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EFFICIENT SYNTHESIS OF 11α,25-DIHYDROXYVITAMIN D_3 AND D_2 -TBDMS ETHER

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Introduction

In recent years it has become apparent, that vitamin D deficiency is connected to a wide variety of disorders, beyond just causing bone diseases [1]. Consequently, the development of appropriate assays to measure the status of medicinally relevant vitamin D metabolites in human blood, serum or appropriate tissue is continuously being improved [2].

Most common, and a rapid way, for assessment of vitamin D deficiency is the measurement of 25-hydroxyvitamin D_3 (25(OH)VitD_3) concentration in blood or serum by immunological assays, using specific antibodies in a competitive binding assay. As a major drawback, specific vitamin D antibodies are challenging to develop. Commonly, an appropriate conjugate of a particular vitamin D metabolite is employed, that is linked at its C3-OH-position of the A-ring to biotin or bovine serum albumin (BSA). Unfortunately, using these conjugates, polyclonal antibodies (AB_3) lack sufficient specificity, particularly to distinguish between metabolites bearing different substituents at the A-ring, such as 25(OH)VitD_3 and 1α,25$(OH)_2$VitD_3 (calcitriol). To overcome this problem, 11α,25-dihydroxyvitamin D_3 conjugates can be used instead [3], in turn to be synthesized from a C3-TBDMS ether, that is suitable to be connected at its C11-OH-position via an appropriate linker to biotin or BSA. Furthermore, the corresponding vitamin D_2 analogs have to be considered as well, because they also appear in human blood and tissue and exhibit significant activity.

We describe a new efficient synthesis of 11α,25-dihydroxyvitamin D_3 C3-TBDMS ether (**1**) and its corresponding D_2 analog (**2**) in 500 mg scale, each starting from readily available vitamin D_2.

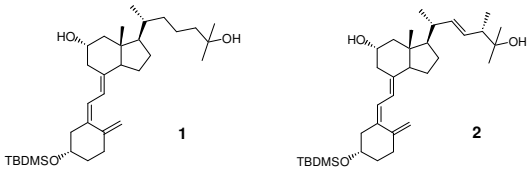
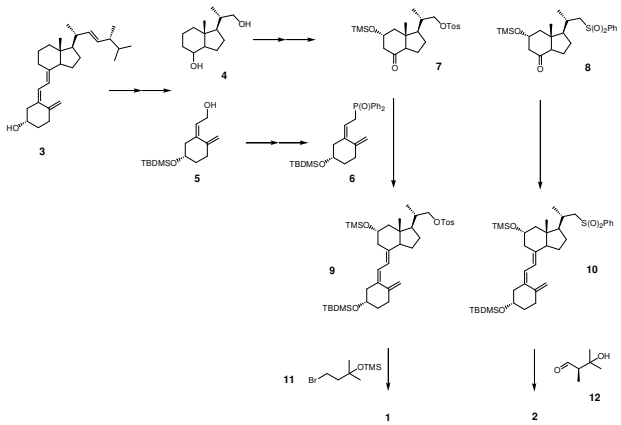


Figure 1: 11α,25-dihydroxyvitamin D_3 and –D_2-TBDMS ether

Methods

Readily available Vitamin D_2 **3** is converted to Inhoffen-Lythgoe diol **4** and allylic alcohol **5**, in turn to be converted in diphenylphosphine oxide **6**, that is connected via a Wittig-Horner reaction [4] with accordingly C11-hydroxylated [5] tosylate **7** and sulfone **8**. The side chains are finally installed by connecting the resulting tosylate **9** with bromide **11**, and sulfone **10** with aldehyde **12** [6], respectively. Desilylation finally leads to **1** and **2**.



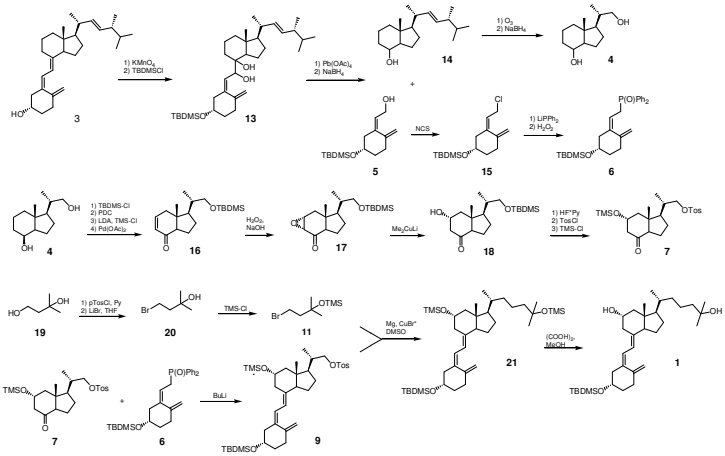
Scheme 1. Strategy to synthesize 1 and 2

Results and Discussion

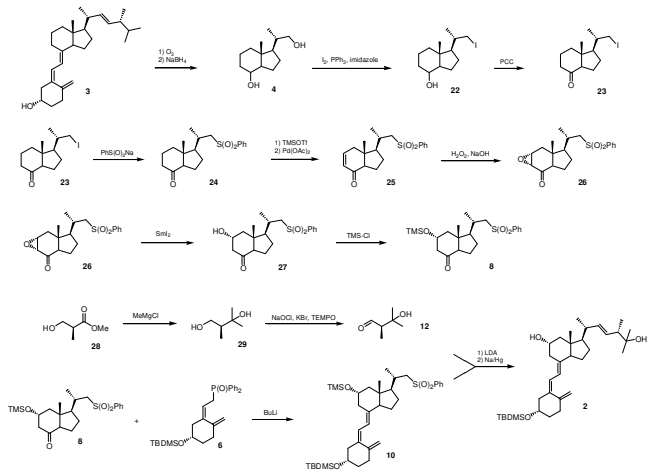
Inhoffen-Lythgoe diol **4** and diphenylphosphine oxide **6** were prepared by application of established standard procedures [4]. For the synthesis of 11α,25-dihydroxyvitamin D_3 (scheme 2), CD ring ketone **7** bearing a C11-OH group is synthesized by a procedure described by Takahashi et al. [5]. Bromide **11** is synthesized from commercially available 3-methyl-1,3-butandiol **19**. Due to their different susceptibility against oxalic acid, the TMS groups could be removed in the presence of the TBDMS group, leading to **1**.

The synthesis of 11α,25-dihydroxyvitamin D_2 C3-TBDMS ether (**2**) is by far more challenging comparing to the synthesis of its D_3 counterpart (scheme 3). A methodology described by Kutner et al. [6] described for the synthesis of 1,25-$(OH)_2$-VitD_2 could finally be applied successfully. Key step is the introduction of the side chain via Julia olefination of aldehyde **12** and phenyl sulfone **10**. CD ring ketone **8** bearing a C11-OH group was again synthesized by a procedure described by Takahashi et al. [5]. The synthesis of enone **25** was carried out starting from **4** in two steps by treatment with trimethylsilyl triflate and subsequent oxidation by palladium (II) acetate. A following epoxidation with hydrogen peroxide led to the corresponding epoxide **26**. Surprisingly, a planned reductive ring opening with Me_2CuLi as successfully applied previously to get **18** in course of the synthesis of **1** failed.

However, epoxide opening of **26** with samarium (II) iodide [7] led to the desired 3-hydroxy ketone **27**, that turned out to be very sensitive against epimerization. Subsequently, the OH group was protected as its TMS ether and the vitamin D skeleton was build up by coupling sulfone **8** with phosphine oxide **6**. In the following step the side chain was connected via a Julia olefination of **10** with aldehyde **12**, in turn synthesized in two steps from commercially available ester **28**. Grignard reaction with methylmagnesium bromide leading to diol **29**, that was oxidized to get **12**. Surpisingly, oxidation with Dess Martin periiodinan, how it is described in the literature, failed. By contrast TEMPO oxidation with sodium hypochlorite as a co-oxidazing agent was successful. In course of the Julia olefination of sulfone **10** with aldehyde **12** the TMS protective group turned out to be unstable, so that **2** was directly formed in the same reaction step.



Scheme 2. Synthesis of 11α,25-dihydroxyvitamin D_3



Scheme 3. Synthesis of 11α,25-dihydroxyvitamin D_2

Conclusion

11α,25-dihydroxyvitamin D_3 (**1**) and 11α,25-dihydroxyvitamin D_2 (**2**) could be prepared in a 500 mg scale to explore their potential for the development of new conjugates to discover specific antibodies for advanced immunological assays and other applications.

References

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