Vitamin D Potency of Human Breast Milk^{*}

ROBERT S. HARRIS, Ph.D., and JOHN W. M. BUNKER, Ph.D., F.A.P.H.A.

Assistant Professor of Nutritional Biochemistry, and Professor of Physiology and Biochemistry, Massachusetts Institute of Technology, Cambridge, Mass.

F OR the prevention and cure of rickets in infants, human breast milk is admittedly superior to cow's milk. One would naturally assume this superiority to be based on a larger content of vitamin D in breast milk. Although quantitative data have not

hitherto been available, there are three investigations which indicate how little vitamin D breast milk contains, for no evidence of healing in rachitic rats was observed after feeding the quantities of human milk shown in Table I. Likewise, no evidence of healing was

TABLE I

	Breast Milk Fed Daily	Total Fed	Authors
<i>cc</i> .	Days	cc.	
25	9	225	Hess, Weinstock and Sherman ¹
40 (condensed)) 7	280	Outhouse, Macy and Brekke ²
20	8	160	Bunker, Harris and Eustis ³

TABLE II

Human Butter Fat Fed Daily *	Total Fat Fed	Equivalent Amount * of Breast Milk	Authors
0.2 cc.	1.6 cc.	50 gm.	Hess and Weinstock ⁴
0.3 gm.	2.4 gm.	73 gm.	Lesne and Vagliani ⁵
0.48 gm.	3.84 gm.	116 gm.	Palmer and Kennedy ⁶
0.72 gm.	5.76 gm.	173 gm.	Sabri and Fikri ⁷
0.36 gm. (Irrad.)	2.88 gm.	86 gm.	Sabri and Fikri ⁸

* Assuming a consumption of 6.0 gm. of diet/rat/day, a feeding period of eight days, and a butter fat content of 3.3 per cent.

recorded after feeding the amounts of butter fat shown in Table II.

In competent bioassay laboratories it has been found that substantially 3 USP XI units of Reference Oil are usually required to induce a unit (2+on the Bill's scale) of healing in rats

^{*}Read by title before the Food and Nutrition Section of the American Public Health Association at the Sixty-seventh Annual Meeting in Kansas City, Mo., October 25, 1938.

Mo., October 25, 1938. Contribution No. 149 from the Biological Research Laboratories, Massachusetts Institute of Technology, Cambridge, Mass. This investigation was aided by a grant from the Permanent Science Fund, American Academy of Arts and Sciences.

TABLE III

Antirachitic Effect of Cream and Butter Fat Prepared from Human Breast Milk

Rat	Supplement		Equivalent Volume Line Test of Breast Milk (Bill's Scale)	
1147	gms.		cc.	
20030		eam	60	0.0
20030	10 (1	"	60	0.0
		"		
20032	10		60	0.0
20033	10	"	60	0.5
20034	20	"	120	0.0
20035	20	"	120	2.5
20036	20	"	120	3.0
20037	20	"	120	1.5
20038	25	"	150	1.5
20039	35	"	210	2.0
20040	20	"	120	0.0
20041	25	"	150	0.0
20700	1.0 bi	itter fat	30	0.0
20701	2.0	"	60	0.0
20702	5.0	""	150	0.0
20703	10.0	"	300	3.0
20704	15.0	"	450	1.5

made experimentally rachitic on the Steenbock 2965 ration. It appears, therefore, that since none of the above feedings of breast milk, or its fat, induced recognizable healing, there is evidence that mother's milk does not contain as much as 10 USP XI units of vitamin D per quart.

The investigation being reported was undertaken to determine the vitamin D potency of winter breast milk.

EXPERIMENTAL PROCEDURE

Sixty quarts of breast milk from wet nurses were obtained through the coöperation of the Boston Dispensary. Cream was obtained by passing this milk through a home-sized De Laval Cream Separator. Ten gm. of this cream, containing 20 per cent butter fat, produced no healing (Table III) when fed in the rachitogenic diet of each of 4 rachitic rats (20030–33) over a period of 6 days, while 20–35 gm. portions produced varying degrees of healing when fed 8 other rats (20034– 41).

The remainder of the cream was extracted with four successive portions of anhydrous ethyl ether and the solvent was removed in vacuo under a stream of carbon dioxide. Graded portions of this ether-soluble fat were fed in the diets of each of 5 rachitic rats (20700-04) over a period of 6 days. The animals readily ingested quantities up to 10 gm., this latter amount producing definite rachitic healing (Table III). The animal fed a 15 gm. quantity showed rachitic healing but failed to gain in weight during the 12 days required for consumption of the supplement.

It is likely that the large feeding of fat prevented an accurate assay. Therefore, the remainder of the butter fat was saponified with 10 per cent aqueous potassium hydroxide and the ether-soluble fraction of the nonsaponifiable material was fed in graded amounts to another series of 8 animals during a period of 6 days. One gm. of this material was equivalent to approximately 2,900 cc. of milk.

Antirachitic Effect of Non-Saponifiable Fraction Prepared from Breast Milk Butter Fat					
Rat	Supplement	Equivalent Volume of Breast Milk	Line Test (Bill's Scale)		
	gm.non-sap.				
21126	. 210	600	1.5		
21127	.225	642	2.0		
21128	. 240	687	1.5		
21129	.255	730	2.5		
21340	.255	730	2.5		
21341	. 255	730	2.5		
21342	.255	730	2.0		
21343	.255	730	2.0		

TABLE IV

The results (Table IV) of the assays of this non-saponifiable material indicate that an equivalent of 730 cc. of human milk contain 3 USP XI units of vitamin D. The potency of the breast milk was therefore of the order of 4 USP XI units per quart.

DISCUSSION

The 15 gallons of breast milk used in these experiments, gathered during December, 1937, and January, 1938, constitute the largest, and therefore most representative, sample of human breast milk ever subjected to bioassay at one time. To conduct this assay it was necessary to prepare the non-saponifiable fraction of the milk fat since a rat will not consume enough breast milk or butter fat daily for a precise assay.

The results indicate that the milk from women in an urban community during the winter months contained not over 4 USP XI units per quart. It has been reported that cow's milk during the summer contains between 23 and 36 USP XI units per quart.

Whence, then, the greater power of breast milk, even in the winter, to protect the human infant against rickets more adequately than modified summer cow's milk? In the bottle feeding of the very young infant, cow's milk is customarily diluted with an equal volume of water. Such modified milk has a vitamin D content of at least 11

USP XI units per quart-nearly 3 times that of human breast milk. As the infant grows and formulae are changed, this disparity widens. The explanation lies elsewhere than in the vitamin D content.

Previously,³ we reported that the milk from mothers fed metabolized milk contained an equivalent of 21 USP XI units per quart. On the basis of the present finding, it may be seen that the vitamin D content of breast milk can be increased at least five-fold.

CONCLUSIONS

1. A composite sample of 15 gallons of human milk contained only 4 USP XI units of vitamin D per quart.

2. The antirachitic superiority of human milk over cow's milk in infant feeding cannot be attributed solely to the quantity of vitamin D which it contains.

3. It is possible to increase the antirachitic potency of human milk at least five-fold by feeding metabolized milk to the lactating mother.

REFERENCES

REFERENCES 1. Hess, A. F., Weinstock, M., and Sherman, E. Antirachitic Properties in Human Milk Developed by Irradiating the Mother. J.A.M.A., 88:24, 1937. 2. Outhouse, J., Macy, I. R., and Brekke, V. Human Milk Studies. V. Quantitative Comparison of the Antiricketic Factor in Human Milk and Cow's Milk. J. Biol. Chem., 78:129, 1928. 3. Bunker, J. W. M., Harris, R. S., and Eustis, R. S. Antirachitic Potency of the Milk of Human Mothers Fed Previously on Vitamin D Milk of the Cow. New Eng. J. Med., 208:313, 1933. 4. Hess, A. F., and Weinstock, M. A Study of

the Antirachitic Factor in Human and in Cow's

the Antirachitic Factor in Human and in Cow's Milk. Am. J. Dis. Child., 35:845, 1927. 5. Lesné, E., and Vagliani, M. Production d'un Lait de Vache Donné de Propriétes Antirachitics. Compt. rend. Acad. d. sc., 179:539, 1924. 6. Palmer, L. S., and Kennedy, C. Antirachitic Properties of Breast Milk. Proc. Soc. Exper. Biol.

& Med., 23:230, 1925.
7. Sabri, F. A., and Fikri, M. M. Experimental Study of the Antirachitic Factor in Human and Animal Milk. Arch. Dis. Childhood, 7:239, 1932.
8. Sabri, F. A., and Fikri, M. M. Further Study of Antirachitic Factor of Human Milk. Arch. Dis. Childhood, 10:377, 1935.