An Efficient Synthetic Strategy for Obtaining 4-Methoxy Carbon Isotope Labeled Combretastatin A-4 Phosphate and Other Z-Combretastatins

ARTICLE *in* JOURNAL OF NATURAL PRODUCTS · DECEMBER 2009

Impact Factor: 3.8 · DOI: 10.1021/np9004486 · Source: PubMed

Published in final edited form as:
J Nat Prod. 2010 March 26; 73(3): 399–403. doi:10.1021/np9004486.

An Efficient Synthetic Strategy for Obtaining 4-Methoxy Carbon Isotope Labeled Combretastatin A-4 Phosphate and Other *Z*-Combretastatins[1]

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Abstract

Human cancer and other clinical trials under development employing combretastatin A-4 phosphate (**1b**, CA4P) should benefit from the availability of a [^{11}C]-labeled derivative for position emission tomography (PET). In order to obtain a suitable precursor for addition of a [^{11}C]methyl group at the penultimate step, several new synthetic pathways to CA4P were evaluated. Geometrical isomerization (*Z* to *E*) proved to be a challenge, but it was overcome by development of a new CA4P synthesis suitable for 4-methoxy isotope labeling.

In 1987, we reported[1] the isolation, structure, and synthesis of combretastatin A-1 (**1c**, CA1) from the subtropical tree *Combretum caffrum* (Eckl. and Zeyh.) Kuntze (Combretaceae)1b collected in southern Africa, and two years later combretastatin A-4 (**1a**, CA4) from the same source was isolated and synthesized.[2] Subsequently, both CA1 and CA4 were converted to phosphate prodrugs (**1d**, CA1P[3] and **1b**, CA4P4) and developed5 to human cancer clinical trials.6 CA1P and CA4P are presently in Phase I/II and III cancer trials, respectively, and CA4P is also in Phase II human macular degeneration (leading cause of blindness)7 clinical trials. The potential of CA4P in treating other eye diseases such as diabetic retinopathy7d and retinoblastoma7b,e is also being developed. Presently, CA4P (a.k.a. Zybrestat) followed by CA1P is the lead among cancer vasculature disrupting drugs.8a–e A large number of other potentially important recent observations concerning medical applications of the leading combretastatins include evidence that CA4P is antiangiogenic,8f increases aberrant organization of metaphase chromosomes in non-small cell lung cancer cells,8g inhibits gastric cancer cell metastasis,8h and improves glucose tolerance in diabetic mice, which in turn suggests a possible new approach to treatment of type 2 diabetes.[8i] The latter evidence provides another mechanistic parallel to resveratrol (**2**)[8j] and suggests many other avenues for research from cancer prevention[8h] to longevity.[8k]

By 2000, positron emission tomography (PET) was already well established as a non-invasive technique for biomedical imaging, and its potential for necessary applications in preclinical and clinical research employing the lead combretastatins, particularly CA4P, was clearly evident. To follow is both a brief outline of the radiolabeling rationale and a practical synthetic route to employ as a model for later introduction of a [^{11}C]-isotope into CA4/CA4P. A [^{11}C]

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Dedicated to the late Dr. John W. Daly of NIDDK, NIH, Bethesda, Maryland and to the late Dr. Richard E. Moore of the University of Hawaii at Manoa for their pioneering work on bioactive natural products.

methyl group was selected as the most efficient method, which required completion of an appropriate new synthesis of CA4 and now remains a most practical option. Meanwhile, a variety of other new syntheses of CA49a,b and its analogues9c–g have been reported, as well as syntheses of CA1P radiolabeled with a [14C]methyl group in high specific activity[10a] and [^{11}C]methyl derivatives[10b] of resveratrol (**2**), augmented by other structural modifications[8j] of this increasingly important naturally occurring stilbene.[8h]

Results and Discussion

When we began this research in 2001, the knowledge of CA4 in vivo metabolism left some uncertainties as to whether [^{11}C]-CA4 was an appropriate target. That has in the interim been resolved. Very recently, it has been confirmed that CA4 is transformed in rat and human liver fractions to a glucuronide metabolite.[11] In the rat, with an approximate therapeutic dose, CA4 is metabolized to both the glucuronide and a previously undetected sulfate derivative at the 4′-phenol position.[11] One or both of these relatively long-lived (compared to the 20.1-min half-life of [^{11}C]) modifications would be useful for measurement of radiation (dosimetry) in the study of absorption, distribution, metabolism, and elimination (pharmacokinetics) of CA4P as part of current human trials. In turn, that would allow a better knowledge of the distribution of CA4 between neoplastic and normal tissue as well as the relative binding potential/effective blockade vs. dose. Those early objectives required a synthesis of CA4P that would allow a short reaction path and high-yield penultimate step for obtaining [^{11}C]-CA4P.

Substitution of a CA4P A-ring methyl group with a [^{11}C]methyl seemed to offer the fewest difficulties[10b] and would also utilize some of the synthetic procedures we previously developed for synthesizing combretastatin A-3, the 3-desmethyl derivative of CA4.[12] When an approach based on the 3-hydroxy-4,5-dimethoxyphenyl A-ring of combretastatin A-3 was discontinued owing to geometrical isomerization problems at the stilbene stage, attention was next focused on a 3,5-dimethoxy-4-hydroxyphenyl A-ring precursor that would lead to phenol **3** and allow methylation without *cis/trans* isomerism. The first such attempt to synthesize phenol **3** began with silyl protection of 3,5-dimethoxy-4-hydroxybenzaldehyde using *tert*-butyldiphenylsilyl chloride (TBDPSCl), which gave a 71% yield of silyl ether **4** (Scheme 1). The benzaldehyde (**4**) was reduced with $NaBH_4$ to afford benzyl alcohol **5** (94%), which was then converted to benzyl bromide **6**, and subsequent treatment with triphenylphosphine led to Wittig salt **7** (62%).

Synthesis of the B ring was begun (Scheme 2) by protection of isovanillin (**8a**) with the *tert*-butyldimethylsilyl (TBDMS) group (**8b**, 84%) as previously reported.[12a] The Wittig coupling of the A and B rings was accomplished by the treatment of phosphonium bromide **7** with *n*-butyl lithium followed by addition of aldehyde **8b** to afford stilbene **9** in 25% yield. Disilyl ether **9** was then selectively deprotected with pyridinium *p*-toluenesulfonate (PPTS) in ethanol to afford 3′-phenol **10** in 65% yield. Stilbene **10** was phosphorylated with dibenzylphosphite to afford the phosphate ester (**11**, 38%).[4,12b,13] Subsequently, the desilylation of phosphorylated stilbene **11** to obtain 4-phenol **3** was attempted, but all efforts failed owing to concomitant cleavage of the phosphate ester.

Since removal of the silyl protecting group was causing difficulty, a different protecting group such as acetate was needed in order to eventually obtain phenol **3**. Because an acetate group is labile under the basic conditions required for a Wittig condensation, it had to be on the A-ring aldehyde unit rather than on the phosphonium salt moiety (ring B). The new A-ring was prepared by the acetylation of 3,5-dimethoxy-4-hydroxybenzaldehyde to give **12** (Scheme 3). The Wittig olefin reaction was accomplished by treatment of phosphonium salt **13**[12b] with *n*-butyllithium followed by the addition of aldehyde **12** to produce stilbene **14** (16%). Desilylation with tetrabutylammonium fluoride (TBAF) afforded 3′-phenol **15** (84%). The phenol group was phosphorylated[4,12b,13] with dibenzylphosphite to produce phosphate ester **16** (68%), and deacetylation by treatment with 5% potassium carbonate in methanol afforded 4-phenol **3**, albeit in moderate yields (30%; 2.4% overall yield from **13**) owing to partial isomerization to the corresponding *trans*-stilbene.

Once the necessary precursor of combretastatin A-4 prodrug was in hand, a suitable methylation procedure was needed that could be easily employed to produce [^{11}C] combretastatin A-4 phosphate prodrug. A selection of procedures for the methylation of 4-phenol **3** were attempted using CH_3I, as this would be a choice methylating agent for producing a [^{11}C]methylated derivative. However, all attempts to methylate the phenol (**3**) with CH_3I resulted in *cis/trans* isomerization.[10a] Therefore, a new approach that would avoid isomerization by way of direct conversion of the silyl ether to a methyl ether was examined.

In a new route to **11** (Scheme 4), Wittig salt **7** was treated with NaH and aldehyde **17**, which was prepared directly from isovanillin (**8a**) in 85% yield. Desilylation of the product, stilbene **11,** with concurrent methylation that could be used to efficiently obtain [^{11}C]combretastatin A-4 was first explored by modification of an existing procedure, in which a TBDMS ether can be converted into a benzyl ether by use of KF and benzyl bromide in tetrahydrofuran. An attempt to apply this method using KF and CH_3I to convert the TBDPS ether directly to a methyl ether did not succeed. However, the approach was successful when TBAF and CH_3I were used and led to combretastatin A-4 3′-dibenzylphosphate (**18**). That result completed a very useful synthetic approach that can now be employed to obtain 4-methoxy isotope-labeled combretastatins and related stilbenes.

Experimental Section

General Experimental Procedures

Abbreviations: TBDMS, *tert*-butyldimethylsilyl; TBDPS, *tert*-butyldiphenylsilyl; DIPEA, diisopropylethylamine; LAH, lithium aluminum hydride; DMAP, dimethylaminopyridine; TBAF, tetrabutylammonium fluoride; DMF, *N*,*N*-dimethylformamide; THF, tetrahydrofuran; DCM, dichloromethane; TLC, thin layer chromatography. All solvents (ether refers to diethyl ether) used in chemical reactions were redistilled and dried. Other reagents were purchased from Sigma-Aldrich Chemical Co., Lancaster-Clariant or Fisher-ACROS Chemical Co. Solvent extracts of aqueous solutions were dried over anhydrous magnesium sulfate unless otherwise noted. Gravity column chromatography (CC) was performed using silica gel (70–230 mesh) from VWR Scientific. All melting points were determined with an Electrochemical digital melting point apparatus, Model IA 9200, and are uncorrected. NMR spectra were recorded using a Varian Gemini 300 or a Varian Unity 400 instrument. Chemical shifts are reported in ppm (δ) downfield from tetramethylsilane as an internal standard. High resolution FAB mass spectra were obtained on a Kratos MS-50 (Midwest Center for Mass Spectrometry, University of Nebraska-Lincoln). Elemental analyses were obtained from Galbraith Laboratories, Inc., Knoxville, TN.

4-(*tert*-Butyldiphenylsilyloxy)-3,5-dimethoxybenzaldehyde (4)

To a solution of imidazole (7.5 g, 109 mmol), 3,5-dimethoxy-4-hydroxybenzaldehyde (10 g, 55 mmol) and DMF (100 mL) was added TBDPSC1[2] (16 mL, 60 mmol). After stirring 16 h, the reaction was terminated by the addition of water and extracted with hexane-EtOAc (1:1, 3 × 75 mL). The extract was washed with brine and, following removal of solvent, the residue was separated by flash chromatography (9:1, hexane-EtOAc) to afford aldehyde **4** as a clear oil (16.25 g, 71%): bp 202–204 °C (0.01 mmHg); ^{1}H NMR ($CDCl_3$, 300 MHz) δ 1.12 (s, 9H), 3.51 (s, 6H), 6.98 (s, 2H), 7.36 (m, 6H), 7.68 (d, *J* = 8.1 Hz, 4H), 9.76 (s, 1H); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 26.62, 55.23, 106.51, 127.15, 127.66, 129.32, 129.55, 133.82, 134.78, 134.97, 140.43, 151.33, 191.01; *anal*. C 71.17%, H 6.90%, calcd for $C_{25}H_{28}O_4Si$, C 71.39%, H 6.71%.

4-*tert*-(Butyldiphenylsilyloxy)-3,5-dimethoxybenzyl Alcohol (5)

Benzaldehyde **4** (5.3 g, 12.6 mmol) was dissolved in ethanol (100 mL) and $NaBH_4$ (0.6 g, 15.1 mmol) was added. After stirring 3 h, the reaction mixture was acidified with dilute HC1 (1 N), and extracted with EtOAc. After solvent removal in vacuo, the resulting oil was purified by CC (7:3 hexane-EtOAc) to afford alcohol **5** (5.01 g, 94%) as a clear oil: bp 218 °C (0.01 mmHg); IR ν_{max} 1134, 1242, 1340, 1427, 1462, 1512, 1593, 2859, 2936, 3420 cm^{-1}; ^1H NMR ($CDCl_3$, 300 MHz) δ 1.091 (s, 9H), 1.99 (bs, 1H), 3.41 (s, 6H), 4.48 (s, 2H), 6.39 (s, 2H), 7.31 (m, 6H), 7.69 (d, *J* = 7.8 Hz, 4H); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 20.05, 26.52, 26.73, 55.13, 65.51, 103.77, 126.96, 127.61, 129.01, 133.36, 133.62, 134.55, 134.76, 135.12, 150.86; *anal*. C 71.22%, H 7.08%, calcd for $C_{25}H_{30}O_4Si$, C 71.05%, H 7.16%.

4-(*tert*-Butyldiphenylsilyloxy)-3,5-dimethoxybenzyl Bromide (6)

To a solution of benzyl alcohol **5** (5 g, 12 mmol) in DCM (100 mL) was added PBr3 (1M in DCM, 6 mL, 6 mmol). After stirring 18 h, the reaction was terminated by the addition of $NaHCO_3$ (10%, 25 mL) and the aqueous phase was extracted with DCM. The combined extract was washed with cold water. Removal of solvent yielded a brown oil (**6**) that was not further purified.

4-(*tert*-Butyldiphenylsilyloxy)-3,5-dimethoxybenzyltriphenylphosphonium Bromide (7)

Benzyl bromide **6** (5.5 g) was dissolved in toluene (200 mL), and triphenylphosphine (3.5 g, 13 mmol) was added. The solution was heated at reflux for 1 h, cooled to rt and allowed to stir an additional 2 h. The resulting precipitate was collected and triturated with ether to afford bromide **7** (5.5 g) as a colorless amorphous solid (62% over 2 steps): mp 152–153 °C (CH_3OH); ^1H NMR (CD_3OD, 300 MHz) δ 1.061 (s, 9H), 3.11 (s, 6H), 4.88 (d, J_{PCH} = 14.4 Hz, 2H), 6.08 (s, 1H), 6.09 (s, 1H), 7.28 (m, 6H), 7.65 (m, 20H), 7.85 (m, 4H); ^{13}C NMR (CD_3OD, 75 MHz) δ 20.83, 27.28, 30.98, 31.60, 55.55, 109.14, 109.22, 118.54, 119.67, 120.35, 120.48, 128.11, 129.20, 129.90, 130.55, 131.14, 131.30, 134.95, 135.44, 135.57, 136.35, 152.35, 152.40; *anal*. C 68.62%, H 6.11%, calcd for $C_{43}H_{44}BrO_3PSi$, C 69.07, H 5.93%.

4-(*tert*-Butyldimethylsilyloxy)-3-methoxybenzaldehyde (8b)

A solution of isovanillin (**8a**, 20 g, 131 mmol) in DCM (400 mL) and DIPEA (25 mL, 145 mmol) was stirred for 10 min, and TBDMSCl (22 g, 145 mmol) was added. After stirring 16 h, the reaction was terminated by the addition of water (50 mL). The organic phase was separated and washed with NaOH (5%, 50 mL) followed by brine. The solvent was removed to provide an orange-brown oil that was separated by vacuum distillation to afford a colorless oil (29.3 g, 84%): bp 140–142 °C (0.01 mmHg): ^1H NMR (CD_3OD, 300 MHz) δ 0.13 (s, 6H), 0.96 (s, 9H), 3.84 (s, 3H), 6.91 (d, *J* = 8.1 Hz, 1H), 7.33 (d, *J* = 2.4 Hz, 1H), 7.43 (dd, *J* = 2.1, 8.1 Hz, 1H), 9.77 (s, 1H).

3′-(*tert*-Butyldimethylsilyloxy)-4-(*tert*-butyldiphenylsilyloxy)-3,4′,5-trimethoxy-*Z*-stilbene (9)

Phosphonium bromide **7** (5 g, 6.7 mmol) was dissolved in THF (100 mL) and cooled to −78 °C. *n*-BuLi (2.5 M in hexane, 2.7 mL, 6.7 mmol) was added, followed after 1 h by aldehyde **8b** (1.95 g, 7.4 mmol) over 5 min. After stirring for 16 h, the reaction was terminated by the addition of water, and the mixture was extracted with EtOAc. The solvent was removed from the organic phase, and the residue was separated by CC to afford a clear oil (**9**, 0.5 g, 25%): bp (dec.) 94–96 °C (0.01 mmHg); ^1H NMR (CD_3OD, 300 MHz) δ 0.15 (s, 6H), 1.02 (s, 9H), 1.15 (s, 9H), 3.32 (s, 6H), 3.79 (s, 3H), 6.41 (s, 2H), 6.97 (d, *J* = 7.8 Hz, IH), 6.82 (s, 1H), 6.85 (s, 1H), 7.04 (s, 1H), 7.37 (m, 10H), 7.75 (m, 4H); ^{13}C NMR (CD_3OD, 75 MHz) δ −4.81, 18.31, 20.05, 25.64, 26.73, 30.27, 30.79, 34.16, 54.92, 55.38, 100.61, 105.79, 111.56, 121.45, 122.56, 125.46, 126.91, 128.57, 128.96, 129.16, 129.73, 130.36, 133.47, 134.55, 135.12, 144.57, 149.95, 150.47; *anal*. C 71.19%, H 7.74%, calcd for $C_{39}H_{50}O_5Si_2$, C 71.52%, H 7.69%.

4-(*tert*-Butyldiphenylsilyloxy)-3′-hydroxy-3,4′,5-trimethoxy-*Z*-stilbene (10)

Stilbene **9** (0.34 g, 0.5 mmol) and PPTS (12 mg, 0.05 mmol) were dissolved in ethanol (25 mL). The mixture was heated to 60 °C and allowed to stir for 100 h. Ethanol was removed and the residue was separated by CC (9:1 hexane-EtOAc) to yield stilbene **10** (0.18 g, 65%): mp 86–87 °C; ^{1}H NMR (CD_3OD, 300 MHz) δ 1.089 (s, 9H), 3.28 (s, 6H), 3.84 (s, 3H), 5.46 (s, 1H), 6.36 (s, 2H), 6.65 (d, *J* = 8.7 Hz, 1H), 6.72 (dd, *J* = 1.8, 8.4 Hz, 1H), 7.33 (m, 10H), 7.70 (m, 4H); ^{13}C NMR (CD_3OD, 75 MHz) δ 19.99, 26.67, 29.10, 30.76, 54.94, 55.79, 105.78, 110.21, 115.06, 120.93, 126.88, 128.44, 128.96, 129.24, 129.68, 130.69, 133.46, 134.39, 135.12, 145.14, 145.60, 150.50; *anal*. C 72.98%, H 6.53%, calcd for $C_{33}H_{36}O_5Si$, C 73.30%, H 6.71%.

3′-*O*-Bis(benzyl)phosphoryl-4-(*tert*-butyldiphenylsilyloxy)-3,4′,5-trimethoxy-*Z*-stilbene (11). From Phenol 10. Method A

To a solution of 3′-phenol **10** (0.18 g, 0.33 mmol) in acetonitrile (10 mL) that was cooled to −10 °C was added CCl_4 (0.32 mL, 3.3 mmol). The mixture was stirred for 10 min before the addition of DIPEA (0.12 mL, 0.68 mmol) and DMAP (4 mg, 0.033 mmol), and after one minute dibenzylphosphite (0.11 mL, 0.49 mmol)[4] was added (dropwise over 5 min). The reaction mixture was stirred for 3 h at −10 °C, treated with KH_2PO_4 (20 mL, 0.5 M), stirred for 10 min, and then extracted with EtOAc. Removal of solvent and separation by CC (1:1 hexane-EtOAc) afforded phosphate **11** (0.10 g, 38%): bp (dec) 148 °C (0.01 mmHg); ^{1}H NMR ($CDCl_3$, 300 MHz) δ 3.64 (s, 6H), 3.76 (s, 3H), 5.11 (s, 2H), 5.14 (s, 2H), 6.44 (d, *J* = 2.1 Hz, 2H), 6.50 (s, 2H), 6.77 (d, *J* = 8.4 Hz, 1H), 7.06 (d, *J* = 8.1 Hz, 1H), 7.12 (m, 1H), 7.32 (s, 10H); *anal*. C 70.22%, H 6.31%, calcd for $C_{47}H_{49}O_8PSi$, C 70.48%, H 6.17%.

4-Acetoxy-3,5-dimethoxybenzaldehyde (12)

A solution prepared from 3,5-dimethoxy-4-hydroxybenzaldehyde (5 g, 27 mmol) and DMAP (0.34 g, 2.7 mmol) in pyridine (50 mL) and acetic anhydride (8 mL, 81 mmol) was stirred for 16 h. Reaction was terminated by the addition of water (100 mL) followed by extraction with EtOAc, washing of the organic phase with HC1 (1 N) and removal of solvent. The residue was purified by CC (7:3 hexane-EtOAc) to give **12** (2.4 g) as a colorless solid that crystallized from EtOAc-hexane: mp 116–117 °C; ^{1}H NMR (CD_3OD) δ 2.36 (s, 3H), 3.90 (s, 6H), 7.15 (s, 2H), 9.91 (s, 1H).

4-Acetoxy-3′-(*tert*-butyldiphenylsilyloxy)-3,4′,5-trimethoxy-*Z*-stilbene (14)

Phosphonium bromide **13** (7.7 g, 1.07 mmol)[12b] in THF (50 mL) was cooled to −78 °C, and *n*-BuLi (2.5 M in hexane, 3.9 mL) was added. The mixture was stirred for 1 h, aldehyde **12** (2 g, 8.9 mmol) was added in four portions (over 5 min), and the resulting mixture was allowed to warm to rt and stirred an additional 3 h. Water (100 mL) was added, and the mixture was extracted with EtOAc. The extract was washed with brine and dried over sodium sulfate. After removal of solvents, the resulting oil was separated by CC (9:1 hexane-EtOAc) to yield stilbene **14** (0.81 g, 16%): mp 81–82 °C (hexane-acetone); ^{1}H NMR (CD_3OD, 300 MHz) δ 1.09 (s, 9H), 2.32 (s, 3H), 3.47 (s, 3H), 3.61 (s, 6H), 6.32 (d, *J* = 1.2 Hz, 2H), 6.47 (s, 2H), 6.58 (d, *J* = 8.1 Hz, 1H), 6.75 (d, *J* = 2.4 Hz, 1H), 6.79 (dd, *J* = 2.4, 8.1 Hz, 1H); *anal*. C 71.87%, H 6.42%, calcd for $C_{35}H_{38}O_6Si$, C 72.14%, H 6.57%.

4-Acetoxy-3′-hydroxy-3,4′,5-trimethoxy-*Z*-stilbene (15)

To a solution of stilbene **14** (0.81 g, 1.4 mmol) in THF (30 mL) was added TBAF (1M in THF, 1.5 mL), stirring was continued for 2 h, 50 mL of 1 N HC1 was added, and the mixture was extracted with EtOAc. The solvent was removed from the organic phase, and the residue was separated by CC (7:3 hexane-EtOAc) to give stilbene **15** as a colorless oil (0.40 g, 84%): bp (dec) 110–112 °C (0.01 mmHg) ^{1}H NMR (CD_3OD, 300 MHz) δ 2.32 (s, 3H), 3.66 (s, 6H), 3.86 (s, 3H), 5.60 (s, 1H), 6.42 (d, *J* = 12 Hz, 1H), 6.50 (d, *J* = 12 Hz, 1H), 6.55 (s, 2H), 6.72 (d, *J* = 8.1 Hz, 1H), 6.79 (dd, *J* = 2.1, 8.1 Hz, 1H), 6.90 (d, *J* = 2.1 Hz, 1H); ^{13}C NMR (CD_3OD, 75 MHz) δ 20.53, 55.95, 56.02, 105.61, 110.27, 115.01, 121.04, 127.57, 128.74, 130.07, 130.26, 135.37, 145.15, 145.74, 151.64, 168.64; *anal*. C 66.33%, H 5.80%, calcd for $C_{19}H_{20}O_6$, C 66.27%, H 5.85%.

4-Acetoxy-3′-*O*-bis(benzyl)phosphoryl-3,4′,5-trimethoxy-*Z*-stilbene (16)

To a solution of 3′-phenol **15** (0.55 g, 1.6 mmol) in acetonitrile (10 mL) that was cooled to −10 °C was added CCl_4 (1.5 mL, 15.8 mmol). The mixture was stirred for 10 min before the addition of DIPEA (0.56 mL, 3.25 mmol) and DMAP (20 mg, 0.15 mmol). After one minute, dibenzylphosphite (0.525 mL, 2.4 mmol) was added (dropwise over 5 min). The reaction mixture was stirred an additional 3 h at −10 °C, treated with KH_2PO_4 (20 mL, 0.5 M), stirred for a further 10 min, and then extracted with EtOAc. The solvent was removed from the organic phase, and the residue was separated by CC (3:2 hexane-EtOAC) to yield phosphate **16** (0.65 g, 68%): mp 82–83 °C; ^{1}H NMR ($CDCl_3$, 300 MHz) δ 3.64 (s, 6H), 3.76 (s, 3H), 5.11 (s, 2H), 5.14 (s, 2H), 6.44 (d, *J* = 2.1 Hz, 2H), 6.50 (s, 2H), 6.77 (d, *J* = 8.4 Hz, 1H), 7.06 (d, *J* = 8.1 Hz, 1H), 7.12 (m, 1H), 7.32 (s, 10H). *anal*. C 66.82%, H 5.79%, calcd for $C_{33}H_{33}O_9P$, C 66.57%, H 5.50%.

3′-*O*-Bis(benzyl)phosphoryl-4-hydroxy-3,4′,5-trimethoxy-*Z*-stilbene (3)

To a solution of stilbene **16** (0.60 g, 0.1 mmol) in CH_3OH (10 mL) was added K_2CO_3 (5 mL, 5% in 1:1 CH_3OH-H_2O). After stirring for 1 h, the reaction was terminated by the addition of HC1 (1N, until pH ~ 7) and extracted with EtOAc. Solvent was removed (in vacuo) from the organic phase, and the residue was separated by CC (3:2 hexane-EtOAc) to afford stilbene **3** as a yellow oil (0.18 g, 30%): bp (dec) 53–55 °C (0.01 mm); ^{1}H NMR ($CDCl_3$, 300 MHz) δ 3.80 (s, 3H), 3.94 (s, 6H), 5.18 (s, 2H), 5.20 (s, 2H), 5.6 (bs, 1H), 6.70 (s, 2H), 6.81 (s, 1H), 6.88 (d, *J* = 8.4 Hz, 1H), 7.21 (d, *J* = 8.4 Hz, 1H), 7.33 (m, 10H); *anal.* C 65.93%, H 5.82%, calcd for $C_{31}H_{31}O_8P$, C 66.19%, H 5.55%.

3-*O*-Bis(benzyl)phosphoryl-4-methoxybenzaldehyde (17)

Isovanillin (5 g, 32 mmol) was phosphorylated using DIPEA (1.7 mL, 67 mmol), DMAP (0.40 g, 3.2 mmol), dibenzylphosphite (10.6 mL, 48 mmol) acetonitrile (200 mL), and CCl_4 (30 mL, 320 mmol) as described above (see **16**). Removal of solvent (in vacuo) and separation by CC (1:1 hexane-EtOAc) afforded phosphate **17** (11.2 g, 85%) as a colorless solid: mp 68–69 °C (from hexane-EtOAc); ^{1}H NMR ($CDCl_3$, 300 MHz) δ 3.85 (s, 3H), 5.18 (d, *J* = 9.3 H, 4H), 7.22 (d, *J* = 8.4 Hz, 1H), 7.34 (s, 10H), 7.62 (s, 1H), 7.77 (m, 1H), 9.75 (s, 1H).

4-(*tert*-Butyldiphenylsilyloxy)-3′-*O*-bis(benzyl)phosphoryl-3,4′,5-trimethoxy-*Z*-stilbene (11). From Salt 7. Method B

To a suspension of Wittig salt **7** (7.6 g, 10 mmol) in toluene (500 mL, freshly distilled) was added NaH (0.53 g, 13 mmol) with stirring (1 h). The solution was cooled to 0 °C before the addition of aldehyde **17** (4.13 g, 10 mmol). Stirring was continued at 0 °C for 6 h and at rt for 16 h before the addition of water (100 mL) and extraction with EtOAc. After removal of solvent (in vacuo) from the organic phase, the oil was separated by CC (7:3 hexane-EtOAc), affording stilbene **11** (2.1 g, 25%) as a colorless oil.

3′-*O*-Bis(benzyl)phosphoryl-combretastatin A-4 (18)

To a solution of $Na_2S_2O_3$ (20 mg, 0.125 mmol), TBAF (125 μL, 1M in THF) and CH_3I (80 L, 1.25 mmol) in THF (1 mL) was added stilbene **11** (0.10 g, 0.125 mmol), and the mixture was stirred for 1 h at rt. The reaction was terminated by the addition of water (5 mL) and the mixture was extracted with DCM. The solvent was removed (in vacuo) from the organic phase, and the residue was separated by CC (3:2 hexane-EtOAc) to yield stilbene **18**[4] (51 mg, 74%): mp 73 °C; (lit4 mp 73 °C); ^{1}H NMR ($CDCl_3$, 300 MHz) δ 3.67 (s, 6H), 3.77 (s, 3H), 3.80 (s, 3H), 5.12 (s, 2H), 5.14 (s, 2H), 6.40 (d, *J* = 12 Hz, 1H), 6.45 (d, *J* = 12 Hz, 1H), 6.48 (s, 2H), 6.78 (d, *J* = 9 Hz, 1H), 7.06 (dd, *J* = 1.8, 9 Hz, 1H), 7.15 (d, *J* = 1.8 Hz, 1H), 7.30 (s, 10H).

Acknowledgments

Financial support we are pleased to record was provided by Outstanding Investigator Grant CA44344-10-12 and RO1 CA90441-01-04, 2R56-CA 09441-06A1, and 5RO1 CA 090441-07 awarded by the Division of Cancer Treatment and Diagnosis, National Cancer Institute, DHHS; the Arizona Biomedical Research Commission; the Robert B. Dalton Endowment Fund; the Caitlin Robb Foundation; Dr. John C. Budzinski; the Eagles Art Ehrmann Cancer Fund; and the Ladies Auxiliary to the Veterans of Foreign Wars. For other helpful assistance, we thank Drs. John C. Knight and Michael D. Williams (NSF grant CHE-9808678).

References and Notes

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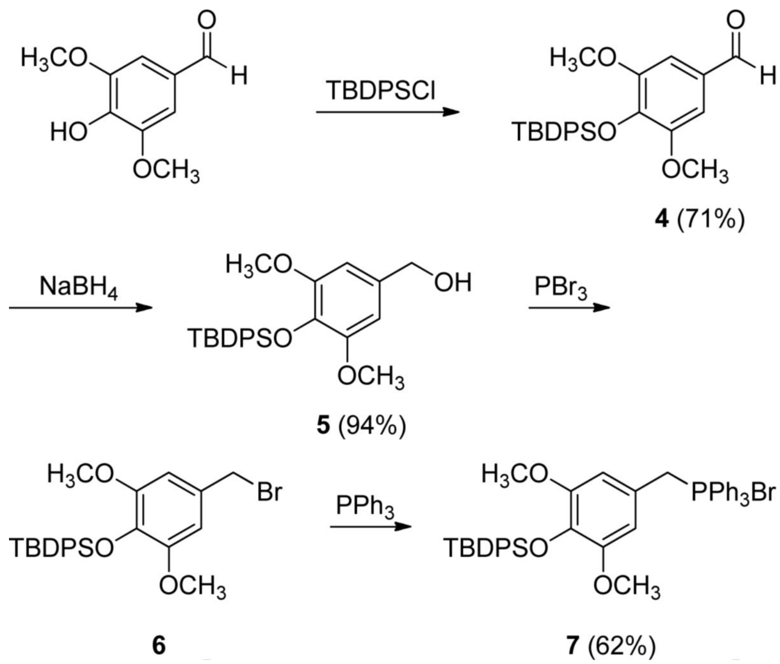
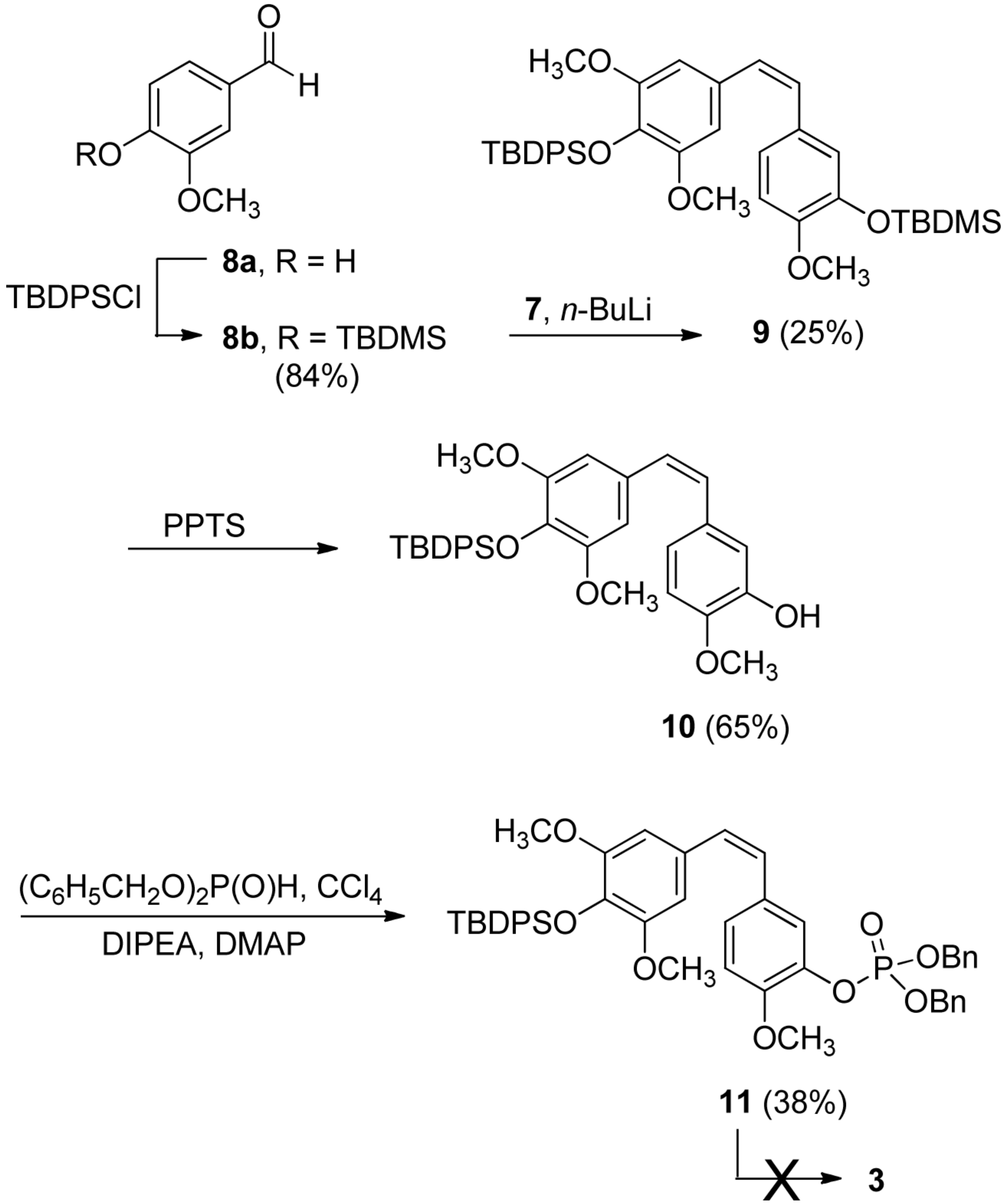
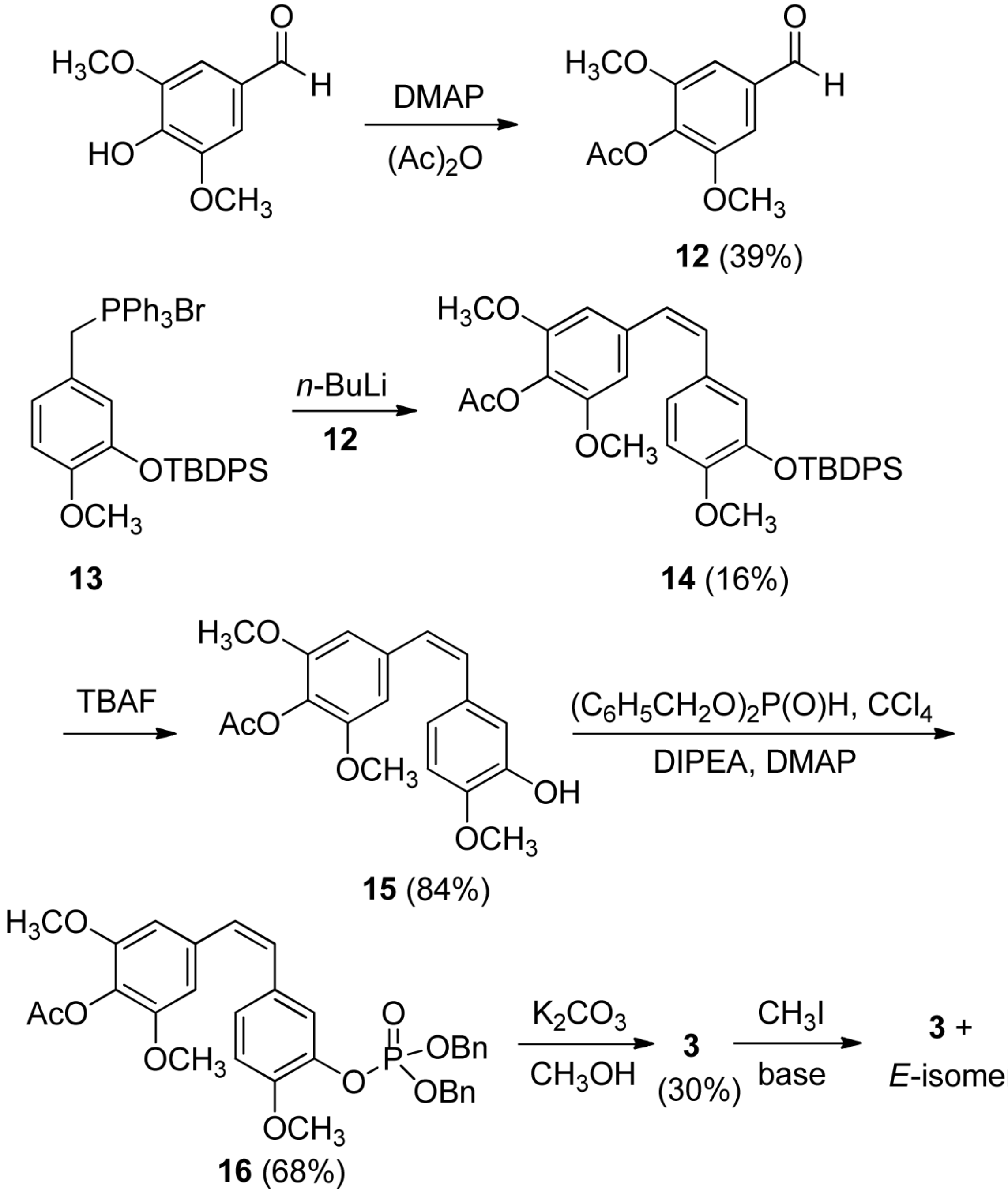


Scheme 1.

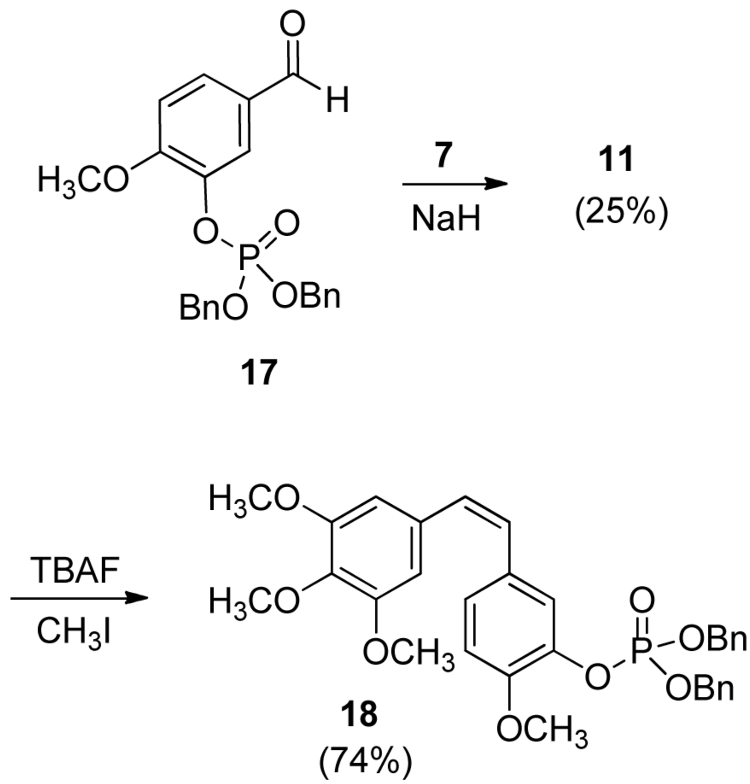


RO
OCH3
H
TBDPSCl
8a, R = H
8b, R = TBDMS
(84%)
7, n-BuLi
H3CO
TBDPSO
OCH3
OTBDMS
OCH3
9 (25%)
PPTS
OH
10 (65%)
(C6H5CH2O)2P(O)H, CCl4
DIPEA, DMAP
OBn
OBn
11 (38%)
3

Scheme 2.

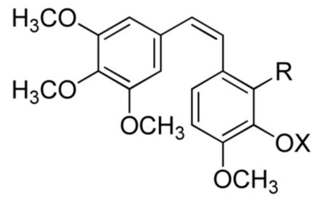

H3CO
HO
OCH3
O
H
DMAP
(Ac)2O
AcO
12 (39%)
PPh3Br
OTBDPS
13
n-BuLi
12
14 (16%)
TBAF
OH
15 (84%)
(C6H5CH2O)2P(O)H, CCl4
DIPEA, DMAP
OBn
16 (68%)
K2CO3
CH3OH
3
(30%)
CH3I
base
3 +
E-isomer

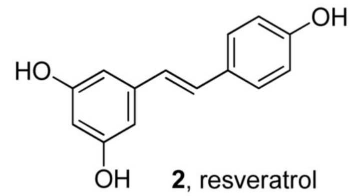
Scheme 3.



H3CO
O
H
O
P
O
BnO
OBn
17
7
NaH
11
(25%)
TBAF
CH3I
H3CO
H3CO
OCH3
O
P
OBn
OBn
OCH3
18
(74%)

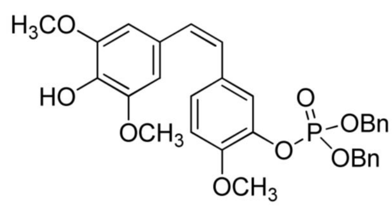
Scheme 4.





2, resveratrol

1a, R = X = H, CA4
b, R = H, X = $P(O)(ONa)_2$, CA4P
c, R = OH, X = H, CA1
d, R = $OP(O)(ONa)_2$, X = $P(O)(ONa)_2$, CA1P



3