Natural Product Synthesis Hot Paper

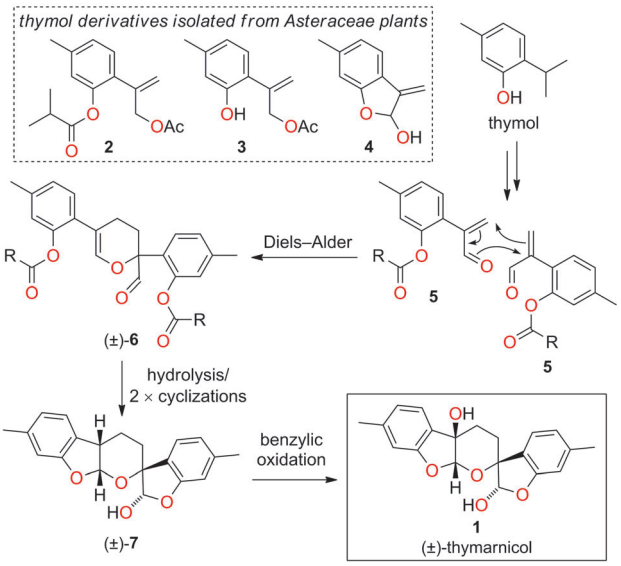
International Edition: DOI: 10.1002/anie.201701481
German Edition: DOI: 10.1002/ange.201701481

Total Synthesis of a Dimeric Thymol Derivative Isolated from *Arnica sachalinensis*

Irene De Silvestro, Samuel L. Drew, Gary S. Nichol, Fernanda Duarte, and Andrew L. Lawrence**

Abstract: *The total synthesis of a dimeric thymol derivative (thymarnicol) isolated from Arnica sachalinensis was accomplished in 6 steps. A key biomimetic Diels–Alder dimerization was found to occur at ambient temperature and the final oxidative cyclization occurs when the substrate is exposed to air and visible light. These results indicate that this natural product is likely the result of spontaneous (non-enzyme-mediated) reactivity.*

In 1999, Passreiter and co-workers isolated a racemic thymol derivative (**1**) from the flower heads of *Arnica sachalinensis* (Asteraceae), a sunflower native to Sakhalin Island off the Pacific Coast of Russia (Scheme 1).[1] For ease of discussion within this manuscript, and for future communications, we suggest "thymarnicol" (a portmanteau of thymol and *Arnica*) as a suitable name for compound **1**. Preliminary biological testing has revealed thymarnicol (**1**) to have potentially useful antifeedant,[1a] phytotoxic,[2] and anti-inflammatory[3] activity. For its small molecular size, thymarnicol (**1**) possesses significant molecular complexity and, despite its dimeric origins, contains no element of symmetry. The dense array of oxygen functionalities and four associated stereogenic centers at the core of the novel spiro[benzofuran-pyranobenzofuran] framework poses a considerable synthetic challenge. The previous isolation of oxidized and acetylated thymol derivatives from other Asteraceae plants (e.g., **2**, **3** and **4**)[4] led Passreiter and co-workers to propose a biosynthetic pathway for thymarnicol (**1**; Scheme 1).[1a] It begins with hetero-Diels–Alder dimerization of enal-thymol derivative **5** to give dihydropyran **6**.[5] Hydrolysis of the phenol ester groups then enables two cyclizations to form pentacycle **7**, with a final oxidation at the benzylic methine position giving thymarnicol (**1**). We decided to embark upon efforts to achieve a concise total synthesis of this complex and compact natural product and to investigate the chemical feasibility of Passreiter's proposal.



Scheme 1. Structure of thymarnicol (**1**) and Passreiter's proposed biosynthetic pathway. Ac = acetyl.

The synthesis began with Wittig methylenation of commercially available acetophenone **8** to give alkene **9**,[6] followed by acetylation using acetic anhydride in pyridine (Scheme 2). Both steps proceeded in excellent yield and could be conducted on a multigram scale. Oxidation of alkene **10** to enal **11**, using stoichiometric SeO_2, was performed in several smaller batches since the yield was found to decline with scale (Scheme 2).[7] The chemical feasibility of Passreiter's proposed hetero-Diels–Alder dimerization was quickly established, with enal **11** being found to undergo dimerization when stored neat at ambient temperatures (ca. 50% conversion after 5 days). Heating neat samples of enal **11** at 80 °C for 42 h resulted in near quantitative dimerization. The crude dimer was immediately subjected to basic conditions to deprotect the two phenols. This did not give pentacycle **7** (see Scheme 1) but instead gave lactol **12**, the product of just one cyclization (Scheme 2). Interestingly, lactol **12** exists as a mixture of two diastereomers, and the ratio between them, determined by analysis of the ^{1}H NMR spectrum, varies with the solvent (d.r. in $CDCl_3$ 72:28, CD_3COCD_3 78:22), presumably as a result of facile lactol epimerization. It was observed that lactol **12** was unstable; new peaks corresponding to thymarnicol (**1**) could be identified in the ^{1}H NMR spectra of older samples. After careful experimentation, we discovered that this fortuitous aerial oxidation is

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Supporting information and the ORCID identification number(s) for the author(s) of this article can be found under: https://doi.org/10.1002/anie.201701481.

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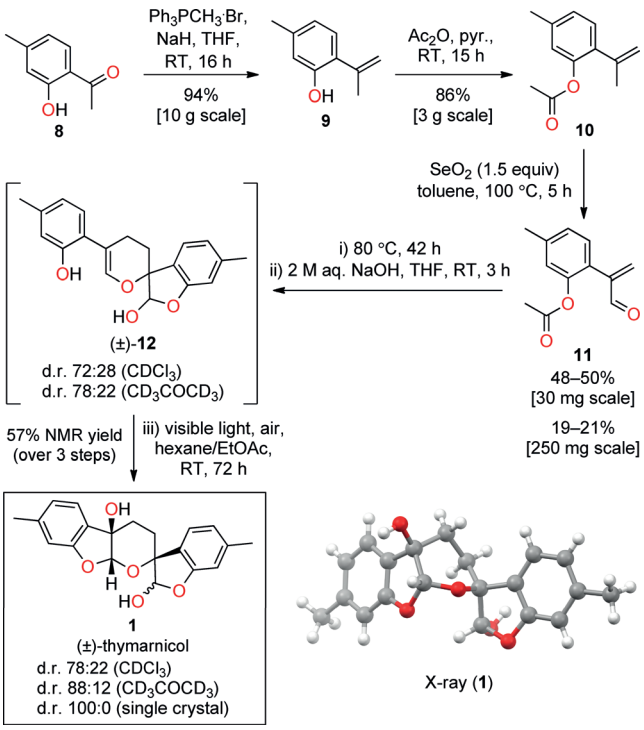

Scheme 2. Six-step synthesis of thymarnicol (**1**). THF = tetrahydrofuran.

promoted by exposure to visible light.[8] Therefore, a hexane/EtOAc solution of crude lactol **12**, open to the atmosphere, was irradiated with visible light from an 11 W compact fluorescent lamp for 72 h. Analysis of the ^{1}H NMR spectrum of the resulting product, with inclusion of an internal standard, indicated a remarkable 57% crude yield of the two lactol epimers of thymarnicol (**1**) over three steps from enal **11**. Work is ongoing in our laboratory to identify other minor products from this aerial oxidation and to interrogate likely mechanisms.[9]

The final three-step sequence, from enal **11** to thymarnicol (**1**), could be conducted on a more than 100 mg scale and without chromatographic purification of intermediates. Column chromatography followed by preparative HPLC could then be used to access analytically pure samples of thymarnicol (**1**; 40 mg prepared so far), but still as an unavoidable mixture of the two lactol epimers. Crystallization from acetonitrile resulted in crystals suitable for single-crystal X-ray diffraction studies.[10] The crystal structure obtained matched that reported for the natural material,[1b] which consists of just one lactol epimer (Scheme 2). Nevertheless, subsequent analysis of these crystals by solution-phase ^{1}H NMR spectroscopy showed the presence of both lactol epimers. It must be concluded that thymarnicol (**1**) is stereodynamic; it exists as a mixture of lactol epimers in solution but can exist as a single epimer in the solid state. Thus, a six-step total synthesis of thymarnicol (**1**) has been achieved, involving the formation of nine new bonds (three C–C, six C–O), three new rings, and four new stereogenic centres.

Density functional theory (DFT) calculations at the ωB97X-D/6-31 + G(d) level of theory[11] were undertaken to investigate the factors that control the reactivity and regioselectivity of the hetero-Diels–Alder dimerization (Scheme 3).[12] For the simplified model compound **11a**, which bears no *ortho* substituent, there are eight distinct transition structures (TSs) possible (Scheme 3a). These TSs are described using a notation where the regiochemical-orientation is *meta* (*m*) or *para* (*p*), the Alder–Stein mode is *endo* (*N*) or *exo* (*X*), and the dienophile adopts either an s-*cis* (*cis*) or s-*trans* (*trans*) conformation. Without exception, the *para* TSs were found to be significantly lower in energy than the *meta* TSs owing to better orbital overlap and lower distortion penalties (Scheme 3a; see the Supporting Information for full distortion/interaction analysis).[13,14] The unexpected *exo* selectivity observed for the majority of the TSs is due to favourable non-covalent (dispersion) interactions between the two aromatic rings (as shown in Scheme 3b for the *meta-X-trans* TS), which outweighs the higher distortion penalty that normally disfavours *exo* TSs.[15] The lowest energy TS, however, is not *exo* orientated; the *para-N-cis* TS is a rare example of a C_2-symmetric bis-pericyclic TS (Scheme 3c).[16] In bis-pericyclic TSs, the [4+2] and [2+4] cycloaddition pathways have fully merged and thus benefit from three primary orbital interactions. Following the bis-pericyclic TS, the pathway then bifurcates to give the degenerate [4+2] and [2+4] cycloadducts. The lowest energy TS for hetero-Diels–Alder dimerization of enal **11** was also found to be a *para-N-cis* TS, which although not C_2-symmetric still has bis-pericyclic character (Scheme 3d; see the Supporting Information for full computational details for enal **11**).

Further insight into the origin and reactivity of thymarnicol (**1**) was acquired through other unsuccessful synthetic studies. For example, different phenol-protecting groups were investigated (Scheme 4). The *tert*-butyldimethylsilyl (TBS)-protected enal **13** underwent a less efficient Diels–Alder dimerization, requiring higher temperatures and giving lower yields of dihydropyran **14** (Scheme 4a). Use of the sterically less demanding MOM (methoxymethyl) ether, however, resulted in dimerization occurring at ambient temperature, with a synthetically useful 77% yield of dihydropyran **16** achieved upon heating enal **15** at 80 °C for 22 h (Scheme 4a). Unfortunately, dihydropyran **16** could not be successfully advanced to give thymarnicol (**1**; see below). The most interesting precursor to investigate, from a biomimetic perspective, was the unprotected monomer **4**, a known natural product that exists primarily as the lactol isomer (Scheme 4b).[4b,17] When compound **4** was stored neat at ambient temperatures, multiple new minor peaks appeared in the ^{1}H NMR spectrum and the Diels–Alder dimer **12** could be identified amongst these new species. Therefore, an alternative biosynthetic pathway involving dimerization of the natural product **4** is chemically feasible. Synthetically speaking, however, this route was not pursued owing to difficulties associated with the preparation and purification of lactol **4** and an apparent lack of selectivity for dimerization.

The greatest synthetic challenge encountered during our synthetic efforts was the propensity of the thymarnicol nucleus to undergo acid-promoted rearrangements. For example, attempts to deprotect dimer **16** under standard

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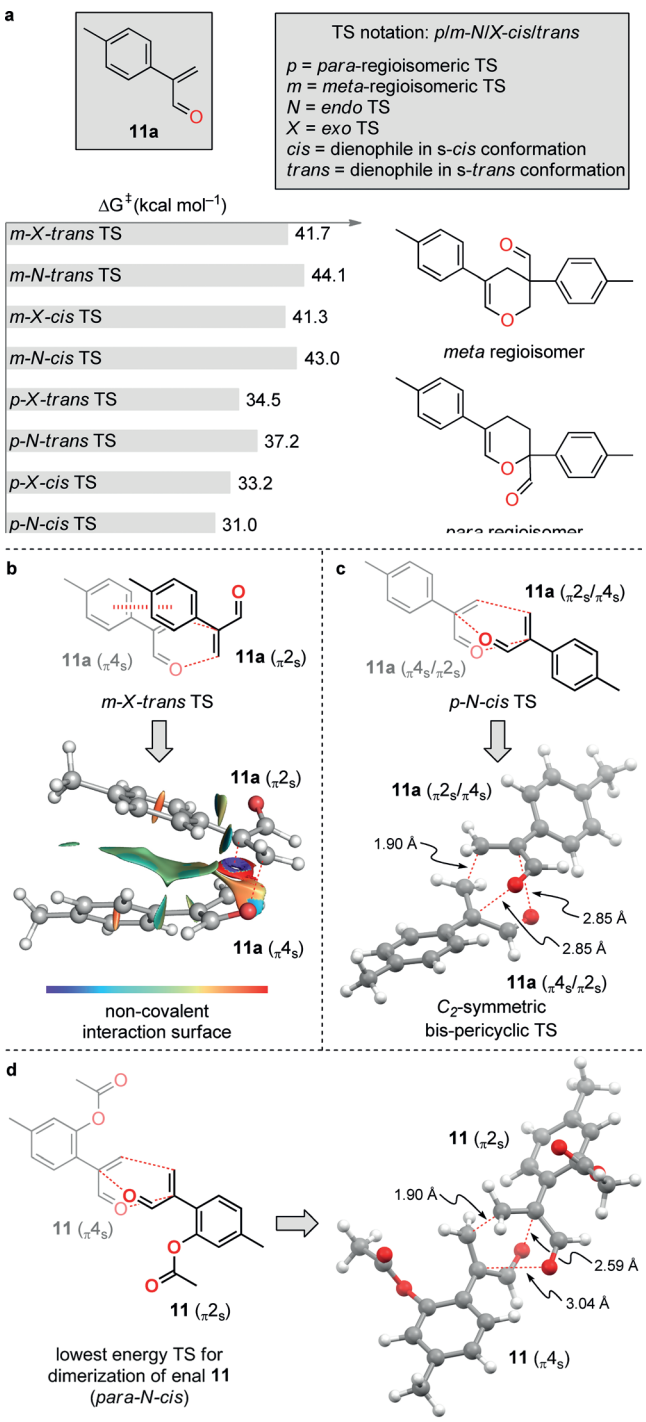
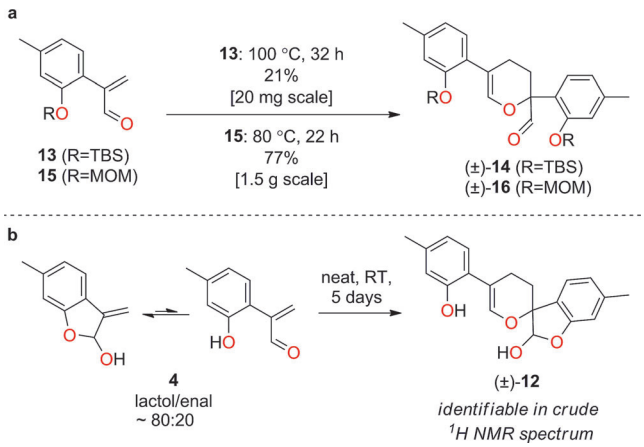


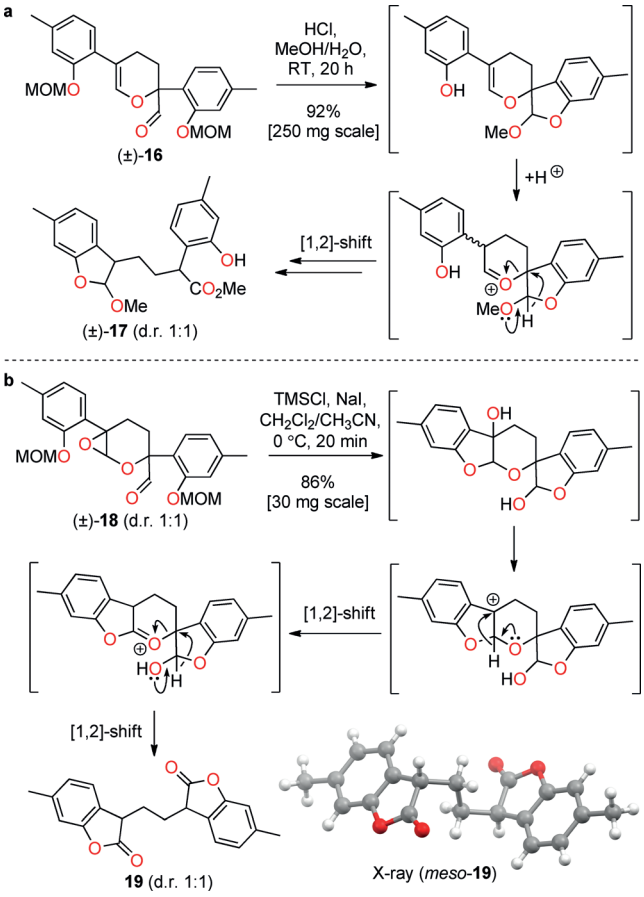
Scheme 3. a) Reaction free energies ($\Delta G^{\ddagger}_{80\,^{\circ}C}$) at the ωB97X-D/6-311++G(d,p)//ωB97X-D/6-31+G(d) level of theory for the eight possible transition structures for hetero-Diels–Alder dimerization of model compound **11a**. b) The ωB97X-D/6-31+G(d) optimized structure of the *m-X-trans* TS for model compound **11a**, with a non-covalent interaction surface (green indicates van der Waals/dispersion interactions, blue indicates strong polar interactions, and red indicates steric repulsion). The ωB97X-D/6-31+G(d)-optimized structures of the *p-N-cis* TS for model compound **11a** (c) and for enal **11** (d).

HCl/MeOH conditions resulted in cleavage of the dihydropyran ring to give dihydrobenzofuran **17** as a mixture of two diastereomers (Scheme 5a).[18] Similarly, attempts to cleave



Scheme 4. a) Hetero-Diels–Alder dimerization of differently protected monomers **13** and **15.** b) Hetero-Diels–Alder dimerization of unprotected monomer **4**. MOM = methoxymethyl, TBS = *tert*-butyldimethylsilyl.

the MOM ethers in epoxide **18** led to the formation of bislactones **19** in high yield (Scheme 5b).[10,19] The predisposed nature of these rearrangements, which presumably occur through [1,2]-shift mechanisms (see Scheme 5), leads us to



Scheme 5. a) Rearrangement observed during deprotection of MOM-protected dimer **16**. b) Rearrangement observed during deprotection of MOM-protected epoxide **18**. TMS = trimethylsilyl.

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speculate that structures akin to **17** and **19** might be isolated as natural products in the future.[20]

In conclusion, through our synthetic efforts, we have been able to investigate the chemical feasibility of Passreiter's suggested biosynthetic pathway for thymarnicol (**1**). We have shown that the proposed hetero-Diels–Alder dimerization is plausible, with both acetyl- and MOM-protected systems (**11** and **15**) undergoing spontaneous dimerization at room temperature. We have also provided evidence in support of an alternative biosynthetic pathway involving dimerization of lactol **4**, a known natural product previously isolated from related Asteraceae plants.[4b] It was discovered that dihydropyran **12** undergoes the final oxidative cyclization when simply exposed to air and visible light. Therefore, our results indicate that the complexity-generating Diels–Alder dimerization and oxidative cyclization likely proceed without the intervention of enzymes to produce natural thymarnicol (**1**).[21]

Acknowledgements

This work was supported by a Royal Society Research Grant and a Marie Curie Career Integration Grant. F.D. acknowledges the Royal Society for a Newton International Fellowship. I.D.S. thanks the University of Edinburgh for the provision of a studentship. Antony Crisp (Australian National University) is thanked for conducting preliminary experiments. Dr. Stephen Moggach (University of Edinburgh), Dr. Nicholas Green (University of Edinburgh) and Prof. Mick Sherburn (Australian National University) are thanked for helpful discussions.

Conflict of interest

The authors declare no conflict of interest.

Keywords: biomimetic synthesis · cycloaddition · dimerization · natural products · terpenoids

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[9] Inclusion of a photosensitizer (rose bengal) did not lead to any appreciable rate enhancement.

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Manuscript received: February 10, 2017
Final Article published: ■■ ■■, ■■■■

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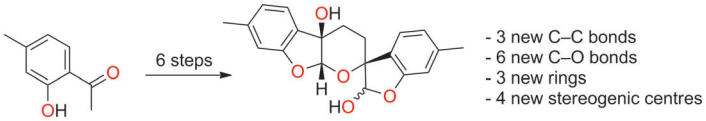
Communications

Natural Product Synthesis

I. De Silvestro, S. L. Drew, G. S. Nichol, F. Duarte,* A. L. Lawrence* _____ ■■■■–■■■■

Total Synthesis of a Dimeric Thymol Derivative Isolated from *Arnica sachalinensis*



(±)-thymarnicol

Poised to (re)act: The total synthesis of a dimeric thymol derivative (thymarnicol) isolated from *Arnica sachalinensis* was accomplished in 6 steps. A key biomimetic Diels–Alder dimerization was found to occur at ambient temperature and the final oxidative cyclization occurs when the substrate is exposed to air and visible light. These results indicate that this natural product is likely the result of spontaneous (non-enzyme-mediated) reactivity.

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