SYNTHESIS OF 26-METHYL CHOLESTANE AND IDENTIFICATION OF CRYOSTANE IN MID-NEOPROTEROZOIC SEDIMENTS

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Introduction

The biomarker distributions of mid-Neoproterozoic sediments (800 and 717 million years) from various locations are characterized by the occurrence, besides cholestane isomers, of a novel series of C_{28} steranes whereas classical C-24 alkylated steranes (i.e. ergostanes and stigmastanes) are absent. The name "cryostane" was ascribed to these unusual C_{28} steranes which seem to be restricted to pre-Snowball Earth sediments. They possibly attest of the particular evolutionary state of the eukaryotes from this period (Brocks et al, 2016). Cryostanes, which have a longer retention time in gas chromatography than ergostanes, have been postulated to correspond to sterane isomers having a C-26 methylated side-chain (Brocks et al, 2016). We report here the conclusive identification of cryostanes as 26-methyl cholestanes (Fig. 1) based on comparison of chromatographic behavior and mass spectrum of the last eluting cryostane isomer with those of a 26-methyl 5α(H),14α(H),17α(H),20*R*-cholestane **1** standard obtained by synthesis.

Results

26-Methyl ααα*R*-cholestane **1** has been synthesized starting from 26-hydroxy ααα*R*-cholestane **2**, a compound previously prepared starting from stigmasterol (Paulus, 1993). The synthetic pathway leading to 26-methyl αααR-cholestane **1** is described in Fig. 1A and comprises three steps: (1) the oxidation of alcohol **2** into its related aldehyde using pyridinium dichromate, (2) the addition of one carbon atom leading to alkene **3** by a rhodium-catalyzed methylenation step following the procedure described by Lebel and Paquet (2004) and involving triphenylphosphine (PPh_3), Wilkinson catalyst, and trimethylsilyldiazomethane ($TMSCHN_2$) as the carbon source, and finally (3) PtO_2 hydrogenation of **3** yielding 26-methyl ααα*R*-cholestane **1**. The identification of the geochemical cryostanes as 26-methyl cholestane isomers is supported by the fact that the mass spectrum (electron impact) of 26-methyl ααα*R*-cholestane **1** is identical to that of the last eluting cryostane isomer and coelutes with it on two different gas chromatography columns (Fig. 1B). Given the similarity with "classical" geological sterane distributions, the early eluting geochemical isomers (Fig. 1B) most likely correspond to, respectively, ααα*S*, αββ*R* and αββ*S* isomers of 26-methyl cholestane. In this respect, it is noteworthy that the first eluting cryostane isomer coelutes with a minor synthetic 20*S* isomer of **1**.

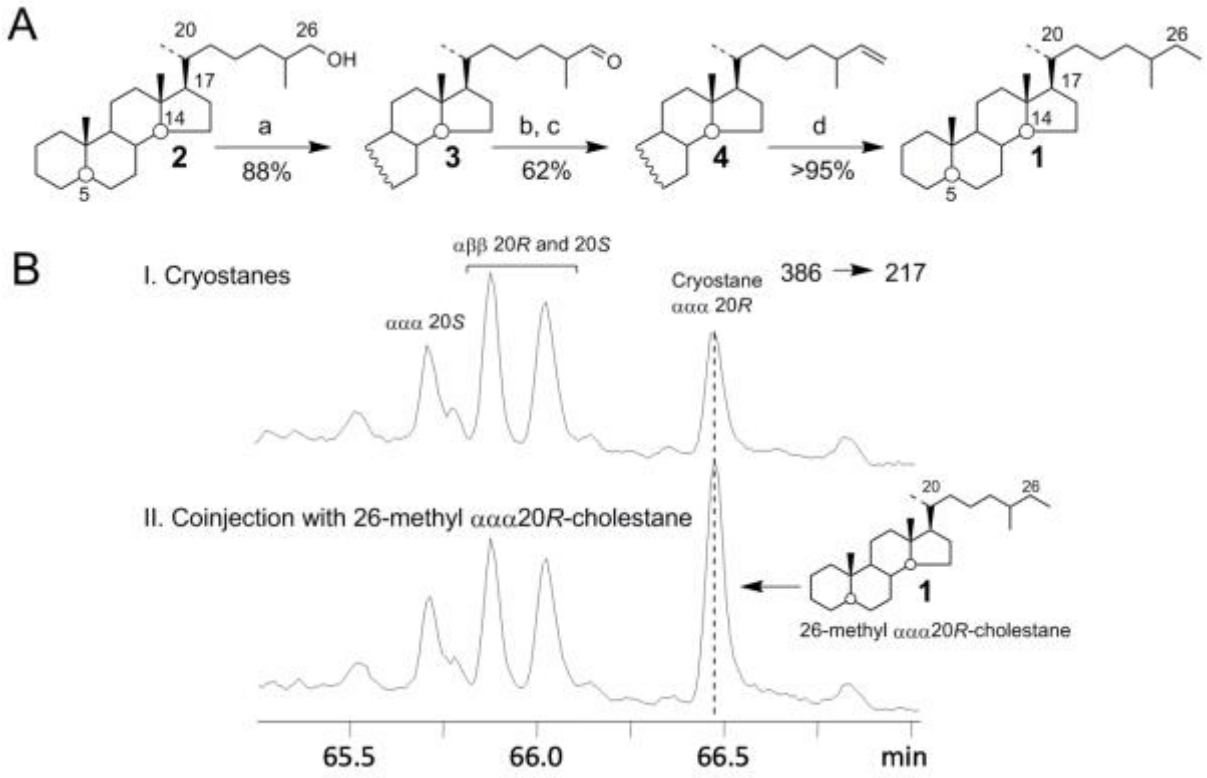

Figure 1 *(A) Synthesis of 26-methyl cholestane* ***1*** *from 26-hydroxy cholestane* ***2*** *(a) Pyridinium dichromate, CH_2Cl_2, (b) iPrOH, PPh_3, Wilkinson catalyst, tetrahydrofuran (THF), (c) $TMSCHN_2$, THF, (d) H_2, PtO_2, EtOAc; (B) MRM chromatograms of 386 → 217 precursor-product transitions showing a co-injection experiment on a DB-5MS chromatographic column for the identification of cryostane. (**I**) Chuar Group sample 10J093 (outcrop, 1544 meters above base of Chuar Group); (**II**) co-injection of the above samples.*

Biological sterols with the cryostane side-chain are not known. Sterols methylated at C-26 and bearing additional alkylations at other positions (e.g. C-24) have however been identified in living organisms, and seem to be restricted to some sponges (Giner, 1993). This leads us to propose sponges as potential precursor organisms for 26-methyl cholestanes. Other possible sources, such as protists, have also been proposed (Brocks et al., 2016).

References

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