Storage of milk powders under adverse conditions

2. Influence on the content of water-soluble vitamins

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1. Storage of milk powder under unfavourable conditions accelerates the normally slow deterioration in nutritional quality. The effects of such storage on the water-soluble vitamin composition were examined.

2. (*a*) Spray-dried whole milk containing 25 g water/kg was stored at 60° and 70° and sampled weekly to 9 weeks. (*b*) Spray-dried whole milk and skimmed milk were adjusted to contain 40 and 100 g water/kg and stored at 37° in nitrogen and in oxygen. Samples were taken for analysis at intervals during storage.

3. The samples were analysed for eight B-complex vitamins and ascorbic acid, and also for total lysine, 'reactive lysine' and 'lysine as lactulosyl-lysine'.

4. Storage at 60° caused rapid destruction of folic acid (53% loss at 4 weeks) and slower loss of thiamin, vitamin B_6 and pantothenic acid (18% at 8 weeks). There was no change in the content of riboflavin, biotin, nicotinic acid and vitamin B_{12}. At 70° the rate of destruction of the four labile vitamins was much increased; 18% or less survived at 4 weeks.

5. At 37° and 40 g water/kg there was little change in total and 'reactive' lysine during storage for 57 d. Lactulosyl-lysine was demonstrably present but at low concentration. There was considerable loss of folate (72%) and ascorbate (91%) during storage for 30 d in O_2, but no significant loss in N_2. Thiamin fell by approximately 12% in 57 d, equally in O_2 and N_2. The content of the remaining vitamins was unchanged. At 100 g water/kg there were progressive Maillard changes. During 27 d in N_2 the colour changed from cream to pale brown, but in O_2 there was no perceptible colour change. Total lysine fell by 20% in 27 d, and 'reactive lysine' by 30%. Folate was stable during 16 d in N_2, but largely (94%) destroyed in O_2. Ascorbic acid was also destroyed in N_2 as in O_2. Thiamin fell by 41% in 27 d, equally in O_2 and N_2. Vitamin B_6 was more labile, especially in N_2, falling by 71% in 16 d.

6. With skimmed-milk powder containing 100 g water/kg, storage at 37° in O_2 and N_2 gave much the same results as for the corresponding whole-milk powder. The presence of milk fat had no marked effect on the stability of the water-soluble vitamins.

7. Destruction of vitamins was clearly linked to the progress of Maillard-type reactions and was strongly influenced by time and temperature of storage, moisture content and, in some instances, by the presence of O_2.

Prolonged storage of whole-milk powder at temperatures below 38° causes little or no change in the content of several of the B-complex vitamins. Thus, Mattick *et al.* (1945) found no loss of riboflavin or thiamin in whole-milk powder packed in air or inert gas, during storage for 12 months at room temperature. Ascorbic acid decreased by < 50% in air-packed samples but to a lesser extent in samples packed in inert gas. Sharp *et al.* (1945) extended these findings, reporting that storage of whole-milk powder for 6 months at 37·8°, in air or inert gas, caused no loss of riboflavin, nicotinic acid, pantothenic acid, biotin or vitamin B_6. The content of ascorbic acid declined, by approximately 32 mg/kg with air-packing and 8 mg/kg with inert gas.

If, however, the conditions of storage are such as to promote Maillard-type reactions, as by an increase in temperature of storage or in moisture content of the milk powder, then considerable damage to the nutritional quality may ensue. The protein nutritional quality is impaired, mainly as a consequence of degradation of the lysine residues, and there are concomitant losses of several vitamins.

The present paper reports the effects of storage of low-moisture milk powder at high temperatures, as described by Hurrell *et al.* (1983), on the B-complex vitamins and vitamin C, and presents for comparison values for 'reactive lysine' and 'lysine as lactulosyl-lysine' (Finot *et al.* 1981). Experiments are also described in which the moisture content of the milk powder was increased to induce Maillard damage. Dried whole milk and dried skimmed milk were adjusted to contain 100 g water/kg and stored at 37° in N_2 and in O_2.

EXPERIMENTAL

Test materials

Spray-dried whole-milk powder containing 25 g water/kg was stored at 60° and 70° and sampled at weekly intervals to 9 weeks, as described in an earlier paper in this series (Hurrell *et al.* 1983).

A sample was obtained of freshly-manufactured spray-dried whole-milk powder containing 40 g water/kg. A portion (50 g) was put into store at −30° in a hermetic container. A further 400 g was distributed in layers approximately 18 mm deep in 100-mm Petri dishes, which were then covered with lids and stacked in two anaerobic jars. The jars were evacuated and filled, one with O_2 and the other with O_2-free N_2. This procedure was repeated to effect a more complete displacement of air and the jars, containing O_2 or N_2 at atmospheric pressure, were incubated at 37°. Petri dishes were removed at intervals, one from each jar, and their contents transferred to screw-stoppered bottles and deep-frozen. On each such occasion the jars were refilled with O_2 or N_2 as described above.

To 500 g of this milk powder was added enough distilled water to increase the moisture content to approximately 100 g/kg. The wetted powder was mixed in a blender jar and rubbed through a 60 mesh stainless-steel wire sieve. A portion (50 g) was bottled and frozen, and the remainder stored at 37° in O_2 or N_2 and sampled at intervals, as described above.

Spray-dried skimmed-milk powder (500 g) was similarly adjusted to approximately 100 g/kg moisture content, stored at 37° in O_2 or N_2, and sampled at intervals to 105 d.

Analytical methods

Lysine. Total lysine, reactive lysine and lactulosyl-lysine from furosine were defined and measured as described by Hurrell *et al.* (1983).

Water-soluble vitamins. Samples of the dried-milk preparations were reconstituted in boiled distilled water by gently mixing in a Potter-Elvehjem type homogenizer (for details, see Ford, 1964).

Total ascorbic acid was measured by the microfluorimetric method of Deutsch & Weeks (1968) adapted to suit the concentrations present in cows' milk.

Riboflavin, nicotinic acid, pantothenic acid and biotin were assayed by standard microbiological procedures (Ford *et al.* 1953; Chapman *et al.* 1957). Folate activity was assayed with *Lactobacillus casei* by an adaptation of the procedure recommended by Herbert (1961) for the assay of folate in blood serum (Ford, 1967). Vitamin B_{12} was assayed with *Lactobacillus leichmannii* as described by Gregory (1954), and thiamin with *Lactobacillus fermenti*; the test medium was that of Banhidi (1958) and the milk samples (1 ml reconstituted milk) were extracted by heating for 30 min at 100° with 20 ml 0·033 M-sulphuric acid. Vitamin B_6 was assayed with *Kloeckera brevis* as described by Barton-Wright (1963), except that the test samples were extracted with 0·055 M-hydrochloric acid as recommended by Gregory (1959).

The B-vitamin assays were done once only. They were interpolated in a larger series of assays on milk, in which reference milk samples were included and assayed repeatedly as a check on 'within assay' and 'between assay' variability. The standard errors given with Tables 1–3 pertain to this wider survey, but are quoted here as being representative.

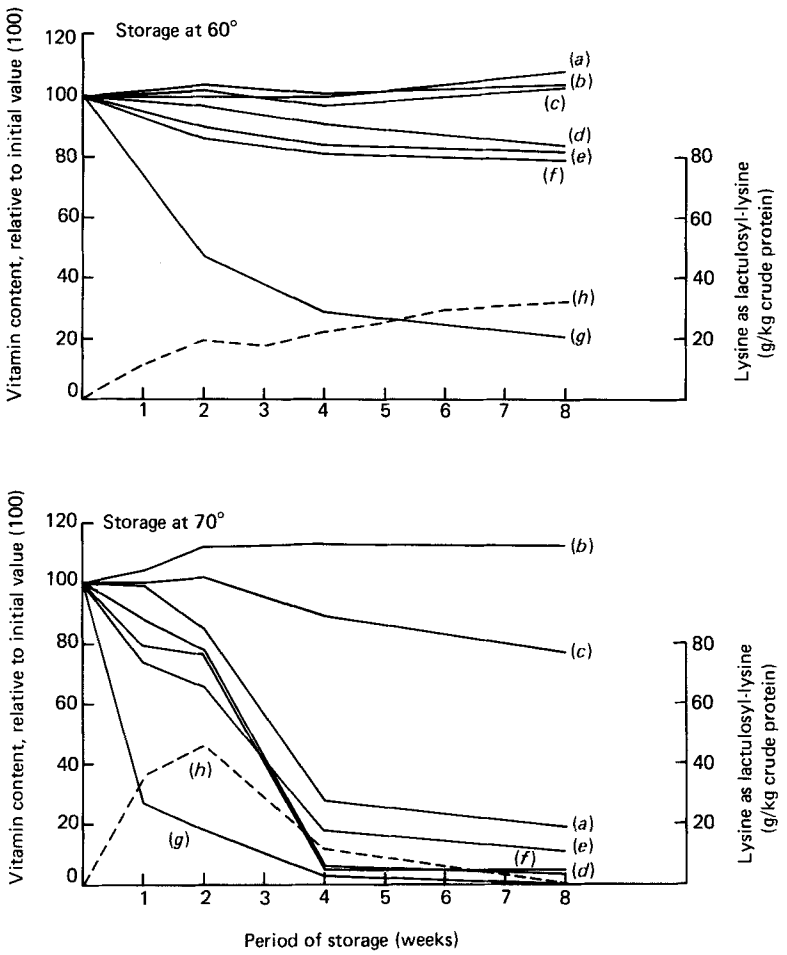

Fig. 1. Content of B-complex vitamins and lactulosyl-lysine in whole-milk powder containing 25 g moisture/kg, as measured at intervals during storage at 60° and 70°. (*a*) Vitamin B_{12}, (*b*) nicotinic acid, (*c*) biotin, (*d*) pantothenic acid, (*e*) vitamin B_6, (*f*) thiamin, (*g*) folate, (*h*) lysine as lactulosyl-lysine.

RESULTS

Storage of whole-milk powder at 60° and 70°

Table 1 and Fig. 1 show the effect of storage of spray-dried whole-milk at 60° and 70° on the content of total and reactive lysine and seven vitamins of the B-complex. During storage for 8 weeks at 60° there was no change in the content of nicotinic acid, biotin and vitamin B_{12}. The same was probably true also for riboflavin, but the results were unaccountably variable and so are held back. The content of thiamin, pantothenic acid and vitamin B_6 declined progressively, by approximately 18% at 8 weeks. Folate was exceptionally labile; more than 50% was lost by 2 weeks, and 80% by 8 weeks.

At 70°, as at 60°, nicotinic acid proved entirely stable. The level of biotin remained constant for the first 2 weeks of storage and thereafter declined, by 25% at 8 weeks. The content of vitamin B_{12}, vitamin B_6, thiamin and pantothenic acid fell sharply after 2 weeks storage, and the rate of loss appeared to accelerate in parallel with the destruction of the

Table 1. *Effect of storage of whole-milk powder containing 25 g moisture/kg, at 60° and 70°, on the content of total and available lysine (mg/g crude protein (nitrogen × 6·25)) and vitamins of the B-complex*

Storage temperature and time (weeks)	Total lysine	Reactive lysine	Lysine as lactulosyl-lysine	Thiamin (as hydrochloride) (μg/g)	Nicotinic acid (μg/g)	Pantothenic acid (as calcium D-pantothenate) (μg/g)	Vitamin B_6 (μg/g)	Biotin (ng/g)	Folate (ng/g)	Vitamin B_{12} (ng/g)
Control (stored at −30°)	85·4	85·4	0	4·2	7·7	32	3·8	130	340	13
60° 2	80·4	72·6	19·5	3·6	8·0	31	3·4	132	160	13
4	73·1	64·2	22·3	3·4	7·8	29	3·2	126	100	13
8	67·8	55·3	31·3	3·3	8·0	27	3·1	134	70	14
70° 1	64·4	49·8	36·5	3·3	8·0	28	2·8	130	90	13
2	56·3	37·9	46·1	3·2	8·6	25	2·5	132	60	11
4	18·9	14·2	11·8	0·2	8·7	2·1	0·7	116	10	4
8	14·6	14·6	0	0·2	8·6	1·3	0·4	100	0	3
SE of analytical value*	1·1	†	†	0·08	0·25	1·1	0·17	7·8	9·3	0·86

* For details, see p. 365. † Single values.

Table 2. *Content of total and available lysine (mg/g crude protein (nitrogen × 6·25)) and of selected water-soluble vitamins in whole-milk powder containing 40 and 100 g moisture/kg and stored at 37° in oxygen and in N_2*

Storage time (d)	Total lysine	Reactive lysine	Lysine as lactulosyl-lysine (mg/g crude protein)	Thiamin (μg/g)	Vitamin B_6 (μg/g)	Folate (ng/g)	Ascorbic acid (μg/g)
			40 g moisture/kg				
Control*	85·8	85·8	0	4·1	4·0	300	86
30							
N_2	81·7	80·1	4·2	4·0	4·1	316	78
O_2	74·2	82·5	4·3	3·8	3·7	83	8
57							
N_2	75·7	74·6	5·3	3·6	3·7	303	nd
O_2	79·2	76·7	5·8	3·6	4·0	57	nd
			100 g moisture/kg				
Control*	86·4	86·4	0	3·1	4·3	308	79
16							
N_2	nd	nd	nd	2·3	1·2	308	nd
O_2	nd	nd	nd	2·1	1·9	19	nd
27							
N_2	66·8	61·7	12·9	1·8	0·8	250	0
O_2	70·3	64·2	14·9	1·8	0·9	19	0
SE of analytical value	†	†	†	0·08	0·17	9·3	†

nd, not determined. *Stored at −30°. † Single values.

previously formed lactulosyl-lysine and the onset of advanced Maillard browning. By 4 weeks there remained approximately 6% of the thiamin and pantothenic acid, 18% of the vitamin B_6 and 28% of the vitamin B_{12}. As at 60°, folate was the most labile; 73% was lost at 1 week, and 97% at 4 weeks.

Storage of whole-milk powder at 37°

Spray-dried whole-milk powders containing 40 and 100 g moisture/kg were stored at 37°, in O_2 and in N_2. At 40 g/kg moisture content the milk powder did not change in colour during 57 d of storage. At 100 g/kg there was a distinct colour change during storage in N_2 for 27 d, from creamy-yellow to pale brown. In O_2, however, there was no perceptible colour change.

Table 2 shows the effects on the content of total and available lysine, ascorbic acid, thiamin, folate and vitamin B_6.

At 40 g/kg moisture content there was only a small fall in total lysine and reactive lysine during storage for 57 d. Lactulosyl-lysine was demonstrably 'present' after storage but at low concentration.

The content of ascorbic acid fell only marginally during 30 d storage in N_2, but in O_2 the vitamin was largely destroyed. Similarly with folate, there was considerable loss during storage in O_2 but no loss in N_2. With thiamin there was an apparent loss of approximately 12% at 57 d, equally in O_2- and N_2-stored milk. The content of the remaining B-complex vitamins, nicotinic acid, pantothenic acid, vitamin B_6, riboflavin, biotin and vitamin B_{12}, was unchanged after storage.

At 100 g/kg moisture content there was evidence of deterioration in protein nutritional quality after 27 d. Total lysine had fallen by approximately 20%, reactive lysine by almost

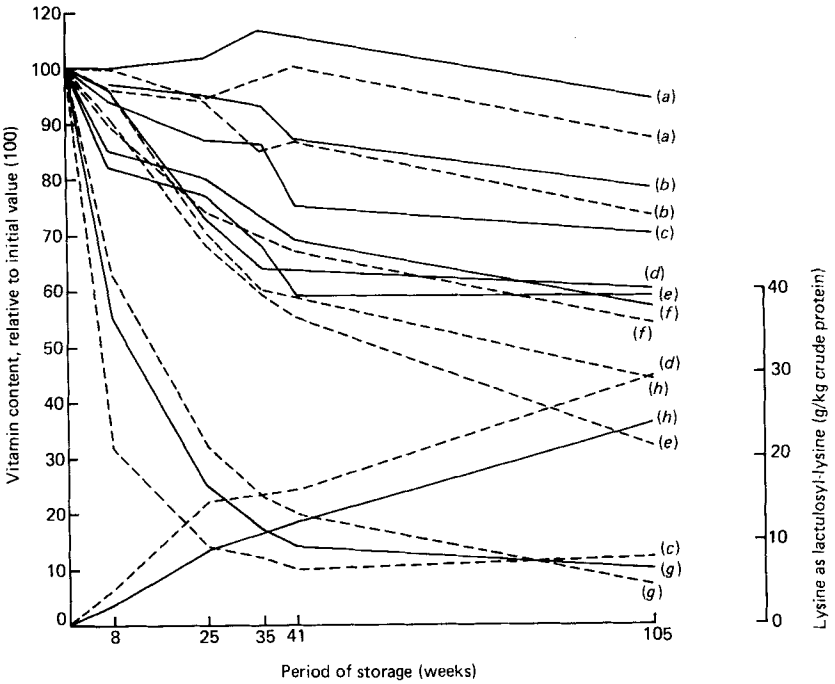

Fig. 2. Content of B-complex vitamins and lactulosyl-lysine in skimmed-milk powder containing 100 g moisture/kg, as measured at intervals during storage at 37° in oxygen (---) or nitrogen (——). (*a*) Riboflavin, (*b*) pantothenic acid, (*c*) folate, (*d*) thiamin, (*e*) vitamin B_{12}, (*f*) reactive lysine, (*g*) vitamin B_6, (*h*) lysine as lactulosyl-lysine.

30% and approximately 14 mg lysine/g crude protein was estimated to be present as lactulosyl-lysine.

Thiamin content fell progressively, by 41% in 27 d storage, and equally with O_2 and N_2. Vitamin B_6 proved even more labile, especially on storage in N_2 which resulted in 71% loss by 16 d. The pantothenic acid content did not fall significantly. Folate showed no change during 16 d storage in N_2 and only 19% loss after 27 d. However in O_2 the rate of destruction was much faster; 94% was lost by 16 d. Ascorbic acid was completely destroyed after 27 d, even with N_2 storage.

Storage of skimmed-milk powder at 37°

Spray-dried skimmed-milk containing 100 g moisture/kg was stored at 37°, in O_2 and in N_2. As with the corresponding whole-milk powder (see p. 359), browning during storage was more advanced in the N_2-stored samples. After 105 d in N_2 the product was deep orange-brown in colour, whereas the O_2-stored milk was yellow-brown.

Table 3 and Fig. 2 show the effects on the contents of total and available lysine, ascorbic acid, and B-complex vitamins.

Total lysine declined progressively, by 31% at 105 d. There was a corresponding and somewhat steeper fall in reactive lysine, and a complementary increase in lactulosyl lysine, which was greater in the O_2-stored milk.

The content of thiamin and vitamin B_{12} declined during storage at approximately the same rate, and marginally faster in O_2. After 105 d the loss with N_2 was approximately 40% and with O_2 approximately 62%. Folate also declined, comparatively slowly in N_2 (30% loss

Table 3. *Effect of storage of skimmed-milk powder containing 100 g moisture/kg at 37° in oxygen and in nitrogen on the content of total and available lysine (mg/g crude protein (nitrogen × 6·25)), ascorbic acid and vitamins of the B-complex*

Storage time at 37° (d)	Total lysine	Lysine as lactulosyl-lysine	Reactive lysine	Ascorbic acid (μg/g)	Thiamin (as hydro-chloride) (μg/g)	Nicotinic acid (μg/g)	Pantothenic acid (as calcium D-panto-thenate) (μg/g)	Vitamin B_6 (μg/g)	Biotin (ng/g)	Folate (ng/g)	Riboflavin (μg/g)	Vitamin B_{12} (ng/g)
Control (stored at 30°)	79·1	0	79·1	59	4·3	7·9	31	4·5	200	326	16	22
8												
N_2	68·3	2·4	67·3	28	4·1	8·1	30	2·5	210	306	16	18
O_2	72·8	4·5	70·8	0	4·1	8·4	30	2·8	220	105	16	20
25												
N_2	66·9	9·0	63·3	nd	3·2	8·1	30	1·1	210	282	16	17
O_2	64·1	14·7	58·3	nd	3·0	8·7	29	1·4	220	46	15	15
35												
O_2	nd	nd	nd	nd	2·8	8·3	29	0·8	200	280	17	15
N_2	nd	nd	nd	nd	2·6	8·4	27	1·0	190	38	16	13
41												
N_2	59·2	12·3	54·3	nd	nd	8·3	27	0·6	210	245	nd	13
O_2	59·1	15·7	52·8	nd	nd	8·4	27	0·9	200	32	16	12
105												
N_2	54·5	24·5	45·1	nd	2·6	8·5	24	0·4	210	229	15	13
O_2	54·9	29·6	43·0	nd	1·9	8·7	23	0·3	210	39	14	7
SE of analytical value*		†	†		0·08	0·25	1·1	0·17	7·8	9·3	0·26	0·86

nd, not determined. *For details, see p. 356. † Single values.

by 105 d) and rapidly in O_2 (86% loss by 25 d). After the rapid initial loss the folate activity in the O_2-stored milk stabilized at approximately 12% of the level in the control. There was no loss of nicotinic acid or biotin, and an apparent small loss of riboflavin after prolonged storage in O_2 was of border-line significance. Pantothenic acid was somewhat less stable and fell by approximately 25% after 105 d. Vitamin B_6 was highly labile, again apparently more so in N_2 than in O_2 (see Table 2), and after 35 d the loss was approximately 80%.

DISCUSSION

It is evident from these results that several water-soluble vitamins may be destroyed during the storage of dried-milk powder under unfavourable conditions. Vitamin B_6, thiamin, folate, vitamin B_{12}, pantothenic acid and ascorbic acid were all labile to different extents. In contrast, nicotinic acid, biotin and riboflavin were comparatively stable.

The destruction of these vitamins appeared to be linked to the progress of Maillard-type reactions, and was strongly influenced by the time and temperature of storage, moisture content and, in some instances, by the presence of O_2. In the preceding paper (Hurrell *et al.* 1983) we reported that on storage of whole-milk powders at 60° there was a progressive increase in lactulosyl-lysine. At 70°, lactulosyl-lysine was formed more rapidly and then degraded. We now find that the destruction of vitamins in these powders coincided with the formation of advanced Maillard reaction products from lactulosyl-lysine degradation (Table 1). It would appear, therefore, that vitamin B_6, thiamin, pantothenic acid and vitamin B_{12} were destroyed by reaction with the advanced Maillard reaction products. However, there were some smaller losses of thiamin, vitamin B_6 and pantothenic acid during storage at 60°, at which temperature the advanced Maillard reactions had not taken place. Pryidoxamine and thiamin both have free amino groups and might have reacted with lactose directly, forming biologically-inactive products. But the loss of pantothenic acid cannot be accounted for in this way and it may have resulted from the influence of heat alone.

Several workers have reported the instability of vitamin B_6 in a variety of foodstuffs. Gregory & Kirk (1978) investigated the stability of B_6 vitamins in model food-systems stored at 37° and 0·6 water activity and identified the binding of pyridoxal to protein, forming ϵ-pyridoxyl lysine, as the prime mechanism of degradation. In the model food-systems studied by Gregory & Kirk (1978), pyridoxamine was rapidly converted to pyridoxal during storage. In milk powder, with its high content of lactose, interaction of pyridoxamine with lactose or advanced Maillard products might be a significant alternative pathway to destruction.

There was no loss of vitamin B_6 from milk powder containing 40 g moisture/kg, during storage at 37° for 57 d (Table 2). Mercurio & Tadjalli (1979) examined tinned milk powder that had been stored for 20 years at room temperature, and found that the vitamin B_6 content was much the same as that in the freshly-canned product from the same manufacturer, although available lysine, riboflavin and thiamine were lower by 30, 50 and 61% respectively. It seems, therefore, that loss of vitamin B_6 during storage of dried milk of low moisture content at normal temperatures is a negligible problem.

The presence of O_2 was the predominant factor in the destruction of vitamin C and folate. O_2 also had some smaller influence on the loss of vitamin B_{12}, vitamin B_6 and thiamin and on the development of browning. Vitamin C was the most sensitive of the vitamins to destruction in the presence of O_2 and, although its mechanism of degradation in foods depends on many factors (Bauernfeind & Pinkert, 1971), it is possible that in these milk powders it was due to oxidation followed by a Maillard-type reaction. Ascorbic acid is an enediol and is readily oxidized to dehydroascorbic acid, the diketo form, which reacts readily with free amino groups (Hodge, 1953). However, at 100 g/kg moisture content, the ascorbate was completely destroyed after storage under N_2 for 27 d at 37°, whereas at 40 g

moisture/kg there was comparatively little loss (Table 2). This suggests that ascorbic acid, like thiamin, vitamin B_6, pantothenic acid and vitamin B_{12} may be destroyed by advanced Maillard-reaction products.

Like thiamin and pyridoxamine, the N-methyl tetrahydrofolate present in milk has a free amino group in the molecule, and destruction of the vitamin in the absence of O_2 might similarly be caused by its interaction with lactose or with advanced Maillard reaction products. However, the milk folate is far more sensitive to destruction in the presence of O_2. Thus, with milk powder containing 40 g/kg moisture there was no loss of folate during storage in N_2 for 57 d at 37°, whereas in O_2 there was a 70% loss after only 30 d. The general picture resembles that reported for liquid milk sterilized by the ultra-high temperature process and aseptically filled into foil-lined cartons. The stability of folate to processing and during subsequent storage was closely associated with the presence of ascorbic acid, and loss of ascorbic acid was determined mainly by the residual concentration of O_2 in the milk after processing (Ford, 1967). Supplementation of the milk with ascorbic acid (Ford *et al.* 1974) or flushing the milk with O_2-free N_2 (Ford, 1967) effectively removed the residual O_2 and stabilized the milk folate.

Our finding that milk powders brown more rapidly during storage under N_2 than under O_2 was unexpected. There are several reports in the literature that aqueous Maillard systems brown more rapidly in O_2 than in N_2 (Lewis *et al.* 1949; Hashiba, 1975, 1976). However, there are also reports to the contrary. Thus, Bohart & Carson (1955) found that solutions containing glucose and glycine (0·45 M; pH 6·8 in phosphate buffer) gave two to three times more colour when heated at 50° under N_2 than under O_2. And Gregory & Kirk (1978) noted that the rate of browning and loss of vitamin B_6 in model food-systems was lower when they used storage cans with a large head-space volume. They attributed this effect to the larger supply of gaseous O_2. A possible explanation is that O_2 transforms the intermediate compounds formed on the degradation of the Amadori compound into less reactive substances that do not polymerize to brown pigments. Thus, if under our 'dry' conditions the main pathway for Amadori product degradation was fragmentation of the sugar moiety to give α-keto aldehydes or α-hydroxyaldehydes such as deoxyglucosone, pyruvaldehyde, glyceraldehyde and glycolaldehyde, then in O_2 they might be converted to the corresponding acids, which are less reactive. In N_2, the α-keto- and α-hydroxyaldehydes could react with free amino groups to give brown products. Another explanation might be that browning is a free-radical reaction (Namiki & Hayashi, 1981) which would be inhibited by O_2.

The moisture content of the milk powders had a marked influence on the stability of several of the water-soluble vitamins during storage. From Table 2 it is evident that losses of vitamin B_6 and thiamin were much greater at 100 g/kg than at 40 g/kg moisture content; and if we compare values for the N_2-stored milk powders the same is true for ascorbate. If these vitamins are assumed to take part in Maillard-type reactions, then such an effect of water content would be expected. It is notable that the vitamins were much more sensitive than lysine to destruction at the higher moisture level.

The moisture content of the milk had no influence on the stability of pantothenic acid. On prolonged storage (105 d at 37°) of skimmed-milk powder containing 100 g moisture/kg there was a progressive slow loss of pantothenic acid, amounting to approximately 25% at 105 d (Table 3), and there was some indication that the rate of loss was greater with N_2 storage. Nicotinic acid and biotin were entirely stable under these storage conditions, and the small decline in riboflavin content was of border-line significance. The presence of milk fat had no marked effect on the stability of the water-soluble vitamins, as may be seen by comparing values for whole-milk powder (100 g moisture/kg; Table 2) stored 27 d at 37°, with those for skimmed-milk powder stored 25 d under the same conditions (Table 3). There was perhaps a slightly greater loss of thiamin and vitamin B_6 from the whole-milk powder.

Our present findings confirm and extend earlier reports (for a review, see Hartman & Dryden, 1974) that several of the B-complex vitamins in low-moisture milk powders are stable over long periods of storage at temperatures up to approximately 40°, so long as access of light and moisture is prevented. In practice moisture is not a serious problem: most powders contain 20–40 g moisture/kg and are hermetically packed, and would be expected to reach the consumer in temperate countries with no significant loss of product quality or nutritional value. In some of the developing countries, however, a combination of higher temperatures and inadequate storage and transport systems might result in the products being subjected to temperatures in excess of 40° (cf. Hurrell *et al.* 1983). Our results indicate that, provided the product retains its natural colour, there should be only small losses of water-soluble vitamins excepting folate and ascorbate, even after storage for several weeks at temperatures up to 60°. In infant formulas the losses would normally be offset by addition of vitamin supplements by the manufacturer. We know of no published information on the fate of folic acid in low-moisture powders stored under N_2, but it is clear that large losses of folate and ascorbate may occur even at normal temperatures if O_2 is not excluded from the package. In view of the doubts expressed by Barford & Pheasant (1981) concerning the general assumption that added folic acid is an adequate substitute for the naturally-occurring derivatives of tetrahydrofolic acid, it would seem wise to ensure that as much as possible of the natural vitamin is retained during processing and storage.

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