) 25,000	(*b*) 26,000	(*a*) 21,750	(*b*) 6,500	48,000
	Percentage loss	(*a*) 33·5	(*b*) 33·5	(*a*) 48·0	(*b*) 46·0	(*a*) 54·5	(*b*) 65·8	
8. Molassed bone meal plus trace elements No. 708	Potency (i.u.)	(*a*) 34,000	(*b*) 27,000	(*a*) 26,000	(*b*) 24,000	(*a*) 20,500	(*b*) 12,000	48,000
	Percentage loss	(*a*) 29·2	(*b*) 44·0	(*a*) 46·0	(*b*) 50·0	(*a*) 57·4	(*b*) 75·0	
9. Dicalcium phosphate No. 233	Potency (i.u.)	(*a*) 32,500	(*b*) 17,000	(*a*) 27,500	(*b*) 12,500	(*a*) 17,500	(*b*) 7,500	48,000
	Percentage loss	(*a*) 32·4	(*b*) 64·5	(*a*) 43·0	(*b*) 74·0	(*a*) 63·0	(*b*) 84·5	
Average (nine licks)	Percentage loss	28·8 (9·0–43·8)	45·0 (29·0–64·5)	44·7 (34·5–66·0)	61·1 (46·0–75·2)	59·4 (47·5–78·8)	74·0 (63·5–84·5)	48,000

TABLE 4.—*Influence of sunlight on the stability of a synthetic Vitamin (A B_2 D_3) concentrate in phosphatic licks.* (a) *Diffused sunlight;* (b) *Direct sunlight control: Vit. A:* 40,000 *i.u.*

Type of lick	Period of exposure	0 Hours		24 Hours		48 Hours		168 Hours	
1. Bone meal salt (2: 1).....	Potency Vit. A (i.u.).....	(*a*) 39,280	(*b*) 39,280	(*a*) 28,400	(*b*) 25,839	(*a*) 24,853	(*b*) 15,700	(*a*) 12,000	(*b*) 7,800
	Percentage loss.....	1·8	1·8	29·0	35·6	38·0	60·6	70·0	83·0
2. Dicalcium phosphate No. 233	Potency.....	(*a*) 37,840	(*b*) 37,840	(*a*) 30,500	(*b*) 25,090	(*a*) 29,000	(*b*) 19,200	(*a*) 24,400	(*b*) 15,800
	Percentage loss.....	4·7	4·7	23·7	37·4	27·5	52·0	39·0	60·5

TABLE 5.—*Influence of sunlight on the stability of a synthetic Vitamin A concentrate (Vitamin A—Palmitate) in phosphatic licks Control:* 40,000 *i.u. Vit. A.*

Conditions: Simultaneous exposure: (a) *Indoors—diffused sunlight;* (b) *Outdoors—direct sunlight;* (c) *Outdoors—direct sunlight, licks offered to sheep*

Type of lick	Period of exposure	0 Hours			24 Hours			48 Hours		
1. Bone meal salt (2: 1) No. 526	Potency Vit. A (i.u.)....	(*a*) 36,500	(*b*) 35,000	(*c*) 36,000	(*a*) 33,200	(*b*) 28,750	(*c*) 27,200	(*a*) 32,600	(*b*) 26,800	(*c*) 18,480,
	Percentage loss of potency	8·75	12·5	10·0	17·0	28·1	32·0	18·5	33·0	53·8
2. Mineralised bone meal salt plus trace elements No. 530	Potency Vit. A (i.u.)....	(*a*) 35,000	(*b*) 34,000	(*c*) 34,000	(*a*) 32,200	(*b*) 28,000	(*c*) 22,800	(*a*) 31,240	(*b*) 26,800	(*c*) 18,000
	Percentage loss of potency	12·5	15·0	15·0	19·5	30·0	43·0	21·9	33·0	55·0

Type of lick	Period of exposure	96 Hours			168 Hours		
1. Bone meal salt (2: 1) No. 526	Potency Vit. A (i.u.).....	(*a*) 28,400	(*b*) 21,120	(*c*) 17,200	(*a*) 24,000	(*b*) 20,640	(*c*) 16,200
	Percentage loss of potency	29·0	47·2	57·0	40·0*	48·4*	59·5
2. Mineralised bone meal salt plus trace elements No. 530	Potency Vit. A (i.u.)... ...	(*a*) 27,800	(*b*) 21,000	(*c*) 16,800	(*a*) 21,000	(*b*) 16,500	(*c*) 14,600
	Percentage loss of potency	30·5	45·0	58·0	47·5*	58·7*	63·5

* Overcast sky

Discussion of Results

In Tables 2, 3, 4 and 5 are given the Vitamin A potencies of enriched phosphatic licks exposed to hot, humid atmospheric conditions. Vitamin A deteriorated in all the experiments. The loss of potencies in the licks (Table 2) were initially from 8·75 to 45·2 per cent, increasing to 26 to 56 per cent after 24 hours' exposure, and 34·6 to 68·8 per cent after 48 hours' exposure. After one week the loss was from 47·3 to 91·5 per cent.

Various factors could have been responsible for this substantial loss of Vitamin A potencies in all the licks: the temperature and humidity high, the presence of phosphates and trace elements, and the possibility of the gelatin coat of synthetic Vitamin A esters (whether acetate or palmitate) not being impervious to atmospheric moisture, air and sunlight.

Davies & Warden (1954), and Halverson & Hendrick (1955), amongst others have shown that certain minerals including those in minute quantities as trace elements in feeds have a destructive action on Vitamin A, but that this loss was partially overcome by the use of encapsulating products. Wall & Kelly (1951) in studies on stability of Vitamin A esters, have stressed the importance of temperature concentration of the vitamin ester and the carrier.

The influence of sunlight becomes apparent in Table 3. The loss of Vitamin A potency of the licks showed a similar trend as shown in Table 2 for diffused sunlight. In the case of direct sunlight the loss of potency was appreciably higher. The average loss was 28·8 per cent in diffused light and 45 per cent in bright sunlight within a period of 24 hours. For the following 24 hour periods up to 168 hours the losses were progressively higher, and attained a high level of 59·4 per cent and 74·0 per cent loss respectively.

The influence of exposure to diffused and bright sunlight on the fortified phosphate licks when the vitamin mixture contained in addition to Vitamin A also riboflavin and Vitamin D_3, is shown in Table 4. It is noteworthy that the initial loss on mixing is small. This favourable trend can be ascribed to the presence of riboflavin, a light sensitive vitamin. As an antioxidant it had protective qualities against Vitamin A loss. These qualities were only temporary as can be seen from further periods of exposure. After one week the losses were 39 per cent and 70 per cent for diffused sunlight and in bright sunlight 60·5 per cent and 80 per cent respectively for the two phosphate licks under consideration.

Further evidence is presented (Table 5) to illustrate the influence of sunlight on the stability of Vitamin A. It becomes evident that under overcast conditions the losses were smaller. The losses at the end of a week under these conditions were 40 per cent indoors compared with 48 per cent outdoors.

It is wellknown that the ultra violet rays affect the stability of Vitamin A and it is acceptable that the ultra violet rays of the sunlight accelerated the loss of potency recorded on exposure.

Evidence is presented below to show to what extent ultra violet rays affected the synthetic concentrate alone and when incorporated in a phosphatic lick. For this purpose a Sterisol Lamp (Model PL 390-generator NN 30/89 V) with a power consumption of 35 watts and 220 volts, was used.

TABLE 5.—*Stability of a synthetic Vitamin A palmitate (gelatin coated) when exposed to ultra violet rays (Sterisol Lamp)*

	Period of Exposure	Potency Vit. A	Original Potency	Loss of Potency
Vit. A. Source	Hours	(i.u.)	(i.u.)	%
1. Synthetic concentrate (gelatin coated)	0·5	16,320	20,000	18·4
	1·0	16,000	20,000	20·0
	2·0	14,400	20,000	28·0
	3·0	14,200	20,000	29·0
	4·0	13,760	20,000	31·2
2. Phosphate lick plus synthetic concentrate	1·0	14,000	20,000	30·0
	2·0	13,000	20,000	35·0

Within four hours of exposure to ultra violet rays the synthetic concentrate, encapsulated in gelatin, lost 31·2 per cent of its original strength.

In the mixed phosphate-synthetic concentrate sample the loss was 30 per cent within one hour and 35 per cent after two hours.

From the evidence presented here it can be concluded that the incorporation of Vitamin A in the so-called stabilised form in phosphatic licks for range animals is unpractical. In South Africa with its abundant sunshine and especially where very hot and humid conditions may prevail at times, the possibility of extensive Vitamin A losses is of real importance. The alternative, practical method is the incorporation of stabilized Vitamin A in a finely divided state in concentrate rations for stall-fed animals. For range animals, however, the feeding of similarly enriched supplement feeds in the form of cubes may be an answer to the problem.

SUMMARY

Stabilised synthetic Vitamin A, either as acetate or palmitate encapsulated in gelatin was incorporated in mineral-rich phosphatic licks and exposed to hot, humid atmospheric conditions which at times prevail in the summer rainfall area of South Africa. The loss of Vitamin A potency under these experimental conditions led to the conclusion that the feeding of stabilised Vitamin A incorporated in licks fed to range animals is unpractical.

REFERENCES

ANDEREGG, L. T. & NELSON, V. E. (1926). Milk powders as food. II. Observations on the existence of vitamin E. *Ind. Eng. Chem.* 18, 620–622.

BAIRD, F. D., RINGROSE, A. L. & MACMILLAN, M. J. (1939). The stability of Vitamins A and D in mixed feed ingredients. Part 2. Vitamin A. *Poultry Sci.* 18, 441–448.

BIERER, B. W. & VICKERS, C. L. (1958). Pelletising effect on Vitamin A and E. *J. Amer. Vet. Med. Ass.* 133, 228.

BETHKE, R. M., RECORD, P. R. & WILDER, O. H. M. (1939). The stability of Carotene and Vitamin A in a mixed ration. *Poultry Sci.* 18, 179–187.

BUXTON, L. O. (1947). Tocopherols as antioxidants for Vitamin A in fish liver oils. *Ind. Eng. Chem.* 39, 225–232.

CAMP, A. A., CARTRITE, H. T., QUISSENBERRY, J. H. & COUCH, J. R. (1955). Supplementary vitamin A and Choline in the diet of the growing chick. *Poultry Sci.* 34, 1134–1138.

CASTANO, F. F., BOUCHER, R. V. & CALLENBACH, E. W. (1951). Utilization by the chick of Vitamin A from different sources. 1. Crystalline Carotene, crystalline Vitamin A acetate and " Black Cod " Liver Oil. *J. Nutrition* 45, 131–141.

DAVIES, A. W. & WARDEN, A. N. (1954). The stability of Vitamin A in animal feeding stuffs. *J. Sci. Food Agric.* 5, 107.

DUNN, L. C. (1924). The effect of cod liver oil in various amounts and forms on the growth of young chickens. *J. Biol. Chem.* 6, 129–136.

FRITZ, J. C., WHARTON JR., F. D., HENLEY, R. M. & SCHOENE, R. B. (1956). Preservation of Vitamin A in feeds. *Fed. Proc.* 15, 551–552.

HALVERSON, A. W. & HART, E. B. (1950). Factors affecting the stability of the Vitamin A from cod liver oil in cereal feeds. *J. Nutr.* 40, 415–427.

HALVERSON, A. W. & HENDRICK, C. M. (1955). Effect of trace elements and other dietary ingredients upon Vitamin A stability in stored poultry feeds. *Poultry Sci.* 34, 355–360.

HARMS, R. H., REID, B. L. & COUCH, J. R. (1955). Storage of Vitamin A in chick livers as a criterion of stability, availability, and dietary level. *Poultry Sci.* 34, 1125–1133.

HOLMES, A. D., PIGOTT, M. G. & MENARD, D. F. (1930). The permanency of cod liver oil vitamins in stored feeds. *Poultry Sci.* 10, 37–52.

LOWEN, L., ANDERSON, L. & HARRISON, R. W. (1937). Cereal flours as antioxidants for fishery products. Halibut liver and salmon oils. *Ind. Eng. Chem.* 29, 151–156.

MATTERSON, L. D., BUNNEL, R. H., STINSON, L. D., SINGSEN, E. P. & POTTER, L. M. (1955). The effect of incorporating diphenyl-p-phenylene diamine into a ration containing various levels of Vitamin A from fish oils. *Poultry Sci.* 34, 1080–1084.

MARCUS, J. K. (1931). The destructive action of finely divided solids on Vitamin A. *J. Biol. Chem.* 90, 507–513.

MILLER, M. W., JOUKOVSKY, V. & HOKENSTAD, N. (1942). The effect of manganese sulphate on the stability of Vitamin A and D of cod liver oil when stored in mixed feeds. *Poultry Sci.* 21, 200–202.

OLSEN, E. M., HARVEY, J. D., HILL, D. C. & BRANION, H. D. (1959). Utilization and stability of commercial Vitamin A supplements. *Poultry Sci.* 38, 929–942.

POWICK, W. C. (1925). Inactivation of Vitamin A by rancid fat. *J. Agric. Res.* 31, 1017–1026.

REID, B. L., DAUGHERTY, H. K. & COUCH, J. R. (1955). The stability of Vitamin A in mixed feeds and premixes. *Poultry Sci.* 34, 603–608.

SIEDLER, A. J., ENZER, E. & SCHWEIGERT, B. S. (1956). Vitamin A and carotene stability in feeds containing antioxidant treated animal fats. *J. Agric. Food. Chem.* 4, 1023–1029.

SIEDLER, A. J. & SCHWEIGERT, B. S. (1954). Vitamin stabilization. Effect of added stabilized animal fats on stability of Vitamin A in feeds. *J. Agric. Chem.* 2, 193–195.

SCHROEDER, C. H., REDDING, G. K. & HUBER, L. J. (1936). Some causes and effects of a high free fatty acid content of the meat scraps in poultry rations. *Poultry Sci.* 15, 104–114.

STEPHENSON, E. L. (1953). Stability of Vitamin A in poultry feeds. *Arkansas Farm Res.* 2 (3).

WALL, M. E. & KELLY, E. G. (1951). Stability of Carotene and Vitamin A in dry mixtures. *Ind. Eng. Chem.* 43, 1146–1150.

VITAMIN A ACTIVITY IN FEED PRODUCTS. Vitamin A acetate in gelatin (Pfizers) Chas. Pfizer & Co. Inc., 630 Flushing Ave., Brooklyn 6, N. York, U.S.A.