AGRICULTURAL MATERIALS

Relative Stability of Selenites and Selenates in Feed Premixes as a Function of Water Activity

SYLVAN EISENBERG
Micro Tracers Inc., 1370 Van Dyke Ave, San Francisco, CA 94124

Sodium selenite is more hygroscopic than sodium selenate. It is, therefore, more likely to dissolve when dispersed in feeds of relatively high water activity. When dissolved, it may form selenious acid and disperse as a vapor. This is easily demonstrated by mounting a filter paper wetted with a reagent such as ascorbic acid over the subject feed, but not in contact with it. The paper turns brown as elemental selenium is formed from reduction of the vapor. Analysis of the paper ensures that the brown is indeed selenium. Though premixes are generally low enough in moisture content to ensure stability of the selenites, this is not true of many feeds. The water activities of a number of feeds, feed premixes, and feed ingredients have been determined instrumentally and compared to those of saturated solutions of sodium selenite and sodium selenate. There is no question that the selenite often dissolves with the potential to react and, in so doing, loses its nutritional function.

The January 9, 1974, U.S. Food and Drug Administration (FDA), Title 21, Code of Federal Regulations, Part 573.920 responded to the demonstrated deficiency of selenium (Se) in animal feeds. It permitted the inclusion of sodium selenite or sodium selenate in premixes to yield 200 ppm Se. Each batch of premix was to be assayed for Se. No less than 1 lb of premix could be added per ton of complete feed to yield 0.1 ppm Se for chickens other than laying hens, beef, and pigs, and 0.2 ppm for turkeys. On April 6, 1987, the regulation was modified, increasing the level in premixes to 600 ppm in all feeds to 0.3 ppm and eliminating the assay of each batch of premix.

The modification was initiated by the continued Se deficiencies found to persist even after the original regulation went into effect. Although sodium selenate was permitted as the Se source, it was seldom used since it was more expensive than sodium selenite. No consideration was given to the relative stability of these Se salts. Sodium selenite is easily reduced to elemental Se by Vitamin C, ferrous salts, iodides,

and, very likely, other reducing agents. The elemental Se is not considered as serving nutritional requirements. For this reduction of selenites to elemental Se to occur, the selenite must be in solution. This underscores the significance of the water activity (a_w), of premixes and feeds.

The a_w of a substance is defined as the vapor pressure of water in equilibrium with the substance divided by that of pure water at the same temperature. Pure distilled water, therefore, has an a_w of exactly 1.

Higher a_w substances tend to support more microorganisms. Bacteria usually require at least 0.91, and fungi at least 0.7. Water migrates from high a_w substances to low a_w substances. For example, when honey ($a_w = 0.6$) is exposed to humid air ($a_w = 0.7$), the honey will absorb water from the air. Chemical reactions generally occur in solution. One factor in the relative stabilities of sodium selenite and sodium selenate may, therefore, be the water activities of their respective saturated solutions. The lower the a_w the more likely the absorption of water from the environment. Sodium selenite in solution reacts with many reducing agents, such as ascorbic acid, ferrous sulfate, and potassium iodide, being reduced to insoluble elemental Se (reddish-brown). This paper explores the respective stabilities of added selenites and selenates as a function of the matrix a_w.

Experimental

Samples

Six feed ingredient samples (Table 1), 10 premix samples (Table 2), and 11 feed samples from various commercial sources (Table 3) were used in this study.

Chemicals

(**a**) *For determining a_w, of saturated solutions of sodium selenite and sodium selenate.*—Two samples of commercial sodium selenite, one of Filipino and another of German origin, and one sample of reagent grade, and 2 samples of sodium selenate, one of Filipino origin and another of reagent grade.

(**b**) *For testing reactivity of sodium selenite with ascorbic acid, ferrous sulfate, and potassium iodide.*—Reagent ferrous sulfate heptahydrate, U.S. Pharmacopeia (USP) ascorbic acid, and USP potassium iodide (Mallinckrodt Laboratory Chemicals, Phillipsburg, NJ).

(**c**) *Microtracer™ sand-Se 4.5% (4.5% Se).*—80 + 150 mesh sand with an adsorbed layer of sodium selenite (MicroTracers, Inc., San Francisco, CA).

Table 1. Water activities and percent moisture in various feed ingredients

Feed ingredient	Temp., °C	Moisture, %	a_w
Soy meal	27.4	11.8	0.633
Soy protein	27.5	11.5	0.616
Wheat meal	27.5	12.8	0.643
Corn	25.3	14.8	0.728
Safflower meal	23.4	7.5	0.499
Limestone	23.4	—[a]	0.468

[a] — = Not determined.

Table 2. Water activities and percent moisture in various feed premixes

Premix	Temp., °C	Moisture, %	a_w
Vitamin-mineral	27.5	6.34 ± 0.003	0.481
Premix feed works	23.4	6.30	0.431
Choline chloride-60	23.4	3.24	0.197
Choline chloride-50	23.4	3.32	0.102
Amoxcilin, 20%	23.3	5.21	0.440
Zinpro-microplex	23.2	3.69	0.225
Biocox 120G	23.2	1.32	0.420
Coccimase-25	23.2	0.25	0.406
Monteban pr.	23.1	6.15	0.448
Rumensin pr.	23.1	8.09	0.449

Table 3. Water activities and percent moisture in various feeds

Feed	Temp., °C	Moisture, %	a_w
Layers feed-1	21.7	10.4 ± 0.05	0.600 ± 0.005
Co-chicken	22.2	10.2 ± 0.06	0.595 ± 0.004
Sow feed-2	23.7	10.5 ± 0.01	0.553 ± 0.032
Sow feed-3	23.7	7.5 ± 0.08	0.425 ± 0.021
Sow feed-4	26.6	13.9 ± 0.05	0.647 ± 0.033
Chicken	24.8	13.5 ± 0.06	0.663 ± 0.002
Turkey 20%	25.1	11.5 ± 0.21	0.676 ± 0.002
Turkey 20%	22.0	11.4	0.664
Turkey 16%	25.1	11.1 ± 0.20	0.657 ± 0.002
Turkey 16%	22.1	11.1	0.646
Dog food	23.4	9.5	0.533

Table 4. Water activities of saturated solutions of selenium compounds, averages of 3 independent determinations

Sample	Temp., °C	Time, min	a_w
Sodium selenite, reagent	23.5	3	0.708 ± 0.002
Sodium selenite, Germany	24.6	5	0.690 ± 0.004
Sodium selenite, Filipino	24.6	6	0.681 ± 0.007
Avg.			0.693 ± 0.014
Sodium selenate, reagent	23.5	4	0.755 ± 0.003
Sodium selenate, Filipino	23.5	4	0.730 ± 0.010
Avg.			0.743 ± 0.018

Instrument

The Rotronic Instrument Corp. (Huntington, NY) Model AwVC AwQuick a_w meter was used throughout, allowing 3–6 min, as required, to attain equilibrium.

Apparatus

(**a**) *Petri dishes*.—9.0 cm diameter × 1.5 cm high, with covers (Part No. 25384-302, VWR Scientific, West Chester, PA, or equivalent).

(**b**) *Plastic boxes*.—4 cm diameter × 1 cm high (WP40, Rotronic Instrument Corp., Huntington, NY).

(**c**) *Plastic rings*.—Cut 1 cm lengths from 6 oz plastic cups obtained at Walgreens Drug Stores, or use 3 dominos or dice as spacers.

(**d**) *Aluminum moisture dishes*.—5.0 cm diameter × 2.3 cm high, with covers (Part No. 72410-006, VWR Scientific, or equivalent).
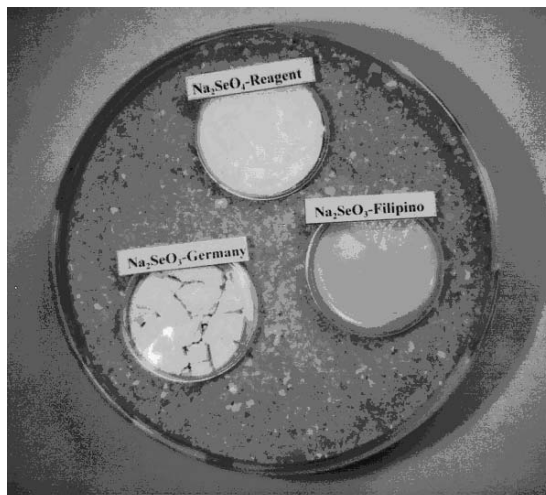

Figure 1. Sodium selenite and sodium selenate exposed to turkey feed.

Table 5.   Percent increase in weight of Se salts[a]

Days	Sodium selenite, Filipino	Sodium selenite, Germany	Sodium selenate, Filipino
	Increase in weight, %		
1	6.89	7.03	1.66
2	11.91	11.12	1.71
5	30.11	21.88	2.74
6	35.00[b]	26.90	2.18
8	44.28	38.46	1.04
9	49.06	42.60[b]	0.84
13	65.23	57.36	0.85
16	71.29	62.27	0.92
20	75.64	74.80	0.75
23	78.05	79.96	0.85

[a] Water transferred from 700 g turkey feed (11.5% moisture and 0.676 a_w) to samples of sodium selenite and sodium selenate in aluminum moisture dishes floating upon the feed contained in a covered plastic container at 25°C.

[b] Drops of free liquid appeared.

(e) *Whatman No. 1 filter paper.*—4.25, 5.5, and 11.0 cm diameter (Part Nos. 1001-042, 1001-055, and 1001-110, respectively, at VWR Scientific, or equivalent).

(f) *Ziplock Twist'n Loc container and lid.*—32 fl. oz (Walgreens Drug Stores; or Rubbermaid 1.3 L Round Storage Container from Rubbermaid Commerical Products, Inc.).

Method

Volatilization of sodium selenite, probably as selenious acid, is demonstrated by placing a 4.25 cm filter paper on the bottom of a Petri dish and, wetting the paper with 1% HCl containing 1% sodium selenite. Another, slightly larger sheet is supported 1 cm above it and this wetted with 1% ascorbic acid. The dish is covered and placed it in an incubator overnight at 45°C. The upper paper would turn light brown due to the formation of elemental selenium. This can be confirmed by analysis of the paper.

Similarly, half-fill a quart container with ground corn to which 600 ppm Se as sodium selenite has been added and mixed. Place a plastic ring on top of the corn to support a filter paper wetted with 1% ascorbic acid. Close the chamber and incubate overnight at 45°C. Volatilization of the selenite would be demonstrated if the paper turned brown, and if its analysis for selenium were positive.

The extent of feed carry-over from one mix to another in feed manufacturing can be estimated by adding a Microtracer to the first mix, and then establishing its presence and relative quantity in the following mix. The Microtracer is removed from samples by a magnetic separator and the concentration of tracer particles is determined by counting the number of particles in retrieved samples. This is done by scattering

Table 6.   Water activities of saturated solutions of the specified compounds, average of 3 independent determinations

Sample	Temp., °C	Time, min	a_w
Manganese sulfate	23.7	6	0.936 ± 0.012
Ferrous sulfate	23.7	6	0.958 ± 0.017
Ascorbic acid	23.7	6	0.938 ± 0.015

known weights on paper wetted with one of the reducing agents, ascorbic acid, ferrous sulfate or potassium iodide. Each particle should leave a dark brown spot of selenium on the paper. The spots can be counted at leisure.

Results and Discussion

The water activities of 6 feed ingredients are reported in Table 1 (0.468 ≤ a_w ≥ 0.728); of 10 premixes in Table 2 (0.102 ≤ a_w ≥ 0.481); and of 10 feeds in Table 3 (0.425 ≤ a_w ≥ 0.676). These may be compared to the water activities of saturated solutions of sodium selenite and sodium selenate as reported in Table 4 (0.69 and 0.74, respectively). Both Se salts should be stable in the premixes. The selenite might well be unstable in certain of the feed ingredients, particularly the corn, and in certain feeds, particularly turkey and chicken feeds. The potential of sodium selenite to pick up moisture from such environments and become reactive is illustrated in Figure 1 and Table 5. Evidence for consequent instability follows.

Volatilization of sodium selenite, probably as selenious acid, was easily demonstrated. Incubating paper wetted with 1% ascorbic acid held above another paper wetted with sodium selenite and 1% HCl overnight at 45°C produced a brown discoloration of the upper paper. Analysis of the
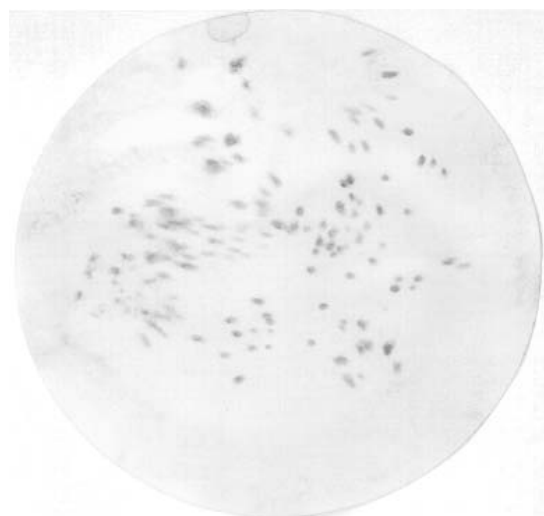

Figure 2.   MT Sand-Se 4.5% scattered on paper wetted with 1% ferrous sulfate heptahydrate.

Table 7.   Number of particles per milligram of MT Sand-Se 4.5%[a]

Reagent	MT Sand-Se, mg	Count, No.	No./mg	Color		
				Front	Back	Background
Ascorbic acid[b]	1.2	152	126	Brown	Brown	White
	2.4	232	101	Brown	Brown	White
	1.5	167	111	Brown	Brown	White
Ferrous sulfate[c]	1.4	157	112	Brown	White	Sl. Yellow
	2.4	333	139	Brown	White	Sl. Yellow
Potassium iodide[d]	1.6	177	111	Brown	Brown	White
	1.0	123	123	Brown	Brown	White
	2.2	139	116	Brown	Brown	White
Avg. count (n = 8)			117 ± 12			

[a] Determined by scattering small quantities on filter papers wetted with various reagents, then counting the brown Se spots formed by reaction of the selenite. Visibility of spots on the back of the paper and the effects on the background color of the white paper are noted in the last column.
[b] 1%.
[c] Heptahydrate, 1%.
[d] 1% in 0.02 M HCl.

floating paper found 0.203 µg Se whereas none was detected on a like blank paper. Detecting level was 0.1 µg (1, 2).

Similarly, a quart container with cover was half-filled with a ground corn to which 600 ppm selenium as sodium selenite had been added and mixed. A plastic ring was placed on top of the corn to support a filter paper disc wetted with 1% ascorbic acid. The chamber was closed and, then incubated overnight at 45°C. The paper turned brown. It contained 3.51 µg selenium, again demonstrating the mobility of sodium selenite as a vapor and its potential to be reduced to the element.

The above magnitudes of selenium movement are merely suggestive of potential movement and loss as a nutrient. In practice, the times are much longer and the conditions quite different, steam used in pelleting for example. The losses probably are significantly greater.

The water activities of saturated solutions of various selenium compounds are reported in Table 4, and manganese sulfate, ferrous sulfate, and ascorbic acid in Table 5.

The number of particles per mg of MT Sand-Se 4.5% can be determined by scattering known weights on paper wetted with one of the reducing agents, ascorbic acid, ferrous sulfate or potassium iodide. Brown discoloration of the entire paper occurs quickly followed by relatively slow formation of dark brown selenium spots in the way of each tracer particle. This is illustrated in Figure 2. The numbers of particles per milligram are reported in Table 6. The average of 8 determinations was 117 ± 12. This standard deviation (SD) conforms to what one would expect since particle counts may be expected to follow the Poisson Distribution for which the SD equals the square root of the mean, or ±11 in this case.

The vapor phase should be carefully checked for the presence of a volatile oxidizing agent. Note that sodium selenite in presence of a dilute acid yields selenious acid.

When heated, this sublimes to yield a yellowish-green vapor with a pungent sour smell (3, 4). Holding an acidified iodide-wetted paper over, but not in contact with, an ascorbate-treated or ferrous-treated paper immediately after dispersing MT Sand-Se on the paper, tests for its presence. A test for a volatile oxidizing agent could be very important, suggesting a source of potential Se loss.

The absorptions of water by selenites and selenates from turkey feed as a function of time are reported in Table 7, and from chicken feed in Table 8. We note that the small difference in water activities of these feeds (0.676 and 0.663); Table 3 has a significant effect upon the rate of water absorption by the selenite; the selenate loses moisture in each case. We note

Table 8.   Percent increase in weight of Se salts[a]

Days	Increase in weight, %		
	Sodium selenite, Filipino	Sodium selenite, Germany	Sodium selenate, reagent
1	3.02	3.86	0.88
4	11.81	14.40	1.58
6	14.37	17.17	1.50
10	26.34	27.63	0.21
11	27.00	27.97	0.15
13	27.56	27.80	0.06
17	30.71	27.94	0.00

[a] Water transferred from 700 g chicken feed (13.5% moisture and 0.663 a_w) to samples of sodium selenite and sodium selenate in aluminum moisture dishes floating upon the feed contained in a covered plastic container at 25°C.

further that the selenite should be stable in any of the premixes with the water activities reported in Table 2: $0.102 \leq a_w \geq 0.481$.

Conclusions

The relative instability of sodium selenite compared to sodium selenate in aqueous environments suggests that the selenite should not be used. Had the selenate been used initially, the need to elevate the permitted amount of Se in premixes from 200 to 600 ppm might have been avoided.

References

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