Organic LETTERS

Letter

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Total Synthesis and Structural Revision of Mycalol, an Anticancer Natural Product from the Marine Source

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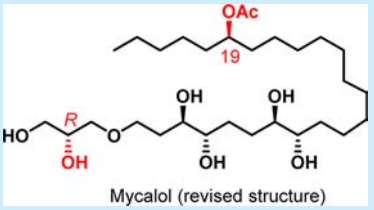
Supporting Information

ABSTRACT: The total synthesis of an anticancer (anaplastic thyroid) natural lipid mycalol has been accomplished for the first time. Synthesis of originally proposed structure necessitated the revision of structure in which the position of acetate group moved from C20 to C19 and a change in stereochemistry of the glycerol unit.



Anaplastic thyroid carcinoma (ATC) is a rare and invasive cancer of the thyroid gland with extremely difficult diagnosis and poor treatment options.[1] Although ATC accounts for a small percentage (~2%) of all cancer patients, it is considered a very serious one. For example, it accounts for more than 50% of thyroid carcinoma-related deaths in the United States.[2] ATC progresses very rapidly with a high degree of invasiveness and early metastasis, which leads to survival of patients for only 2−6 months and rarely one year.[3] Recommended medication as treatment for this cancer is a combination of doxorubicin and cisplatin along with radiotherapy.

Nature has been a great source for several drugs which are on the market for various cancers.[4] Along these lines, mycalol (**1**), a novel polyoxygenated ether lipid isolated from a marine sponge by Fontana and co-workers, attracted our attention owing to its promising specific activity against different cell lines derived from ATC.[5] It is an acyclic linear structure with six chiral centers, of which the C20 center was completely isolated from the rest. Although the structure looks deceptively simple, its flexible nature with multiple chiral centers makes this molecule a very challenging one. Much painstaking effort (by a combination of chemical and chiroptical methods) from Fontana's group resulted in a molecular structure as drawn in Figure 1.[5] Because of its important biological activity and interesting structural features, we have initiated a program on this scaffold. Our goals in this project are (i) total synthesis of mycalol to access material for further biological profiling and (ii) confirmation of assigned structure including relative and absolute stereochemistry. Our efforts and findings from this program are described here.

The key disconnections and sources for chiral centers present in mycalol (**1**) are shown in Figure 1. Our plan was to utilize readily accessible starting materials to construct two key fragments, **A** and **B**, and then join them using a cross-metathesis reaction. Our synthesis commenced with a known allyl ether **2**[6] prepared from (*R*)-solketal, which was subjected to hydroboration[7] to give alcohol **3**. The aldehyde obtained from PDC oxidation reaction of **3** was immediately treated with vinylmagnesium bromide to give the diastereomeric mixture of allylic alcohols **4** (1:1). Using the well-established protocols[8] of the Sharpless enantioselective epoxidation [(−)-DIPT, titanium isopropoxide and *tert*-butyl hydroperoxide], the diastereomeric alcohols were resolved and the desired chiral epoxide **5** was obtained. Epoxide **5** was converted to compound **6** (fragment **A**) using trimethylsulfonium ylide[9] followed by protection of the resulting diol (Scheme 1). The absolute configuration of alcohol in **5** was independently further confirmed by synthesizing compound **6** using a chiral pool approach (unpublished results; details are provided in the Supporting Information).

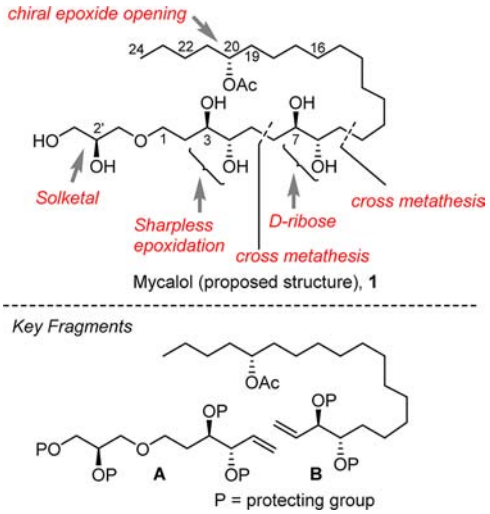


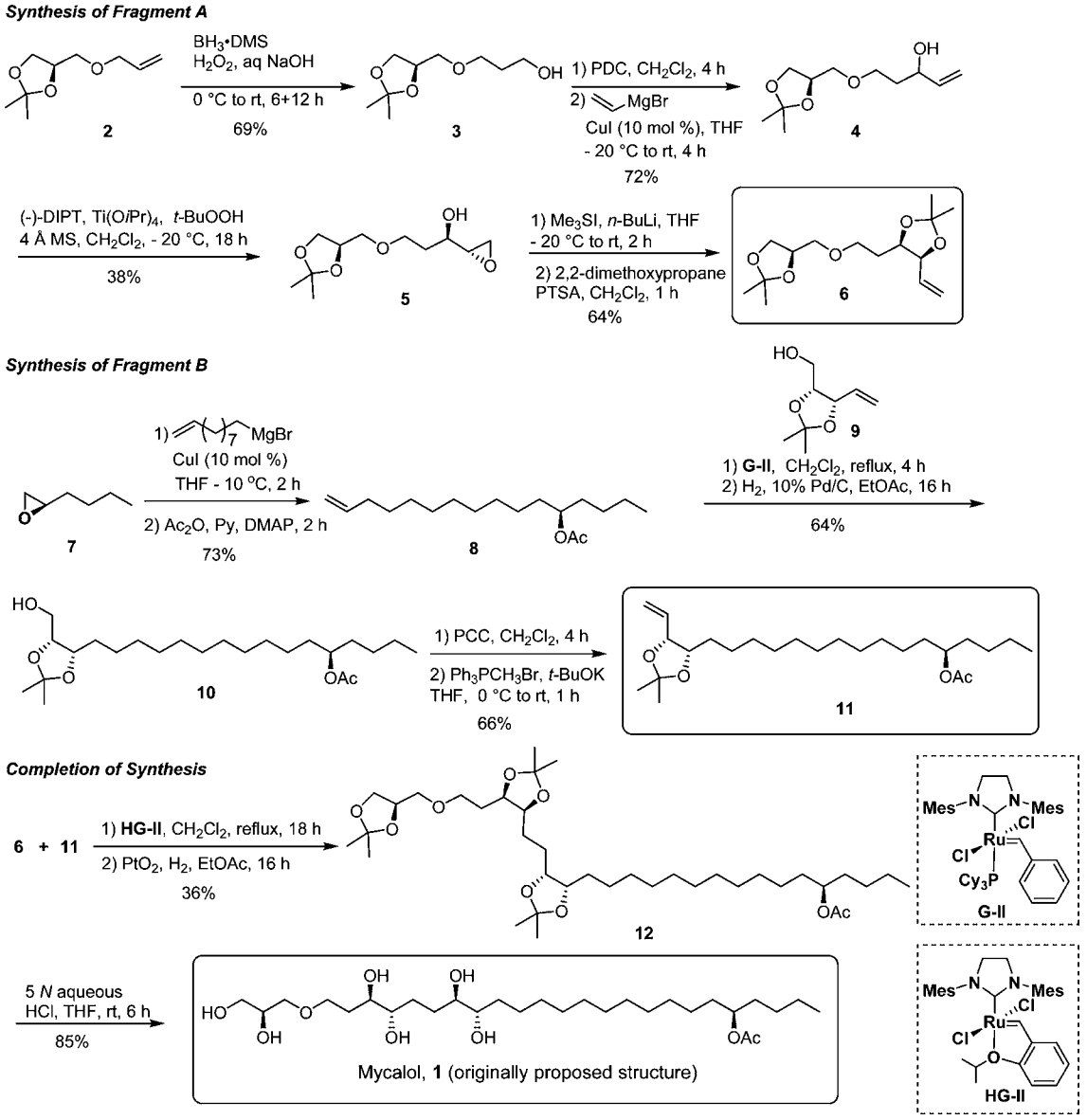
Figure 1. Planned strategies and key fragments to access mycalol.

To access fragment **B**, readily available (*R*)-epoxide **7**[10] was regioselectively opened with the C10-alkene Grignard reagent,[11] and it was immediately protected to give acetate **8**. The known

Received: February 3, 2015
Published: March 12, 2015

DOI: 10.1021/acs.orglett.5b00345
Org. Lett. 2015, 17, 1652−1655

Scheme 1. Synthesis of Key Fragments and Total Synthesis of Mycalol (Proposed Structure)



alcohol **9**[12] prepared from D-ribose was subjected to a cross-metathesis reaction[13] with the acetate **8** followed by hydrogenation resulting in compound **10**. Oxidation and one-carbon Wittig reaction in **10** gave 20-carbon unit **11**, which is nothing but a fragment **B**. Having both key fragments in hand, the next task was to join them together through another cross-metathesis reaction. For this purpose, a few trials were made, and finally, we were pleased to find that Hoveyda–Grubbs catalyst (**HG-II**)[14] gave the desired result. Compounds **6** and **11** underwent a cross-metathesis reaction to produce the olefin which was hydrogenated (PtO_2, H_2) to furnish the proposed structure of mycalol in protected form. Finally, deprotection of all three acetonide groups using aqueous HCl (5 N) resulted in **1**.

Comparison of NMR spectra of both the synthesized compound **12** and the protected form of mycalol[5] showed subtle variations. In particular, ^{13}C NMR spectra showed minor differences in the region of hydrocarbons, and the most noticeable one was the absence of a peak at ∼32 ppm in the spectrum of **12**. Various high-field 2D-NMR analyses of compound **12** suggested that there could be a problem with the position of acetate group on the lipid chain. We observed that the terminal methyl group protons of **12** (0.85 ppm) showed HMBC cross-peaks (Figure 2a) to carbons at 22.9 (C23) and 27.9 (C22), whereas the reported NMR data for the triacetonide of mycalol suggested correlations to carbons at 22.9 and 32 ppm. In addition, the C22 carbon at 27.9 ppm in compound **12** also showed expected HMBC correlation to the proton attached to the carbon bearing the acetoxy group. The observed shielding of C22, compared to a paraffinic methylene, is also in agreement with the γ-substituent effect of an acetoxy group.[15] Hence, the reported chemical shift of ∼32 ppm for C22 suggested that the acetoxy substituent is likely to be further away from the terminus in the natural product.

Further careful analysis of ^{13}C NMR data revealed that there is some additional difference with respect to two peaks at 75 ppm (correspond to C2′ and C3) in compound **12** which can be attributed to the glycerol configuration (see Figure 2b). To support this hypothesis, we have synthesized triacetonide **15** to compare NMR spectra in which glycerol stereochemistry was reversed. For this purpose, compound **14** (fragment **A**) was

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Org. Lett. 2015, 17, 1652–1655

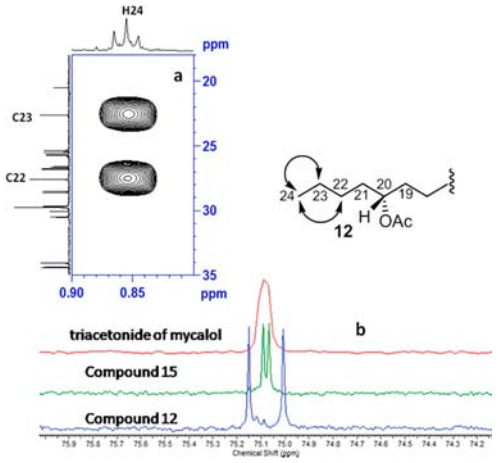
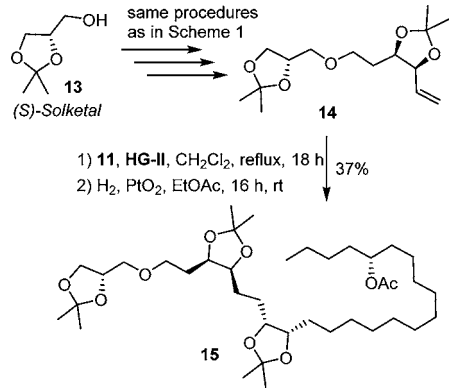


Figure 2. Key observations in NMR spectra: (a) $^{1}H-^{13}C$ HMBC correlations for compound **12**. (b) ^{13}C NMR differences in the region at ~75 ppm of compounds **12**, **15**, and triacetonide of mycalol.

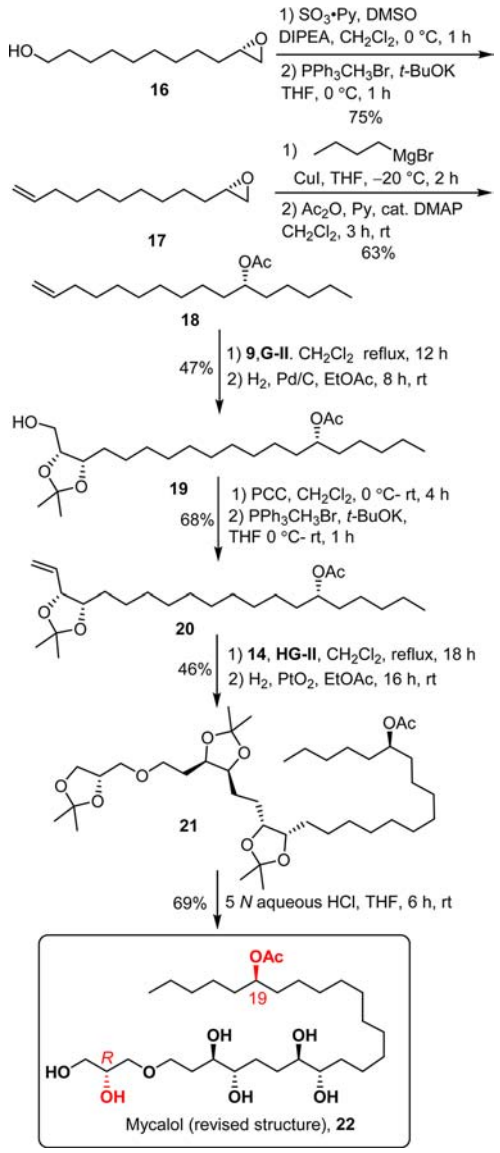
prepared by following the same procedures described for the synthesis of compound **6** except that (*R*)-solketal was replaced with (*S*)-solketal (Scheme 2). The exact scheme and complete

Scheme 2. Synthesis of Triacetonide 15 with (*S*)-Configuration at C2′



experimental details are provided in the Supporting Information. Compounds **14** and **11** were then subjected to a cross-metathesis reaction followed by hydrogenation to result in compound **15**. The ^{13}C NMR spectra (Figure 2b) of three compounds, **12**, **15**, and triacetonide of mycalol, were compared, and it was found that compound **15** has a better match. Based on these findings, we put forward the likely structure of mycalol with two changes: (a) position of the acetate moiety on C19 (instead of C20) and (b) configuration of glycerol moiety (*R*-configuration instead of *S*- configuration). With this structure in mind, we set out for a synthesis to prove our hypothesis. A synthesis of the newly proposed mycalol was planned broadly in a manner similar to that of **1**. Synthesis of fragment **B** began with an epoxy alcohol **16**,[16] which on Parikh–Doering oxidation[17] followed by one-carbon Wittig olefination resulted in an epoxy alkene **17** in good yields. The terminal epoxide present in **17** was regioselectively opened with *n*-butyl Grignard reagent in the presence of CuI to furnish the alcohol which was immediately acetylated to give compound **18** with desired length of carbon chain. The alcohol **9** and acetate **18** were subjected to a cross-metathesis reaction followed by hydrogenation to give the desired compound **19**. Oxidation of the alcohol and one-carbon Wittig reaction resulted in 20-carbon unit **20** (fragment **B** in the present case). Compounds **14** and **20** underwent a cross-metathesis reaction under similar conditions (as delineated in Scheme 1) to produce the desired olefin, which was again hydrogenated (PtO_2, H_2) to furnish the revised structure of mycalol **21** in protected form (Scheme 3). We were delighted to find that it was an exact match

Scheme 3. Total Synthesis of Mycalol with Revised Structure



when we compared the NMR spectral data of synthesized compound **21** and the triacetonide of mycalol (reported from Fontana's group[5]).[18,19] In particular, ^{13}C NMR spectra were found to be very clear and identical. The ^{13}C NMR spectrum of **21**, indeed, showed the presence of a methylene carbon at 32 ppm which also exhibited HMBC correlation to the terminal methyl protons and thus confirming its position at C22 (Figure 3). Finally, deprotection of all three acetonide groups in **21** furnished the mycalol **22** with the revised structure. All the spectral data (^{1}H, ^{13}C, HRMS) of synthesized mycalol were compared with that of the isolated natural product mycalol and found to be identical.[5,19] Optical rotation of synthetic mycalol was measured at $[\alpha]_D^{24}$ +2.69 (*c* 0.16, MeOH), and the reported value was at $[\alpha]_D^{20}$ +3.45 (*c* 0.10, MeOH).[5]

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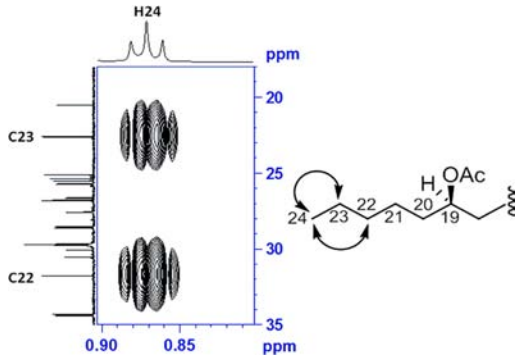

Figure 3. $^{1}H-^{13}C$ HMBC correlations for compound **21**.

Thus, we have achieved the first total synthesis of putative and actual natural product mycalol. The total synthesis necessitated the revision of the originally proposed structure. Based on careful analysis of high-field 2D NMR spectra, we have proposed the new structure to mycalol with two changes in the originally proposed structure by Fontana's group.[20] Further optimization of the present route and generation of a focused library of molecules around the mycalol skeleton, in particular, synthesis of various stereoisomers and their biological evaluation will be investigated in due course.

■ ASSOCIATED CONTENT

Supporting Information

Experimental details, data comparison tables of synthesized vs natural product, and copies of NMR spectra including 2D NMR analysis. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

We acknowledge the CSIR, New Delhi, for support through XII Five Year Plan (CSC0108: ORIGIN, BSC0123: GenCODE and CSC0405: NMRCAR). We thank Prof. Angelo Fontana (Istituto di Chimica Biomolecolare, Consiglio Nazionale delle Ricerche, Italy) for his timely help by providing FIDs which helped us in detailed comparative analysis of NMR data. We thank Dr. Madhuri V. Shinde, Dr. Sibanarayan Tripathy, and Mr. Pankaj Khairnar of CSIR-NCL for their help with some experiments. B.S. thanks CSIR for the award of fellowship.

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(19) See the Supporting Information for comparison tables of NMR data.

(20) Although the CD spectrum of the 1,2-dibenzoyl derivative of natural mycalol was comparable to those reported for (2*S*)-glycerol derivative by Uzawa et al., the present revision of the stereochemistry at C2′ of mycalol is compatible with the experimental data. Uzawa, H.; Nishida, Y.; Ohrui, H.; Meguro, H. *J. Org. Chem.* **1990**, *55*, 116.

DOI: 10.1021/acs.orglett.5b00345
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