Summary

The clinical manifestations of 104 cases of Weil's disease occurring between 1934 and 1939 are described.

In the paper are discussed some of the bacteriological and serological investigations carried out.

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THE ANTIRACHITIC VALUE OF HUMAN MILK

BY

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It is a surprising fact that the extensive literature on vitamin D does not contain precise information giving the range of antirachitic potency of human milk. It is a matter of particular importance when the character of the diet consumed by a large proportion of the people in this country is borne in mind. Our knowledge regarding the assimilation of bone-forming elements in the diet of mammalian species has been largely built up on the results of experiments on animals; these have taught us how great an importance is to be attached not only to vitamin D but to the total daily intakes of calcium and of phosphorus in one form or another, as well as the ratio (Ca/P) between these intakes. An admirable summary of the existing information regarding the significance of the level of intake of these mineral elements has recently been provided by Shohl (1938), but, broadly speaking, it can be said that, at any rate one level of Ca or P intake—assimilation—in the absence of supplementary factors such as members of the vitamin D group, is most satisfactory within a certain and rather narrow range of values for the Ca/P ratio. Moreover, at any one Ca/P ratio the assimilation tends to improve as the total intake of the two components rises. Finally, it should be noted that the administration of vitamin D at any level of intake or Ca/P ratio improves the deposition of bone mineral, but, in general, its influence is most pronounced when the former is low and when values of the ratio are outside the desirable range.

In days when fish-liver oils, antirachitic concentrates, and even the pure vitamins of the D group are so readily available, it is easy to forget that until the introduction

in England in the late eighteenth century of cod-liver oil for the treatment of bone affections the ordinary person had no such means of protection against rickets. Accustomed as we are to-day to think in terms of doses of hundreds and thousands of international units of vitamin D, the amounts supplied by ordinary foods must strike us as curiously insignificant (milk 0.5 to 4, butter 20 to 100, egg yolk 150 to 500, liver 10 to 50 I.U. per 100 grammes) (Fixsen and Roscoe, 1937–8). What, then, was the position in England before the virtues of cod-liver oil were recognized? In a recent publication Drummond and Wilbraham (1939) examined this question in some detail, with the result, it is thought, that evidence has been obtained to show that the striking outburst of rickets which appears to have occurred in Southern England and Ireland towards the end of Tudor times, and which attracted great interest, was due primarily to a sharp fall in the consumption of dairy produce. It is there suggested that the outbreak of the disease was more related to a decreased intake of calcium in many diets than to a reduction in the amount of natural vitamin D, although doubtless both played a part.

Calcium Requirements

The remarkable facts revealed by the discovery of vitamin D and the investigation of its role have tended to direct attention rather unduly, so far as the study of human rickets is concerned, from the significance of the intake of bone-forming elements. The surveys of Sir John Orr (1936) and of others have given convincing proof how large a proportion of our population to-day are living on diets which fail to supply the estimated physiological requirement for calcium. These diets are in striking contrast to those of the relatively poor Englishman of centuries ago, when, thanks to wholemeal breads and large consumption of milk and cheese, the daily intake of calcium was often as much as four to six times that of the poor to-day. Seventeenth century diets of simple folk—for example, sailors or the patients of St. Bartholomew's Hospital—provided no less than 2 grammes of calcium a day, and, let it be noted, the Ca/P ratios were often excellent. It is not unreasonable to think that the human requirement for vitamin D would probably be at its minimum under such conditions. They are rarely found to-day. Hundreds of thousands of the poorer sections of the community are living at the present time on diets supplying from 0.2 to 0.5 gramme of calcium daily—intakes which are all too often associated with relatively high intakes of phosphorus, giving ratios of below 0.5 and not infrequently as low as 0.2.

Experiments have already clearly demonstrated the beneficial effect of supplementing diets of this character with additional calcium, thereby not only raising the total intake of bone-forming salts but also bringing the figure for the Ca/P ratio nearer the optimum range of 1 to 2. The improvement particularly affected the bony structures, and in this connexion it is important to remember that no additional vitamin D was supplied (Coward *et al.*, 1938; Gaunt *et al.*, 1938).

Factors leading to Rickets

The surprisingly small amount of vitamin D in cows' milk (17 to 26 I.U. per quart in summer and a mere 5 to 8 I.U. per quart in winter) and the discovery of the fact that the higher values in summer are due not to a change of diet but to the direct effect of exposure to sunshine and skyshine (Campion *et al.*, 1937) raised the very important question whether the smaller incidence of

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rickets among breast-fed infants, as compared with those reared on cows' milk, is to be ascribed to a more efficient utilization of the bone-forming salts, to a richer content of antirachitic vitamin in mothers' milk, or perhaps to both factors.

The literature on the antirachitic value of human milk is unsatisfactory. Hess and Weinstock (1927) found that 25 to 30 ml. of human milk was required to prevent rickets developing in rats fed on a rachitogenic diet, whereas 20 to 25 ml. of cows' milk was effective. The results of this investigation, and of similar experiments subsequently reported by other workers, are misleading because striking improvements in the condition of the bones of animals fed on certain types of rachitogenic diets can be brought about merely by improving the Ca/P ratio. The apparent superiority of cows' milk as an antirachitic agent revealed by such experiments could be ascribed more correctly to its higher content of these elements than to any greater concentration of vitamin D.

In order to assay the vitamin D content of milks it is advisable to separate the fat and administer it to the

entirely satisfactory in that they were for the most part random samples collected during the first two months of pregnancy from patients attending the infant welfare department of University College Hospital. Unfortunately, it was not practicable to obtain samples fully representative of the twenty-four-hours yield. On receipt of the specimens in the laboratory sufficient was retained for the calcium and phosphorus analyses (see later), the remainder being treated with alcohol and a mixture of ether and light petrol in order to separate the fat. The isolated fat was administered in carefully measured doses to groups of rats in which rickets had been produced by a rachitogenic diet. The degree of rickets at the beginning of the experiment and after ten days' dosage with the milk fats was assessed by x-ray examinations of the leg bones.

The milks were also analysed for total and dialysable calcium and phosphorus, the method for the phosphorus analyses being that of Kuttner and Lichtenstein (1930). For determining the dialysable Ca and P we used a suitable modification of the "static" dialysis method employed by Lampitt and his co-workers (1933, 1934, 1937). Usually about 120 ml. of milk, from which as much fat had been removed as possible by centrifugation, was dialysed against 3.3 ml. of water.

This provided about 2 ml. of dialysate suitable for analytical purposes. The results as a whole are set out in Table I. An attempt was made to ascertain whether any correlation could be traced between the results of the analyses of the individual milks and such information as was available regarding the nutritive state of the mother. Unfortunately it was not found possible to obtain precise details regarding the everyday diet of the women, but, by questioning, a certain amount of information was gathered which permitted a rough classification to be made, partly on the basis of the general character of the diet and partly on statements regarding the total income, number in the family, rent, etc.

Factors influencing the Vitamin D Content of Milk

An attempt has been made to correlate the results with various factors which are believed to influence the vitamin D content of cows' milk. In the first place, there is no evidence of a seasonal variation. The mean value of sixteen samples taken during the winter months (October to March, inclusive) is 5.9 I.U. per 100 ml., which compares well with that of 6.2 for the ten summer samples. This is a reasonable conclusion, for one would scarcely expect an effect from the influence of sunshine or skyshine to be noticeable in working-class women in London. The mean value of the whole twenty-six samples is 6 I.U. per 100 ml.

What seem to be significant differences appear when the results are classified according to estimates of the intakes of vitamin D by the women (Table II). The value of these results is open to question because, in the first place, the numbers in the groups are small, and, secondly, because we had not the information to provide an accurate estimate of the intake of vitamin D in the mothers' diet. With this reservation, however, we are inclined to believe that the results show that the vitamin D content of human milk is increased by raising the vitamin D content of the diet. Bunker, Harris, and Eustis (1933) have presented evidence that the administration to women of cows' milk artificially treated to increase its vitamin D value will raise the antirachitic value of their milk.

We are inclined to think—although again caution is necessary in view of the small number of samples analysed—that approximation between the mean antirachitic

TABLE I.—Analyses of Human Milks for Vitamin D, Calcium, and Phosphorus

No.	Month of Test	% Fat	I.U. Vitamin D		Mg. % Ca		Mg. P %	
			Per gm. Fat	Per 100 ml. Milk	Total	Dialysable	Total	Dialysable
1	Jan.	4.0	1	4	25	—	10	—
2	Feb.	3.3	2.5	8	26	6	13	—
3	"	4.0	0.5	2	34	8	14	5.2
4	Mar.	3.75	2	7.5	35	11	15	6.4
5	"	4.0	2	8	37	18	15	8.8
6	Apr.	4.2	1	4	26	17	15	6.3
7	"	3.9	1	4	32	23	16	7.5
8	"	3.9	1	4	33	21	15	7.5
9	"	3.5	1.2	4	33	20	15	6.4
10	May	7.1	1.3	9	30	16	14	3.6
11	"	6.1	0.7	4	26	14	15	6.6
12	"	2.4	1.1	2.5	—	—	—	—
13	June	5.9	3	18	31	21	17	9.3
14	"	4.0	1	4	29	15	12	8.2
15	July	4.5	1	4.5	38	22	14	7.7
16	Sept.	3.0	2.7	8	—	—	—	—
17	Oct.	3.4	2.7	9	—	—	—	—
18	Nov.	5.5	0.7	5.5	26	23	18	6.0
19	"	3.8	1	4	36	—	17	—
20	"	4.1	1.8	7	34	22	13	11.0
21	Dec.	5.1	1.7	9	25	18.7	13	8.4
22	"	3.3	1.6	5	41	39	13	8.0
23	Jan.	4.5	1.5	6.5	28	22	10	7.0
24	"	4.0	1	4	—	—	—	—
25	"	4.2	1.2	5	—	—	—	—
26	Feb.	3.5	1	3.5	—	—	—	—
27	"	3.0	1.5	6.5	—	—	—	—

animals fed on a rachitogenic diet. Few investigators have troubled to do this, and none, so far as we are aware, has attempted to make assays of the vitamin D content of human milk-fat by the modern method of evaluation against the approved international standard of vitamin D. The specimens examined by us were not

potency of the milk yielded by women on diets supplying in all probability between 100 to 250 I.U. of vitamin D daily and that of the milk from women dosed during the latter half of pregnancy with much larger amounts of vitamin D (approximately 1,000 I.U. daily) in the form of halibut-liver oil supplement indicates that 10 I.U. per 100 ml. of breast milk represents about the normal maximum value. Possibly the same evidence can be put forward to indicate that an intake of about 200 I.U. represents the *minimum* daily requirement of the nursing mother. Our results appear to show that the large doses of vitamin D given to some of the mothers over several months of pregnancy failed to bring about anything approaching a proportional increase in the vitamin D content of the milk. It may be, therefore, that there is an optimum concentration of this substance regulated by the mammary gland. It will be recalled that Gerstenberger, Hartman, and Smith (1927) failed to increase the antirachitic potency of breast milk by giving a teaspoonful of cod-liver oil (250 I.U.?) daily to a woman already receiving an adequate diet.

Our estimate of 200 I.U. of vitamin D as the *minimum* intake of the pregnant and lactating woman is in reasonable agreement with the daily intake of 300 I.U. recently recommended by the Committee of the Health Organization of the League of Nations.

TABLE II.—Attempt to correlate Vitamin D Content of Diet and of Milk

Estimated Vitamin D in Diet. I.U. Daily	Vitamin D Content of Milk Fat. I.U. per gramme	Vitamin D Content of Milk. I.U. per 100 ml.
<i>Group A. Daily D intakes probably less than 100 I.U.</i>		
35	1	4
50	1	4
50	1	4
70	1	4
70	0.5	4
85	0.5	2
Mean	0.8	3.7
<i>Group B. Daily intake probably above 100 I.U.</i>		
125	3	18
130	2.7	8
145	1.2	4
155	2.5	8
180	1.3	9
200	1	4
220	1.2	4
Mean	1.8	7.9
<i>Group C. Women receiving vitamin D therapy (1,000 I.U. daily as halibut-liver oil) during latter half of pregnancy (figures in first column refer to diet only).</i>		
36	0.7	5.5
60	1.8	7
90	2.7	9
90	1.7	9
190	1.0	4
200	1.5	6.5
Mean	1.5	6.9

Vitamin D Requirement for Bone Formation in Infants

It is necessary now to consider our results from the standpoint of bone formation in infants. There is, or, at all events, there has been until recently, no very clear

idea how much vitamin D is required by an infant. For some time past it has been rather generally accepted that artificial feeding with cows' milk should be supplemented with vitamin D therapy. All sorts of doses have been recommended, but a considerable proportion of paediatricians have been in favour of daily doses of the order of 1,000 I.U. It now appears that in the majority of cases this is too much.

In a series of careful studies, in which cows' milk which had been artificially "fortified" in respect to vitamin D was used, Jeans and Stearns (1937, 1938a) were able to show that about 70 to 100 I.U. a day during the first three months will prevent the appearance of rachitic symptoms in the majority of full-term infants. This level of intake, provided by approximately 120 ml. per kg. of body weight of milk containing 12 I.U. per 100 ml., was not found adequate for premature babies, nor did it ensure optimum growth. Full protection was given when the intake of vitamin D was three to four times as great. The very large doses—for example, 1,000 to 5,000 I.U. daily—which were so popular in the early days of enthusiasm for vitamin D therapy and which are still not infrequently employed, are probably unnecessary and actually appear sometimes to exert a definitely inhibitive action on growth (Jeans and Stearns, 1938b).

Influence of Calcium-Phosphorus Ratio

It is interesting that a satisfactory intake of cows' milk providing 12 I.U. of vitamin D per 100 ml. has been found to produce good calcification in normal full-term infants, because the amount of vitamin D taken by the young child would be of the same order as that provided by the milk of a well-nourished mother. This probability has to be considered together with the fact that the assimilation of the bone-forming elements is better (40 to 60 per cent.) when the child is fed on mother's milk than when cows' milk is used (20 to 30 per cent.). The reason for this better utilization has not yet been determined, but it may be important that the Ca/P ratio of human milk approximates to that of bone mineral, whereas the Ca/P ratio of cows' milk is considerably smaller.

	Ca (mg.%)	P (mg.%)	Ca/P Ratio
Human milk	27-34 ..	13-17 ..	2.0
Cows' milk	100-160 ..	70-100 ..	1.5
Bone mineral (dahlite) $2Ca_3(PO_4)_2 \cdot CaCO_3$			2.2

The relatively small concentration of calcium and phosphorus in human milk as compared with cows' milk raises several curious problems, of which the most important has been revealed by the careful studies of Leitch (1937). She has shown that the *total* intake of calcium when mother's milk is taken, much less the quantity actually *retained*, is often insufficient to provide the amount calculated as being requisite to maintain the composition of the body either at the maximum figure of 10 grammes of calcium per kilogram of body weight or at the smaller estimate of 8.13 grammes of calcium per kilogram (Hamilton, 1922).

Consideration of this extraordinary finding has led Dr. Leitch to consider the possibility that breast-fed infants must often exhibit not only imperfect calcification of new matrix but also osteoporotic changes in bones which at birth were relatively firm. It may be a fact that undetected rachitic changes are much more common in breast-fed infants than is suspected (Maddox, 1932), and it may also be true that if it were possible to make histological examinations we would be surprised at the frequency with which osteoporotic changes occur in the first half of lactation (Zarfl *et al.*, 1923). There seem to be only two ex-

planations of the figures Dr. Leitch has produced. Either the newly born infant carries a reserve store of calcium sufficient to provide for adequate calcification during the months when the mother's milk does not provide enough—and it must be borne in mind that there is such a protective storage of iron—or the data of the metabolism experiments which Dr. Leitch has used for her analysis are misleading, perhaps because the milk was not of such a quality as to favour optimum calcium retention.

It remains uncertain in view of the evidence reviewed by Leitch whether changes in the dietary intake of calcium or phosphorus are reflected to any significant extent in the composition of a mother's milk. Our own results, admittedly scanty, suggest that such an effect is not detectable, at all events in the first two or three months of lactation. Some of the mothers we investigated had been receiving about 2 grammes of calcium lactate daily during the latter half of pregnancy, but this supplement to calcium-deficient diets did not appreciably influence the calcium content of their milks.

Conclusion

There is certainly need for more information, not only about the composition but also about the *yield* of milk from mothers whose diet, adequate in all other respects, provides at least 2 grammes of calcium, 1.5 grammes of phosphorus, and 300 I.U. of vitamin D daily. If such milk were made the subject of metabolic investigation on infants it is possible that some of the disturbing discrepancies would be dispelled.

Summary

The literature on vitamin D does not contain precise information as to the range of antirachitic potency of human milk.

The results of an analysis of human milks for vitamin D, calcium, and phosphorus are here recorded and an attempt made to correlate the vitamin D content of diet with that of the milk.

The conclusions derived from the analysis are given.

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THE PATHOLOGY OF CHLORIDE METABOLISM IN MAN*

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During the past ten years considerable significance has been attached to disturbances of the metabolism of chlorides in man. The information gained from observations on patients and experimental work in animals has applications in diseases so diverse as intestinal obstruction, pyloric stenosis, chronic nephritis, oedema, obesity, hypertension, and Addison's disease. The principles which control the chloride balance of the body and determine the nature of the processes of absorption, secretion, re-absorption, and excretion of chloride are of importance in these diseases, and their significance is already reasonably well established. The phenomena which control the exchange of chlorides in tissue cells and fluids are not yet fully explained.

Normal Cycle of Chloride Metabolism

The average normal adult on an ordinary diet ingests about 12 grammes of chloride daily. This can be easily verified by estimating the daily excretion of chloride in the urine. In addition to the urinary excretion, small quantities of chloride are excreted in the faeces and limited amounts in the skin secretions (probably not more than 2 grammes daily in the absence of active perspiration). In forced sweating the skin loss in chloride may amount to 10 or 12 grammes daily (McCance, 1938). It should be borne in mind that clinical analogies to such abnormal skin losses occur in conditions where there is intense sweating, in acute febrile disturbances, in the sweating of tuberculosis and acute rheumatism, in pneumonia, in thyrotoxic crises, and in heavy muscular exertion in hot surroundings.

Apart from external losses by these routes—skin, urine, and faeces—there occurs a continual circulation of chloride to and from the intestinal lumen. The total quantities of chloride in circulation in this way daily in the normal person are somewhat indefinite. Approximate measurements of volume and chloride content of the various alimentary secretions give some basis for calculation, as shown in Table I (McCance, 1936).

TABLE I.—*Intestinal Secretions: Chloride Content*

Secretion	Vol. (c.cm.) in 24 Hours	Authority	Chloride Content (grammes)
Saliva	1,500	Bidder and Schmidt	7.5
Gastric juice ..	2,000-3,000	Nicol and Lyall	17
Bile	300-500	Pfaff and Balch	4
Pancreatic juice	500-800	Wohlegemuth	8
Succus entericus	3,000	Pregl	18
Total chloride			54.5

These figures are quite approximate, and probably form a conservative estimate.

Nicol and Lyall (1939a and 1939b), for example, as shown in Table II, were able to remove 29 grammes NaCl by gastric aspiration from a male patient in the first day, and a total of 93 grammes in six days' aspiration, the amount on the first day representing the uninfluenced gastric secretion of chloride in this individual.

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