cal College) also reported some low serum $1,25(OH)_2D_3$ values. He measured 10 pg ml^{-1} in subjects with D-dependent rickets but in D-resistant rickets the measurements were 32 pg ml^{-1} and were similar to control subjects—a surprising finding, for the low serum phosphate levels in these subjects might have been expected to produce higher $1,25(OH)_2D_3$ values. Nevertheless, Haussler reported that treatment with $1,25(OH)_2D_3$ cured the rachitic lesions of D-resistant rickets and was similarly effective in the treatment of a patient with tumour-induced osteomalacia with a serum $1,25(OH)_2D_3$ of 17–18 pg ml^{-1}.

One of the more curious twists to the $1,25(OH)_2D_3$ story is Haussler's observation that female spawning trout have levels of the metabolite in excess of 75 pg ml^{-1}. The metabolite is said to be important for phosphate regulation in the eel (McIntyre *et al. J. clin. Endocr.* **5**(Supp.) 85S; 1976) yet neither D. R. Fraser (see *News & Views* **273,** 426; 1978) nor M. R. Holick (Massachusetts General Hospital) has been able to detect this or other dihydroxy metabolites of vitamin D_3 in fish receiving isotopically-labelled vitamin. In view of the specificity of Haussler's assay many would assume that it is indeed $1,25(OH)_2D_3$ he is measuring. However, the observations of Fraser and Holick suggest that it may not be this metabolite, but perhaps one of a closely related structure.

Several authors stressed that $24,25(OH)_2D_3$ may be important for calcium action in bone but not everyone is convinced that it is. S. Edelstein (Weizmann Institute, Israel) presented evidence that $24,25(OH)_2D_3$ seems to be necessary for endochondral ossification of cartilage and bone. J. Kanis (University of Oxford) says the metabolite increases calcium retention in man and that this may be associated with increases in skeletal calcium accretion. And H. M. Malluche (University of Southern California School of Medicine) reported that in chicks neither $1,25(OH)_2D_3$ nor $24,25(OH)_2D_3$ would maintain bone integrity on their own, but that both metabolites were required for this purpose, a view put forward earlier by Ornoy *et al.* (*Nature* **276,** 517; 1978). S. W. Stanbury (University of Manchester) questions whether $24,25(OH)_2D_3$ is so important for calcification of bone. In subjects treated for vitamin D deficiency with 1,000 IU of ^3H-D_3 Stanbury observed $1,25(OH)_2D_3$ and $25,26(OH)_2D_3$ in the serum in the first few days but no $24,25(OH)_2D_3$. In some cases $24,25(OH)_2D_3$ was not seen until 6 or 10 days after the start of vitamin D therapy. In view of the fact that changes in the calcification front of bone are seen 3–5 days after the administration of vitamin D, Stanbury queried whether $24,25(OH)_2D_3$ was the principal metabolite responsible for mineralisation. Could it be that $25,26(OH)_2D_3$ is a more important metabolite for mineralisation he asked. Stanbury also reported that serum levels of $25,26(OH)_2D_3$ were 0.5–0.8 ng ml^{-1} and about a third of the $24,25(OH)_2D_3$ values in D-replete subjects. J. L. H. O'Riordan (Middlesex Hospital) on the other hand found much lower $25,26(OH)_2D_3$ values (178–336 pg ml^{-1} using a radioimmunoassay. The antibody used by O'Riordan has been developed for use in measuring $1,25(OH)_2D_3$, however, in view of its high, and as yet unexplained, cross-reactivity with the other dihydroxymetabolites of vitamin D_3 it is being used to measure $25,26(OH)_2D_3$.

Measurements of $24,25(OH)_2D_3$ still tend to be controversial chiefly because of the twofold difference reported for UK and US subjects. C. M. Taylor (University of Manchester) says that $24,25(OH)_2D_3$ values are about 8% of the $25(OH)D_3$ values whereas Haddad *et al.* (*Arch. Biochem. Biophys.* **182,** 390; 1977) claim that in the US they are about 20%. Furthermore, Taylor says that there is no detectable $24,25(OH)_2D_3$ in anephric subjects whereas Haddad, H. F. DeLuca (in *Proc. VII International Congress of Nephrology,* Montreal, 1978) and R. L. Horst (University of Wisconsin) say there is. However, it seems that these differences between the UK and US laboratories may not be irreconcilable. There are two possible explanations.

The first is one of three new metabolites of vitamin D_3 reported by H. F. DeLuca (University of Wisconsin) one of which comigrates with $24,25(OH)_2D_3$ on Sephadex LH 20 chromatography, but which is separated from it on high performance liquid chromatography (HPLC). The metabolite is a lactone involving carbons 23 and 25 of the D_3 side chain. Although the lactone accounted for a sizeable portion of the $24,25(OH)_2D_3$ peak on Sephadex when chick serum was used, Horst said that he had not seen it in human plasma. The other alternative is the 25,26-dihydroxymetabolite of vitamin D_2 which we (Hay & Jones *Clin. Chem.* **25,** 473; 1979) have reported also to comigrate with $24,25(OH)_2D_3$ on Sephadex LH 20. Taylor confirmed this observation, but reported that this comigration also occurs on HPLC. It seems, therefore, that where vitamin D_2 is consumed in appreciable quantities in the diet—as it is in the US—that $25,26(OH)_2D_2$ may be assayed together with $24,25(OH)_2D_3$. This D_2 metabolite may also account for the $24,25(OH)_2D_3$ values recorded in anephric subjects in the US.

Measurements of both $25,26(OH)_2D_2$ and $24,25(OH)_2D_3$ will be required before this can be answered definitively. Perhaps when these results are available the UK and US results will be seen to agree and investigators can turn once more to elucidate the actual functions of the 24,25 and 25,26 dihydroxy metabolites of vitamin D, and indeed that of all the other metabolites discovered over the past year, for it is quite clear that many doubts exist about some of the theories put forward so far. □

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A hundred years ago

A STRIKING and highly promising line of research has been recently adopted by Mr. Muybridge, of San Francisco, at the instance of Governor Stanford, viz., the instantaneous photography of animals in motion. Some of his earlier photographs of a fast-trotting horse presented attitudes wholly unexpected, and they were even thought absurd. The method latterly adopted seems to have been making the horse trot or gallop past twelve cameras, arranged in series, and by breaking threads stretched across its path, release an electric current, which effected exposure of the plates. Thus the flying steed was obtained in every position. Mr. Muybridge gave public exhibitions of what had been done. The small negatives were magnified into life size, and projected on a screen, so that every motion was visible. These exhibitions do not seem to have been appreciated by the San Franciscans. The *Scientific American,* however, and afterwards *La Nature,* have published cuts taken from the photographs, and much general interest has been awakened in these researches. Among those specially interested is Prof. Marey, who desired to be put in communication with Mr. Muybridge, as he wanted to ask his aid in solving certain physiological problems, so difficult to solve otherwise; *e.g.,* questions connected with the flight of birds. He had been dreaming of a kind of photographic gun, to seize the bird in an attitude or series of attitudes of flight. What beautiful zootropes, too, might be had! Mr. Muybridge's cartoons representing the fast gallop give a key to the breaking down of so many horses. It appears as though one foreleg had to sustain the whole weight of horse and rider while the body is moved along five feet. And just before the foot is raised, a perpendicular from it would strike the back of the saddle; so that there is immense leverage, the centre of gravity being thrown so far forward of its support, and the tendons must have a terrible tension. These inquiries are being further developed by the liberality of Governor Stanford and the skill of Mr. Muybridge, and valuable results may doubtless be looked for.

From *Nature* **19,** 3 April. 517; 1879.

0028–0836/79/0278–0510$01.00