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# A new metabolite of Paricalcitol: stereoselective synthesis of (22*Z*)-isomer of 1α,25-dihydroxy-19-norvitamin $D_2$

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ABSTRACT

Stereoselective synthesis of (22*Z*)-isomer of Paricalcitol, an analog of 1,25-dihydroxyergocalciferol, an active form of vitamin $D_2$ (Ergocalciferol) has been described. The two key critical synthetic steps involved are Julia–Lythgoe's Wittig–Horner coupling of aldehyde functionality of CD-ring system with benzothiazolyl sulfone, and Horner–Wadsworth–Emmons reaction of phosphine oxide with a Windaus–Grundmann's ketone to build a diene motif between the A and CD-ring system of Paricalcitol.


## Introduction

The natural hormones 1α,25-dihydroxyvitamin $D_3$ (**1**, Calcitriol), 1α,25-hydroxyvitamin $D_3$ (**2**, Alfacalcidol), 1α,25-hydroxyvitamin $D_2$ (**3**, Doxercalciferol) (Fig. 1), are members of steroid/thyroid/androgen nuclear receptor super-family, and act as endogenous ligands for the nuclear vitamin D receptor (VDR), and show significant biological activities.[1] Calcitriol (**1**) is well known as a primary regulator for calcium and phosphate homeostasis,[2,3] and plays a critical role in regulation of the proliferation of malignant cells.[4,5]

In recent times, the non-natural vitamin $D_2$ (**6**, Ergocalciferol) has been administrated to humans and domestic animals[6] in parallel to vitamin D to influence metabolism and biological activities. There is also a strong evidence on vitamin $D_2$ undergoing double hydroxylation[7,8] to produce 1α,25-dihydroxyvitamin $D_2$.

Further, most of analogs examined so far belong to natural vitamin $D_3$ series. In comparison to vitamin $D_3$ derivatives, vitamin $D_2$ derivatives are challenging to synthesize due to an additional chiral center at C-24 and a double bond (*E*-geometry) at C-22. Among these Paricalcitol (**4**) is being used to treat secondary hyperparathyroidism (SHPT) in patients with chronic kidney disease (CKD).[9] Several synthesis routes have been published for the synthesis of Paricalcitol.[10]

However, these synthetic approaches focus on the synthesis of the *E*-configuration at C-22. For verification of analytical methods for Paricalcitol it is necessary to provide reference material of the corresponding *Z*-isomer. This requirement will become even more important with the expected implementation of the new USP method specifying this compound specifically.[11]

Various structural modifications (Fig. 1) explored on 1α,25-dihydroxy 19-nor vitamin D are in side chain viz. reduced double bond or substituents around double bond, surprisingly researchers failed to evaluate the potential of isomerization at C-22, 23 position. Herein, we report the first total synthesis of *Z*-stereoisomer of Paricalcitol at C-22, 23 by maneuvering the coupling of A-ring system with a Windaus–Grundmann's keto-acetal under Wittig–Horner condition,[12–14] and subsequently Julia–Lythgoe[15] olefination on ACD-ring system. (Strategy II, Fig. 2.)

## Results and discussion

As a part of our quest on the stereoselective synthesis of vitamin $D_2$ metabolites, we approached our scientific efforts in two ways: (i) synthesis of either precursor **7** (Strategy I) or **12** (Strategy II) (Fig. 2) and (ii) preparation of benzothiazolyl sulfone **8**, a key intermediate for bringing side chain under Julia–Lythgoe olefination condition.

Our studies began with the synthesis of intermediate **13**[16] from D-(−)-Quinic acid,[17] and its conversion to phosphine oxide **11** in 10

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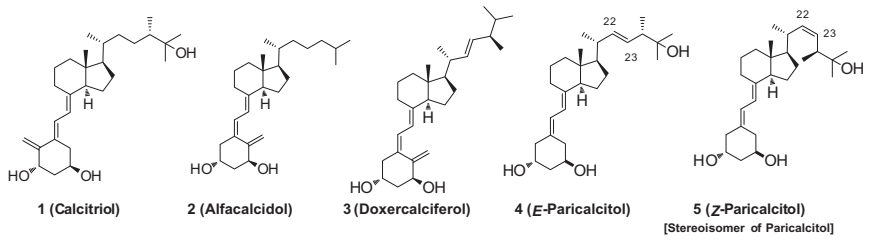



**Figure 1.** Structures of vitamin D receptor analogs.

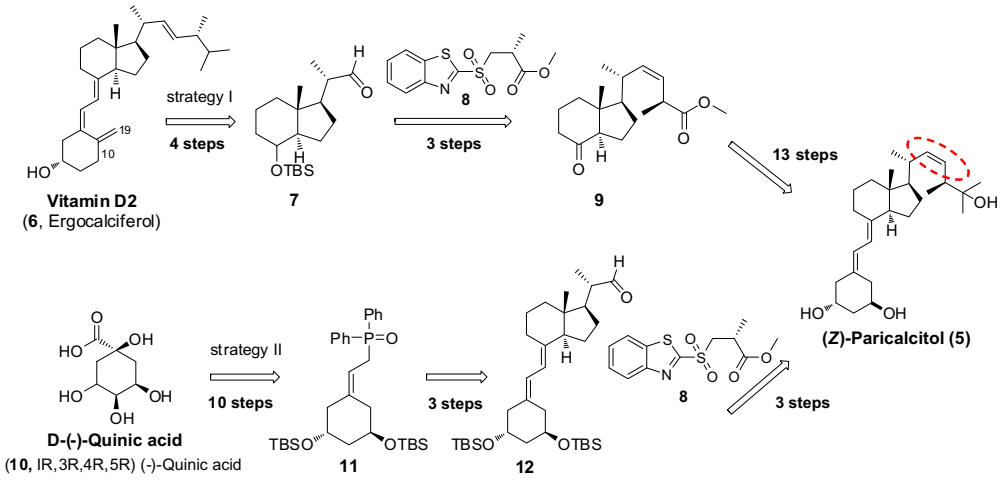
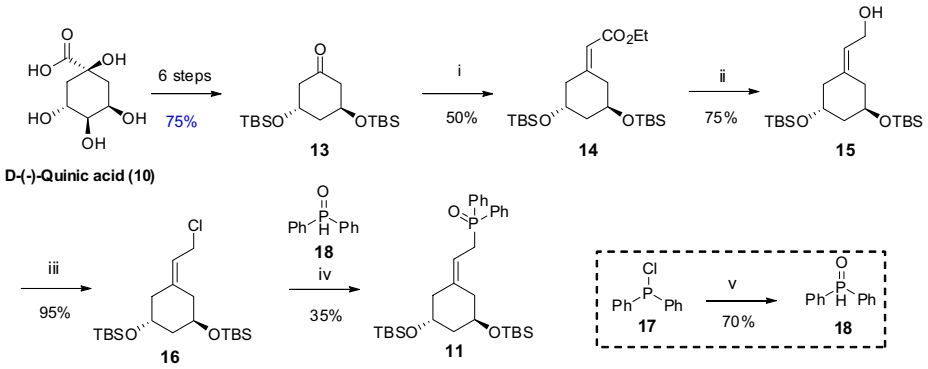



**Figure 2.** Retro synthetic approach of (*E*)-Paricalcitol and its stereoisomer (*Z*)-Paricalcitol.

steps (Scheme 1). The preparation of **11** was critical for the success of this method, as this will participate in constructing A-ring system[18] in Paricalcitol (Fig. 2; Strategy II). Various attempts were made to convert **15** to **11**, either through the preparation of tosylate of **15** under standard condition[19] or chloride **16** with NCS as reported by Uskokovic group,[20] and subsequently their displacement with lithium phenylphosphine. The preparation of **16** was challenging in the above condition, thereof, we shifted our attention to triphosgene (in hexane) condition.[21] Intermediate **16** was displaced with diphenyl phosphine oxide to get **11**. The main advantage of this reaction condition was the preparation of air stable diphenylphosphine oxide from inexpensive chlorodiphenylphosphine **17** and its usage. Another crucial intermediate **7** was prepared from Ergocalciferol (**6**) in multiple steps (Scheme 2) as per literature report.[22]

Julia–Lythgoe olefination of **7** and sulfone **8** in the presence of NaHMDS/or LiHMDS in THF (−78 °C, 30 min) afforded prominently *trans*-olefin **24** (*E*/*Z*-isomer ratio 82:18) in 59% yield. We further explored the potential of other reagents like methyl 2-[bis(2,2,2-trifluoroethoxy)phosphoryl]acetate and diphenoxy phosphonate to obtain the desired result but were unsuccessful. The intermediate **24** was taken forward to synthesize Paricalcitol, thinking to get separation of these isomers at some stage. However, our efforts were not encouraging and isomers (*E*- and *Z*-isomer) were separated only by preparative HPLC purification.

Unpromising results on first strategy (Scheme 2) impelled us to look for alternative plan to achieve the desired results. The critical intermediate **22** of CD-ring system was conveniently prepared by ozonolytic cleavage of vitamin $D_2$ (**6**) in methanol, using $CHCl_3$ as co-solvent (Scheme 3). The residual acid in $CHCl_3$ was sufficient




**Scheme 1.** Reagents and conditions: (i) $(TMS)CH_2CO_2Et$ (1.5 equiv), LiHMDS (1 M in THF, 1.5 equiv), THF, 0 °C–rt, 15 min; then cooled to −78 °C, add **13**, −78 °C, 3 h; (ii) DIBAL-H (2.5 equiv); toluene, −78 °C to rt, 3 h; (iii) Triphosgene (0.5 equiv), pyridine (2 equiv), hexane, 0 °C–rt, 30 min; (iv) **18** (1 equiv), NaH (1 equiv), DMF, 0 °C–rt, 30 min; then cooled to −60 °C, add **16** (1 equiv), −60 °C to 1 h; rt to 1 h; (v) 1 N aq HCl (2 vol), rt, 18 h.

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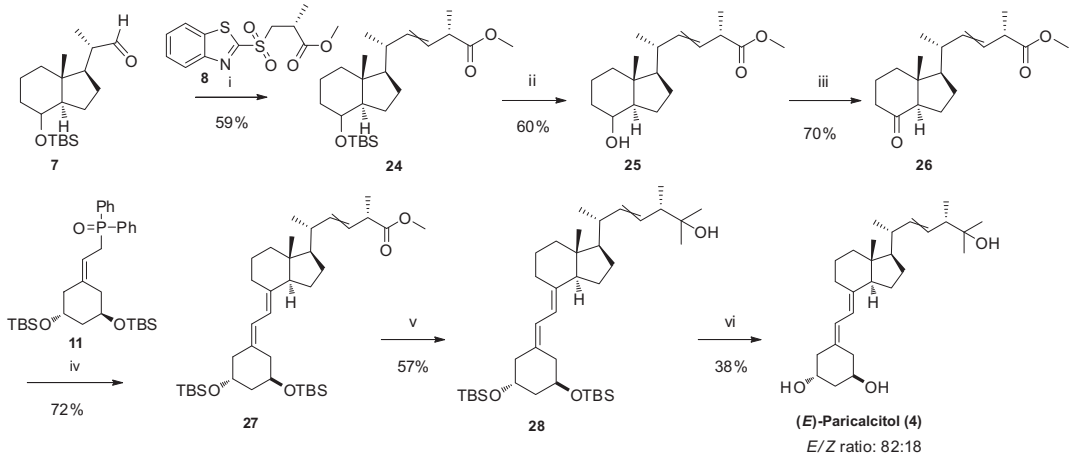
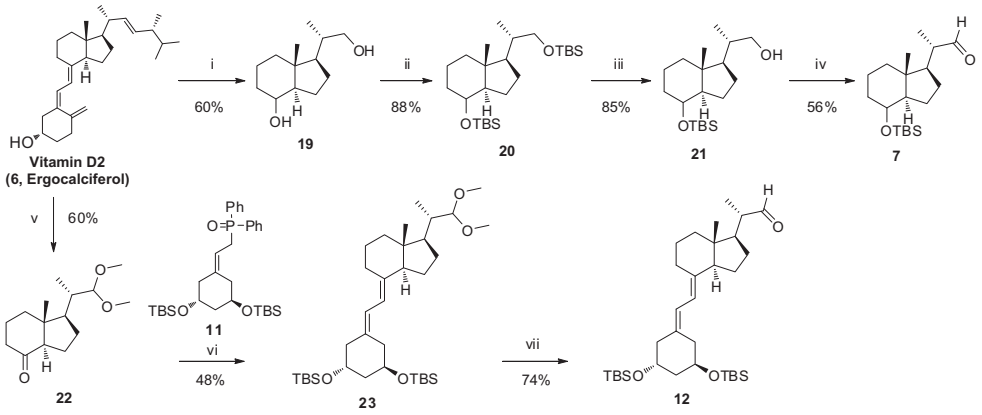
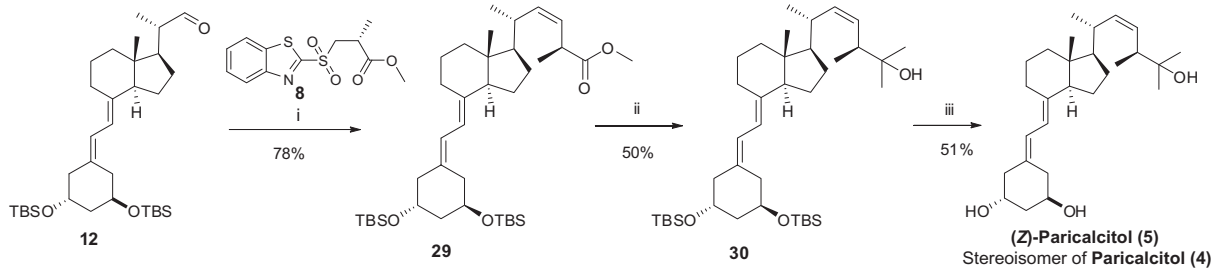



**Scheme 2.** Reagents and conditions: (i) **8** (1.5 equiv), LiHMDS (1 M in THF; 1.5 equiv), THF, −78 °C to 15 min; then add **7** (1 equiv), −78 °C to rt, 5 h; (ii) TBAF (1 M in THF), THF, 80 °C, 24 h; (iii) PDC (2.5 equiv), 4 A MS, $CH_2Cl_2$; 0 °C, 4 h; (iv) **11** (1 equiv), NaHMDS (1 M in THF, 1.1 equiv), THF, −78 °C, 10 min; then add **26** (1 equiv), −78 °C, 1 h; (v) MeMgBr (3 M in $Et_2O$, 5 equiv), THF, 0 °C, 4 h; (vi) $NH_4F$ (5 equiv), MeOH, reflux, 5 h.




**Scheme 3.** Reagents and conditions: (i) (a) $O_3$, $NaHCO_3$, MeOH, 8 h; (b) $NaBH_4$, rt, 18 h; (ii) TBSCl (3 equiv), Imidazole (4 equiv), DMAP (cat), DMF, rt, 18 h; (iii) TBAF (1 M in THF), THF, 0 °C–rt, 18 h; (iv) PCC (2.2 equiv), Celite, $CH_2Cl_2$, 0 °C–rt, 18 h; (v) (a) $O_3$, pyridine, MeOH, −78 °C, 8 h; $Me_2S$ (6 equiv), −78 °C to 1 h; rt to 1 h; (vi) **11** (1 equiv), NaHMDS (1 M in THF), THF, −78 °C to 5 min, then add **22** (1 equiv), −78 °C to 15 min; (vii) $CHCl_3/H_2O$/TFA (4:2:1), 0 °C–rt, 1 h.




**Scheme 4.** Reagents and conditions: (i) **8** (1.1 equiv), LiHMDS (1 M in THF), THF, −78 °C to 15 min; then add **12** (1 equiv), −78 °C, 15 min; (ii) MeMgBr (3 M in $Et_2O$, 3 equiv), THF, 0 °C, 4 h; (iii) TBAF (1 M in THF, 5 equiv), THF, 60 °C, 2 h.

to catalyze acetalization of the aldehyde functionality to provide keto-acetal **22** during a reductive workup with dimethyl sulfide in 60% yield. Wittig–Horner coupling of keto-acetal **22** with phosphine oxide **11** (ring A synthon) afforded the diene keto-acetal **23**, which on deprotection provided aldehyde **12**.[23] However, Julia–Lythgoe olefination on **12** with sulfone **8** in the presence of LiHMDS in THF (−78 °C, 30 min) gave ester **29** in good yield (78%) with desired stereoselectivity, a *Z*-isomer. Grignard reaction on ester **29** with MeMgBr (3 M in $Et_2O$) in ether (0 °C, 4 h), followed by removal of silyl protecting group on **30** gave target *Z*-isomer of Paricalcitol **5** in 51% yield (Scheme 4). The overall yield of this sixteen step synthesis is 0.1% starting from Quinic acid (**10**).

The characteristic difference in *E* and *Z*-isomer of Paricalcitol was in chemical shift of protons on C-22 and C-23 atoms. These protons were observed as multiple peaks in the region 5.27–5.40 for *E*-isomer, however, in case of *Z*-isomers these protons were well distinguished at 5.162–5.216 (m, 1H), 5.33–5.33 (m, 1H) respectively by $^1H$ NMR (Fig. 3). The other protons are in

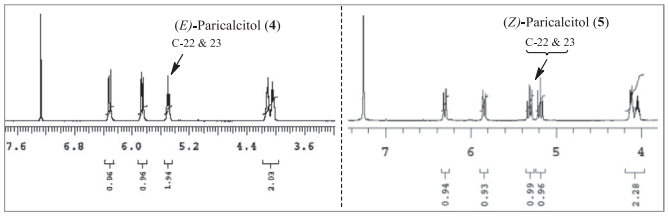



**Figure 3.** Characteristic protons of (*E*)-Paricalcitol (**4**) and its stereoisomer (*Z*)-Paricalcitol (**5**) in $CDCl_3$ solvent (400 MHz).

agreement with reported value[24] for *E*-isomer. These isomers are separable on HPLC.

The solution form of *Z*-isomer of Paricalcitol **5** was found to be stable under the thermal condition (60–180 °C) in various solvents (viz. EtOH, acetonitrile, Dowtherm).

## Conclusion

In conclusion, we have succeeded in developing an efficient synthetic approach for the synthesis of *Z*-isomer of Paricalcitol from Quinic acid under Julia–Lythgoe's olefination condition. The key intermediates in this synthesis effort were phosphine oxide **11** and benzothiazolyl sulfone **8**.

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## Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.tetlet.2016.01.110.

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