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# Total Synthesis of (+)-Medicarpin

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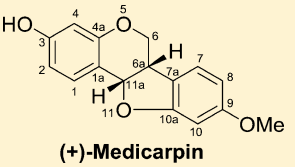
Supporting Information

**ABSTRACT:** (+)-Medicarpin has been synthesized asymmetrically for the first time in a linear scalable process with an overall yield of 11%. The two chiral centers were constructed in one step via condensation using a chiral oxazolidinone auxiliary. This method will likely accelerate research on medicarpin as an erythropoietin inducer for erythropoietin-deficient diseases.




Medicarpin (9-methoxy-6a,11a-dihydro-6*H*-benzo[4,5]-furo-[3,2-*c*]chromen-3-ol) (Figure 1) is a pterocarpan,

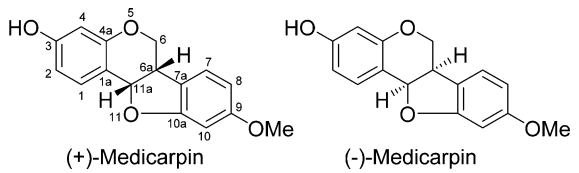



**Figure 1.** Structures of (+)-medicarpin and (−)-medicarpin.

a type of isoflavonoid. (+)-Medicarpin has been isolated from several medicinal plant species with various biological effects, including *Sophora japonica*[1] as a phytoalexin, *Zollernia paraensis*[2] and *Platymiscium yucatamun*[3] with antifungal properties, *Machaerium aristulatum*,[4] *Platymiscium floribundum*,[5] and Brazilian red propolis[6] with cytotoxic effects, and *Dalbergia oliveri*[7] as a larvicidal compound.

Medicarpin and the flavonoids formononetin and ononin have been isolated as constituents of a major compound group from "Radix Hedysari", the dried roots of *Hedysarum polybotrys*. Radix Hedysari ("Hongqi" in mainland China) has been used commonly as a substitute for "Radix Astragali" (*Astragalus membranaceus*, "Huangqi" in mainland China). The latter, one of the most important and widely used tonics in Traditional Chinese Medicine (TCM), is utilized as a restorative food with estrogenic, erythropoietic, and osteogenic properties and as an herbal decoction to reinforce "Qi" (vital energy). Formononectin and ononin act as erythropoietin (EPO) inducers via the activation of hypoxia-inducible factor-1alpha (HIF-1$\alpha$).[8] However, our team has found that, while medicarpin is also an EPO inducer, it does not act through HIF-1$\alpha$ (unpublished results). Most EPO inducers that do act through the HIF-1$\alpha$ pathway have failed in clinical trials due to cardiovascular side effects; thus, medicarpin merits study as a replacement for existing EPO inducers.

Medicarpin, mostly as the racemate, has also been investigated as a treatment for postmenopausal osteoporosis. The compound potently inhibits osteoclastogenesis and prevents estrogen-deficient bone loss but does not display uterine estrogenicity.[9] It also promotes bone healing[10] and increases bone mass by osteoblast differentiation with estrogen receptor (ER) $\beta$-mediated osteogenic action.[11] In a comparative study of racemic and both enantiomerically pure medicarpins, (+)-medicarpin was determined to be the most potent species regarding increased levels of two osteogenic genes (Runx-2 and BMP-2).[12]

In addition, medicarpin has been associated with potential anticancer properties and may be a chemotherapy sensitizer for multi-drug-resistant P388 leukemia cancer cells.[13] The combination of medicarpin and tumor necrosis factor $\alpha$-related apoptosis-inducing ligand (TRAIL) achieved enhanced apoptosis without significantly influencing cytotoxicity in primary normal human peripheral blood mononuclear cells.[14] This contrast between medicarpin-mediated apoptotic regulation in normal and cancer cells is also related to medicarpin's osteogenic activity. In recent studies, medicarpin down-regulated GRP78, an ER chaperone with antiapoptotic effects, thereby leading to osteoblast differentiation and increased osteoblast survival.[15]

Several synthetic routes to racemic pterocarpans, including (±)-medicarpin,[9,10,12] have been reported. The most common key step in generating the pterocarpan four-ring system has involved hydrogenative cyclization[16] or borohydride reductive cyclization of isoflavones,[9,10,12,17] while other approaches

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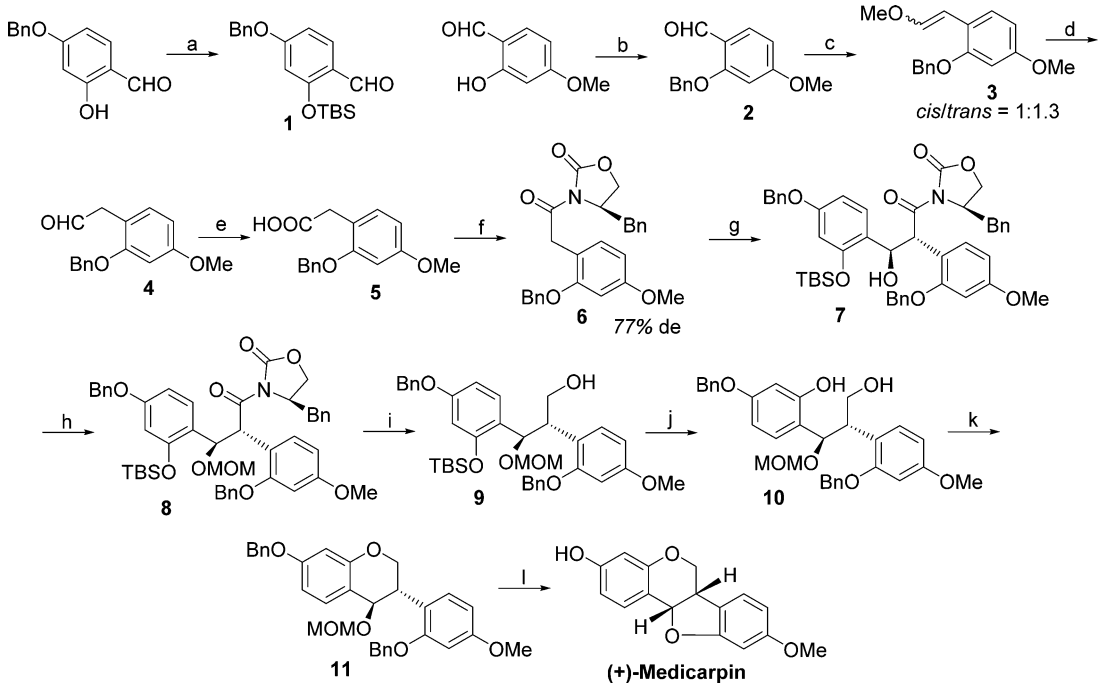
**Scheme 1. Synthetic Route to (+)-Medicarpin**[a]




[a]Reagents and condition: (a) TBSCl, imidazole, DMF, rt, 3 h, 99%; (b) BnBr, $K_2CO_3$, acetonitrile, reflux overnight, 93%; (c) *t*-BuOK, $Ph_3P^+CH_2OMeCl^-$, THF, ice bath, 1 h, 93%; (d) 3 N HCl, THF, reflux, 1 h, 86%; (e) 2-methyl-2-butene, $NaClO_2$, $NaH_2PO_4$, THF, *t*-BuOH, rt, 3 h, 81%; (f) (i) oxalyl chloride, $CH_2Cl_2$; (ii) (*R*)-4-benzyl-2-oxazolidinone, *n*-BuLi, THF, −78 °C to rt, 2 h, 79%; (g) $Bu_2BOTf$, DIPEA, $CH_2Cl_2$, −78 to 0 °C, then **1**, −78% °C to rt, 76%; (h) MOMCl, DIPEA, rt, 48 h, 83%; (i) $LiBH_4$, aq $Et_2O$, 0 °C to rt, 1 h, 61%; (j) TBAF, THF, rt, 1 h, 96%; (k) TPP, DEAD, THF, rt, 30 min, 96%; (l) (i) Pd/C, $H_2$, MeOH, EtOAc, rt, 24 h; (ii) CSA, $CH_2Cl_2$, rt, 30 min, 55% for two steps.

involved 3 + 2 cycloaddition of 2*H*-chromenes with 2-alkoxy-1,4-benzoquinones,[18] Heck arylation of bischromenes with *o*-chloromercuriphenols,[19] and 1,3-Michael−Claisen condensation of α-methylene-γ-butyrolactones prepared from 2,3-dihydro-7-hydroxy-4*H*-1-benzopyran-4-one with α-sulfur-substituted ketones.[20] However, pure optical pterocarpan isomers could only be obtained via optical resolution using chiral high-performance liquid chromatography (HPLC).[12] Some enantioselective approaches[12] have been described, including a recent enantioselective total synthesis of (−)-medicarpin in a total overall yield of 4% in nine steps.[21] The key diastereoselective step in this preparation involved an *ortho*-quinone methide Diels−Alder reaction, while the benzopyran system was assembled using an oxidative cyclization presumably involving a *para*-quinone intermediate.

Ultimately, stereoselectivity is a key issue in the synthesis of all naturally occurring flavonoids,[22] including the formation of the $C_{6a}$ and $C_{11a}$ of pterocarpans. Some enantioselective approaches have been described,[12] including a recent enantioselective total synthesis of (−)-medicarpin in a total overall yield of 4% in nine steps.[21] The key diastereoselective step in this preparation involved an *ortho*-quinone methide Diels−Alder reaction, while the benzopyran system was assembled using an oxidative cyclization likely involving a *para*-quinone. Earlier, Ferreira et al.[23−25] employed an aldol condensation of phenylacetates with benzaldehydes, by which stereoselectivity could be introduced. The resulting 2,3-diaryl-3-hydroxypropanoate products could then undergo stepwise deprotection and cyclization to the desired pterocarpans.

## RESULTS AND DISCUSSION

As shown in Scheme 1, the present asymmetric strategy for the synthesis of (+)-medicarpin is based on the modification of Ferreira's synthesis. It was envisioned that the desired stereochemistry at $C_{6a}$ and $C_{11a}$ of (+)-medicarpin could be readily generated by an asymmetric Evans' aldol reaction. This synthesis can be accomplished using a chiral oxazolidone auxiliary, specifically, (*R*)-4-benzyl-2-oxazolidinone.

At first, the initial intermediates **1** and **2** were obtained by protecting the hydroxy groups in 4-benzyloxy-2-hydroxybenzaldehyde and 4-methoxy-2-hydroxybenzaldehyde as the *t*-butyldimethylsilyl and benzyl ethers, respectively. The former compound is the source of the A-ring and $C_{11a}$ of medicarpin, while the latter compound supplies the D-ring and $C_{6a}$. A classic Wittig olefination with (methoxymethyl)triphenylphosphonium chloride converted the aldehyde in **2** to a methoxyvinyl group in **3** and provided the additional carbon to become the $C_6$ moiety of medicarpin. Compound **3** was obtained in a mixture of *cis* and *trans* isomers with a ratio of ca. 1:1.3 (based on the $^1$H NMR integral) and used in subsequent reactions without further separation. Acidic hydrolysis of **3** with dilute HCl gave 2-benzyloxy-4-methoxyphenylacetaldehyde (**4**) in very high yield. A Pinnick oxidation (sodium chlorite, 2-methyl-2-butene, monosodium phosphate) of the aldehyde group in compound **4** provided the carboxylic acid functionality in compound **5**. Treatment of **5** with oxalyl chloride gave the acyl chloride, which was reacted without column purification with (*R*)-4-benzyl-2-oxazolidinone using *n*-butyllithium as a base to give imide **6** in 79% yield. An Evans' asymmetric aldol addition between aldehyde **1** and the boron enolate of imide **6** gave aldol adduct **7** with a good value of diastereomeric excess


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Table 1. Comparison of the $^1$H NMR Data of Synthetic (+)-Medicarpin and Natural Sample (Measured at 400 MHz in $CDCl_3$, $\delta$ in ppm)

| synthetic (+)-medicarpin | natural (+)-medicarpin |
|---|---|
| 7.39 (1H, d, $J$ = 8.8 Hz) | 7.40 (1H, d, $J$ = 8.5 Hz) |
| 7.13 (1H, d, $J$ = 8.8 Hz) | 7.16 (1H, d, $J$ = 8.8 Hz) |
| 6.55 (1H, dd, $J$ = 8.0 Hz, 2.4 Hz) | 6.58 (1H, dd, $J$ = 8.5 Hz, 2.4 Hz) |
| 6.46−6.44 (2H, m) | 6.50 (2H, br s) |
| 6.41 (1H, d, $J$ = 2.4 Hz) | 6.45 (1H, d, $J$ = 2.4 Hz) |
| 5.50 (1H, d, $J$ = 6.8 Hz) | 5.53 (1H, d, $J$ = 6.7 Hz) |
| 4.24 (1H, dd, $J$ = 11.2 Hz, 4.8 Hz) | 4.26 (1H, dd, $J$ = 10.9 Hz, 4.8 Hz) |
| 3.76 (3H, s) | 3.80 (3H, s) |
| 3.62 (1H, t, $J$ = 10.8 Hz) | 3.65 (1H, dd, $J$ = 10.9, 10.9 Hz) |
| 3.55−3.50 (1H, m) | 3.55 (1H, m) |

(de 77%) and the desired stereochemistry at position $C_{6a}$. Protection of the hydroxy group in **7**, giving the methoxymethyl ether in compound **8**, was achieved readily by treating **7** with chloromethyl methyl ether in the presence of *N*,*N*-diisopropylethylamine. Subsequently, the oxazolidinone auxiliary was removed with lithium borohydride to generate the required primary hydroxy group in **9**. Selective deprotection of the silyl ether was achieved by using tetrabutylammonium fluoride (TBAF) to give the diol **10**. The B-ring of medicarpin was formed by using Mitsunobu conditions [triphenylphosphine, diethyl azodicarboxylate (DEAD)] to produce compound **11**, a bicyclic alkyl aryl ether (chromane) in excellent yield, from **10**. Finally, regioselective palladium-catalyzed hydrogenation followed by treatment with camphorsulfonic acid resulted in debenzylation and stereoselective cyclization to form the C-ring in the dihydrobenzofuran moiety of (+)-medicarpin in a moderate yield in two steps. The $^1$H NMR, $^{13}$C NMR, and optical data of the synthetic (+)-medicarpin were in good agreement with previous values reported in the literature.[7,12,26,27] The comparison of the $^1$H NMR spectroscopic data of synthetic (+)-medicarpin with that of a natural sample is outlined in Table 1.

In summary, starting from commercially available 4-methoxy-2-hydroxybenzaldehyde, a new asymmetric synthesis of (+)-medicarpin was achieved in 11 steps with an overall yield of 11%. Compared with previous synthetic works, the present method was more efficient in terms of overall reaction yields and could be readily scaled up to produce gram quantities of (+)-medicarpin. The developed synthetic method may be expected to find applications in the synthesis of relevant medicarpin analogues by the variation of starting materials. Currently, elaboration of the protocol to the synthesis of (−)-medicarpin with the minor modification of the chiral oxazolidone auxiliary is under investigation.

## EXPERIMENTAL SECTION

**General Experimental Procedures.** All reagents and solvents were used as received from Sigma-Aldrich or other commercial sources. The solvent used, unless otherwise indicated, was $CDCl_3$. Thin-layer chromatography (TLC) was performed on Merck percolated silica gel 60 F-254 plates. To purify all synthetic compounds, silica gel chromatography was carried out on an ISCO CombiFlash Rf flash chromatograph system with prepacked Redi Sep Rf Si gel column (Teledyne ISCO). NMR spectroscopic data were measured on an Inova-400 instrument with $Me_4Si$ (TMS) as internal standard. High-resolution mass spectra were measured on a Thermo LTQ-FT-ICR-MS-7T spectrometer, and data were recorded as $m/z$ values. Melting points were measured using an electrothermal instrument. Analytical HPLC resolution was performed on a HPLC Shimadzu system consisting of a LC-20AT HPLC pump, a SIL-20A$_{HT}$ autosampler, CBM-20A and SPD-M20A detectors, and CHIRALPAK IB (3 $\mu$m) column with hexane and ethyl acetate (4:1) as eluent (25 °C, 1 mL/min).

*4-Benzyloxy-2-(tert-butyldimethylsilyloxy)benzaldehyde (**1**).* 4-Benzyloxy-2-hydroxybenzaldehyde (11.40 g, 50 mmol), imidazole (3.74 g, 55 mmol), and *t*-butyldimethylsilyl chloride (TBSCl) (8.39 g, 54 mmol) were mixed in dimethylformamide (DMF) (100 mL) and stirred at rt for 3 h. MeOH was then added, and the mixture was stirred for another 30 min. Water was added, and the resultant mixture was extracted with $Et_2O$. The organic layers were combined, washed with $H_2O$ and brine, dried over $MgSO_4$, and purified by flash chromatography using EtOAc and hexane to give compound **1** in quantitative yield as a colorless oil: $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 10.28 (1H, s), 7.79 (1H, d, $J$ = 8.8 Hz), 7.40−7.33 (5H, m), 6.68 (1H, dd, $J$ = 8.8 Hz, 2.4 Hz), 6.37 (1H, d, $J$ = 2.4 Hz), 5.10 (2H, s), 0.99 (9H, s), 0.22 (6H, s); $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 188.5, 164.7, 160.6, 135.9, 130.0, 128.7, 128.2, 127.2, 121.4, 108.8, 105.9, 70.2, 60.3, 25.6, 18.2, −4.4; HRESIMS $m/z$ 357.1886 $[M + CH_3 + H]^+$ (calcd for $C_{21}H_{29}O_3Si$ 357.1886).

*2-Benzyloxy-4-methoxybenzaldehyde (**2**).* To a solution of 4-methoxy-2-hydroxybenzaldehyde (15.2 g, 100 mmol) in acetonitrile (200 mL) were added $K_2CO_3$ (16.56 g, 120 mmol) and BnBr (13.07 mL, 110 mmol), and the resultant solution was refluxed overnight. The mixture was cooled, and the solvent was then removed under a vacuum. $H_2O$ (100 mL) was added to the residue, which was then extracted with $Et_2O$ and brine and dried over $MgSO_4$. Recrystallization from MeOH gave compound **2** (22.5 g, 93%). Spectroscopic data were in agreement with literature values.[28]

*Methyl Vinyl Ether **3**.* *t*-BuOK (15.66 g, 139.5 mmol) was added in portions to (methoxymethyl)triphenylphosphonium chloride (47.82 g, 139.5 mmol) in 300 mL of anhydrous THF at 0 °C under Ar. After 30 min, aldehyde **2** (22.50 g, 93 mmol) in 100 mL of anhydrous THF was added dropwise over 30 min, and the mixture was stirred for another 30 min before saturated $NH_4Cl$ was added to quench the reaction. EtOAc was used for extraction, and the organic layers were washed with brine and dried over $MgSO_4$. A mixture of *cis*- and *trans*-vinyl ether **3** (based on the $^1$H NMR integration, the *cis*/*trans* ratio was determined as ca. 1:1.3) was obtained (25.06 g, 93%) as colorless oil by flash chromatography using EtOAc and hexane: $^1$H NMR (400 MHz, $CDCl_3$) *cis* + *trans* isomers $\delta$ 7.95 (0.360H, d, $J$ = 8.0 Hz), 7.44−7.31 (4.40H, m), 7.15 (0.52H, d, $J$ = 8.0 Hz), 7.02 (0.48H, d, $J$ = 12.0 Hz), 6.50−6.44 (2H, m), 6.08 (0.40H, d, $J$ = 8.0 Hz), 6.01 (0.49H, d, $J$ = 12.0 Hz), 5.63 (0.38H, d, $J$ = 8.0 Hz), 5.07 (1.12H, s), 5.04 (0.88H, s), 3.36−3.37 (3H, m), 3.73 (1.32H, s), 3.62 (1.68H, s); $^{13}$C NMR (100 MHz, $CDCl_3$) *cis* + *trans* isomers $\delta$ 158.8, 158.6, 156.0, 155.8, 148.3, 146.3, 137.1, 137.0, 130.2, 128.5, 128.4, 127.8, 127.7, 127.2, 127.2, 127.2, 126.7, 118.4, 118.1, 105.1, 104.7, 100.7, 100.2, 99.6, 98.6, 70.3, 70.2, 60.3, 56.4, 55.3, 55.3; HRESIMS $m/z$ 271.1330 $[M + H]^+$ (calcd for $C_{17}H_{19}O_3$ 271.1334).

*2-Benzyloxy-4-methoxyphenylacetaldehyde (**4**).* To a solution of **3** (13.52 g, 50 mmol) in 200 mL of THF was added 3 N HCl (15 mL), followed by reflux for 1 h. The mixture was cooled to rt, and


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saturated $NaHCO_3$ (100 mL) was added. $Et_2O$ was used for extraction, and the organic layers were washed with brine and dried over $MgSO_4$. The solvent was removed in vacuo, and the residue was purified by flash chromatography using EtOAc and hexane to afford **4** (11.05 g, 86%) as a light yellow oil: $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 9.67 (1H, s), 7.37−7.29 (5H, m), 7.05 (1H, d, $J$ = 8.0 Hz), 6.55 (1H, s), 6.48 (1H, d, $J$ = 8.0 Hz), 5.04 (2H, s), 3.77 (3H, s), 3.61 (2H, s); $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 200.3, 160.4, 157.5, 136.5, 131.6, 128.6, 127.9, 127.2, 127.2, 113.7, 104.8, 99.9, 70.1, 55.3, 44.8; HRESIMS $m/z$ 257.7738 $[M + H]^+$ (calcd for $C_{16}H_{17}O_3$ 257.7718).

*2-Benzyloxy-4-methoxyphenylacetic Acid (**5**).* At 0 °C, $NaH_2PO_4$ (6.21 g, 51.7 mmol) in 40 mL of $H_2O$ was added to aldehyde **4** (11.05 g, 43.1 mmol), 2-methyl-2-butene (18.14 g, 258.7 mmol), and $NaClO_2$ (tech. 80%, 5.85 g, 51.74 mmol) in *t*-BuOH/THF (1:1, 200 mL). The ice bath was removed after 30 min, and the mixture was stirred for 3 h. The organic solvents were removed in vacuo, and the aqueous layer was extracted with $CH_2Cl_2$ and then dried over $MgSO_4$. After solvent was evaporated, the crude compound was purified by recrystallization from EtOAc−hexane to give compound **5** (9.50 g, 81%) as a white solid: mp 114.0−116 °C; $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 7.35−7.26 (5H, m), 7.10 (1H, d, $J$ = 8.0 Hz), 6.50 (1H, d, $J$ = 2.4 Hz), 6.46 (1H, dd, $J$ = 8.0, 2.4 Hz), 3.76 (3H, s), 3.63 (2H, s); $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 177.6, 160.2, 157.3, 136.6, 131.3, 128.5, 127.8, 127.0, 115.1, 104.6, 99.9, 70.0, 55.3, 35.2; HRESIMS $m/z$ 273.1120 $[M + H]^+$ (calcd for $C_{16}H_{17}O_4$ 273.1127).

*Imide **6**.* To a solution of acid **5** (10.64 g, 39.1 mmol) in anhydrous $CH_2Cl_2$ (100 mL) were added oxalyl chloride (7 mL) and DMF (catalytic). The mixture was then stirred at rt for 1 h, followed by evaporation of solvent. The residue was dissolved in anhydrous THF to give an acyl chloride solution. In another flask, *n*-BuLi (2.5 M, 16.4 mL, 41 mmol) was added to (*R*)-4-benzyl-2-oxazolidinone (7.09 g, 40 mmol) in anhydrous THF (160 mL) at −78 °C. The mixture was warmed to 0 °C, stirred for an additional 30 min, and then recooled to −78 °C. The acyl chloride solution was added dropwise to the reaction mixture and stirred at the same temperature for 1 h before being warmed to rt and stirred for 2 h. Saturated aqueous $NH_4Cl$ was added, and the mixture was extracted with EtOAc, which was washed with brine, dried over $MgSO_4$, and triturated with EtOAc and hexane to give **6** (13.3 g, 79%) as a white solid: mp 108.0−110.0 °C; $[\alpha]_D^{23}$ −52.2 (*c* 0.12, MeOH); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 7.40−7.25 (8H, m), 7.13−7.09 (3H, m), 6.55 (1H, d, $J$ = 2.4 Hz), 6.50 (1H, dd, $J$ = 8.0 Hz, 2.4 Hz), 5.03 (2H, s), 4.54−4.48 (1H, m), 4.22 (2H, d, $J$ = 2.8 Hz), 4.04 (1H, dd, $J$ = 8.8 Hz, 2.8 Hz), 3.97 (1H, t, $J$ = 8.0 Hz), 3.79 (3H, s), 3.19 (1H, dd, $J$ = 13.4 Hz, 2.8 Hz), 2.47 (1H, dd, $J$ = 13.4 Hz, 10.0 Hz); $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 171.5, 160.2, 157.4, 153.5, 136.9, 135.5, 131.5, 129.3, 128.8, 128.8, 128.5, 127.9, 127.4, 127.1, 115.7, 104.5, 99.8, 70.8, 66.1, 55.4, 55.3, 37.6, 36.8; HRESIMS $m/z$ 432.1817 $[M + H]^+$ (calcd for $C_{26}H_{26}NO_5$ 432.1811).

*Alcohol **7**.* To a stirred solution of imide **6** (7.56 g, 17.52 mmol) in anhydrous $CH_2Cl_2$ (100 mL) was added *N*,*N*-diisopropylethylamine (DIPEA) (3.70 mL, 21 mmol) and dibutylboron triflate (1 M in $CH_2Cl_2$, 20 mL) at −78 °C. The mixture was warmed to 0 °C and stirred for 1 h. The aldehyde **1** (6.60 g, 19.27 mmol) in anhydrous $CH_2Cl_2$ was added dropwise, and the resultant mixture was stirred at −78 °C for 1 h. The reaction was warmed to rt, stirred overnight, and quenched by addition of pH 7 buffer (25 mL), followed by slow addition of MeOH−$H_2O$ (2:1, 50 mL) with ice bath cooling followed by further stirring at rt for 1 h. The organic solvent was removed under vacuum, and $Et_2O$ was used for extraction. The organic layers were washed with saturated $NaHCO_3$ and dried over $MgSO_4$. Flash chromatography (hexane−EtOAc 9:1) gave **7** (10.3 g, 76%) as a white solid: mp 74.0−76.0 °C; $[\alpha]_D^{23}$ +70.3 (*c* 0.15, MeOH); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 7.39−7.24 (13H, m), 7.11−7.09 (3H, m), 6.76 (1H, d, $J$ = 8.0 Hz), 6.41 (2H, ddd, $J$ = 15.6, 8.4, 2.4 Hz), 6.30 (1H, d, $J$ = 2.4 Hz), 6.12 (1H, d, $J$ = 2.4 Hz), 5.59 (2H, s), 4.92 (2H, s), 4.81 (1H, d, $J$ = 12.0 Hz), 4.64−4.58 (1H, m), 4.53 (1H, d, $J$ = 12.0 Hz), 3.96−3.95 (2H, m), 3.71 (3H, s), 3.67 (1H, d, $J$ = 2.4 Hz), 3.28 (1H, dd, $J$ = 13.4 Hz, 2.8 Hz), 2.42 (1H, dd, $J$ = 13.4 Hz, 10.0 Hz), 0.94 (9H, s), 0.13 (6H, s); $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 175.2, 160.2, 158.7, 158.6, 153.2, 152.1, 137.5, 137.4, 135.6, 131.2, 129.6, 129.1, 128.7, 128.5, 128.2, 128.1, 127.7, 127.6, 127.5, 127.4, 127.2, 124.6, 114.6, 106.9, 105.4, 104.2, 100.0, 70.4, 70.1, 68.8, 66.1, 55.4, 55.4, 48.1, 37.8, 26.0, 18.3; HRESIMS $m/z$ 796.3276 $[M + Na]^+$ (calcd for $C_{46}H_{51}NNaO_8Si$ 796.3286).

*MOM-Protected Alcohol **8**.* MOMCl (2.00 mL) was added dropwise to alcohol **7** (5.96 g, 7.70 mmol) and DIPEA (4.34 mL) in anhydrous $CH_2Cl_2$ (50 mL) at 0 °C, and the mixture was warmed to rt overnight. After TLC indicated the reaction was not complete, additional amounts of MOMCl (2.00 mL) and DIPEA (4.34 mL) were added, and stirring was continued overnight. $H_2O$ was added, and the mixture was stirred for 10 min, extracted with $CH_2Cl_2$, and washed with brine. Flash chromatography (hexane−EtOAc 9:1) gave **8** (5.23 g, 83%) as a light yellow oil: $[\alpha]_D^{23}$ +77.4 (*c* 0.09, MeOH); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 7.59 (1H, d, $J$ = 8.0 Hz), 7.44 (2H, d, $J$ = 8.0 Hz), 7.36−7.26 (8H, m), 7.24−7.19 (4H, m), 7.00 (2H, dd, $J$ = 7.2, 1.2 Hz), 6.54 (2H, td, $J$ = 9.0, 2.4 Hz), 6.45 (1H, d, $J$ = 2.4 Hz), 6.25 (1H, d, $J$ = 2.4 Hz), 6.04 (1H, d, $J$ = 8.8 Hz), 5.63 (1H, d, $J$ = 8.8 Hz), 5.04−4.96 (2H, m), 4.94 (2H, s), 4.45 (1H, d, $J$ = 8.8 Hz), 4.36 (1H, d, $J$ = 8.8 Hz), 4.17−4.11 (1H, m), 3.75 (3H, s), 3.71 (2H, dd, $J$ = 8.8, 2.4 Hz), 3.46 (1H, t, $J$ = 8.0 Hz), 3.09 (1H, dd, $J$ = 13.2 Hz, 3.2 Hz), 3.03 (3H, s), 2.11 (1H, dd, $J$ = 13.2, 10.4 Hz), 1.05 (9H, s), 0.22 (6H, d, $J$ = 2.8 Hz); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ 171.8, 159.8, 159.0, 158.1, 154.6, 152.2, 137.2, 136.9, 135.9, 130.1, 129.9, 129.3, 128.7, 128.5, 128.3, 127.9, 127.5, 127.5, 127.4, 126.9, 122.8, 118.0, 107.1, 104.9, 104.5, 99.9, 94.1, 71.9, 70.4), 69.9, 65.4, 55.9, 55.6, 55.2, 46.8, 37.3, 34.6, 25.8, 18.2; HRESIMS $m/z$ 840.3528 $[M + Na]^+$ (calcd for $C_{48}H_{55}NNaO_9Si$ 840.3544).

*Alcohol **9**.* $LiBH_4$ (4 M in THF, 2.00 mL) was added to alcohol **8** (5.23 g, 6.40 mmol) in aqueous $Et_2O$ (50 mL, plus 0.1 mL $H_2O$) at rt, and the mixture was stirred for 1 h. The reaction was quenched with 1 N NaOH (30 mL) and stirred for an additional 1 h. The solution was extracted with $Et_2O$, washed with brine, and dried over $Mg_2SO_4$. Flash chromatography (hexane−EtOAc 4:1) gave **9** (2.51 g, 61%) as a colorless oil: $[\alpha]_D^{23}$ +141.5 (*c* 0.07, MeOH); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 7.42−7.29 (11H, m), 7.00 (1H, d, $J$ = 8.4 Hz), 6.50 (2H, dt, $J$ = 8.8, 2.4 Hz), 6.39 (1H, d, $J$ = 2.4 Hz), 6.34 (1H, d, $J$ = 2.4 Hz), 5.42 (1H, d, $J$ = 6.4 Hz), 4.98 (2H, s), 4.93 (1H, d, $J$ = 12.4 Hz), 4.81 (1H, d, $J$ = 12.4 Hz), 4.40 (2H, dd, $J$ = 16.4 Hz, 8.4 Hz), 3.81−3.78 (2H, m), 3.75 (3H, s), 3.73−3.71 (1H, m), 3.13 (3H, s), 1.01 (9H, s), 0.18 (6H, d, $J$ = 4.4 Hz); $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 159.3, 158.8, 158.1, 154.5, 137.5, 137.1, 129.8, 129.2, 128.7, 128.5, 128.1, 127.7, 127.6, 127.3, 123.7, 120.9, 107.8, 105.7, 104.8, 100.1, 94.2, 72.1, 70.3, 70.2, 64.5, 55.6, 55.4, 44.8, 29.9, 26.0, 18.4; HRESIMS $m/z$ 667.3055 $[M + Na]^+$ (calcd for $C_{38}H_{48}NaO_7Si$ 667.3067).

*Diol **10**.* To a solution of alcohol **9** (2.51 g, 3.89 mmol) in THF (20 mL) was added tetrabutylammonium fluoride (TBAF) (1 M in THF, 5 mL). After 1 h, water was added, and EtOAc was used for extraction. The organic layers were combined, washed with brine, and dried over $MgSO_4$. Flash chromatography (hexane−EtOAc 1:1) gave diol **10** (1.99 g, 96%) as a colorless oil: $[\alpha]_D^{23}$ +188.0 (*c* 0.08, MeOH); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 7.41−7.30 (10H, m), 7.20 (1H, d, $J$ = 8.4 Hz), 6.85 (1H, d, $J$ = 8.4 Hz), 6.53−6.48 (3H. m), 6.42 (1H, dd, $J$ = 8.4, 2.4 Hz), 5.15 (1H, d, $J$ = 8.4 Hz), 5.00−4.98 (4H, m), 4.54 (1H, d, $J$ = 6.8 Hz), 4.37 (1H, d, $J$ = 6.8 Hz), 3.76 (3H, s), 3.67−3.62 (3H, m), 2.96 (3H, s); $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 159.8, 159.7, 157.8, 156.7, 136.9, 136.6, 129.7, 128.6, 128.5, 128.0, 127.9, 127.6, 127.5, 120.4, 116.8, 106.9, 104.7, 103.4, 100.1, 94.1, 70.4, 69.9, 63.4, 55.8, 55.3; HRESIMS $m/z$ 553.2188 $[M + Na]^+$ (calcd for $C_{32}H_{34}NaO_7$ 553.2202).

*Ether **11**.* To diol **10** (1.99 g, 3.73 mmol) and $PPh_3$ (1.18 g, 4.50 mmol) in 30 mL of anhydrous THF was added DEAD (40 wt % in toluene, 2.10 mL, 4.60 mmol), and TLC indicated the reaction was complete after about 30 min. The mixture was loaded onto silica gel and purified by flash chromatography (hexane−EtOAc 1:1) to give ether **11** (1.83 g, 96%) as a colorless oil: $[\alpha]_D^{24}$ −21.5 (*c* 0.13, MeOH); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 7.46−7.29 (10H, m), 7.14 (1H, d, $J$ = 8.4 Hz), 7.04 (1H, d, $J$ = 8.4 Hz), 6.57 (1H, dd, $J$ = 8.4 Hz, 2.4 Hz), 6.50 (2H, dd, $J$ = 9.6 Hz, 2.4 Hz), 6.32 (1H, dd, $J$ = 8.4 Hz, 2.4 Hz), 5.08 (2H, s), 5.01 (2H, s), 4.70−4.68 (2H, m), 4.62 (1H, d, $J$ = 8.4 Hz), 4.47 (1H, dd, $J$ = 11.2 Hz, 3.2 Hz), 4.45 (1H, dd, $J$ = 11.2,


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3.2 Hz), 3.72 (3H, s), 3.62−3.60 (1H, m), 3.30 (3H, s); $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 160.3, 159.6, 157.1, 151.3, 136.8, 136.7, 130.2, 128.5, 127.9, 127.9, 127.7, 127.5, 127.2, 123.9, 119.9, 115.8, 108.7, 105.4, 103.0, 100.6, 98.6, 90.9, 70.5, 70.1, 55.4, 55.0, 25.6, 14.4; HRESIMS $m/z$ 451.1908 $[M - MOM - H_2O + H]^+$ (calcd for $C_{30}H_{27}O_4$ 451.1909).

*(+)-Medicarpin.* A solution of ether **11** (1.83 g, 3.57 mmol) and 10% Pd/C (400 mg) in EtOAc−MeOH (1:3, 30 mL) was hydrogenated at balloon pressure overnight. The catalyst was filtered off using Celite, and the solvent was removed under vacuum to give a white foam, which was dissolved in anhydrous $CH_2Cl_2$ (30 mL). Camphorsulfonic acid (20 mg) was added, and the mixture was stirred at rt for 30 min. Saturated $NaHCO_3$ was added, and the mixture was extracted with $CH_2Cl_2$ and then dried over $MgSO_4$. Flash chromatography (hexane−EtOAc 1:1) gave (+)-medicarpin (533 mg, 55% over two steps) as a white solid: mp 131.0−133.0 °C (lit. 132.0−133.5 °C,[7,27] 125.0−127.0 °C[5]); $[\alpha]_D^{23}$ +223.6 (*c* 0.15, MeOH) [lit. $[\alpha]_D^{20}$ +223.1 (*c* 0.16, acetone)];[7,27] $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 7.39 (1H, d, $J$ = 8.8 Hz), 7.13 (1H, d, $J$ = 8.8 Hz), 6.55 (1H, dd, $J$ = 8.0 Hz, 2.4 Hz), 6.46−6.44 (2H, m), 6.41 (1H, d, $J$ = 2.4 Hz), 5.50 (1H, d, $J$ = 6.8 Hz), 4.24 (1H, dd, $J$ = 11.2 Hz, 4.8 Hz), 3.76 (3H, s), 3.62 (1H, t, $J$ = 10.8 Hz), 3.55−3.50 (1H, m); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ 161.3, 160.8, 157.3, 156.8, 132.4, 125.0, 119.3, 112.8, 110.0, 106.7, 103.9, 97.1, 78.8, 66.7, 55.7, 39.7; HRESIMS $m/z$ 269.0819 $[M - H]^-$ (calcd for $C_{16}H_{19}O_4$ 269.0814).

## ■ ASSOCIATED CONTENT

### Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.jnatprod.7b00741.

Mass spectroscopy methods, $^1$H and $^{13}$C NMR spectra and MS analyses for compounds **1**, **3**−**11**, and (+)-medicarpin (PDF)

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### Notes

The authors declare no competing financial interest.

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## ■ REFERENCES


(1) VanEtten, H. D.; Matthews, P. S.; Mercer, E. H. *Phytochemistry* **1983**, *22*, 2291−2295.

(2) Ferrari, F.; Botta, B.; Alves de Lima, R. *Phytochemistry* **1983**, *22*, 1663−1164.

(3) Reyes-Chilpa, R.; Gómez-Garibay, F.; Moreno-Torres, G.; Jiménez-Estrada, M.; Quiroz-Vásquez, R. I. *Holzforschung* **1998**, *52*, 459−462.

(4) Seo, E. K.; Kim, N. C.; Mi, Q.; Chai, H.; Wall, M. E.; Wani, M. C.; Navarro, H. A.; Burgess, J. P.; Graham, J. G.; Cabieses, F.; Tan, G. T.; Farnsworth, N. R.; Pezzuto, J. M.; Kinghorn, A. D. *J. Nat. Prod.* **2001**, *64*, 1483−1485.

(5) Militão, G. D. G.; Dantas, I. N. F.; Pessoa, C.; Falcão, M. J. C.; Silveira, E. R.; Lima, M A. S.; Curi, R.; Lima, T.; Moraes, M. O.; Costa-Lotufo, L. V. *Life Sci.* **2006**, *78*, 2409−2419.

(6) Awale, S.; Li, F.; Onozuka, H.; Esumi, H.; Tezuka, Y.; Kadota, S. *Bioorg. Med. Chem.* **2008**, *16*, 181−189.

(7) Pluempanupat, S.; Kumrungsee, N.; Pluempanupat, W.; Ngamkitpinyo, K.; Chavasiri, W.; Bullangpoti, V.; Koul, O. *Ind. Crops Prod.* **2013**, *44*, 653−658.

(8) Zheng, K. Y.; Choi, R. C.; Cheung, A. W.; Guo, A. J.; Bi, C. W.; Zhu, K. Y.; Fu, Q.; Du, Y.; Zhang, W. L.; Zhan, J. Y.; Duan, R.; Lau, D. T.; Dong, T. T.; Tsim, K. W. *J. Agric. Food Chem.* **2011**, *59*, 1697−1704.

(9) Tyagi, A. M.; Gautam, A. K.; Kumar, A.; Srivastava, K.; Bhargavan, B.; Trivedi, R.; Saravanan, S.; Yadav, D. K.; Singh, N.; Pollet, C.; Brazier, M.; Mentaverri, R.; Maurya, R.; Chattopadhyay, N.; Goel, A.; Singh, D. *Mol. Cell. Endocrinol.* **2010**, *325*, 101−109.

(10) Dixit, M.; Raghuvanshi, A.; Prakash Gupta, C.; Kureel, J.; Nizam Mansoori, M.; Shukla, P.; John, A. A.; Singh, K.; Purohit, D.; Awasthi, P.; Singh, D.; Goel, A. *PLoS One* **2015**, *10*, e0144541.

(11) Bhargavan, B.; Singh, D.; Gautam, A. K.; Mishra, J. S.; Kumar, A.; Goel, A.; Dixit, M.; Pandey, R.; Manickavasagam, L.; Dwivedi, S. D.; Chakravarti, B.; Jain, G. K.; Ramachandran, R.; Maurya, R.; Trivedi, A.; Chattopadhyay, N.; Sanyal, S. *J. Nutr. Biochem.* **2012**, *23*, 27−38.

(12) Goel, A.; Kumar, A.; Hemberger, Y.; Raghuvanshi, A.; Jeet, R.; Tiwari, G.; Knauer, M.; Kureel, J.; Singh, N. K.; Gautam, A.; Trivedi, R.; Singh, E.; Bringmann, G. *Org. Biomol. Chem.* **2012**, *10*, 9583−9592.

(13) Gatouillat, G.; Magid, A. A.; Bertin, E.; El Btaouri, H.; Morjani, H.; Lavaud, C.; Madoulet, C. *Phytomedicine* **2015**, *22*, 1186−1194.

(14) Trivedi, R.; Maurya, R.; Mishra, D. P. *Cell Death Dis.* **2014**, *5*, e1465.

(15) Kureel, J.; John, A. A.; Raghuvanshi, A.; Awasthi, P.; Goel, A.; Singh, D. *Mol. Cell. Biochem.* **2016**, *418*, 71−80.

(16) Krishna Prasa, A. V.; Kapil, R. S.; Popli, S. P. *J. Chem. Soc., Perkin Trans. 1* **1986**, 1561−1563.

(17) Cocker, W.; McMurry, T. B. H.; Staniland, P. A. *J. Chem. Soc.* **1965**, 1034−1037.

(18) Engler, T. A.; Reddy, J. P.; Combrink, K. D.; Vander Velde, D. *J. Org. Chem.* **1990**, *55*, 1248−1254.

(19) Narkhede, D. D.; Iyer, P. R.; Iyer, C. S. A. *Tetrahedron* **1990**, *46*, 2031−2034.

(20) Ozaki, Y.; Mochida, K.; Kim, S. W. *J. Chem. Soc., Perkin Trans. 1* **1989**, 1219−1224.

(21) Feng, Z. G.; Bai, W. J.; Pettus, T. R. R. *Angew. Chem., Int. Ed.* **2015**, *54*, 1864−1867.

(22) Marais, J. P. J.; Ferreira, D.; Slade, D. *Phytochemistry* **2005**, *66*, 2145−2176.

(23) van Aardt, T. G.; van Heerden, P. S.; Ferreira, D. *Tetrahedron Lett.* **1998**, *39*, 3881−3884.

(24) van Aardt, T. G.; van Rensburg, H.; Ferreira, D. *Tetrahedron* **1999**, *55*, 11773−11786.

(25) van Aardt, T. G.; van Rensburg, H.; Ferreira, D. *Tetrahedron* **2001**, *57*, 7113−7126.

(26) Chang, L. C.; Gerhäuser, C.; Song, L.; Farnsworth, N. R.; Pezzuto, J. M.; Kinghorn, A. D. *J. Nat. Prod.* **1997**, *60*, 869−873.

(27) Deesamer, S.; Chavasiri, W.; Chaichit, N.; Muangsin, N.; Kokpol, U. *Acta Crystallogr., Sect. E: Struct. Rep. Online* **2009**, *65*, o2387.

(28) Lin, C. F.; Yang, J. S.; Chang, C. Y.; Kuo, S. C.; Lee, M. R.; Huang, L. J. *Bioorg. Med. Chem.* **2005**, *13*, 1537−1544.




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