Available online at www.sciencedirect.com
SCIENCE DIRECT
Tetrahedron
Tetrahedron 61 (2005) 927–939

RuO_4-catalyzed oxidative polycyclization of squalene. Determination of the configuration of the penta-tetrahydrofuranyl diol product

Teresa Caserta,[a] Vincenzo Piccialli,[a,*] Luigi Gomez-Paloma[b] and Giuseppe Bifulco[b,*]

[a]*Dipartimento di Chimica Organica e Biochimica, Università degli Studi di Napoli "Federico II", Via Cynthia 4, 80126, Napoli, Italy*
[b]*Dipartimento di Scienze Farmaceutiche, Università di Salerno, Via Ponte Don Melillo, 84084 Fisciano (Salerno), Italy*

Received 1 July 2004; revised 12 October 2004; accepted 5 November 2004

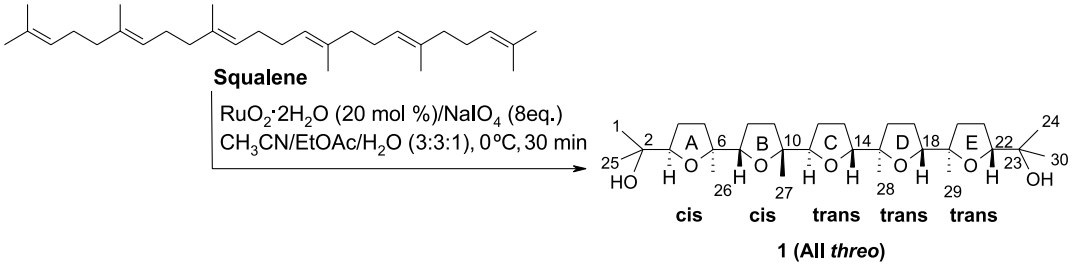
Available online 8 December 2004

Abstract—The configuration of the penta-tetrahydrofuranyl diol (penta-THF) product obtained by a single-step, RuO_4-catalyzed oxidative polycyclization of squalene, has been determined as *cis–threo–cis–threo–trans–threo–trans–threo–trans*. The *cis–cis–trans–trans–trans* sequence for the five contiguous THF rings has been established through extensive 2D-NMR spectroscopic studies carried out both on the intact molecule and on some of its derivatives, including the oxidative cleavage products obtained by degradation of the penta-THF with PCC/AcOH. Four different chemical approaches were devised to determine each of the four *threo* relationships within each carbon pair connecting adjacent THF rings in the molecule. To this aim, studies have been carried out either on some intermediates of the process leading to penta-THF, obtained by stopping the oxidation of squalene prior to completion, or on a degradation product of the penta-THF, obtained from the latter through a bidirectional double oxidative degradation with PCC.
© 2004 Elsevier Ltd. All rights reserved.

1. Introduction

2,5-Bonded polytetrahydrofurans (poly-THF) are key structural fragments in natural[1–3] and non-natural[4] products. They are embodied, for example, in the structure of bioactive squalene-derived natural products such as glabrescol,[1] longilene peroxide[2] and teurilene,[3] in polyether antibiotics such as monensin and ionomicin as well as in many *Annonaceous* acetogenins[5] a class of natural products with remarkable antitumoral and pesticide activity that has stimulated synthetic efforts from many research groups.[6]

Recently, we have discovered a novel, RuO_4-catalyzed, oxidative process that allows, in a single step, the stereoselective synthesis of adjacently linked poly-tetrahydrofuranyl diol products starting from isoprenoid polyenes.[7] Among the studied cases, the oxidation of squalene appears to be very intriguing since it furnishes the penta-THF **1** (Scheme 1; configuration as determined in this work), as a single diastereomer, in a remarkable 50% yield. This is the sole example, so far known, of a single-step *quintuple* oxidative polycyclization leading to a poly-THF product.

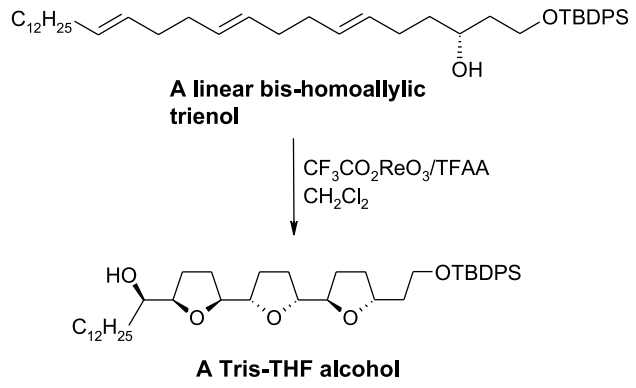


Scheme 1. Oxidative polycyclization of squalene with cat. RuO_4.

Keywords: RuO4; Squalene; Oxidative polycyclization; Penta-THF configuration; PCC; Oxidative cleavage.
* Corresponding authors. Tel.: +39 81 674111; fax: +39 81 674393 (V.P.); fax: +39 89 96282 (G.B.); e-mail addresses: vinpicci@unina.it; bifulco@unisa.it

0040–4020/$ - see front matter © 2004 Elsevier Ltd. All rights reserved.
doi:10.1016/j.tet.2004.11.018

The new process appears to be related to the polycyclization of bis-homoallylic hydroxypolyenes mediated by Re(VII) oxo species, mostly Re_2O_7 and $CF_3CO_2ReO_3$, that allows the assembly of up to three contiguous THF rings in a single step or in a sequential way. As an example, the synthesis of an advanced intermediate in the synthesis of 17,18-*bisepi*-goniocin, through a single-step triple oxidative cyclization reaction in the presence of a Re(VII) reagent, is shown in Scheme 2.[8] A notable difference between the two processes stands in the catalytic nature of the Ru-mediated process; also to be noted is the structure of the poly-THF product: a poly-THF alcohol for the process induced by rhenium, a poly-THF diol for the Ru-mediated one.



Scheme 2. An example of rhenium(VII)-mediated polycyclization of a bis-homoallylic trienol.

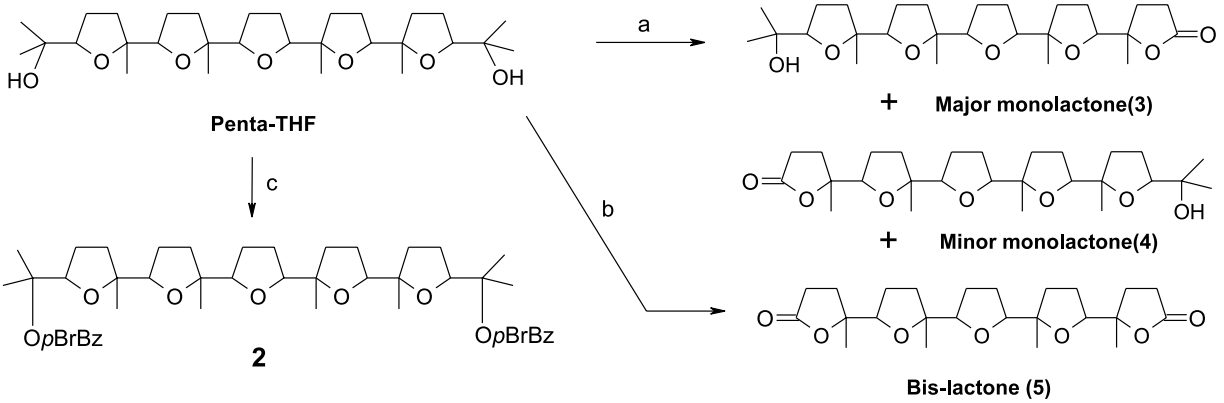
While for the Re-mediated process Sinha and co-workers have proposed simple stereoselectivity rules capable to predict its stereochemical outcome, mostly for linear polyenes,[8] the factors affecting the stereochemical course of the RuO_4-induced process are still unclear mainly due to the scarcity of available data, a fact that strongly hampers a close comparison of the two related processes. Therefore, with the aim of gaining further insight into the above Ru-mediated process the determination of the configuration of penta-THF **1** was accomplished as described in this paper; this proved to be a challenging task that required extensive 2D-NMR and chemical work, carried out both on **1** and on some of its derivatives.

2. Results and discussion

As a first step towards this goal we attempted the crystallization of **1**. Unfortunately, this product could not be induced to crystallize in a variety of organic solvents or solvent mixtures. Nor the bis-*p*-bromobenzoyl derivative of **1** (**2**, 80%, Scheme 3) obtained by reaction with *p*-bromobenzoyl chloride/DMAP in CH_2Cl_2, gave crystals suitable for X-ray analysis. Our attention was then turned to NMR studies. However, the full NMR assignment on compound **1** was intrinsically complicated by the high degree of superimposition of ^1H and ^{13}C resonances which occurred even in the two-dimensional experiments. For instance, all the methines belonging to THF rings A–E showed a severe overlapping and, in particular, all the tetrhydrofuran protons possess ^1H chemical shift values falling in the very narrow range of 0.13 ppm. Due to the high repetitive character of this structure, the same situation applies to most nuclei belonging to the five THF rings.

This particularly unfavourable situation required, together with the routine 2D-COSY, TOCSY, HSQC, and HMBC experiments, the acquisition of a set of three experiments which turned to be crucial for the complete assignment of **1**. In particular, the 2D-HSQC-TOCSY allowed to extrapolate the ^1H and the ^{13}C resonances for each of the five THF rings in **1**; subsequently 2D-INEPT-INADEQUATE and 2D-INADEQUATE experiments were employed in the resonance specific assignment of all ^1H and the ^{13}C signals, respectively.

However, the relative configuration of **1** could not be assigned just by using NMR data collected for **1**, due to the presence of some ambiguous cross-peaks in its ROESY spectrum. This prompted us to seek suitable derivatives of **1**, that could be obtained with the minimum stereochemical sacrifice, to be used for further NMR studies. In doing so, we were attracted by the PCC-mediated oxidative degradation of some bis-THF compounds, sharing with **1** a 2-hydroxypropyl terminus, previously carried out by McDonald and Towne.[9] We were pleased to find that treatment of **1** in the reported conditions [PCC (5 equiv), AcOH (70 equiv), Celite in CH_2Cl_2] cleanly gave a 6:1:6 mixture (52% overall yield) of the two mono-lactones **3** and



Scheme 3. Some derivates of penta-THF 1. Reagents and conditions: (a) PCC (5 equiv), AcOH (70 equiv), celite (10×weight of **1**), CH_2Cl_2, 24 h (**3**: 22%; **4**: 4%; **5**: 26%); (b) PCC (10 equiv), AcOH (150 equiv), celite (10×weight of **1**), CH_2Cl_2, 60 h (70%); (c) *p*BrBzCl (15 equiv), DMAP (30 equiv), CH_2Cl_2 (80%).

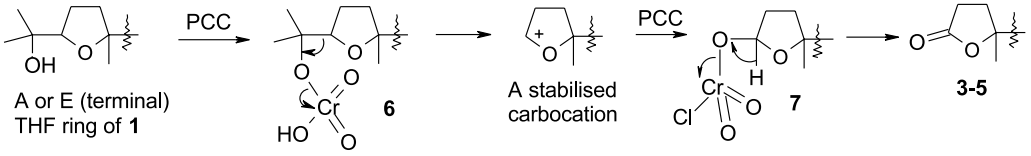
4 and the bis-lactone **5** (Scheme 4), derived from the oxidative cleavage of one or both the three-carbon 2-hydroxypropyl termini. Under forcing conditions (PCC: 10 equiv, AcOH 150 equiv, 60 h) bis-lactone **5** was obtained as the sole reaction product in a 70% yield.

^{1}H NMR spectra of derivatives **3**–**5** were simplified by the lack of one THF proton and two methyl signals in **3** and **4**, and two THF protons and four methyl signals in **5**. Collection of a new set of NMR data for these compounds finally allowed to unambiguously assign a *cis–cis–trans–trans–trans* sequence to the five adjacent THF rings in compound **1**.

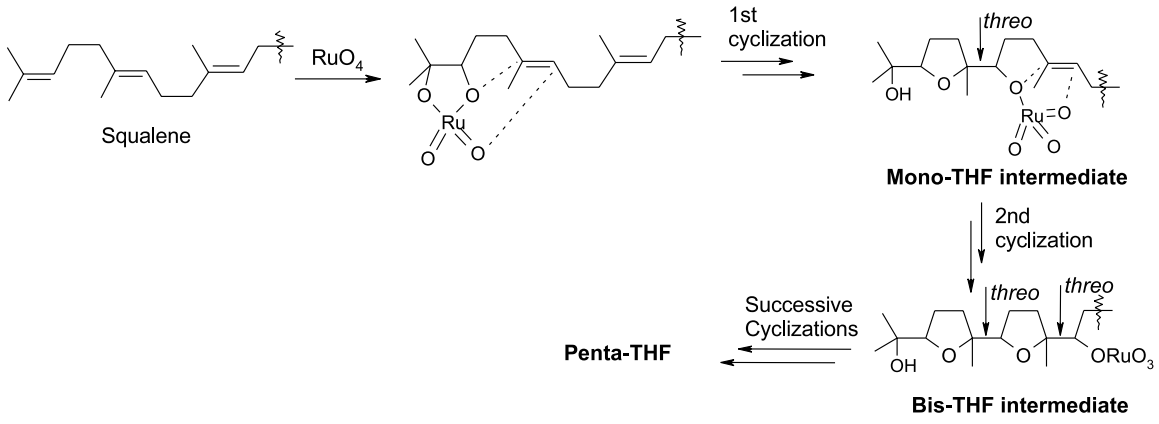
In particular, the application of the same assignment strategy employed for **1** afforded the sequence specific attribution of all crucial methine and methyl resonances belonging to rings A–E of compounds **2**–**5**. The comparative analysis of the ROESY spectra of compounds **1**–**5** allowed to establish the relative configuration indicated in Scheme 1. The *cis*-configuration of ring A was suggested by a strong ROESY correlation between H-3 and Me-26 present in the spectra of compounds **1**–**3** (**4** and **5** lack H-3). Likewise, ROESY correlations between proton H-7 and Me-27 were detected in the spectra of all compounds **1**–**5**, allowing to determine a *cis*-arrangement for B ring as well. Ring C is characterized by a *trans*-arrangement suggested primarily by the lack of ROESY contacts between H-11 and H-14 in the spectra of **1**–**5**. Further support came from key ROESY correlations, across ring functions, between H-11 and Me-28 of ring D, and H-14 and Me-27 of ring B. On the basis of a similar analysis, the configuration of rings D and E was also established. Particularly, diagnostic for determining the *trans*-arrangement for both rings D and E were the expected lack of ROESY correlations between Me-28 and H-18, and between Me-29 and H-22. Again the latter configurational assignment was corroborated by a dipolar coupling between Me-28 and Me-29, observed in the ROESY spectrum of compound **2**.

A plausible mechanism explaining the oxidative cleavage of the two terminal isopropyl portions in **1** is shown in Scheme 4 that also agrees with related PCC chemistry.[10] Thus, the initially formed chromium (VI) ester **6** fragments to form a stabilised carbocation species that is then intercepted by another PCC molecule to form **7**. Subsequent elimination of a chromium species generates the lactone function found in compounds **3**–**5**. The markedly different yields of monolactones **3** (22%) and **4** (4%) is evidence of a greater propensity of the side chain next to the *trans*-THF ring to undergo cleavage. This is a relatively unknown process.

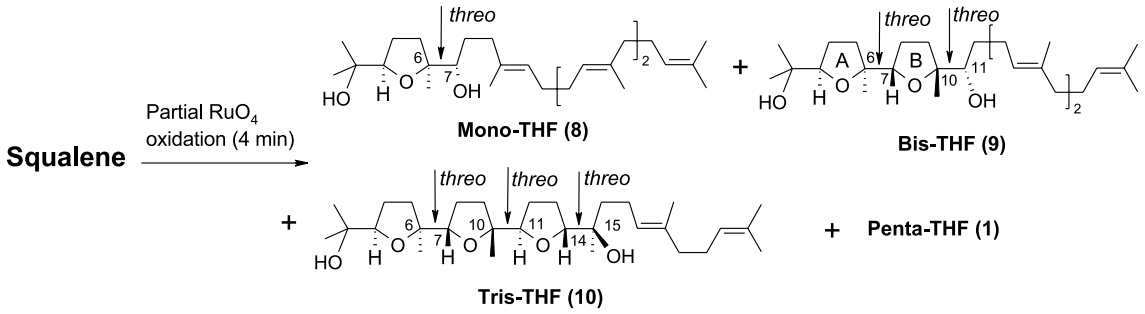
In the next step towards the elucidation of the configuration of **1**, we had to establish the stereochemical relationship within each of the four carbon pairs connecting the adjacent THF rings in **1**. As previously experienced in similar studies from our own group[7] and of other researchers,[11] this could not be accomplished by NMR spectroscopic analysis alone. In fact, the pattern of dipolar contacts across adjacent THF rings can be hardly converted into useful information on their relative orientation, as a simple inspection of molecular models would confirm. Though, in principle, both an *erythro* and *threo* relationship could subsist for each THF pair, closure of each THF ring likely takes place by *syn*-addition[12] of two oxygens through the [3+2] cycloaddition of a RO-Ru=O portion on each of the six double bonds of the polyene, as shown in Scheme 5 for the first two ring-closing steps. This would agree with previous results from the cyclization of farnesyl acetate (FA) and geranyl geranyl acetate (GGA)[7] and related Re(VII) chemistry,[8] and would imply that an all-*threo* penta-THF should be obtained from the *all-trans* squalene. A definitive confirmation of this appeared, nevertheless, necessary to put our understanding of the process on a firm basis.



Scheme 4. Proposed mechanism for the PCC-mediated oxidative cleavage of **1**.



Scheme 5. Mechanistic hypothesis for the oxidative polyciclization of squalene with cat. RuO_4.



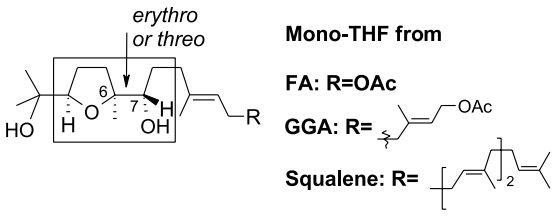
Scheme 6. Partial oxidation of squalene with cat. RuO_4.

Proof for the all-*threo* arrangement of **1** was gained through extensive chemical work carried out on the intermediates of the polycyclization process, obtained by partial oxidation of squalene, as well as on some degradation products of **1**. In particular, when the oxidation of squalene was stopped prior to completion (4 min), the mono-, bis-, and tris-THF compounds **8**–**10** (Scheme 6), derived from the partial cyclization of squalene, could be isolated by HPLC, along with penta-THF **1** and the unreacted polyene. Rather surprisingly, the tetra-THF diol intermediate, possessing the A/B/C/D ring sequence (see structure **22** in Scheme 11), could not be detected among the reaction products even after a careful HPLC analysis of the reaction mixture.

Based on the assumption that compounds **8**–**10** possess an all-*threo* arrangement, as the penta-THF to which they eventually give rise, a determination of the C_6–C_7 and C_{10}–C_{11} stereorelationships in **1** was made on compounds **8** and **9**. Though this furnishes an indirect evidence of the above configurations in **1**, we are confident that compounds **8**–**10** are indeed intermediates of the process, leading to the penta-THF formation based on the following considerations. Firstly, they are the sole mono-, bis- and tris-THF diol products isolated during the partial oxidation of squalene obtained as main products of the reaction mixture; secondly, we have previously demonstrated that the mono- to tris-THF diols isolated from the partial oxidation of FA and GGA with RuO_4 possess configuration that matched that of the poly-THF product of which they are precursors. Determination of the relationship between the carbon pairs connecting B/C and C/D rings (C_{14}–C_{15}, and C_{18}–C_{19} carbon pairs, respectively) was carried out on two products derived from degradation of **1** as depicted in Schemes 9 and 11 (later in the discussion), also taking into account evidence collected on compounds **8**–**10** and the above assumption.

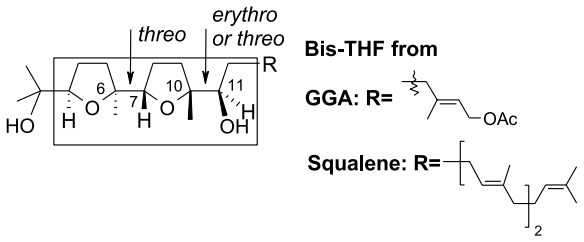
NMR evidence from the proton spectra of compounds **8** and **9** pointed to an all-*threo* arrangement for both. In fact, chemical shift values of the C*H*–O and Me proton resonances belonging to the mono- or bis-THF alcohol portions in these compounds (Tables 1 and 2, see the structural portions in the boxes) showed a very good agreement with those observed for the corresponding protons in the structurally related *cis–threo* mono-THF and *cis–threo–cis–threo* bis-THF analogues, respectively, previously isolated from the partial oxidation of FA and GGA.[7] In particular, diagnostic for the configuration at C-7 (squalene numeration) in **8** is the H-7 proton resonance that in the *threo*-isomers falls in the narrow range δ 3.38–3.39 (Table 1) while is downfield shifted at δ 3.54–3.56 in the *erythro*-isomers. Analogously, in the bis-THF series (Table 2) the H-11 resonance is a probe for the configuration at the C-11 centre. This proton exhibited a chemical shift value in the range δ 3.38–3.41 (virtually the same observed for H-7 in the mono-THF series) for the C_{10}–C_{11} *threo* compounds while it is once again downshifted, at δ 3.52–3.54, in the C_{10}–C_{11} *erythro* isomers. Also noticeable, on passing from *threo* to *erythro* (C_6–C_7 and C_{10}–C_{11}) isomers, is the small but diagnostic downfield shift (from 1.24–1.25

Table 1. Comparison of diagnostic ^{1}H NMR data of some isoprenoidic mono-THF diols (squalene numeration)



Proton	Mono-THF from squalene (**8**) (*cis–threo*)	Mono-THF from FA (*cis–threo*)	Mono-THF from GGA (*cis–threo*)	Mono-THF from squalene (*cis–erythro*)	Mono-THF from FA (*cis–erythro*)
C*H*-O(THF)	3.85	3.85	3.85	3.82	3.82
C*H*-OH	3.39	3.38	3.38	3.56	3.54
3×Me	1.12	1.12	1.13	1.13	1.14
	1.16	1.16	1.16	1.15	1.16
	1.24	1.24	1.24	1.26	1.26

Table 2. Comparison of diagnostic ^{1}H NMR data of some isoprenoidic bis-THF diols (squalene numeration)



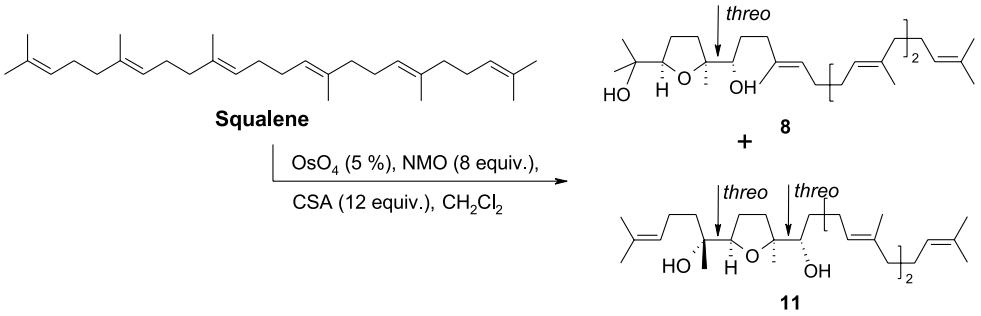
Proton	Bis-THF from squalene (**9**) (*cis–threo–cis–threo*)	Bis-THF from GGA (*cis–threo–cis–threo*)	Bis-THF from squalene (*cis–threo–cis–erhytro*)	Bis-THF from GGA (*cis–threo–cis–erhytro*)
2×C*H*-O(THF)	3.82	3.81	3.82	3.82
	3.92	3.93	3.91	3.92
C*H*-OH	3.41	3.38	3.54	3.52
4×Me	1.09	1.09	1.11	1.11
	1.13	1.13	1.13	1.14
	1.13	1.15	1.14	1.14
	1.25	1.24	1.26	1.26

to 1.26 ppm), observed in both series, of one of the methyl signals.

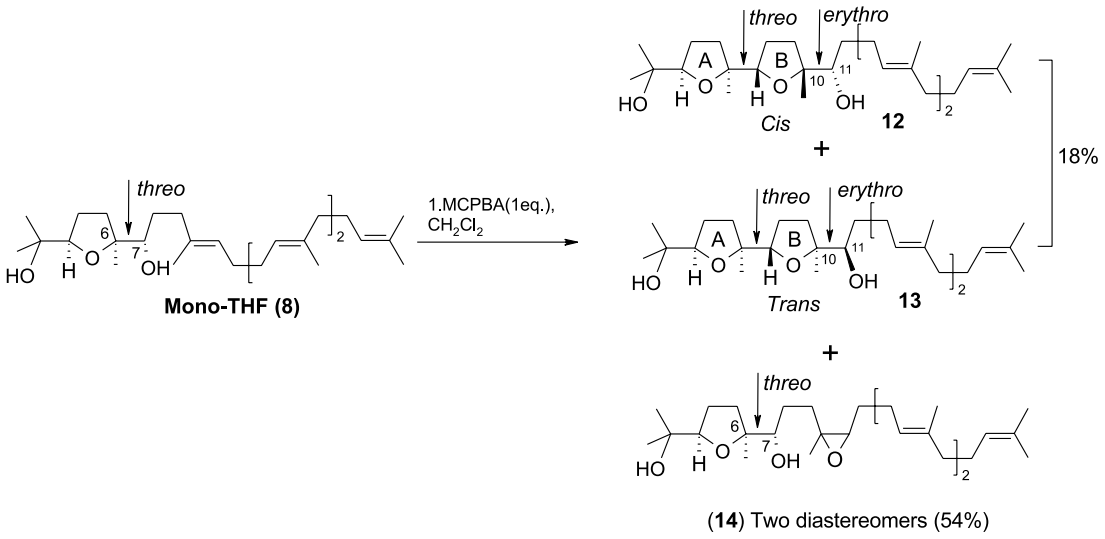
Confirmation of the *cis–threo* configuration of **8** was obtained by synthesis via *cis*-streoselective OsO_4-mediated cyclization of squalene. This reaction has previously been carried out in high yields on 1,5-dienes.[13] Extension of the process to a polyene possessing more than two double bonds has never been carried out before and was expected to give lower cyclization yields of the expected mono-THF **8**. In fact, in principle, three isomeric mono-THF diols could be obtained from the process involving the three different 1,5-dienic substructures present in the polyene; in addition, once formed these products could likely undergo further attack of the oxidant to the other olefinic double bonds of the molecule. Thus, treatment of squalene with catalytic OsO_4 (10 mol%) in the presence of NMO (8 equiv) and CSA (12 equiv) (Scheme 7) afforded only two out of the three expected isomers namely mono-THF **8** and **11**, in a ca. 1:1 ratio (**8**: 7%; **11**: 6% respect to reacted squalene) along with unreacted squalene (ca. 10%). The low yields of these two products were mainly due to the presence of some more polar substances (not further studied), probably obtained, as above anticipated, from the further evolution of the first-formed monocyclized species. It is also to be noted that compounds **8** and **11** are the sole noticeable mono-THF products obtained from the process and the HPLC profile of the reaction mixture showed no peaks other than those relative to these products in the region corresponding to the polarity expected for squalene-derived mono-THF diols.

Unambiguous evidence of the all-*threo* configuration for bis-THF **9** was gained by comparison with the two isomeric C_{10}–C_{11} *erythro* bicyclic tetrahydrofuranyl diol products (*cis–threo–cis–erythro* **12** and *cis–threo–trans–erythro* **13**) obtained from mono-THF **8** by MCPBA epoxidations (1 equiv in CH_2Cl_2, Scheme 8). In particular, this reaction afforded a mixture of the two diastereomeric hydroxy-epoxides **14** (54%) along with the expected two bis-THF compounds **12** and **13** (18% overall), derived from the acid-catalyzed intramolecular *anti*-opening of the hydroxy-epoxides **14**. As expected spectral data and chromatographic (HPLC) properties of these compounds were different from those exhibited by the bis-THF **9** confirming it to possess a C_{10}–C_{11} *threo* relationship.

As for the *threo* relationship within the C_{14}–C_{15} carbon pair it was demonstrated by symmetry considerations on a degradation product obtained from **1** as shown in Scheme 9. In particular, LAH reduction of the pentacyclic bis-lactone **5**, followed by acetylation gave the tris-THF **15**. This compound possesses two tertiary alcohol functions flanking the B and D THF rings, a structural feature also found in **1** that appears to be responsible for the observed reactivity of this compound towards PCC. When compound **15** was subjected to the same PCC degradation conditions employed for the conversion **1**→**5**, the



Scheme 7. Oxidative cyclization of squalene with OsO_4.

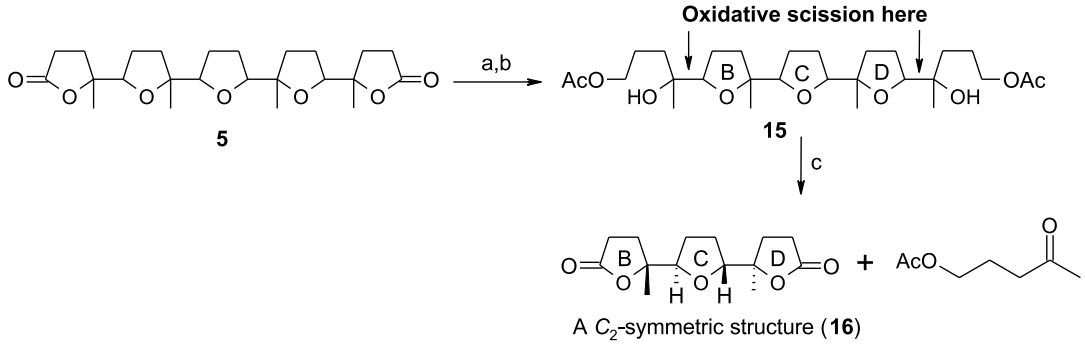


Scheme 8. Synthesis of squalene-derived, C_{10}–C_{11} *erythro*, bis-THF diols **12** and **13**.

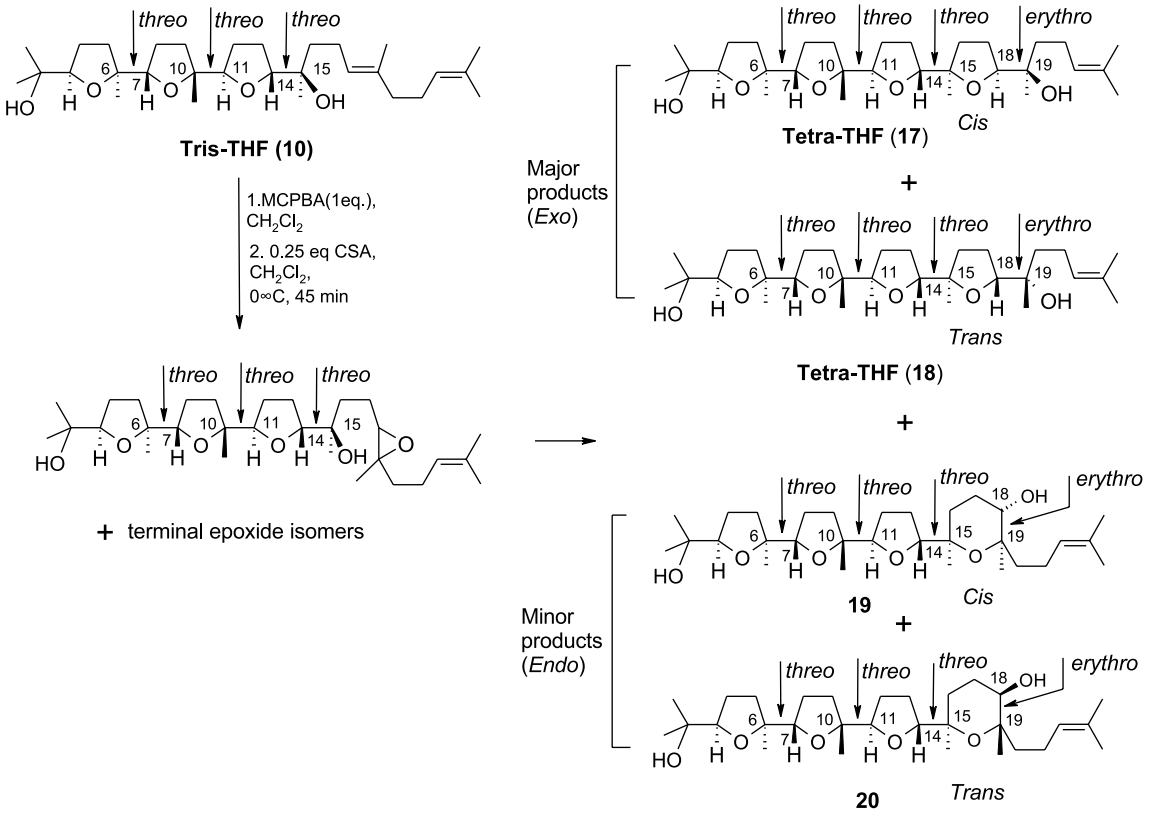
C_2-symmetric bis-lactone **16** was cleanly obtained along with 5-acetoxypentan-2-one.[14] The isolation of the latter compound, derived from the oxidative cleavage of the two side chains bonded to rings B and D, adds further support to the mechanism proposed in Scheme 4, for the PCC-mediated oxidative cleavage of **1**. As required for a C_2-symmetric, *threo–trans–threo*, structure (remind that the C_{10}–C_{11} *threo* had already been established), compound **16** displayed half of the expected NMR signals in both the ^{1}H and ^{13}C NMR spectra.

Finally, determination of the C_{18}–C_{19} *threo* relationship was based on the same reasoning employed to establish the C_{10}–C_{11} relationship. Therefore, tris-THF **10** (Scheme 10), was transformed into the isomeric C_{18}–C_{19} *erythro* tetra-THF **17** and **18** via MCPBA epoxidation, followed by acid-catalyzed (CSA) cyclization. In this case, the acidic treatment was necessary because the first-formed hydroxyepoxides did not spontaneously cyclize to THF. This reaction afforded, besides expected **17** and **18** (major products; *exo*-oxacyclizations) the two ring-D tetrahydropyrane isomers **19** and **20**, derived from *endo*-oxacyclization processes. Their structure was in agreement with the presence of an acetylatable OH group. In addition, the chemical shift and *J*-values for the H-18 proton in the *trans–erythro* isomer **20** (J=10.8, 5.2 Hz; axial proton in a chair conformation) well agree with those reported for a recently synthesized product embodying a 2-hydroxy-*trans*-THP ring in the structure.[15] This proton in the *cis–erythro* isomer **19** displayed *J* values (3 and 4 Hz) in accordance with those expected for an equatorial proton in the chair conformation.

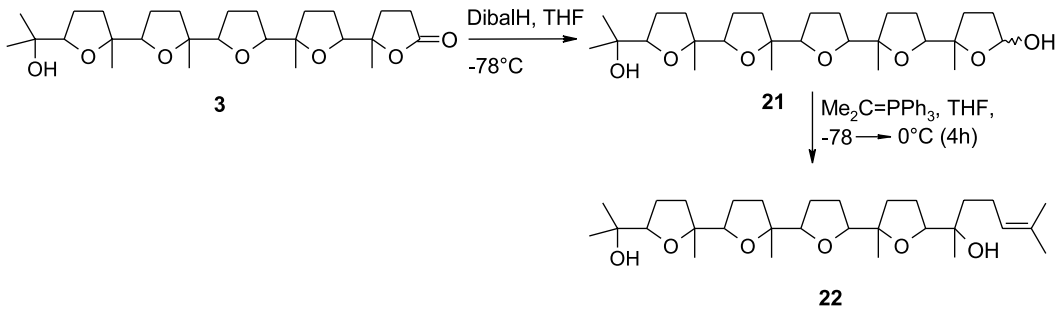
As above mentioned, the partial RuO_4 oxidation of squalene did not furnish the tetra-THF intermediate of the process, even stopping the process at different reaction times (1.5, 3, 6, 7.5 and 10 min). However, this compound (**22**) could be obtained from mono-lactone **3**, in turn derived from penta-THF **1** (Scheme 3), as shown in Scheme 11. In particular, reduction of **3** with DIBALH, followed by Wittig olefination reaction of lactol **21**, in the presence of excess isopropyltripehenylphosphonium iodide, afforded the desired tetra-THF **22**, though in a low 8–10% yield (HPLC). This compound displayed spectral and chromatographyc properties different from those exhibited by the pair **17**/**18**, indicating it to be the all-*threo*-isomer. A double-Wittig reaction of the above type has recently been carried out in high yields on the bis-lactol possessing a gross structure corresponding to bis-lactone **5**, a compound strictly similar to **21**.[16] The low yield in our case is probably ascribable to the presence in **21** of an unprotected alcohol function in the



Scheme 9. Bidirectional PCC oxidative degradation of bis-lactone **5**. Reagents and conditions: (a) $LiAlH_4$, Et_2O, rt, 1 h; (b) Ac_2O, pyridine (**15**, 72% for two steps); (c) PCC (1 equiv), AcOH (150 equiv), celite, CH_2Cl_2 (**16**, 47%).



Scheme 10. Synthesis of squalene-derived, C_{18}–C_{19} *erythro*, tetra-THF diols **17** and **18**.



Scheme 11. Synthesis of squalene derived tetra-THF **22**.

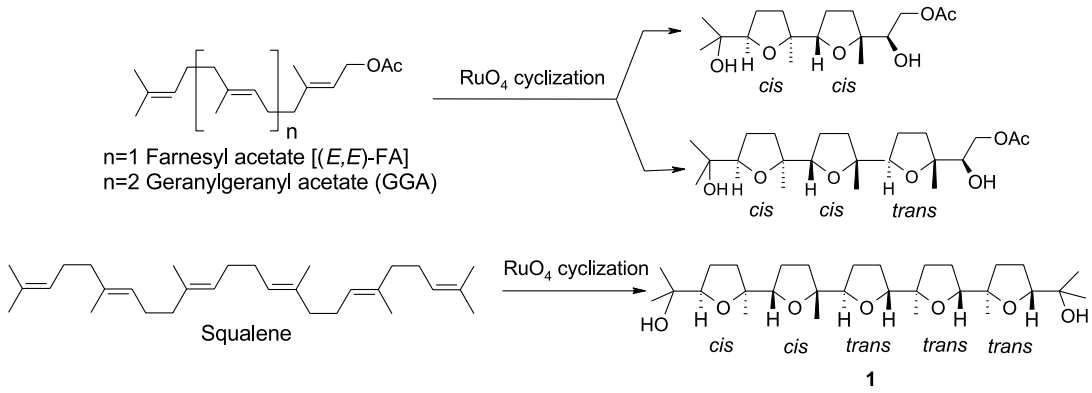
structure, as previously experienced by others in similar cases.[17]

Previous studies on the synthesis of squalene-derived penta-THF products have been reported[18] mostly addressed to the synthesis of glabrescol, a naturally occurring isomer of **1** isolated from *Spatelia glabrescens.*[1] An approach based on exo-selective oxacyclizations of polyepoxides was generally used in these syntheses allowing to obtain all-*erythro* penta-THF materials. The easy access to poly-THF compounds **1**, **8**–**10** and **22** from the low-price, commercially available, squalene opens the way to the preparation of other penta-THF materials, isomeric with **1**, through mixed RuO_4-based and epoxidation/acid-catalysed *anti* cyclization, chemistry, as well as through synthetic strategies previously reported by others.[19] At least some of these materials are expected to possess ionoforic activity[20] or could be used in other studies.

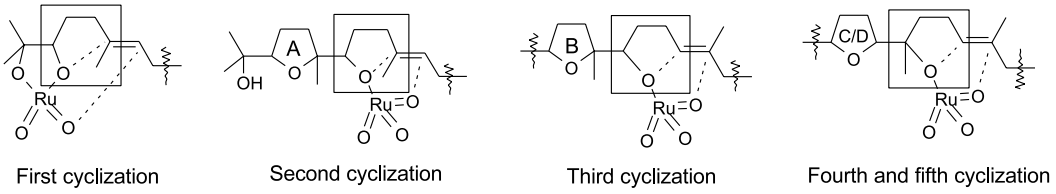
3. Conclusion

The elucidation of the configuration of the above penta-THF gives a further insight into the stereocontrol of the oxidative polycyclization process mediated by RuO_4. However, at the present level of knowledge it seems still hazardous trying to rationalize, either the stereochemical outcome for the oxidation of squalene, or to trace out general rules (if any) to predict the streochemical course of this kind of process.

Some observation should, nevertheless, be made. With reference to **1**, we can notice that the first two cyclizations lead to *cis*-THF rings as it happens for the oxidation of the structurally related polyenes FA and GGA (Scheme 12).[7] Thus, it appears that three consecutive isoprene units bonded head-to-tail invariably give rise to two consecutive *cis*-THF rings. As for the third cyclization step leading to the central cycle in **1**, it proceeds in a *trans* manner as it



Scheme 12. Summary of the poly-THF diol products obtained from the oxidation of isoprenoid polyenes.



Scheme 13. Supposed cyclizing species in the oxidation of squalene.

occurs in the third cyclization of GGA, though a structural difference exists between this compound and squalene (head-to-tail versus tail-to-tail bond of the fourth isoprene unit).

When considering the supposed cyclizing species for each ring-closing step, it should be noted that, apart from the first cyclization, three different, but strictly similar, structural situations subsist which refer to the second, third and fourth/fifth cyclizations (Scheme 13, see boxes). Can only the structural change in the cyclizing portion be responsible for the observed sterochemical control? The formation of *trans*-THF rings from the last two cyclizations, proceeding through similar cyclizing substructures, would seem to back up this hypothesis. However, if this is the case, it remains to be explained why the third cyclization in GGA proceeds with the formation of a *trans*-THF ring, despite the cyclizing portion being similar to that involved in the second cyclizations of FA, GGA and squalene, where a *cis*-ring is formed. Therefore, other factors should be considered such as the THF-*O*-coordination to ruthenium[21] of the THF ring adjacent to the cyclizing portion, as well as its configuration which could arrange the molecule in a conformation facilitating either of the two competing cyclization routes.

4. Experimental

4.1. General methods

All reagents and anhydrous solvents were purchased (Aldrich and Fluka) at the highest commercial quality and used without further purification. Where necessary, flame-dried and argon-charged glassware was used. Reactions were monitored by thin-layer chromatography carried out on precoated silica gel plates (Merck 60, F_{254}, 0.25 mm thick). Merck silica gel (Kieselgel 40, particle size 0.063–0.200 mm) was used for column chromatography. HPLC separations were carried out on a Varian 2510 apparatus equipped with a Waters R403 dual cell differential refractometer using phenomenex 250×10 mm and 250×4.6 mm (both 5μ) columns. NMR experiments were performed on Bruker DRX-600, Bruker WM 400, Varian 300, and Gemini 200 spectrometers in $CDCl_3$. All the 2D-NMR spectra were acquired at 600 MHz in the phase-sensitive mode with the transmitter set at the solvent resonance and TPPI (Time Proportional Phase Increment) used to achieve frequency discrimination in the ω_1 dimension. Standard pulse sequence and phase cycling were used for DQF-COSY, 2D-TOCSY, HSQC, 2D-HSQC-TOCSY, HMBC, 2D-INEPT-INADEQUATE, 2D-INEDAQUATE and ROESY spectra. The NMR data were processed on a Silicon Graphic Indigo2 Workstation using UXNMR software. Proton chemical shifts were referenced to the residual $CHCl_3$ signal (7.26 ppm); ^{13}C NMR chemical shifts were referenced to the solvent (77.0 ppm). Abbreviations for signal coupling are as follows: s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet. IR spectra were collected on a Jasco FT-IR-430 spectrometer. ESI mass spectrometric analyses were recorded on a Waters Micromass ZQ mass spectrometer equipped with an Electrospray source used in the positive mode. HRMS spectra were recorded on a Voyager DE-PRO mass spectrometer using MALDI-TOF ionization.

For all the reported products the numeration of penta-THF **1** is used.

4.2. Oxidation of squalene. Synthesis of penta-THF 1 and isolation of the mono-, bis-, tris-THF diols intermediates of the process

Squalene (500 mg, 1.22 mmol) was dissolved in the biphasic mixture EtOAc/CH_3CN/H_2O (3:3:1) (140 mL),

then $NaIO_4$ (2.09 g, 9.76 mmol) and a catalytic amount of $RuO_2 \cdot 2H_2O$ (32.5 mg, 0.244 mmol, 20 mol%) were added in sequence under vigorously mechanic stirring at 0 °C. After 30 min excess of a saturated $Na_2S_2O_3 \cdot 5H_2O$ solution was added (50 mL) until the mixture turned to black; then the mixture was filtered and extracted with EtOAc (3× 150 mL) and the combined organic phase was dried over Na_2SO_4 and evaporated. The oily residue was purified by silica gel column chromatography (eluent $CHCl_3$/MeOH, 97:3) followed by HPLC (hexane/ethyl acetate 1:1) to yield **1** (320 mg, 50%, $t_R = 11.5$ min).

When the oxidation of squalene (1.49 g, 3.6 mmol) was stopped after 4 min, the crude reaction mixture was separated by column chromatography (silica, gradient elution from 9:1 hexanes/ethyl ether to ethyl ether) to yield, in the first-eluted fractions, unreacted squalene (450 mg, 30%), the C-7 ketone corresponding to mono-THF **8** (40 mg, 4%) and the C-11 ketone corresponding to bis-THF **9** (55 mg, 5%). A fraction at an intermediate polarity (330 mg) contained mono-THF diol **8**, bis-THF diol **9** and tris-THF diol **10**. A slightly more polar fraction gave pure penta-THF **1** (320 mg, 28%). Compounds **8–10** were separated by HPLC (hexane/EtOAc, 75:25) to yield **8** (45 mg, 5%; $t_R = 21$ min), **9** (52 mg, 4.5%; $t_R = 17.5$ min) and **10** (50 mg, 4.3%; $t_R = 14$ min).

4.2.1. Compound 8. IR (neat): ν_{max} 3396 (br) cm^{-1}. ^1H NMR (200 MHz): δ 5.20 (1H, m, olefinic proton), 5.12 (3H, m, olefinic protons), 3.85 (1H, t, $J = 6.8$ Hz, H-3), 3.39 (1H, dd, $J = 7.7$, 3.6 Hz, H-7), 1.68 (3H, s, Me), 1.59 (12H, s, 4× Me), 1.24, 1.16, 1.12 (3H each, s's, 3×C(O)Me). ^{13}C NMR (100 MHz): δ 135.2, 135.0, 134. 9, 131.2, 124.8, 124.4, 124.25, 124.19, 85.8, 85.2, 71.5, 39.7, 36.6, 35.1, 30.4, 28.3, 28.2, 27.3, 26.8, 26.7, 26.6, 25.6, 24.8, 21.0, 17.6, 16.0. LRMS *m/z* 499 (78, $[M+K]^+$), 483 (100, $[M+Na]^+$). HRMS Calcd for $C_{30}H_{52}O_3Na$ 483.3816, found 483.3831.

4.2.2. Compound 9. IR (neat): ν_{max} 3447 (br) cm^{-1}. ^1H NMR (200 MHz): δ 5.10 (3H, m, olefinic protons), 3.92, 3.82 (1H each, both t, $J = 7.2$ Hz for both, H-3 and H-7), 3.41 (1H, dd, $J = 9.2$, 2.6 Hz, H-11), 1.67 (3H, s, Me), 1.62 (3H, s, Me), 1.59 (6H, s, 2×Me), 1.25 (3H, s, C(O)Me), 1.13 (6H, s, 2×C(O)Me), 1.09 (3H, s, C(O)Me). ^{13}C NMR (100 MHz): δ 135.7, 134.9, 131.3, 124.4, 124.3, 124.1, 86.0, 85.1, 83.4, 83.0, 76.1, 71.6, 39.8, 39.7, 35.1, 34.4, 32.3, 27.9, 27.2, 26.8, 26.7, 25.8, 25.7, 25.0, 23.9, 20.1, 17.7, 16.1, 16.0. LRMS *m/z* 515 (100, $[M+K]^+$), 499 (77, $[M+Na]^+$). HRMS Calcd for $C_{30}H_{52}O_4Na$ 499.3765, found 499.3778.

4.2.3. Compound 10. IR (neat): ν_{max} 3420 (br) cm^{-1}. ^1H NMR (200 MHz): δ 5.16, 5.10 (1H each, both bt, $J = 6.9$ Hz for both, olefinic protons), 3.93–3.77 (4H, overlapped multiplets, H-3, H-7, H-11 and H-14), 1.68 (3H, s, Me), 1.60 (6H, s, 2×Me), 1.24, 1.18, 1.12, 1.05, 0.98 (3H each, s's, 5×C(O)Me). ^{13}C NMR (100 MHz): δ 134.3, 131.4, 125.1, 124.5, 85.5, 85.4, 85.1, 84.7, 83.2, 83.1, 72.9, 71.7, 40.0, 39.7, 35.3, 34.8, 28.98, 28.1, 27.0, 26.8, 26.6, 25.6, 25.2, 24.6, 22.4, 20.9, 19.7, 17.6, 15.9. LRMS *m/z* 531 (100, $[M+K]^+$), 515 (45, $[M+Na]^+$). HRMS Calcd for $C_{30}H_{52}O_5Na$ 515.3714, found 515.3725.

4.2.4. Compound 1. IR (neat): ν_{max} 3466 (br) cm^{-1}. ^1H NMR (600 MHz, attribution by 2D-NMR): δ 3.93 (1H, t, $J = 6.5$ Hz, H-11), 3.87 (1H, t, $J = 7.5$ Hz, H-7), 3.83 (1H, dd, $J = 8.0$, 2.5 Hz, H-18), 3.81, 3.80, (3H, overlapped m's, H-3, H-14, H-22), 2.20, 1.56 (2H, m's, H_2-16), 2.19, 1.51, (2H, m's, H_2-5), 2.10, 1.58 (2H, m's, H_2-20), 1.76, 1.70 (2H, m's, H_2-21), 2.00, 1.56, (2H, m's, H_2-9), 1.90, 1.85 (2H, m's, H_2-17), 1.90, 1.70 (2H, m's, H_2-13), 1.94, 1.92 (2H, m's, H_2-4), 1.88 (4H, m, H_2-8, H_2-12), 1.22 (3H, s, Me-25), 1.20 (3H, s, Me-30), 1.15 (3H, s, Me-26), 1.14 (3H, s, Me-27), 1.11 (3H, s, Me-29), 1.10 (3H, s, Me-24), 1.07 (6H, s, Me-28, Me-1). ^{13}C NMR (150 MHz): δ ^{13}C NMR (150 MHz, attribution by 2D-NMR): δ 87.4 (CH-22), 86.4 (CH-18), 85.4 (CH-3), 85.1 (C-15, CH-11), 84.7 (CH-14), 84.1 (C-6), 83.9 (C-19), 83.7 (C-10), 82.6 (CH-7), 72.0 (CH-3), 70.7 (C-23), 34.9 (CH_2-20), 34.8 (CH_2-16), 34.6 (CH_2-9), 34.1 (CH_2-5), 28.1 (CH_3-30, CH_3-25), 28.2 (CH_2-13), 27.4 (CH_2-17), 27.3 (CH_2-8), 27.2 (CH_2-12), 26.7 (CH_2-21), 26.1 (CH_2-4), 25.1 (CH_3-28), 24.7 (CH_3-29), 24.4 (CH_3-24, CH_3-1), 24.1 (CH_3-26), 23.2 (CH_3-27). LRMS: *m/z* 563 (18, $[M+K]^+$), 547 (24, $[M+Na]^+$), 525 (100, $[M+H]^+$). HRMS Calcd for $C_{30}H_{52}O_7Na$ 547.3612, found 547.3621.

4.3. Bis-*p*-bromobenzoate ester (2)

To a solution of **1** (30 mg, 0.06 mmol) in CH_2Cl_2 (0.5 mL) was added DMAP (221.7 mg, 1.81 mmol) and *p*-bromobenzoyl chloride (219.5 mg, 1.00 mmol) dissolved in CH_2Cl_2 (2 mL) and the resulting solution was stirred at room temperature for 60 h. The mixture was diluted with $CHCl_3$, filtered through a 4 cm silica gel pad. Elution with ethyl ether and evaporation gave a residue that was purified by preparative TLC (hexanes/ethyl ether 75:25) to yield **2** (43 mg, 80%) as a colorless oil.

4.3.1. Compound 2. IR (neat): ν_{max} 1716 cm^{-1}. ^1H NMR (600 MHz, attributions by 2D-NMR): δ 7.83, 7.82, (2H each, d's, $J = 8.8$ Hz for both, aromatic protons), 7.53 (4H, d, $J = 8.4$ Hz, aromatic protons), 4.22 (1H, dd, $J = 7.0$, 7.0 Hz, H-3), 4.18 (1H, dd, $J = 9.2$, 5.8 Hz, H-22), 3.93 (1H, dd, $J = 8.5$, 6.6 Hz, H-7), 3.85, 3.83, 3.73 (3H, overlapped m's, H-11, H-14, H-18), 2.22, 2.01, 1.99, 1.97, 1.94, 1.89, 1.87, 1.84, 1.74, 1.59 (20H, overlapped m's, 10×CH_2), 1.62 (3H, s, Me-30), 1.59 (3H, s, Me-1), 1.58 (3H, s, Me-25), 1.57 (3H, s, Me-24), 1.22 (3H, s, Me-26), 1.15 (3H, s, Me-27), 1.14 (3H, s, Me-29), 1.09 (3H, s, Me-28). ^{13}C NMR (150 MHz, attributions by 2D-NMR): δ 166.4 (C), 165.3 (C), 131.6–131.2 (10 CH, aromatic methines), 128.2 (2 C, aromatic quaternary), 87.0 (C) 86.7 (CH-18), 86.1 (CH-22), 85.6 (CH-11), 85.4 (CH-14), 85.1 (C), 84.8 (C), 84.4 (C), 83.9 (C), 83.7 (C), 82.8 (CH-3), 82.6 (CH-7), 72.2 (C), 70.8 (C), 34.8 (CH_2), 34.7 (CH_2), 33.1 (CH_2), 27.9 (CH_2), 27.3 (2×CH_2), 27.2 (CH_2), 27.1 (CH_2), 26.9 (CH_2), 25.1 (CH_3-29), 24.4 (CH_3-28), 23.7 (CH_3-27), 23.4 (CH_3-26), 23.3 (CH_3-30), 23.0 (CH_3-1), 21.8 (CH_3-25), 21.7 (CH_3-24). LRMS *m/z* 931/929/927 (18, $[M+K]^+$), 915/913/911 (30, $[M+Na]^+$). HRMS Calcd for $C_{44}H_{58}Br_2O_9Na$ 911.2346, found 911.2328.

4.4. Bis-lactone 5 and monolactones 3 and 4

To a solution of **1** (200 mg, 0.38 mmol) in CH_2Cl_2 (8 mL)

was added celite (2 g), PCC (1.9 mmol, 409.5 mg) and AcOH (26.6 mmol, 1.5 mL) and the resulting heterogeneous mixture was stirred at room temperature for 24 h and then loaded on a silica gel column chromatography. Elution with $CHCl_3$/MeOH 9:1 afforded a colourless oil (150 mg). HPLC separation (hexane/EtOAc 60:40) gave unreacted **1** (28 mg, 14%, $t_R=15$ min), monolactones **3** (40 mg, 22%, $t_R=$ 18 min) and **4** (7.5 mg, 4%, $t_R=17$ min), and bis-lactone **5** (43 mg, 26%, $t_R=20.5$ min). When the reaction was carried out with 10 equiv PCC and 150 equiv AcOH, the yield of bis-lactone **5** raised to 70%, yields for compounds **3** and **4** were 6 and 2%, respectively, and a 2% amount of unreacted **1** was recovered.

4.4.1. Compound 3. IR (neat): ν_{max} 3467 (br), 1772 cm^{-1}. ^1H NMR (600 MHz, attributions by 2D-NMR): δ 3.85, 3.87, 3.89 (3H, overlapped, m's, H-7, H-11, H-18), 3.78 (1H, dd, $J=7.6$, 5.4 Hz, H-3), 3.74 (1H, dd, $J=9.7$, 5.5 Hz, H-14), 2.75 (1H, ddd, $J=17.2$, 10.8, 10.8 Hz, H_a-21), 2.40, 2.38, 2.20, 2.16, 1.94, 1.90, 1.87, 1.66, 1.58, 1.52, (19H, overlapped m's, $9\times CH_2$ and H_b-21), 1.30 (3H, s, Me-29), 1.19 (3H, s, Me-25), 1.12 (3H, s, Me-26), 1.11 (3H, s, Me-27), 1.03 (3H, s, Me-1), 1.01 (3H, s, Me-28). ^{13}C NMR (150 MHz, attributions by 2D-NMR): δ 178.2 (C), 86.6 (C) 86.3 (CH-18), 86.2 ((C), 85.3 (CH-3), 85.2 (C), 85.1 (CH-11), 84.8 (CH-14), 84.1 (C), 82.3 (CH-7), 71.8 (C), 34.5 (CH_2), 34.4 (CH_2), 34.3 (CH_2), 32.6 (CH_2), 30.2 (CH_2), 28.3 (CH_2), 28.1(CH_3-25), 27.0 (CH_2), 26.8 (CH_2), 26.6 (CH_2), 26.0 (CH_2), 24.7 (CH_3-1), 23.9 (CH_3-29), 23.8 (CH_3-26), 23.6 (CH_3-28), 22.3 (CH_3-27). LRMS *m/z* 519 (22, $[M+K]^+$), 503 (48, $[M+Na]^+$), 481 (30, $[M+H]^+$). HRMS Calcd for $C_{27}H_{44}O_7Na$ 503.2986, found 503.2971.

4.4.2. Compound 4. IR (neat): ν_{max} 3497 (br), 1772 cm^{-1}. ^1H NMR (600 MHz, attributions by 2D-NMR): δ 3.75, 3.80, 3.83, 3.87, 3.91 (5H, overlapped m's, H-7, H-11, H-14, H-18, H-22), 2.80 (1H, ddd, $J=17.4$, 10.8, 10.8 Hz, H_a-4), 2.45, 2.42, 2.17, 1.95, 1.91, 1.89, 1.88, 1.79, 1.61, 1.59, 1.56, (19H, overlapped m's, $9\times CH_2$ and H_b-4), 1.35 (3H, s, Me-26), 1.20 (3H, s, Me-30), 1.14 (3H, s, Me-27), 1.11 (3H, s, Me-29), 1.10 (3H, s, Me-24), 1.07 (3H, s, Me-28). ^{13}C NMR (150 MHz, attributions by 2D-NMR): δ 178.2 (C), 87.7 (C) 87.4 (CH, C-22), 86.6 (CH-18), 86.0 (C), 85.7 (CH-11), 85.3 (C), 85.1 (CH-14), 84.3 (C), 84.1 (CH-7), 71.1 (C), 34.8 ($2\times CH_2$), 34.6 (CH_2), 32.2 (CH_2), 30.2 (CH_2), 28.1 (CH_2), 27.2 ($2\times CH_2$), 26.6 ($2\times CH_2$), 28.0 (CH_3-30), 24.7 (CH_3-26), 24.7 (CH_3-29), 24.4 (CH_3-24), 24.3 (CH_3-28), 22.2 (CH_3-27). LRMS *m/z* 519 (23, $[M+K]^+$), 503 (47, $[M+Na]^+$), 481 (12, $[M+H]^+$). HRMS Calcd for $C_{27}H_{44}O_7Na$ 503.2986, found 503.2988.

4.4.3. Compound 5. IR (neat): ν_{max} 1770 cm^{-1}. ^1H NMR (600 MHz, attributions by 2D-NMR): 3.85, 3.89, 3.93 (3H, overlapped m's, H-7, H-11, H-18), 3.72 (1H, dd, $J=9.6$, 5.8 Hz, H-14), 2.90, 2.77, 2.43, 2.40, 1.95, 1.89, 1.87, 1.57 1.55, (20H, overlapped m's, $10\times CH_2$),.1.02 (3H, s, Me-28), 1.11 (3H, s, Me-27), 1.33 (6H, s, Me-26, Me-29), ^{13}C NMR (150 MHz, attributions by 2D-NMR): δ 179.6 (C), 178.7 (C), 87.3 (C) 87.1 (C), 86.8 (C), 86.7 (C), 86.3 (CH-18), 85.6 (CH-14), 85.5 (CH-11), 84.3 (CH-7), 34.2 ($2\times CH_2$), 32.5 ($2\times CH_2$), 30.3 ($2\times CH_2$), 28.2 (CH_2), 27.0 (CH_2), 26.7 (CH_2), 26.3 (CH_2), 24.2 (CH_3-29, CH_3-26), 23.7 (CH_3-28), 20.7 (CH_3-27). LRMS *m/z* 475 (100, $[M+K]^+$), 459 (88, $[M+Na]^+$). HRMS Calcd for $C_{24}H_{36}O_7Na$ 459.2359, found 459.2358.

4.5. Mono-THF diols 8 and 11

A solution of squalene (160 mg, 0.39 mmol) in CH_2Cl_2 (40 mL) was treated with *N*-methylmorpholine *N*-oxide monohydrate (420 mg, 3.12 mmol) and CSA (1.09 g, 4.7 mmol) followed by osmium tetroxide (10 mg, 0.039 mmol, 10%) and the solution was stirred at room temperature for 24 h. The reaction was quenched with saturated aqueous sodium thiosolfate and $NaHCO_3$ and the biphasic solution was extracted with CH_2Cl_2 (3×20 mL), then the organic phase was dried with Na_2SO_4 and evaporated. The oily residue was purified by HPLC (hexane/EtOAc 70:30) to give unreacted squalene (16 mg, 10%, $t_R=6$ min), mono-THF diol **8** (13 mg, 7%, $t_R=$ 15.4 min) and mono-THF diol **11** (11 mg, 6%, $t_R=9.6$ min).

4.5.1. Compound 11. IR (neat): ν_{max} 3412 (br) cm^{-1}. ^1H NMR (200 MHz): δ 5.11 (4H, m, olefinic protons), 3.88 (1H, t, $J=7.5$ Hz, H-7), 3.39 (1H, dd, $J=8.7$, 2.7 Hz, H-11), 1.68 (6H, s, $2\times$Me), 1.63, 1.62 (3H each, s's, $2\times$Me), 1.60 (6H, s, $2\times$Me), 1.14, 1.08, (3H each, s's, $2\times$C(O)Me). LRMS *m/z* 499 (97, $[M+K]^+$), 483 (100, $[M+Na]^+$). HRMS Calcd for $C_{30}H_{52}O_3Na$ 483.3816, found 483.3822.

4.6. Bis-THF diols 12 and 13

To a solution of **8** (25 mg, 0.054 mmol) in CH_2Cl_2 (2.5 mL) was added MCPBA (10.2 mg, 0.059 mmol) at 0 °C and the mixture was stirred at this temperature for 1.5 h. Then the reaction was quenched with saturated aqueous $NaHCO_3$ and extracted with CH_2Cl_2 (3×10 mL). The organic phase was dried over Na_2SO_4 and evaporated. The residue was purified by HPLC (hexane/EtOAc 65:35) to yield unreacted **8** (6.2 mg, 25%, $t_R=12.5$ min), the two diasteroisomeric C_{10}–C_{11} epoxides **14** (major isomer: 7.7 mg, 30%, $t_R=18.2$ min; minor isomer: 6.1 mg, 24%, $t_R=15.7$ min) and bis-THF diols **12** (2.3 mg, 9%, $t_R=24.6$ min) and **13** (2.3 mg, 9%, $t_R=26.8$ min).

4.6.1. Major epoxide. IR (neat): ν_{max} 3420 (br) cm^{-1}. ^1H NMR (200 MHz): δ 5.12 (3H, m, olefinic protons), 3.85 (1H, t, $J=7.0$ Hz, H-3), 3.38 (1H, dd, $J=5.8$, 4.4 Hz, H-7), 2.70 (1H, t, $J=5.8$ Hz, H-11), 1.67 (3H, s, Me), 1.60, 1.59 (overall 9H, s's, Me), 1.25, 1.24, 1.15, 1.11 (3H each, s's, $4\times$C(O)Me). LRMS *m/z* 515 (48, $[M+K]^+$), 499 (100, $[M+Na]^+$).

4.6.2. Minor epoxide. IR (neat): ν_{max} 3407 (br) cm^{-1}. ^1H NMR (200 MHz): δ 5.18 (2H, m, olefinic protons), 5.07 (1H, t, $J=7.0$ Hz, olefinic proton), 3.85 (1H, t, $J=7.0$ Hz, H-3), 3.38 (1H, dd, $J=5.8$, 4.4 Hz, H-7), 2.70 (1H, t, $J=$ 5.8 Hz, H-11), 1.68 (3H, s, Me), 1.60 (9H, s, $3\times$Me), 1.24 (6H, s, $2\times$C(O)Me), 1.16, 1.12 (3H each, s's, $2\times$C(O)Me). LRMS *m/z* 515 (59, $[M+K]^+$), 499 (100, $[M+Na]^+$).

4.6.3. Compound 12. IR (neat): ν_{max} 3447 (br) cm^{-1}. ^1H NMR (200 MHz): δ 5.12 (3H, m, olefinic protons), 3.85 (2H, m, H-3 and H-7), 3.52 (1H, dd, $J=7.7$, 1.2 Hz, H-11), 1.67, 1.62 (3H each, s's, $2\times$Me), 1.59 (6H, s, $2\times$Me), 1.25, 1.19, 1.12, 1.08 (3H each, s's, $4\times$C(O)Me). LRMS *m/z* 515

(69, $[M+K]^+$), 499 (100, $[M+Na]^+$), 477 (27, $[M+H]^+$).HRMS Calcd for $C_{30}H_{52}O_4Na$ 499.3765, found 499.3771.

4.6.4. Compound 13. IR (neat): ν_{max} 3446 (br) cm^{-1}. ^1H NMR (200 MHz): δ 5.09 (3H, m, olefinic protons), 3.91, 3.82 (1H each, t's, $J=7.4$, 7.0 Hz, respectively, H-3 and H-7), 3.54 (1H, dd, $J=9.6$, 2.8 Hz, H-11), 1.67, 1.62 (3H each, s's, 2×Me), 1.59 (6H, s, 2×Me), 1.26, 1.14, 1.13, 1.11 (3H each, s's, 4×C(O)Me). LRMS *m/z* 515 (49, $[M+K]^+$), 499 (100, $[M+Na]^+$), 477 (12, $[M+H]^+$). HRMS Calcd for $C_{30}H_{52}O_4Na$ 499.3765, found 499.3752.

4.7. Diacetate 15

$LiAlH_4$ (45.5 mg, 1.20 mmol) was added in portions to a solution of bis-lactone **5** (240 mg, 0.55 mmol) in dry ethyl ether (5 mL) at 0 °C. The mixture was allowed to warm to room temperature over 1 h and then quenched by dropwise addition of wet ethyl ether and water. After all inorganic materials were precipitated, the solid was filtered and washed with EtOAc and the organic phase was evaporated to give an oily products that was treated with Ac_2O in pyridine at room temperature for 16 h. Then the mixture was partitioned between EtOAc and 0.1 M HCl (20 mL) and the organic phase was washed with saturated aqueous $NaHCO_3$ (20 mL), dried over Na_2SO_4 and evaporated to give a colorless oil. HPLC purification (hexane/EtOAc 35:65) yielded diacetate **15** (209 mg, 72% for two steps, $t_R=$ 11.0 min).

4.7.1. Compound 15. IR (neat): ν_{max} 3403 (br), 1733 cm^{-1}. ^1H NMR (400 MHz): δ 4.02 (4H, m, 2×CH_2OAc), 3.90 (1H, dd, $J=8.1$, 3.9 Hz, THF proton), 3.87 (1H, dd, $J=7.9$, 5.1 Hz, THF proton), 3.78, (1H, m, THF proton), 3.74 (1H, dd, $J=8.8$, 5.9 Hz, THF proton), 2.05 (6H, s, 2×OAc), 1.12, 1.11, 1.06, 1.02, (3H each, s's, 4×C(O)Me). ^{13}C NMR (100 MHz): δ 171.1, 171.1, 85.8, 85.13, 85.09, 84.7, 84.1, 83.2, 73.6, 71.9, 65.3, 65.0, 36.5, 36.3, 35.1, 34.7, 27.9, 27.2, 26.4, 25.7, 24.7, 24.0, 23.3, 23.1, 21.8, 21.1, 21.0, 20.9. LRMS *m/z* 567 (50, $[M+K]^+$), 551 (100, $[M+Na]^+$), 477 (26, $[M+H]^+$). HRMS Calcd for $C_{28}H_{48}O_9Na$ 551.3197, found 551.3200.

4.8. Bis lactone 16

To a solution of **15** (30 mg, 0.056 mmol) in CH_2Cl_2 (1.5 mL) was added celite (300 mg), PCC (120.7 mg, 0.56 mmol) and AcOH (480 μL, 8.4 mmol) and the resulting heterogeneous mixture was stirred at room temperature for 24 h and the loaded on a silica gel column. Elution with $CHCl_3$/MeOH 9:1 afforded a colorless oil. HPLC purification gave **16** (6.7 mg, 47%, $t_R=$23.6 min), 2-oxo-penten-1-yl acetate (5.6 mg, 70%, $t_R=$11.3 min)[15] along with a monolactone acetate product derived from the cleavage of only one of the two side chains (1.7 mg, 7.5%, $t_R=$27.8 min).

4.8.1. Compound 16. IR (neat): ν_{max} 3527 (br), 1770 cm^{-1}. ^1H NMR (200 MHz): δ 3.88 (2H, ddd, $J=9.3$, 5.2, 3.2 Hz, H_{11} and H_{14}), 2.68 (2H, ddd, $J=17.8$, 10.3, 9.3 Hz, H_a-8 and H_a-17), 2.50 (2H, ddd, $J=17.7$, 10.8, 3.7 Hz, H_b-8 and H_b-17), 2.36 (2H, ddd, $J=16.5$, 9.9, 3.7 Hz, H_a-9 and H_a-16), 1.97 (6H, m, H_b-9 and H_b-16, H_2-12 and H_2-13), 1.31 (6H, s, 2×C(O)Me). ^{13}C NMR (50 MHz): δ 177.2, 85.9, 85.7, 32.4, 29.7, 26.5, 23.5. LRMS *m/z* 307 (43, $[M+K]^+$), 291 (100, $[M+Na]^+$), 269 (60, $[M+H]^+$). HRMS Calcd for $C_{14}H_{20}O_5Na$ 291.1209, found 291.1210.

4.8.2. Monolactone acetate. IR (neat): ν_{max} 3412 (br), 1772, 1737 cm^{-1}. ^1H NMR (200 MHz): δ 4.08 (2H, m, CH_2OAc), 3.96–3.78 (3H, m, THF protons), 2.04 (s, acetate), 1.33, 1.09, 1.02 (3H each, s's, 3×C(O)Me). LRMS *m/z* 437 (43, $[M+K]^+$), 421 (100, $[M+Na]^+$), 399 (35, $[M+H]^+$).

4.9. Tetra-THF diols 17 and 18

To a solution of tris-THF **10** (25 mg, 0.051 mmol) in CH_2Cl_2 (2.5 mL) was added MCPBA (10 mg, 0.056 mmol) at 0 °C and the mixture was stirred at the same temperature for 1.5 h. After the reaction was quenched with saturated aqueous $NaHCO_3$ the mixture was extracted with CH_2Cl_2 (3×10 mL), the organic phase was dried over Na_2SO_4 and evaporated. The residue was purified by HPLC (hexane/EtOAc 80:20) to give unreacted **10** (7.5 mg, 30%, $t_R=$ 10.2 min), a mixture of unseparable C_{18}–C_{19} diastereomeric epoxides contaminated by a 20% amount of terminal epoxides (7.3 mg, 47%, $t_R=30.5$ min) along with a mixture of the two diastereomeric terminal epoxides (3.6 mg, 25%, $t_R=29$ min).

4.9.1. Mixture of C_{18}–C_{19} epoxides. IR (neat): ν_{max} 3423 (br) cm^{-1}. ^1H NMR (200 MHz, data from the mixture contaminated by terminal epoxides): δ 5.07 (t, $J=7.0$ Hz, olefinic protons), 3.95–3.70 (m, H-3, H-7, H-11 and H-14), 2.70 (t, $J=6.2$ Hz, H-18), 1.67, 1.61, (s's, 2×Me), 1.27, 1.24, 1.17, 1.12 1.02, 0.97 (s,s, 6×C(O)Me). LRMS: *m/z* 547 (43, $[M+K]^+$), 531 (100, $[M+Na]^+$).

4.9.2. Mixture of terminal epoxides. IR (neat): ν_{max} 3420 (br) cm^{-1}. ^1H NMR (200 MHz): δ 5.20 (bt, $J=7.0$ Hz, olefinic protons), 4.09 (bs, OH), 3.94–3.78 (m, H-3, H-7, H-11 and H-14), 2.70 (t, $J=6.4$ Hz, H-22), 1.62, (s, Me), 1.29, 1.25, 1.24, 1.17, 1.12 1.04, 0.98 (3H each, s's, 7×C(O)Me). LRMS: *m/z* 547 (32, $[M+K]^+$), 531 (100, $[M+Na]^+$).

To the mixture of tris-THF epoxides obtained as above (7 mg, 0.013 mmol) in CH_2Cl_2 (1 mL) was added CSA (0.75 mg, 0.0032 mmol) at 0 °C and the resulting solution was stirred at the same temperature for 45 min. Then the reaction was quenched with saturated aqueous $NaHCO_3$ and extracted with CH_2Cl_2 (3×5 mL). The organic phase was dried over Na_2SO_4 and evaporated. The residue was purified by HPLC (hexane/EtOAc 75:25) to give two diastereomeric tetra-THF diols (**17**/**18**, *exo* products) (major isomer: 2.9 mg, 45%, $t_R=14.5$ min; minor isomer: 1.5 mg, 23%, $t_R=16.5$ min) along with the tetrahydropyrane-containing isomers **19** and **20** *(endo products)* (major isomer: 1.2 mg, 18%, $t_R=17.7$ min; minor isomer: 0.9 mg, 14%, $t_R=$ 19.5 min).

4.9.3. Major tetra-THF diol. IR (neat): ν_{max} 3403 (br) cm^{-1}. ^1H NMR (600 MHz): δ 5.12 (1H, bt, $J=6.7$ Hz, olefinic proton), 3.95 (1H, dd, $J=7.0$, 7.0 Hz, THF proton),

3.88 (1H, dd, $J=7.5$, 7.5 Hz, THF proton), 3.80, 3.77, 3.75 (3H, overlapped m's, 3×THF protons), 1.69, 1.62 (3H each, s's, 2×Me), 1.23, 1.18, 1.15, 1.14, 1.08 1.07 (3H each, s's, 6×C(O)Me). ^{13}C NMR (150 MHz): δ 131.6 (C), 124.9 (CH), 86.7 (CH), 85.2 (C), 85.1 (CH), 84.8 (CH), 84.6 (CH), 84.2 (C), 84.0 (C), 82.3 (CH), 72.1 (C), 71.7 (C), 34.5 (CH_2), 33.8 (CH_2), 29.7 (CH_2), 29.5 (CH_2), 29.1 (CH_2), 28.0 (Me), 27.4 (CH_2), 26.8 (CH_2), 26.7 (CH_2), 26.0 (CH_2), 25.6 (CH_3), 25.1 (CH_3), 24.2 (CH_3), 24.0 (CH_2), 23.9 (2×CH_3), 22.7 (CH_3), 17.6 (CH_3). LRMS: *m/z* 547 (40, $[M+K]^+$), 531 (100, $[M+Na]^+$), 509 (15, $[M+H]^+$). HRMS Calcd for $C_{30}H_{52}O_6Na$ 531.3663, found 531.3673.

4.9.4. Minor tetra-THF diol. IR (neat): ν_{max} 3395 (br) cm^{-1}. ^{1}H NMR (600 MHz): δ 5.11 (1H, bt, $J=7.9$ Hz, olefinic proton), 3.98 (1H, bdd, $J=8.4$, 8.4 Hz, THF proton), 3.89–3.71 (4H, overlapped m's, 4×THF protons), 1.68, 1.61 (3H each, s's, 2×Me), 1.20, 1.19, 1.17, 1.13, 1.08, 1.06 (3H each, s's, 6×C(O)Me). ^{13}C NMR (150 MHz): δ 131.1 (C), 124.6 (CH), 84.2 (C), 83.9 (C), 83.7 (C), 72.7 (C), 72.2 (C), 35.2 (CH_2), 34.7 (CH_2), 29.9 (2×CH_2), 27.7 (2×CH_2), 27.5 (2×CH_2), 26.8 (2×CH_2), 85.5 (CH), 85.4(CH), 85.1 (CH), 84.9 (CH), 82.9 (CH), 28.3 (CH_3), 24.9 (CH_3) 24.6 (CH_3), 24.5 (CH_3), 24.0 (2×CH_3), 20.9 (CH_3), 17.4 (CH_3). LRMS: *m/z* 547 (20, $[M+K]^+$), 531 (100, $[M+Na]^+$), 509 (78, $[M+H]^+$). HRMS Calcd for $C_{30}H_{52}O_6Na$ 531.3663, found 531.3649.

4.9.5. Major THP-containing isomer (19). IR (neat): ν_{max} 3412 (br) cm^{-1}. ^{1}H NMR (400 MHz): δ 5.08 (1H, bt, $J=$ 7.0 Hz, olefinic proton), 4.03–3.68 (4H, overlapped m's, 4×THF protons), 3.39 (1H, dd, $J=4.0$, 3.0 Hz, H-19), 1.68, 1.61 (3H each, s's, 2×Me), 1.24, 1.21, 1.16, 1.12, 1.09, 1.08 (3H each, s's, 6×C(O)Me). LRMS: *m/z* 547 (27, $[M+K]^+$), 531 (100, $[M+Na]^+$), 509 (56, $[M+H]^+$). HRMS Calcd for $C_{30}H_{52}O_6Na$ 531.3663, found 531.3674.

4.9.6. Minor THP-containing isomer (20). IR (neat): ν_{max} 3407 (br) cm^{-1}. ^{1}H NMR (400 MHz): δ 5.09 (1H, bt, $J=$ 7.0 Hz, olefinic proton), 3.94–3.70 (4H, overlapped m's, 4×THF protons), 3.47 (1H, dd, $J=10.8$, 5.2 Hz, H-19), 1.67, 1.60 (3H each, s's, 2×Me), 1.22, 1.18, 1.14, 1.13, 1.10, 1.06 (3H each, s's, 6×C(O)Me). LRMS: *m/z* 547 (14, $[M+K]^+$), 531 (100, $[M+Na]^+$), 509 (55, $[M+H]^+$). HRMS Calcd for $C_{30}H_{52}O_6Na$ 531.3663, found 531.3667.

4.10. Synthesis of tetra-THF diol 22

To a solution of monolactone **3** (100 mg, 0.21 mmol) in dry THF (1.5 mL) DIBALH (1.0 M in THF, 0.42 mmol, 450 μL) was added at −78 °C and the solution was stirred at the same temperature for 1 h. Saturated aqueous NH_4Cl was added and the mixture was extracted with $CHCl_3$ (3× 10 mL) then dried over Na_2SO_4 and evaporated to give crude lactol **21** (70 mg, 70%). Further HPLC purification ($CHCl_3$, $t_R=22$ min) afforded pure **21**.

4.10.1. Compound 21. IR (neat): ν_{max} 3420 (br) cm^{-1}. ^{1}H NMR (400 MHz): δ 5.33 (1H, d, $J=3.8$ Hz, H-22), 3.98–3.74 (5H, overlapped m's, 5×THF protons), 1.22 (3H, s, C(O)Me), 1.14, 1.13 (6H each, s's, 4×C(O)Me), 1.06 (3H, s, C(O)Me). ^{13}C NMR (100 MHz): δ 98.8, 85.0, 84.9, 84.9, 84.6, 83.9, 83.7, 83.6, 82.4, 71.6, 34.5, 34.3, 34.2, 33.9, 31.3, 28.0, 27.8, 27.0, 27.0, 26.6, 25.8, 24.7, 24.2, 24.0, 23.8, 23.7, 22.6. LRMS: *m/z* 521 (12, $[M+K]^+$), 505 (16, $[M+Na]^+$). HRMS Calcd for $C_{27}H_{46}O_7Na$ 505.3142, found 505.3129.

n-Butyllithium (1.6 M in hexane, 0.46 mmol) was dropwise added through syringe to a solution of isopropyltriphenilphosphonium iodide (200 mg, 0.46 mmol) in THF (2 mL) at 0 °C. After stirring at this temperature for 20 min, the deep red solution was brought to −78 °C and then transferred via cannula to a solution of **21** (63 mg, 0.13 mmol) in THF (2 mL) at −78 °C. The reaction mixture was allowed to warm to room temperature over 4 h. Then aqueous NH_4Cl was added and the mixture was extracted with ethyl ether (3×30 mL), the organic phase was dried over Na_2SO_4 and evaporated. The residue was purified by HPLC (hexane/EtOAc 30:70) to give unreacted **21** (19 mg, 30%, $t_R=$ 36 min) and terta-THF **22** (6 mg, 8–10% respect to reacted **21**, $t_R=16.5$ min).

4.10.2. Compound 22. IR (neat): ν_{max} 3461 (br) cm^{-1}. ^{1}H NMR (600 MHz): δ 5.11 (1H, bt, $J=7.0$ Hz, olefinic proton), 3.95 (1H, t, $J=7.1$ Hz, THF proton), 3.88 (1H, t, $J=7.6$ Hz, THF proton), 3.82–3.80 (3H, overlapped m's, 3×THF protons), 1.68, 1.61 (3H each, s's, 2×Me), 1.22, 1.15, 1.12, 1.08, 1.07, 1.06 (3H each, s's, 6×C(O)Me). ^{13}C NMR (150 MHz) δ 131.1 (C), 124.6 (CH), 86.2 (C), 85.3 (CH), 85.0 (C), 84.7 (CH), 84.6 (2×CH), 84.5 (C), 82.2 (CH), 71.7 (C), 71.2 (C), 34.3 (CH_2), 34.2 (CH_2), 33.7 (CH_2), 29.1 (CH_2), 27.7 (CH_3), 27.6 (CH_2), 26.7 (2×CH_2), 26.3 (CH_2), 26.2 (CH_2), 25.6 (CH_3), 25.5 (CH_2), 24.4 (CH_3), 24.2 (CH_3), 23.9 (CH_3), 23.7 (CH_3), 20.9 (CH_3), 17.4 (CH_3). LRMS: *m/z* 547 (15, $[M+K]^+$), 531 (20, $[M+Na]^+$), 509 (16, $[M+H]^+$). HRMS Calcd for $C_{30}H_{52}O_6Na$ 531.3663, found 531.3675.

Acknowledgements

We are grateful to MURST, Italy (PRIN 2003), for financial support in favour of this investigation and to the 'Centro di Metodologie Chimico-Fisiche dell'Università di Napoli 'Federico II' ' for NMR and MS facilities.

References and notes

1. Harding, W. W.; Lewis, P. A.; Jacobs, H.; McLean, S.; Reynolds, W. F.; Tay, L.-L.; Yang, J.-P. *Tetrahedron Lett.* **1995**, *36*, 9137–9140.
2. Itokawa, H.; Kishi, E.; Morita, H.; Takeya, K.; Iitaka, Y. *Chem. Lett.* **1991**, 2221–2222.
3. Suzuki, T.; Suzuki, M.; Kurusaki, A.; Matsumoto, T.; Kato, A.; Imanaka, Y.; Kurosawa, E. *Tetrahedron Lett.* **1985**, *38*, 1329–1332.
4. (a) Koert, U.; Stein, M.; Harms, K. *Angew. Chem., Int. Ed. Engl.* **1994**, *33*, 1180–1182. (b) Koert, U.; Stein, M.; Wagner, H. *Liebigs Ann.* **1995**, 1415. (c) Wagner, H.; Harms, K.; Koert, U.; Meder, S.; Boheim, G. *Angew. Chem., Int. Ed.* **1996**, *35*, 2643–2646. (d) Cross, G. G.; Fyles, T. M.; James, T. D.; Zojaji, M. *Synlett* **1993**, 449–480.

5. (a) Zeng, L.; Oberlies, N. H.; Shi, G.; Gu, Z.-M.; He, K.; McLaughlin, J. *Nat. Prod. Rep.* **1996**, *13*, 275–306. (b) Alali, F. Q.; Liu, X.-X.; McLaughlin, J. *Nat. Prod. Rep.* **1999**, *62*, 504–540.
6. For some selected examples of *Annonaceous* acetogenin syntheses see: (a) Cecil, A. R. L.; Brown, R. C. D. *Org. Lett.* **2002**, *4*, 3715–3718. (b) Koert, U. *Tetrahedron Lett.* **1999**, *40*, 5979–5982. (c) Trost, B. M.; Calkins, T. L.; Bochet, C. G. *Angew. Chem., Int. Ed.* **1997**, *36*, 2632–2635. (d) Marshall, J. A.; Hinkle, K. W. *J. Org. Chem.* **1997**, *62*, 5989–5995. (e) Marshall, J. A.; Chen, M. *J. Org. Chem.* **1997**, *62*, 5996–6000.
7. (a) Bifulco, G.; Caserta, T.; Gomez-Paloma, L.; Piccialli, V. *Tetrahedron Lett.* **2002**, *43*, 9265–9269. (b) Bifulco, G.; Caserta, T.; Gomez-Paloma, L.; Piccialli, V. *Tetrahedron Lett.* **2003**, *44*, 5499–5503.
8. Keinan, E.; Sinha, S. C. *Pure Appl. Chem.* **2002**, *74*, 93–105 and in particular Ref. 21 therein.
9. Towne, T. B.; McDonald, F. E. *J. Am. Chem. Soc.* **1997**, *119*, 6022–6028.
10. (a) Baskaran, S.; Islam, I.; Chandrasekaran, S. *J. Org. Chem.* **1990**, *55*, 891–895. (b) Waddel, T. G.; Carter, A. D.; Miller, T. J. *J. Org. Chem.* **1992**, *57*, 381–383.
11. See Ref. 10 andSinha, S. C.; Sinha, A.; Sinha, S. C.; Keinan, E. *J. Am. Chem. Soc.* **1997**, *119*, 12014–12015.
12. McDonald, F. E.; Schultz, C. C. *Tetrahedron* **1997**, *53*, 16435–16448 and references therein.
13. Donohoe, T. J.; Butterworth, S. *Angew. Chem., Int. Ed.* **2003**, *42*(8), 948–951.
14. Hawskley, D.; Griffin, D. A.; Leeper *J. Chem. Soc., Perkin Trans 1* **2001**, 144–148.
15. Bravo, F.; McDonald, F. E.; Neiwert, W. A.; Do, B.; Hardcastle, K. I. *Org. Lett.* **2003**, *5*, 2123–2126.
16. Morimoto, Y.; Iwai, T.; Kinoshita, T. *Tetrahedron Lett.* **2001**, *44*, 6307–6309.
17. Maerchker, A. In *Organic Reactions*, Wiley: New York, 1965; pp 270–490.
18. (a) Xiong, Z.; Corey, J. *J. Am. Chem. Soc.* **2000**, *122*, 4831–4832. (b) Xiong, Z.; Corey, J. *J. Am. Chem. Soc.* **2000**, *122*, 9328–9329. (c) Morimoto, Y.; Iwai, T.; Kinoshita, T. *J. Am. Chem. Soc.* **2000**, *122*, 7124–7125. (d) Hioki, H.; Kanehara, C.; Ohnishi, Y.; Umemori, Y.; Sakai, H.; Yoshio, S.; Matsushita, M.; Kodama, M. *Angew. Chem., Int. Ed.* **2000**, *39*, 2552–2554.
19. See Ref. 8 and: Koert, U.; Stein, M.; Wagner, H. *Chem. Eur. J.* **1997**, *3*, 1170–1179 and references cited therein.
20. (a) Morimoto, Y.; Iwai, T.; Yoshimura, T.; Takamasa, K. *Bioorg. Med. Chem. Lett.* **1998**, *8*, 2005–2010. (b) Wang, X.; Erickson, S. D.; Iimori, T.; Still, W. C. *J. Am. Chem. Soc.* **1992**, *114*, 4128–4137 and references cited therein.
21. (a) Sinha, S. C.; Keinan, E.; Sinha, S. C. *J. Am. Chem. Soc.* **1998**, *120*, 9076–9077. (b) Morimoto, Y.; Iwai, T. *J. Am. Chem. Soc.* **1998**, *120*, 1633–1634. (c) Morimoto, Y.; Iwai, T.; Kinoshita, T. *J. Am. Chem. Soc.* **1999**, *121*, 6792–6797.