Cyclolobus is confined to the upper Chhidru. All the Salt Range representatives of *Cyclolobus* for which accurate occurrence data are available came from 10 to 23 m below the top of the Chhidru (*5*, p. 73). There is no conclusive ammonoid evidence for the precise age of the Kalabagh.

Four additional ammonoids have been reported from the Zewan Formation in the vicinity of Srinagar. "*Xenaspis* cf. *carbonaria* Waagen" was recorded by Diener (*17*) as being represented by three specimens (India Geological Survey, Nos. 11117 to 11119) from the "Spur 2 miles N. of Barus," a well-known locality (*12*) approximately 11 km south-southeast of Guryul Ravine. The parent horizon is "Unit 3½" of Middlemiss and probably corresponds to the basal few meters of the Triassic black shales at Guryul Ravine rather than the underlying Zewan Formation. Our recent study of these three specimens reveals that they are at least specifically distinct from the widespread Late Permian *Xenodiscus carbonarius*, and are best regarded as indeterminate Triassic ophiceratins, possibly *Glyptophiceras*. An associated bivalve [plate 6, figure 1, in (*17*)] is *Claraia stachei* (Bittner). The fourth ammonoid reported from the Zewan Formation (India Geological Survey, No. 11120) was collected near Pahlgam (Pailgam) approximately 95 km east of Srinagar. Stratigraphic occurrence of the specimen is uncertain (*12*) and Diener's assignment (*17*) to the Permian genus *Popanoceras* is certainly incorrect. Our examination leads us to regard the specimen as generically indeterminate, but Triassic in age. Unquestionably Permian ammonoids, including *Xenodiscus* and *Stacheoceras*, have been collected recently from Pahlgam and adjacent areas, but the details of occurrence are not yet available.

The discovery of *Cyclolobus walkeri* near the top of the Zewan Formation of Kashmir strengthens the correlation of the parent beds with the upper Kuling Shale of the central Himalayas, the upper Chhidru Formation of the Salt Range, and the Ankitohazo beds of Madagascar. All occurrences are interpreted as representing the Chhidruan Stage of the middle Dzhulfian Series (*18*). It follows that if no major hiatus occurs in the upper Zewan, the latest Dzhulfian Changhsingian Stage is represented in the top few meters of the Zewan Formation at Guryul Ravine. Detailed collecting and comparison of the fauna from this interval with that from the type Changhsingian of South China and the correlative *Paratirolites* beds of Armenia and possibly Madagascar are proceeding (*19*).

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1α-Hydroxy Derivative of Vitamin D_3: A Highly Potent Analog of 1α,25-Dihydroxyvitamin D_3

Abstract. *The 1α-hydroxy derivative of vitamin D_3 has been chemically synthesized and tested for its biological activity. This analog has comparable biological activity on a weight basis to 1,25-dihydroxyvitamin D_3 in the stimulation of intestinal calcium transport and bone calcium mobilization in normal and anephric rats. Because the 1α-hydroxy derivative is synthesized from cholesterol, it is easier and less expensive to prepare than 1α,25-dihydroxy derivative, making it attractive as a drug in the treatment of renal osteodystrophy and hypoparathyroidism.*

For vitamin D to carry out its biological functions in stimulating intestinal calcium transport and mobilizing bone calcium, it must be hydroxylated on C-25 in the liver (*1*) and subsequently on C-1 by the kidney (*2*). The resulting metabolite, 1α,25-dihydroxyvitamin D_3 [1α,25-$(OH)_2D_3$] (Fig. 1) was isolated in pure form, identified (*3*), and recently synthesized (*4*). This metabolite is the most potent form of vitamin D known (*5*, *6*), and it is the only known

Table 1. Intestinal calcium transport and bone calcium mobilization response of rats to 1α-OH-D_3. Results are means of six rats; S.E., standard error.

1α-OH-D_3 (pmole)	^{45}Ca serosal/ ^{45}Ca mucosal (mean ± S.E.)	Serum Ca (mg/ml) (mean ± S.E.)
	Normal	
None*	1.5 ± 0.2	4.3 ± 0.1
6.2	1.8 ± 0.2	4.8 ± 0.1
62.5	2.8 ± 0.3	5.8 ± 0.1
625	2.9 ± 0.2	6.7 ± 0.1
1250	2.9 ± 0.2	6.9 ± 0.1
	Anephric	
None*	1.5 ± 0.2	4.1 ± 0.1
312	2.3 ± 0.2	5.2 ± 0.1

* The control dose consisted of 50 μl of 95 percent ethanol alone.

vitamin D metabolite active in anephric rats (*7*).

During the course of the 1α,25-$(OH)_2D_3$ synthesis we explored the various reactions utilizing a less expensive starting material, namely cholesterol instead of homocholenic acid. As a result, 1α-hydroxyvitamin D_3 (1α-OH-D_3) (Fig. 1) was prepared.

The possibility that such an analog of 1α,25-$(OH)_2D_3$ might be biologically active was of obvious interest and consequently was examined. The analog proved to be approximately equal to 1α,25-$(OH)_2D_3$ in inducing intestinal calcium transport and bone calcium mobilization in normal and anephric rats.

The synthesis of 1α-OH-D_3 was accomplished by the methods described for 1α,25-$(OH)_2D_3$, except that cholesterol was used as starting material (*4*). The resulting product showed the expected ultraviolet absorption spectrum (λ_{max} 265 nm, λ_{min} 228 nm) for the 5,6-*cis* triene system and mass spectrum [*m/e* 400 (M^+), 287, 152, and 134].

Furthermore, the compound chromatographed as one peak on gas-liquid chromatography, demonstrating its purity (it is interesting that, although this analog has a 5,6-*cis* triene system, it does not cyclize in the gas chromatograph to the pyro and isopyro forms as do other vitamin D metabolites, probably because of the close proximity of 1α-OH to the triene system).

Weanling male rats (Holtzman) were fed for 2 weeks a vitamin D–deficient diet containing adequate calcium and phosphorus (*8*) and then fed a low calcium (0.02 percent), vitamin D–deficient diet for another week (*9*). Groups of rats, either bilaterally nephrectomized or normal, received the appropriate dose via the jugular vein in 0.05 ml of 95 percent ethanol (Fig. 2 and Table 1). Intestinal calcium transport and bone mobilization assays were performed as described (*9*).

The results shown in Fig. 2 demonstrate that 1α-OH-D_3 is capable of stimulating intestinal calcium transport and bone calcium mobilization. This analog elicits an intestinal calcium transport response as early as 3 hours after its administration and shows a maximum response at 6 hours. 1,25-$(OH)_2D_3$ also gives a maximum response 6 hours after intravenous administration (*6*). Although 1α-OH-D_3 does not show as large a transport response as 1,25-$(OH)_2D_3$ at 6 hours, it sustains the response for at least 24

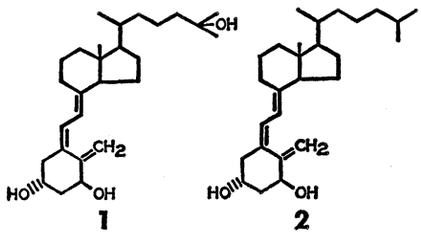

Fig. 1. Structures of 1α,25-$(OH)_2D_3$ (**1**), and 1α-OH-D_3 (**2**).

hours, while the response to 1,25-$(OH)_2D_3$ decreases after 6 hours (*6*). The 1α-OH-D_3 is capable of mobilizing calcium from bone as early as 3 hours after its administration and shows a maximum response at 14 hours.

Table 1 shows that as little as 62.5 pmole of 1α-OH-D_3 induces intestinal calcium transport and bone mineral mobilization at 14 hours after administration. A similar response to 62.5 pmole of 1,25-$(OH)_2D_3$ has been demonstrated (*10*). Of great importance is the fact that 1α-OH-D_3 stimulates both intestinal calcium transport and bone mineral mobilization in anephric rats (Table 1). However, as the dose of 1α-OH-D_3 is increased from 625 pmole to 1250 pmole, there is no significant increase in the intestinal calcium transport response.

Because the 1α-OH-D_3 is less expensive and less difficult to prepare chemically as compared to 1α25-$(OH)_2D_3$, but has similar activity in both anephric and intact vitamin D–deficient rats, it is potentially attractive for the treatment of renal osteodystrophy and hypoparathyroidism. It has been suggested that the 5,6-*trans* form of vitamin D_3 may be a suitable substitute for 1,25-$(OH)_2D_3$ because it can induce both intestinal calcium transport and bone calcium mobilization in anephric rats (*9*). Although this analog is easy and inexpensive to make it is about 50 to 100 times less active than either the 1,25-$(OH)_2D_3$ or 1α-OH-D_3.

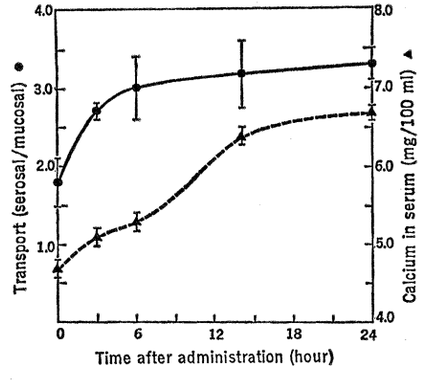

Fig. 2. Intestinal calcium transport (——) and bone calcium mobilization (- - - - -) response of vitamin D–deficient rats on a low calcium diet to 625 pmole of 1α-OH-D_3. The vertical bars represent the standard error of the means; six animals were used for each measurement.

It is not possible to predict at this time whether 1α-OH-D_3 is hydroxylated on C-25 in the liver before it is active. It is interesting that the time it takes to produce a maximum response in the intestine parallels that for 1α,25-$(OH)_2D_3$, which would argue against further hydroxylation. On the other hand the 1α-OH-D_3 does not attain the high intestinal calcium transport response seen for 1α,25-$(OH)_2D_3$ but can sustain its response for longer periods, which could be consistent with the idea that the 1α-OH-D_3 is further hydroxylated presumably on C-25. Additional investigation will be necessary to answer these questions, but the possible utility of this compound in medicine seems clear.

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