Four-step access to the sesquiterpene natural product presilphiperfolan-1β-ol and unnatural derivatives via supramolecular catalysis

Leonidas-Dimitrios Syntrivanis,[†] Shani Levi,[§] Alessandro Prescimone,[†] Dan T. Major,[§] Konrad Tiefenbacher*[,†,‡]

[†] Department of Chemistry, University of Basel, Mattenstrasse 24a, 4058 Basel, Switzerland

[‡] Department of Biosystems Science and Engineering, ETH Zurich, Mattenstrasse 24, 4058 Basel, Switzerland

[§] Department of Chemistry, Bar-Ilan University, Ramat-Gan 52900, Israel

Supporting Information Placeholder

ABSTRACT: Terpenes constitute one of the most structurally varied classes of natural products. A wide range of these structures are produced in nature by type I terpene cyclase enzymes, but such reactivity has proven difficult to reproduce in solution with man-made systems. Herein we report the shortest synthesis of the tricyclic sesquiterpene presilphiperfolan-1β-ol to date, utilizing the supramolecular resorcinarene capsule as catalyst for the key step. This synthetic approach also allows access to unnatural derivatives of the natural product, which would not be accessible through the biosynthetic machinery. Additionally, this study provides useful insight into the biosynthesis of the presilphiperfolanol natural products, including the first direct experimental evidence for the proposed biosynthetic connection between caryophyllene and the presilphiperfolanols.

The tail-to-head terpene (THT) cyclization is one of the most complex reactions observed in nature, giving rise to a vast amount of complex terpene structures starting from a handful of linear precursors.[1] Despite its great synthetic potential, the THT cyclization has proven very difficult to reproduce in solution, as premature quenching of reactive intermediates, for instance by the cleaved leaving group, gives rise to complex mixtures mainly favoring monocyclic products.[2] The Shenvi group succeeded in forming a few polycyclic sesquiterpene structures by covalently linking the leaving group to the molecule.[3] Our group employed a different approach, by using the hexameric resorcinarene capsule **I**,[4,5] formed from the assembly of monomers **1**, as a reaction chamber (Figure 1a). This supramolecular container is able to catalyse the THT cyclization, likely via the stabilization of cationic transition states within its cavity.[6] We also demonstrated that the limited selectivity encountered in the capsule-catalysed cyclizations of linear sesquiterpene precursors could be overcome through the use of a conformationally restricted substrate, in this way achieving the selective preparation of the natural product isolongifolene.[7] However, isolongifolene is a commercially available compound, and it is not known to possess any biological activity. Identifying capsule-catalyzed cyclizations that lead to complex, biologically active terpenes, hard to synthesize by other means, would be highly desirable and showcase the applicability of supramolecular catalysis.

Cyclase enzymes exercise control over the terpene cyclization pathway via their cavity shape, which facilitates specific substrate folding,[8] as well as via specific interactions with active site amino acid residues and cofactors.[9] However, as recently argued by Tantillo,[10] evidence from gas-phase computational studies indicates that not every step is enzyme catalysed, as the intrinsic reactivity of the cation itself can dictate the reaction outcome in some cases. We recognized that the resorcinarene capsule **I** could be utilized to exploit this inherent reactivity: identifying a key cationic intermediate and generating it within its confines should enable the downstream reactions. To probe this possibility in practice, we chose to investigate the postulated biosynthetic connection between caryophyllene and the presilphiperfolanol family of natural products.[11] Three members of this family have been isolated to date: (−)-presilphiperfolan-9α-ol [(−)-**2**],[12] (−)-presilphiperfolan-8α-ol [(−)-**3**],[13] and (−)-presilphiperfolan-1β-ol [(−)-**4**] (Figure 1 b).[14,15] These natural products hold significant importance in the study of terpene biosynthesis, as their strained framework[16] is believed to represent a branching point that links together a number of different biosynthetic pathways.[11] They also exhibit interesting biological activities like antimycobacterial and insect antifeedant properties.[17,18] Their complex tricyclic skeleton has proven to be a challenging target for total synthesis; hitherto reported total syntheses of the presilphiperfolanols have required 13-17 linear steps from commercially available materials.[15,19,20]

The current proposal for the presilphiperfolanol biosynthesis (Figure 1b)[21,22] posits that the initial cyclization of farnesyl pyrophosphate, according to density functional theory (DFT) calculations via its allylic isomer nerolidyl pyrophosphate,[23] leads to the humulenyl cation **5**. After cyclization to the caryophyllenyl cation **6**, a concerted 1,2-alkyl shift/cyclization cascade leads to structure **8**, the direct precursor to presilphiperfolan-9α-ol **2**. From here, isotopic labelling[22] and computational studies[23] support a 1,3-hydride shift (path a, red) as the path that leads to cation **11**; capture of this intermediate by water leads to presilphiperfolan-8α-ol **3**, while further rearrangements are believed to generate a number of other sesquiterpene classes.[13,21,24] The reassignment of the C-9 methyl configuration by Stoltz et al.[15] allowed an alternative 1,2-hydride shift (path b, blue) to be proposed as the pathway that leads to presilphiperfolan-1β-ol (**4**), however so far no further evidence to support this has appeared in the literature. The proposed intermediacy of the caryophyllenyl cation **6**[24c,25] in their biosynthesis has led to a number of studies aimed at a biomimetic synthesis of the presilphiperfolanols from caryophyllene and related compounds in solution; however no such approach has so far provided a natural presilphiperfolanol (see SI for more details).[26,27,28] With this context in mind, we theorized that generation of the caryophyllenyl cation **6**

within the stabilizing environment of the resorcinarene capsule could allow the cascade just described to take place, leading to the formation of the presilphiperfolanol framework.

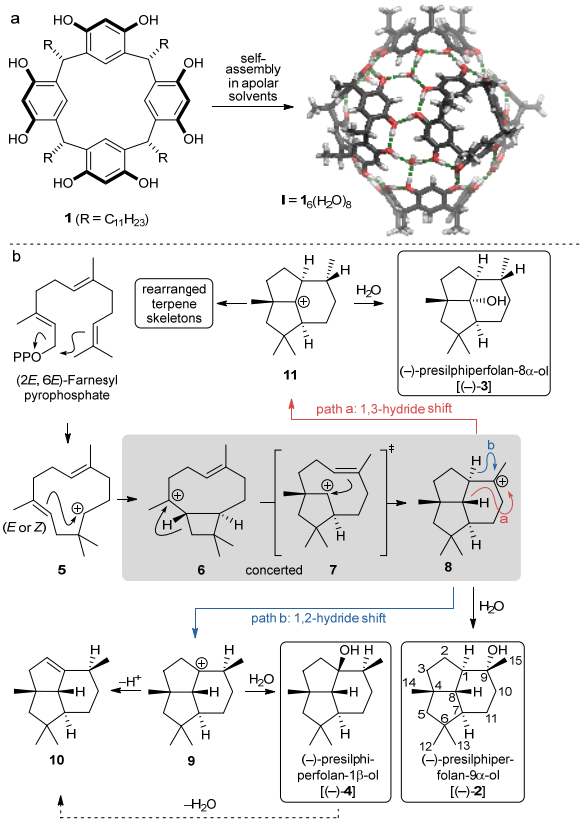
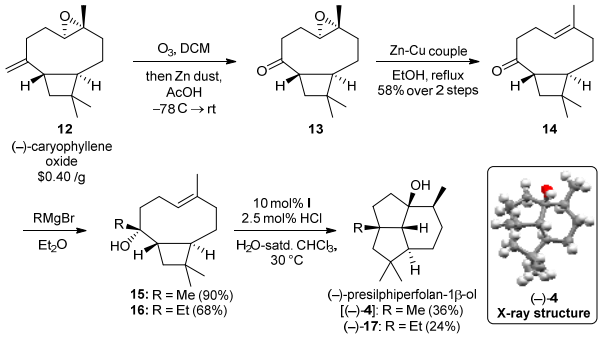


Figure 1. a. Self-assembly of monomer **1** into hexameric capsule **I**; b. Proposed biosynthesis of the presilphiperfolanol family of natural products.

Alcohol **15** (Scheme 1) was chosen as the substrate for our experiments. This was prepared in three steps from inexpensive ($0.4 /g)[29] commercially available (–)-caryophyllene oxide (**12**) following literature procedures;[26] multigram quantities of this alcohol can be readily accessed. In a first experiment, **15** was subjected to reaction with 10 mol% of capsule **I** and 3 mol% of HCl in $CDCl_3$, conditions previously identified by us as optimal for the capsule-catalyzed THT cyclization of sesquiterpenes.[6a-d] Gratifyingly the formation of presilphiperfolan-1β-ol (**4**) as a major product (13% GC yield after 4 days) was observed, along with rearranged alkene **20** (11% GC yield, Figure 2a). The latter is presumably formed through a 1,3-hydride shift of cation **9** (Figure 2c), followed by a methyl shift and elimination. To confirm the structure of the rearranged alkene **20**, a sample isolated after preparative scale reaction (see SI) was subjected to allylic oxidation; further oxidation to the aldehyde and formation of the corresponding semicarbazone gave a crystalline derivative which provided further proof of the compound's structure via X-ray crystallography (Figure 2c). Interestingly, this compound presents, to our knowledge, a previously unknown substitution pattern of the tricyclic presilphiperfolanol skeleton. Other components of the reaction mixture were identified as elimination product **10** and caryophyllene, the latter presumably deriving from dehydration of the starting material.

Scheme 1. Synthesis of (–)-presilphiperfolan-1β-ol and of ethyl substituted analogue (–)-17 utilizing the capsule-catalysed cyclization cascade as the key step.



We next sought to identify optimal conditions for the formation of presilphiperfolan-1β-ol and its derivatives. Pleasingly, it was found that carrying out the reaction in H_2O-saturated $CDCl_3$ slowed down the formation of rearranged alkene **20**, while at the same time increasing the yield of desired presilphiperfolan-1β-ol to synthetically very useful 42% (Figure 2b). The reaction rate was slower than with untreated $CDCl_3$, with conversion reaching a plateau at ~85%; addition of further HCl at this point was ineffective as it increased the rate of formation of rearranged alkene **20**. Carrying out the reaction with higher amounts of HCl favoured formation of the rearranged alkene **20** and of caryophyllene. Aromatic media (benzene and toluene) were also assayed but proved inferior to $CDCl_3$. Interestingly when H_2O-saturated DCM was used, the main byproduct observed was the direct elimination product **10**; only traces of rearrangement product **20** were formed, while the yield of presilphiperfolan-1β-ol was comparable to that observed in the reaction in $CDCl_3$. Under these conditions, prolonging the reaction time eventually led to complete conversion of presilphiperfolan-1β-ol into elimination product **10** (39% GC yield after 20 d). Lastly, rearranged alkene **20** could be obtained as the main product by prolonging the reaction time when using untreated $CDCl_3$ as the solvent, with complete conversion observed after 15 days (21% GC yield).

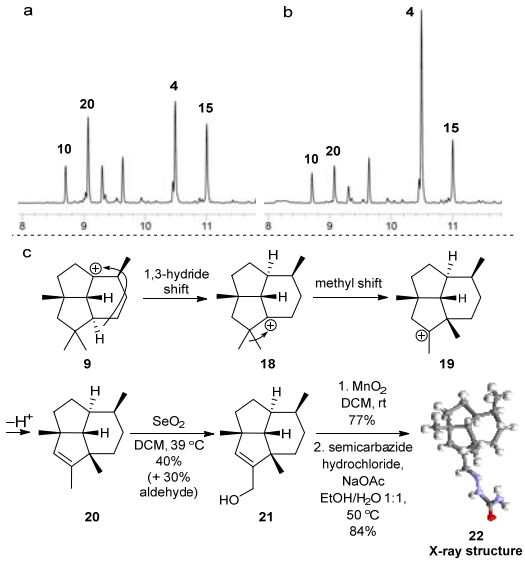


Figure 2. a. Gas chromatogram of reaction in untreated $CDCl_3$ after 4 d; b. Gas chromatogram of reaction in H_2O-satd. $CDCl_3$ after 7 d; c. Proposed mechanism for the formation of rearranged alkene **20** and synthesis of a crystalline derivative.

Control experiments were carried out to confirm that the reaction was taking place inside the cavity of capsule **I**. No product formation was observed when the reaction was carried out in the absence of capsule, or without any added HCl. Similarly, no reaction was observed

when the cavity of the capsule was blocked with a strong-binding guest (Bu_4NBr, 1.5 eq with respect to capsule). Taken together, these results indicate that the synergistic action between capsule and acid is essential for the catalytic activity, in accordance with what was observed in our previous studies.[6a-d] Interestingly, conversion of presilphiperfolan-1β-ol to either **10** or **20** also failed to take place in the absence of capsule, indicating that these are also capsule catalysed processes.

To demonstrate the applicability of this approach to a total synthesis of the natural product, a preparative scale reaction was carried out, using optimized conditions (Scheme 1). A slightly lower (2.5 mol%) loading of HCl was used as this was found to provide a better reaction profile with regards to formation of rearranged alkene **20** on this scale. The reaction was stopped after 9 days (approximately 80% conversion), providing presilphiperfolan-1β-ol in 36% isolated yield. Optical rotation measurement was consistent with the literature value[15] (within experimental error, see SI for more detailed discussion), as expected given enantiomerically pure starting material was utilized. This constitutes the shortest synthesis of (−)-presilphiperfolan-1β-ol to date, in four steps from inexpensive commercially available caryophyllene oxide. Additionally, X-ray crystallographic analysis of crystals of the natural product thus obtained (Scheme 1) confirmed that its structure is identical to that of 9-*epi*-presilphiperfolan-1-ol reported by Joseph-Nathan et al.,[14c] therefore supporting Stoltz's structural reassignment.[15] One potential advantage of resorcinarene capsule **I** over natural enzymes is its capability to convert unnatural substrates; we therefore decided to also investigate the potential of this methodology to provide an unnatural, hitherto unknown presilphiperfolanol analogue. In the event, it was found that reaction successfully took place with ethyl-substituted precursor **16**, providing a C-4 ethyl analogue of presilphiperfolan-1β-ol in 24% isolated yield (32% GC yield) after 10 days.

This study reports the first formation of a presilphiperfolanol natural product from the caryophyllenyl cation **6**, thereby providing experimental evidence for its intermediacy in the biosynthesis of this family of natural products. Additionally, we recognized that no presilphiperfolan-8α-ol **3**, or any products presumed to derive from it, were identified in the reaction mixture. This suggests that in the cavity of the capsule, the 1,2-hydride shift that leads to **9** (Figure 1b, path b) is favored over the 1,3-hydride shift that leads to **11** (path a). To our knowledge the 1,2-hydride shift pathway had not been computationally investigated before, in contrast to the 1,3-hydride shift.[23] Therefore, to probe this issue, the relative energies of the two pathways were calculated (Figure 3, Table S-1). In agreement with our experimental observations, transition state **TSb** was found to be lower in free energy to transition state **TSa** by 3.3 kcal/mol, while the resulting carbocation **9** was also found to be more stable than carbocation **11** by 7.8 kcal/mol. Also the final product **4** is more stable than its counterpart **3** from path a (Table S-2). This supports our hypothesis that the resorcinarene capsule can be used to exploit the inherent reactivity of terpene frameworks, and in this way find application in the biomimetic synthesis of complex terpenes.

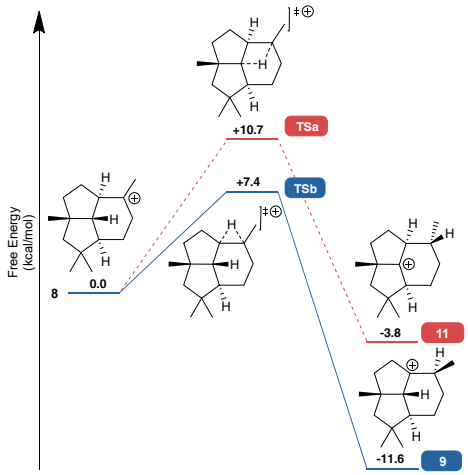


Figure 3. Calculated free energy profile for the pathways leading to presilphiperfolan-8α-ol (path a, red) and presilphiperfolan-1β-ol (path b, blue) [gas phase, M06-2X/6-311++G(2d,p)].

In conclusion, we have shown that the resorcinarene capsule **I** can catalyse the rearrangement of caryophyllenyl alcohol **15** into (−)-presilphiperfolan-1β-ol. This constitutes the shortest synthesis of this natural product to date, in four steps and 18.8% overall yield, while also allowing for the preparation of an unnatural analogue. Modification of the reaction conditions leads to the formation of an unnatural presilphiperfolene-like compound bearing a unique substitution pattern. Additionally this study provides the first direct experimental evidence for the biosynthetic connection between caryophyllene and the presilphiperfolanols, and gives useful insight into the pathways that lead to the different members of the presilphiperfolanol family. Rightfully, one could ask why the development of man-made terpene cyclase mimics would be required, when natural enzymes could be utilized instead. It is certainly conceivable that if the enzyme that produces presilphiperfolan-1β-ol in nature was identified, it could be used for this purpose. However, access to the hitherto unknown rearranged product **20** and to the ethyl derivative of presilphiperfolan-1β-ol is only possible via the supramolecular catalyst **I** so far, demonstrating for the first time the potential advantage of supramolecular catalysis over natural enzymes.

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website.
Experimental details and NMR spectra of new compounds (PDF)

AUTHOR INFORMATION

Corresponding Author

*konrad.tiefenbacher@unibas.ch; tkonrad@ethz.ch

Notes

The authors declare no competing financial interests.

ACKNOWLEDGMENT

This work was supported by funding from the European Research Council Horizon 2020 Programme [ERC Starting Grant 714620-TERPENECAT] and the Swiss National Science Foundation as part of the NCCR Molecular Systems Engineering.

REFERENCES

1. Christianson, D. W., Structural and Chemical Biology of Terpenoid Cyclases. *Chem. Rev.* **2017**, *117* (17), 11570-11648.
2. (a) Gutsche, C. D.; Maycock, J. R.; Chang, C. T., Acid-catalyzed cyclization of farnesol and nerolidol. *Tetrahedron* **1968**, *24* (2), 859-876; (b) Sakane, S.; Fujiwara, J.; Maruoka, K.; Yamamoto, H., Chiral leaving group. Biogenetic-type asymmetric synthesis of limonene and bisabolenes. *J. Am. Chem. Soc.* **1983**, *105* (19), 6154-6155; (c) de Meireles, A. L. P.; Costa, M. d. S.; da Silva Rocha, K. A.; Gusevskaya, E. V., Heteropoly acid catalyzed cyclization of nerolidol and farnesol: Synthesis of α-bisabolol. *Appl. Catal., A* **2015**, *502*, 271-275.
3. Pronin, S. V.; Shenvi, R. A., Synthesis of highly strained terpenes by non-stop tail-to-head polycyclization. *Nat. Chem.* **2012**, *4* (11), 915-920.
4. MacGillivray, L. R.; Atwood, J. L., A chiral spherical molecular assembly held together by 60 hydrogen bonds. *Nature* **1997**, *389* (6650), 469-472.
5. Avram, L.; Cohen, Y., Spontaneous Formation of Hexameric Resorcinarene Capsule in Chloroform Solution as Detected by Diffusion NMR. *J. Am. Chem. Soc.* **2002**, *124* (51), 15148-15149.
6. (a) Zhang, Q.; Tiefenbacher, K., Terpene cyclization catalysed inside a self-assembled cavity. *Nat. Chem.* **2015**, *7* (3), 197-202; (b) Zhang, Q.; Catti, L.; Pleiss, J.; Tiefenbacher, K., Terpene Cyclizations inside a Supramolecular Catalyst: Leaving-Group-Controlled Product Selectivity and Mechanistic Studies. *J. Am. Chem. Soc.* **2017**, *139* (33), 11482-11492; (c) Zhang, Q.; Catti, L.; Tiefenbacher, K., Catalysis inside the Hexameric Resorcinarene Capsule. *Acc. Chem. Res.* **2018**, *51* (9), 2107-2114; (d) Zhang, Q.; Catti, L.; Syntrivanis, L.-D.; Tiefenbacher, K., En route to terpene natural products utilizing supramolecular cyclase mimetics. *Nat. Prod. Rep.* **2019**, Advance Article; (e) Pahima, E.; Zhang, Q.; Tiefenbacher, K.; Major, D. T., Discovering Monoterpene Catalysis Inside Nanocapsules with Multiscale Modeling and Experiments. *J. Am. Chem. Soc.* **2019**, *141* (15), 6234-6246.
7. Zhang, Q.; Rinkel, J.; Goldfuss, B.; Dickschat, J. S.; Tiefenbacher, K., Sesquiterpene Cyclisations Catalysed inside the Resorcinarene Capsule and Application in the Short Synthesis of Isolongifolene and Isolongifolenone. *Nat. Catal.* **2018**, *1* (8), 609-615.
8. Deligeorgopoulou, A.; Allemann, R. K., Evidence for Differential Folding of Farnesyl Pyrophosphate in the Active Site of Aristolochene Synthase: A Single-Point Mutation Converts Aristolochene Synthase into an (E)-β-Farnesene Synthase. *Biochemistry* **2003**, *42* (25), 7741-7747.
9. (a) Major, D. T.; Freud, Y.; Weitman, M., Catalytic control in terpenoid cyclases: multiscale modeling of thermodynamic, kinetic, and dynamic effects. *Curr. Opin. Chem. Biol.* **2014**, *21*, 25-33; (b) Zhou, K.; Peters, R. J., Electrostatic effects on (di)terpene synthase product outcome. *Chem. Commun.* **2011**, *47* (14), 4074-4080; (c) Driller, R.; Janke, S.; Fuchs, M.; Warner, E.; Mhashal, A. R.; Major, D. T.; Christmann, M.; Brück, T.; Loll, B., Towards a comprehensive understanding of the structural dynamics of a bacterial diterpene synthase during catalysis. *Nat. Commun.* **2018**, *9* (1), 3971.
10. Tantillo, D. J., Importance of Inherent Substrate Reactivity in Enzyme-Promoted Carbocation Cyclization/Rearrangements. *Angew. Chem. Int. Ed.* **2017**, *56* (34), 10040-10045.
11. Hong, A. Y.; Stoltz, B. M., Biosynthesis and chemical synthesis of presilphiperfolanol natural products. *Angew. Chem. Int. Ed.* **2014**, *53* (21), 5248-5260.
12. Alberto Marco, J.; Sanz-Cervera, J. F.; Morante, M. D.; García-Lliso, V.; Vallès-Xirau, J.; Jakupovic, J., Tricyclic sesquiterpenes from Artemisia chamaemelifolia. *Phytochemistry* **1996**, *41* (3), 837-844.
13. Bohlmann, F.; Zdero, C.; Jakupovic, J.; Robinson, H.; King, R. M., Eriolanolides, eudesmanolides and a rearranged sesquiterpene from Eriophyllum species. *Phytochemistry* **1981**, *20* (9), 2239-2244.
14. (a) Melching, S.; König, W. A., Sesquiterpenes from the essential oil of the liverwort Conocephalum conicum. *Phytochemistry* **1999**, *51* (4), 517-523; (b) Pinto, S. C.; Leitão, G. G.; Bizzo, H. R.; Martinez, N.; Dellacassa, E.; dos Santos, F. M.; Costa, F. L. P.; Amorim, M. B. d.; Leitão, S. G., (−)-epi-Presilphiperfolan-1-ol, a new triquinane sesquiterpene from the essential oil of Anemia tomentosa var. anthriscifolia (Pteridophyta). *Tetrahedron Lett.* **2009**, *50* (33), 4785-4787; (c) Joseph-Nathan, P.; Leitão, S. G.; Pinto, S. C.; Leitão, G. G.; Bizzo, H. R.; Costa, F. L. P.; Amorim, M. B. d.; Martinez, N.; Dellacassa, E.; Hernández-Barragán, A.; Pérez-Hernández, N., Structure reassignment and absolute configuration of 9-epi-presilphiperfolan-1-ol. *Tetrahedron Lett.* **2010**, *51* (15), 1963-1965.
15. Hong, A. Y.; Stoltz, B. M., Enantioselective total synthesis of the reported structures of (-)-9-epi-presilphiperfolan-1-ol and (-)-presilphiperfolan-1-ol: structural confirmation and reassignment and biosynthetic insights. *Angew. Chem. Int. Ed.* **2012**, *51* (38), 9674-9678.
16. Osawa, E.; Aigami, K.; Takaishi, N.; Inamoto, Y.; Fujikura, Y.; Majerski, Z.; Schleyer, P. v. R.; Engler, E. M.; Farcasiu, M., The mechanisms of carbonium ion rearrangements of tricycloundecanes elucidated by empirical force field calculations. *J. Am. Chem. Soc.* **1977**, *99* (16), 5361-5373.
17. Pinto, S. C.; Leitao, G. G.; de Oliveira, D. R.; Bizzo, H. R.; Ramos, D. F.; Coelho, T. S.; Silva, P. E.; Lourenco, M. C.; Leitao, S. G., Chemical composition and antimycobacterial activity of the essential oil from Anemia tomentosa var. anthriscifolia. *Nat. Prod. Commun.* **2009**, *4* (12), 1675-1678.
18. González-Coloma, A.; Valencia, F.; Martín, N.; Hoffmann, J. J.; Hutter, L.; Marco, J. A.; Reina, M., Silphinene sesquiterpenes as model insect antifeedants. *J. Chem. Ecol.* **2002**, *28* (1), 117-129.
19. Weyerstahl, P.; Marschall, H.; Schulze, M.; Schwope, I., Synthesis of rac-Presilphiperfolan-9-ol. *Liebigs Ann.* **1996**, (5), 799-807.
20. Hu, P.; Snyder, S. A., Enantiospecific Total Synthesis of the Highly Strained (-)-Presilphiperfolan-8-ol via a Pd-Catalyzed Tandem Cyclization. *J. Am. Chem. Soc.* **2017**, *139* (14), 5007-5010.
21. Bohlmann, F.; Jakupovic, J., Neue Sesquiterpen-Kohlenwasserstoffe mit anomalen Kohlenstoffgerüst aus Silphium-arten. *Phytochemistry* **1980**, *19* (2), 259-265.
22. (a) Bradshaw, A. P. W.; Hanson, J. R.; Nyfeler, R., Studies in terpenoid biosynthesis, Part 24. The formation of the carbon skeleton of the sesquiterpenoid, dihydrobotrydial. *J. Chem. Soc., Perkin Trans. 1* **1981**, 1469-1472; (b) Bradshaw, A. P. W.; Hanson, J. R.; Nyfeler, R.; Sadler, I. H., Use of2H–13C n.m.r. coupling patterns in terpenoid biosynthesis. *J. Chem. Soc., Chem. Commun.* **1981**, (13), 649-650; (c) Bradshaw, A. P. W.; Hanson, J. R.; Nyfeler, R.; Sadler, I. H., Studies in terpenoid biosynthesis. Part 25. The fate of the mevalonoid hydrogen atoms in the biosynthesis of the sesquiterpenoid, dihydrobotrydial. *J. Chem. Soc., Perkin Trans. 1* **1982**, 2187-2192; (d) Hanson, J. R., The biosynthesis of some sesquiterpenoids. *Pure Appl. Chem.* **1981**, *53* (6), 1155-1162; (e) Lin, X.; Cane, D. E., Biosynthesis of the sesquiterpene antibiotic albaflavenone in Streptomyces coelicolor. Mechanism and stereochemistry of the enzymatic formation of epi-isozizaene. *J. Am. Chem. Soc.* **2009**, *131* (18), 6332-3.
23. Wang, S. C.; Tantillo, D. J., Prediction of a new pathway to presilphiperfolanol. *Org. Lett.* **2008**, *10* (21), 4827-4830.
24. (a) Coates, R. M.; Ho, Z.; Klobus, M.; Wilson, S. R., Stereochemistry and Reactions of Presilphiperfolanol: A Branch Point Marker in Triquinane Sesquiterpene Biogenesis. *J. Am. Chem. Soc.* **1996**, *118* (39), 9249-9254; (b) Davis, C. E.; Duffy, B. C.; Coates, R. M., Total synthesis of (+/-)-cameroonan-7alpha-ol and biomimetic rearrangements to related nopsane sesquiterpenes. *J. Org. Chem.* **2003**, *68* (18), 6935-6943; (c) Weyerstahl, P.; Marschall, H.; Seelmann, I.; Jakupovic, J., Cameroonane, Prenopsane and Nopsane, Three New Tricyclic Sesquiterpene Skeletons. *Eur. J. Org. Chem.* **1998**, *1998* (6), 1205-1212; (d) Barquera-Lozada, J. E.; Cuevas, G., Role of carbocation's flexibility in sesquiterpene biosynthesis: computational study of the formation mechanism of terrecyclene. *J. Org. Chem.* **2011**, *76* (6), 1572-1577.
25. (a) Bohlmann, F.; Zdero, C., Caryophyllene derivatives and a hydroxyisocomene from Pulicaria dysenterica. *Phytochemistry* **1981**, *20* (11), 2529-2534; (b) Bohlmann, F.; Zdero, C.; King, R. M.; Robinson, H., The first acetylenic monoterpene and other constituents from Senecio clevelandii. *Phytochemistry* **1981**, *20* (10), 2425-2427.
26. Shankar, S.; Coates, R. M., Solvolysis of Caryophyllen-8β-yl Derivatives: Biomimetic Rearrangement−Cyclization to 12-Nor-8α-presilphiperfolan-9β-ol. *J. Org. Chem.* **1998**, *63* (25), 9177-9182.
27. (a) Collado, I. G.; Aleu, J.; Macias-Sanchez, A. J.; Hernandez-Galan, R., Synthesis and antifungal activity of analogues of naturally occurring botrydial precursors. *J. Chem. Ecol.* **1994**, *20* (10), 2631-2644; (b) Collado, I. G.; Aleu, J.; Macías-Sánchez, A. J.; Hernández-Galán, R., Inhibition of Botrytis cinerea by New Sesquiterpenoid Compounds Obtained from the Rearrangement of Isocaryophyllene. *J. Nat. Prod.* **1994**, *57* (6), 738-746; (c) Cameron, A. F.; Hannaway, C.; Robertson, J. M., Crystal and molecular structures of 8,9-dibromo-1,4,4,8-tetramethyltricyclo[5,4,0,0]undecane and 8,9-dibromo-2,2,4,8-tetramethyltricyclo[5,3,1,0]undecane, derivatives of rearrangement products of caryophyllene dihydrochloride and isocaryophyllene respectively. *J. Chem. Soc., Perkin Trans. 2* **1973**, (14), 1938-1942; (d) Gollnick, K.; Schade, G.; Cameron, A. F.; Hannaway, C.; Roberts, J. S.; Robertson, J. M., The structure of an isocaryophyllene rearrangement product, 1,5,9,9-tetramethyltricyclo[6,2,1,04,11]undec-5-ene: X-ray analysis of the dibromo-derivative. *J.*

Chem. Soc. D **1970**, (4), 248-249; (e) Gollnick, K.; Schade, G.; Cameron, A. F.; Hannaway, C.; Robertson, J. M., The structure of a hydrocarbon, 2,6,10,10-tetramethyltricyclo[7,2,0,02,7]undec-5-ene, obtained from caryophyllene dihydrochloride: X-ray analysis of the dibromo-derivative. *J. Chem. Soc. D* **1971**, (1), 46-46; (f) Khomenko, T. M.; Bagryanskaya, I. Y.; Gatilov, Y. V.; Korchagina, D. V.; Gatilova, V. P.; Dubovenko, Z. V.; Barkhash, V. A., Khomenko 1. *Russ. J. Org. Chem.* **1985**, *21*, 614-615; (g) Khomenko, T. M.; Korchagina, D. V.; Gatilov, Y. V.; Bagryanskaya, Y. I.; Tkachev, A. V.; Vyalkov, A. I.; Kun, O. B.; Salenko, V. L.; Dubovenko, Z. V.; Barkhash, V. A., Khomenko 2. *Russ. J. Org. Chem.* **1990**, *26*, 1839-1852.

28. (a) Wallach, O.; Wallach, W., Zur Kenntniss der Terpene und der ätherischen Oele. Einundzwanzigste Abhandlung. I. Zur Charakteristik der Sesquiterpene. *Liebigs Ann.* **1892**, *271* (3), 285-299; (b) Henderson, G. G.; McCrone, R. O. O.; Robertson, J. M., CLXXVIII.—The chemistry of the caryophyllene series. Part II. Clovene and isoclovene. *J. Chem. Soc.* **1929**, *0* (0), 1368-1372; (c) Aebi, A.; Barton, D. H. R.; Burgstahler, A. W.; Lindsey, A. S., Sesquiterpenoids. Part V. The stereochemistry of the tricyclic derivatives of caryophyllene. *J. Chem. Soc.* **1954**, 4659-4665; (d) Barton, D. H. R.; Nickon, A., Sesquiterpenoids. Part VI. The absolute configuration of caryophyllene. *J. Chem. Soc.* **1954**, 4665-4669; (e) Parker, W.; Raphael, R. A.; Roberts, J. S., Neoclovene a novel rearrangement product of caryophyllene. *Tetrahedron Lett.* **1965**, *6* (27), 2313-2316; (f) Parker, W.; Raphael, R. A.; Roberts, J. S., Bridged ring systems. Part XV. Structure of neoclovene (2,6,8,8-tetramethyltricyclo[5,2,2,01,6]undec-2-ene). *J. Chem. Soc. C* **1969**, (20), 2634-2643.

29. Price obtained from Sigma-Aldrich (www.sigmaaldrich.com) for 5 Kg of compound