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Further Evidence of Destruction of Vitamin B in Evaporated Milk.

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Hartwell's¹ method of determining the Vitamin B content of foods differs from that of other workers, in that it consists in measuring the effects of a given ration on the suckling young. Either just before or just following parturition the mother animal is put on a high protein ration containing all the known necessary constituents, with the exception of Vitamin B. In the absence of an adequate amount of Vitamin B the suckling young gain normally for about 13 days, when they suddenly develop convulsions, manifested by general muscular incoordination. This condition is frequently accompanied by unusual outcries, and dragging of the hind legs. The baby animal gasps for breath, and finally dies within a few hours. That these animals do not die of insufficient food is indicated by the fact that milk is very frequently found in the stomachs. When adequate amounts of Vitamin B are added to the ration the young animals develop normally and are weaned at the usual time.

By this method,² comparative studies of fresh and evaporated milks have indicated that evaporated milk is quite deficient in Vitamin B. Since these findings are not in accord with results reported by Gibson³ on pigeons, dogs, and pigs, nor with the recent work of Dutcher,⁴ arrived at by the more usual method of feeding the growing young directly, it seemed possible that the untoward symptoms in Hartwell's animals might be caused by a calcium deficiency, or a lack of Vitamin D. Previous experiments showed that in the process of evaporation the calcium and phosphorus in milk is made less available.⁵ We have, therefore, repeated Hartwell's experiments, making certain additions and modifications in the ration. In our experiments the pregnant animals, which had received our stock ration throughout the gestation period, were given the experimental ration just previous to parturition. When the young were 4 days old all but 4 of the litter were killed. From this time the suckling young were weighed every 4 days.

The ration modifications and the results of the experiments are given in Table I.

TABLE I.
Comparison of Whole and Evaporated Milk Rations as a Source of Vitamin B for the Suckling Young.

Ration	Group	Wt. of litter	Age at onset	Wt. at onset	Ave. wt. of	Total No.
		4 days	of spasms	of spasms	young at 1 mo.	of young weaned
		gm.	days	gm.	gm.	
Control ¹	A	24	13	62+		0
	B	26	12	72		0
100 cc. of whole milk substituted for 50 cc. evaporated milk	A	20			52	4
Control + 0.2 gm. Ca ₃ (PO ₄) ₂	A	37	14	100	41	1
	B	28	15	93		0
Control + 1 gm. Ca ₃ (PO ₄) ₂ + 5 cc. cod liver oil	A	39	13	94		0
Irradiated evaporated milk used in ration	A	29	13	64		0
	B	21	14	60		0
Control + 0.2 gm. Ca ₃ PO ₄) ₂ + 20 cc. wheat embryo extract ²	A	33			42	3 ⁴
	B	33			50	4
Control + 30 cc. wheat embryo extract ³	A	39			45	5 ⁵

¹ This ration consisted of 30 gm. of dried wheat bread, 12 gm. of casein, 50 cc. of evaporated milk, .05 cc. ferric citrate (6% solution) and 0.1 cc. of potassium iodide (2% solution).

² Alcoholic extract of 10 gm. of wheat embryo.

³ Alcoholic extract of 15 gm. of wheat embryo.

⁴ One died at 21 days. Cause unknown.

⁵ This animal suckled 6 young. One was lost at 3 weeks.

Neither the addition of calcium phosphate nor of cod liver oil to the evaporated milk ration, nor the substitution of irradiated evaporated milk modified the untoward symptoms in the young; whereas the addition of wheat embryo extract to the evaporated milk ration, or the substitution of quickly boiled milk for evaporated milk resulted in normal young. It would, therefore, appear that some substance in milk is destroyed by the evaporation processes. This substance is contained in the wheat embryo extract.

These results seem to be verified by tests with pigeons.

The daily administration of 50 cc. of raw milk, when given in conjunction with 20 grams of polished rice was found sufficient to protect pigeons against polyneuritis; whereas 25 cc. of evaporated milk was insufficient. When the amount of evaporated milk was increased to 30 cc. the untoward symptoms were only slightly manifest.

These results are less striking than those with the suckling rats. It is possible that the very young are more sensitive to an antineuritic deficiency. The experiences with beri-beri in the nursing infant are comparable.

Sherman and Axtmayer⁶ and Chick and Rosco⁷ have recently

TABLE II.
Polynuritic Symptoms in Pigeons Receiving Evaporated Milk and Fresh Milk.

Initial Wt.	Forced feeding	Milk* Kind	Amt. Milk	Untoward Symptoms	Gain in Wt.	
					gm.	days
235	33	Fresh	50		65	33 ¹
265	20	Evap. + 1 gm. Ca ₃ (PO ₄) ₂	25	Unable to stand. Regurgitates food. Head hangs on side. Crop hard.	32	30 ²
262	22	Evap. + 1 gm. Ca ₃ (PO ₄) ₂	25	Food not retained. Unable to swallow. Breathing heavily; head down.	5	22 ³
270	30	Evap.	30	Tired; unable to stand erect; crop soft	50	30 ⁴
302	30	Fresh	60		68	30 ⁵
	30	Fresh	60			

* During forced feeding period each bird was given daily in addition to milk 20 gm. polished rice. Gravel available in pens.

¹ Pigeon vigorous, active. No signs of polyneuritis.

² Given 15 cc. wheat embryo extract subcutaneously. Following day bird very much alive; active; crop soft.

³ Bird died. Enlargement of heart questionable; no pericardial fluid.

⁴ Less vigorous than Pigeon 5, but no outstanding signs of polyneuritis.

⁵ Pigeon vigorous; active. No signs of polyneuritis.

* Pigeon stands erect; walks, flies; no signs of polyneuritis.

shown that Vitamin B contains both an antineuritic substance, Vitamin F, and a growth promoting factor, Vitamin G, the former being less thermostable than the latter. Milk is reported as being richer in Vitamin G than it is in F. Our results would seem to indicate that the antineuritic factor is destroyed in part in the evaporated milk.

¹ Hartwell, G. A., *Biol. Chem. J.*, 1922, xvi, 825.

² Hartwell, G. A., *Biol. Chem. J.*, 1925, xix, 227.

³ Gibson, R. B., and Concepcion, I., *Philippine J. Sc. B.*, 1916, xi, 119.

⁴ Dutcher, R. A., Frances, E., and Combs, W. E., *J. Dairy Sc.*, 1926, ix, 379.

⁵ Daniels, A. L., and Laughlin, R., *J. Biol. Chem.*, 1920, xlv, 380.

* Sherman, H. C., and Axtmayer, J. H., *J. Biol. Chem.*, 1927, lxxv, 207.

⁷ Chich, H., and Roseo, M. H., *Biol. Chem. J.*, 1927, xxi, 698.