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A Model Study toward the Concise Synthesis of Bromotyrosine Derived Spiroisoxazoline Natural Products and Analogous Core Structures

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Abstract

A model study of the first non-aromatic ring based approach toward α -hydroxyspiroisoxazolines resembling the bromotyrosine derived natural product and analogous spiroisoxazoline core structures were implemented. The desired molecular architecture was achieved through the multifunctionalization of a key 1,3-diketo spiroisoxazoline. Our strategy could serve as an efficient alternative of previously developed approaches that utilize an aromatic ring oxidation as the essential step to synthesize this class of natural products.

Keywords

Heterocycles; Natural Product Analogues; Spiroisoxazolines; Multifunctionalization; Intramolecular Cyclization

Introduction

Over the past 50 years, marine sponges of the order *Verongida* have been distinguished as affluent sources of α -oximinotyrosine derived marine natural products (Figure 1).^[1] Most recently, the Red Sea sponge *Suberea mollis* were also found to be a source of two new bromotyrosine derived natural products, subereamollines A and B, as secondary metabolites containing the spiro-cyclohexadienyl isoxazoline moiety.^[2] These widespread spiroisoxazoline natural products can easily be distinguished by three major structure categories, where the brominated spiroisoxazoline core contains a cyclohexadiene, bromo epoxy ketone, or a bromohydrin moiety (Figure 1). The structural diversity arising from a unique spiro linkage between a brominated cyclohexadiene, an isoxazoline along with a wide

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Supporting Information (see footnote on the first page of this article): ... Characterization data (¹H and ¹³C NMR spectra) for all new compounds, 2D NMR (NOESY and HMBC) spectra for **14** and **15** and CCDC 967652 for X-ray crystal structures of **10**.

range of amine and diamine linkages, brings about a broad spectrum of pharmacological activities including antiviral,^[3] antimicrobial,^[4] anti-HIV,^[5] antifungal,^[6] antifouling,^[7] Na⁺/K⁺ ATPase inhibition,^[8] histone deacetylase (HDAC) inhibition,^[9] histamine H₃ antagonism,^[10] mycothiol *S*-conjugate amidase inhibition,^[11] isoprenylcysteine carboxy methyltransferase (Icmt) inhibition,^[12] and antineoplastic properties.^[13]

Therefore, constructing the unique and synthetically challenging spiro-skeleton has been of great interest to many chemists.^{[14],[15]} However, continuing efforts resulted in only a few synthetic strategies for the syntheses of functionalized carbocyclic spiroisoxazolines.^[15] Although significant progress in the oxidation of an aromatic ring with thallium(III) trifluoroacetate,^[15a,b] electro-organic oxidation^[15c,d] and *N*-bromosuccinimide^[15f] followed by the intramolecular cyclization of a pendant oxime has been achieved in past decades, the oxidative methods for spiroisoxazoline synthesis remain restricted to aromatic systems where the desired products are sometimes isolated in moderate yields and often required the use of toxic oxidants.^[15a-d] However, the oxidative cyclization of oxime ester using diacetoxyiodo benzene was proven to be the most suitable reagent to date.^[15e,g-i]

In this context, our continuing efforts and success in developing a new methodology^[16] enabled us to generate a novel synthetic strategy toward the bromotyrosine class of natural products (Scheme 1). Herein, we report the multifunctionalization approach of a 1,3-cyclohexane-dione to furnish the desired spiroisoxazoline as a model study for the core structure of bromotyrosine derived spiroisoxazoline natural products and their analogues (Scheme 1). Furthermore, this methodology could potentially serve as an alternative synthetic strategy toward the targeted natural products and closely related spiroisoxazolines as novel analogues of biological interest.

Results and Discussion

Our synthesis basically relies on two approaches, a non aromatic approach towards the spiro-diketone followed by a multifunctionalization approach to furnish the desired molecular architecture resembling the bromotyrosine derived natural product core. The retrosynthetic analysis of the aforementioned natural product core model study shows that the most synthetically interesting core spiroisoxazolines **1** and **2** could be obtained from their corresponding spiro-methoxyenone derivatives **4** and **5** which in turn could be achieved from the same spiro-1,3-diketone moiety **3**. The spiro-diketone **3** would be easily prepared from a base mediated intramolecular condensation of the corresponding ketoester **6** (Scheme 2).

In our synthesis, acyclic isoxazole derivative **6**^[16b,17] was used as a key precursor for this model study. After reacting **6** with sodium hydride, the diketone spiroisoxazoline **3** was isolated in 80% yield (Scheme 3). The ¹H NMR study showed that compound **3** exists as an enol which results in the fast exchange of one of the protons between the two carbonyl groups. It is worth noting that the ready availability of cyclohexane-1,3-dione, and its diverse reactivity, often render cyclohexane-1,3-dione, and its analogues, as suitable starting materials for a number of natural product syntheses.^[18] However, for our synthetic strategy,

we need to convert the diketone into a conjugated diene system for additional functionalization.

We subjected **3** to a variety of reported reaction conditions^[19] to determine the best reagent that would afford spiroisoxazoline regioisomers **4** and **5** in the highest isolated yields (Table 1). Based upon the data shown in Table 1, entry 4, which features 5 mol% TiCl₄ in methanol^[19d] with triethyl amine, both spiroisoxazolines, **5** and **4**, were isolated in 97% yield in a 1: 2.5 ratio favoring the spiroisoxazoline **4** after 15 minutes (Table 1). Due to the fact that both regioisomers were isolated, divergent synthetic pathways were explored where spiroisoxazoline **4** was utilized as a precursor for the natural product core structure (analogous to 11-deoxyfistularin-3), while spiroisoxazoline **5** was developed into a spiroisoxazoline that is more comparable to the agelarin natural products where the carbonyl and isoxazoline moieties have a 1,4-relationship.^[21]

With the anticipation of furnishing a conjugated double bond, we treated the predominant isomer **4** with LHMDs, PhSeCl followed by H₂O₂.^[20,22] As a result, deprotonation of the allylic proton of **4** and subsequent formation and elimination of the selenoxide afforded **8** in 75% yield (Scheme 4). In order to accomplish the core structure, the spiro-diene was then exposed to a number of reaction conditions to bring about di-bromination and concomitant dehydrohalogenation. Unfortunately, after a number of attempts to introduce bromine to **8** through a variety of bromine sources (pyridinium tribromide (PTB)/K₂CO₃, Br₂/CCl₄, NBS/CCl₄), none of the desired product **1** was isolated, instead a decomposition of the starting material was realized (Scheme 4).

Isomer **5** was also treated under a comparable reaction series as shown in Scheme 4 in order to introduce the other double bond to furnish the natural product derivative as a model study for the synthesis of the spiroisoxazoline analogue (Scheme 5). While the isomer **5** was treated with LHMDs, PhSeCl followed by H₂O₂, we isolated spiro-derivative **9** in 71% yield (Scheme 5). Bromination of **9** with bromine in the absence of light afforded the vicinal dibromo derivative, **10**^[23] in 90% yield, as result of over bromination followed by dehydrohalogenation. We did not realize any dibromination of each α -carbon with respect to the ketone as an expected product **2**. The feasibility of introducing the third bromine at α -position of **10** was further experienced under PTB/K₂CO₃ conditions to afford the **11** in 82% yield (Scheme 5). However, our target was to synthesis 1,3-dibromo derivative of spiroisoxazoline **2**.

After this setback, we then decided to switch the electrophile used in the synthesis of **1** from phenylselenium chloride to bromine. We anticipated that after reacting **4** with excess (2.5 equiv.) LHMDs followed by treatment with excess bromine would provide the allylic geminal dibromo compound **12** *via* monobromination. Therefore, we followed this reaction pathway, and, without further purification, the crude allylic geminal dibromo compound **12** was reacted with DABCO *in situ* to afford **13** in 76% yield over two steps in one reaction vessel (Scheme 6). Bromination of **13** adjacent to the carbonyl was achieved in 76% with NBS in the dark. Diastereoselective reduction of **1** with Zn(BH₄)₂^[24] afforded the desired spiroisoxazoline core structures ($\pm$)-**14** and ($\pm$)-**15** in a 4:1 diastereomeric ratio (Scheme 6).

Purification of the crude mixture gave ($\pm$)-**14** and ($\pm$)-**15** in a yield of 64% and 16%, respectively. The diastereomeric ratio favoring the desired *trans*-isomer **14** over the undesired *cis*-isomer **15** was rationalized by intramolecular hydride delivery through a 5-membered zinc chelate that avoids the steric demands of the isoxazoline methylene group, resulting in the delivery of the hydride from the *alpha* face of the ketone.^[25] Though we have synthesized the racemic **14** and **15**, the rigid scaffold of the spiro-isoxazolines showed signs of distinct nOe interactions between the diagnostic protons which enabled us to establish the relative stereochemical assignment of the hydroxyl group on the basis of the NOESY spectrum (Scheme 6). The distinct nOe interactions indicated strong cross peaks between H₅ and H₇ for compound **14**, whereas, in the case of **15**, strong cross peaks were observed between H₅ and H₇, and also between H₁ and H₈, which suggested the formation of *trans*-isomer **14** and *cis*-isomer **15** as a result of diastereoselective reduction (Scheme 6).

Having succeeded in constructing the desired spiroisoxazoline **1** through our model studies, we then focused on transforming the minor isomer **5** to its corresponding dibromo derivative **2** (Scheme 7). Following a similar protocol that was used in Scheme 6, compound **5** was treated with excess LHMDS and Br₂ followed by DABCO under reflux conditions to provide the desired mono-bromo-derivative **17** in 78% yield. Further bromination of **17** using PTB/K₂CO₃ provided the desired α -dibromo derivative **2** in 80% yield (Scheme 7).

Conclusions

In conclusion, we have successfully accomplished the synthesis of the core skeleton that is very similar to naturally available spiroisoxazolines, such as 11-deoxyfistularin-3, as well as the isomeric spiroisoxazoline core where the isoxazoline moiety and carbonyl are positioned in a similar fashion as agelotin A and B. The diverse reactivity of the key 1,3-diketo-spiroisoxazoline precursor enabled us to quickly furnish the desired molecular structures in good to very good isolated yields. Due to the success of this model study, we are positioned to imminently apply the reported synthetic methodology from this model system toward the syntheses of spiroisoxazoline containing natural products as well as their synthetic analogues for biological evaluation.

Experimental Section

General Considerations

Unless otherwise stated, all solvents and reagents were commercially obtained and were used without prior purification. The progression of reaction was monitored by analytical thin-layer chromatography using 60Å silica gel medium and 250µm layer thickness, and compounds were visualized by 254 nm light, basic KMnO₄ (40g of K₂CO₃ + 6g of KMnO₄ in 600mL of water, then 5mL of 10% NaOH added) and subsequent development with either gentle or no heating. The crude products were purified by flash column chromatography over silica gel (60 Å, 0.060–0.200 mm) by using the required hexanes and ethyl acetate ratio as the eluent system. All NMR spectra were measured at 25 °C in the indicated deuterated solvents. ¹H NMR and ¹³C NMR spectra were recorded in 500 MHz and 125 MHz respectively. The NMR data are reported as follows: proton and carbon chemical shifts (δ) in ppm using tetramethylsilane as an internal standard, coupling constants (*J*) in hertz (Hz), and

resonance multiplicities (br = broad, s = singlet, d = doublet, t = triplet, and m = multiplet). The residual protic solvent of CDCl₃ (¹H, δ 7.26 ppm); (¹³C, δ 77.0 ppm central resonance of the triplet), and C₃D₆O (¹H, δ 2.05 ppm; ¹³C, δ 29.84 ppm) were used as the internal references in the ¹H and ¹³C NMR spectra. Melting points are uncorrected. Fourier transform infrared (FTIR) spectra were measured on neat NaCl. The absorptions are given in wave numbers (cm⁻¹). High-resolution mass spectrometry (HRMS) analyses were performed on the basis of positive electrospray ionization. Either protonated molecular ions [*M* + *nH*]^{*n*+} or sodium adducts [*M* + Na]⁺ were used for empirical formula confirmation.

1,3-Diketo-spiroisoxazoline (3)—To a stirred solution of the isoxazoline **6** (1.0 g, 3.45 mmol) in anhydrous toluene (80 mL) was added a 60% dispersion of sodium hydride (0.42 g, 9.94 mmol) in mineral oil. The reaction mixture was then heated to 80 °C and stirred overnight. After the disappearance of starting material, as evidenced by TLC, the reaction mixture was quenched with 1N HCl (20 mL). The reaction mixture was extracted with ethyl acetate (3 × 100 mL), and the combined organic layers were washed with brine (20 mL), dried over MgSO₄, filtered, and the solvent was evaporated *in vacuo*. The crude product was purified by flash column chromatography over silica gel using 1:1 hexanes–ethyl acetate as an eluant to provide dione **3** as a pale yellow solid (0.672 g, 80% yield). *R*_f = 0.33 (1:1 hexanes/EtOAc). mp 163–165 °C. ¹H NMR (500 MHz, C₃D₆O): δ = 7.73–7.72 (m, 2H), 7.45–7.44 (m, 3H), 5.45 (s, 1H), 3.97 (d, *J* = 16.8 Hz, 1H), 3.20 (d, *J* = 16.8 Hz, 1H), 2.82–2.76 (m, 1H), 2.48–2.44 (m, 1H), 2.4–2.35 (m, 1H), 2.24–2.18 (m, 1H). ¹³C NMR (125 MHz, C₃D₆O): δ = 206.3, 156.8, 130.8, 130.7, 129.5, 127.4, 104.0, 86.4, 41.3, 32.7, 27.8. IR (KBr): ν_{max} = 3200, 3072, 3023, 2945, 2913, 1728, 1645, 1609, 1357, 1322, 1247, 1203, 918, 764, 692 cm⁻¹. HRMS (EI): *m/z* calcd for C₁₄H₁₃NO₃ (*M*+Na⁺): 266.0787; found 266.0787.

Spiro-methoxyenones (4 and 5)^{16b}—To a well stirred solution of the cyclic 1,3-diketone **3** (1.15 g, 4.75 mmol) in MeOH (5 mL) was added TiCl₄ (0.23 mL, 1M in CH₂Cl₂) in one portion with a syringe, at room temperature. The reaction mixture was then stirred for an additional 30 min before the addition of Et₃N (0.8 mL, 0.57 mmol). The stirring was continued for additional 45 min before addition of H₂O (3 mL). The reaction mixture was extracted with ethyl acetate (3 × 50 mL) and the combined organic layers were successively washed with H₂O (3 × 100 mL), brine, dried over MgSO₄ filtered and evaporated under reduced pressure. The crude mixture was purified by flash column over silica gel using 3:1 hexanes–ethyl acetate as an eluent to afford **5** and **4** as a separable mixture (1:2.5) of two diastereomers in 97% isolated yield.

Spiro–diene (8)—To a solution of methoxyenone **4** (0.285 g, 1.1 mmol) in anhydrous THF (35 mL) at –78 °C was added slowly a solution of LHMDs (1.65 mL, 1M in THF). The resulting solution was allowed to warm to –10 °C over a period of 1 h. After re-cooling to –78 °C, a solution of PhSeCl (0.316 g, 1.65 mmol) in anhydrous THF was added slowly. The solution was allowed to warm to 0 °C over a period of 30 min. At this stage TLC observation showed the formation of a less polar intermediate selenide. Once this conversion was complete, saturated aq. NH₄Cl (1.0 mL) and 31% aq. H₂O₂ (2.8 mL) were added, and the mixture was stirred for 1 h at 45 °C. The mixture was diluted with EtOAc (50 mL), and

the layers were separated. The organic phase was washed with H₂O (50 mL) and brine (50 mL) and then dried (MgSO₄). The solvent was removed *in vacuo*, and the crude product was purified by flash column chromatography over silica gel using 4:1 hexanes–ethyl acetate as an eluent to obtain diene **8** as yellow oil (0.212 g, 75% yield). R_f = 0.56 (1:1 hexanes/EtOAc). ¹H NMR (500 MHz, CDCl₃): δ = 7.67–7.65 (m, 2H), 7.41–7.40 (m, 3H), 6.50 (d, J = 10.0 Hz, 1H), 6.18 (dd, J = 10.0, 2.3 Hz, 1H), 5.50 (d, J = 2.3 Hz, 1H), 3.83 (s, 3H), 3.73 (d, J = 16.5 Hz, 1H), 3.34 (d, J = 16.5 Hz, 1H). ¹³C NMR (125 MHz, CDCl₃): δ = 196.2, 170.6, 155.1, 139.5, 130.4, 128.8, 128.7, 127.0, 123.7, 98.4, 82.9, 56.3, 46.0. IR (thin film): $\nu_{\max}$ = 3015, 2949, 2923, 2851, 1733, 1576, 1457, 1356, 1249, 1214, 898, 756 cm⁻¹. HRMS (EI): m/z calcd for C₁₅H₁₃NO₃ (M+Na⁺): 278.0787; found 278.0787.

Spiro-derivative (9)—To a solution of methoxyenone **5** (0.100 g, 0.388 mmol) in anhydrous THF (15 mL) at –78 °C was added slowly a solution of LHMDs (0.6 mL, 1M in THF). The resulting solution was allowed to warm to –10 °C over a period of 1h. After re-cooling to –78 °C, a solution of PhSeCl (0.110 g, 0.574 mmol) in anhydrous THF was added slowly. The solution was allowed to warm to 0 °C over a period of 30 min. At this stage TLC observation showed the formation of a less polar intermediate selenide. Once this conversion was complete, saturated aq. NH₄Cl (1.0 mL) and 31% aq. H₂O₂ (1.0 mL) were added and the mixture was stirred for 1 h at 45 °C. The mixture was diluted with EtOAc (50 mL) and the layers were separated. The organic phase was washed with H₂O (50 mL) and brine (50 mL) and then dried (MgSO₄). The solvent was removed *in vacuo* and the crude product was purified by flash column chromatography over silica gel using 3:1 hexanes–ethyl acetate as an eluent to obtain diene **9** as yellow oil (0.070 g, 71% yield). R_f = 0.50 (1:1 hexanes/EtOAc). ¹H NMR (500 MHz, CDCl₃): δ = 7.68–7.66 (m, 2H), 7.46–7.45 (m, 3H), 6.71 (d, J = 9.8 Hz, 1H), 6.21 (d, J = 9.8 Hz, 1H), 5.61 (s, 1H), 3.75 (s, 3H), 3.75 (d, J = 16.8 Hz, 1H), 3.45 (d, J = 16.8 Hz, 1H). ¹³C NMR (125 MHz, CDCl₃): δ = 186.7, 171.2, 155.8, 141.8, 130.6, 128.9, 128.5, 128.0, 126.9, 102.3, 80.9, 56.2, 44.5. IR (thin film): $\nu_{\max}$ = 3015, 2949, 2921, 2855, 1728, 1572, 1458, 1254, 1217, 898, 747 cm⁻¹. HRMS (EI): m/z calcd for C₁₅H₁₃NO₃ (M+Na⁺): 278.0787; found 278.0787.

1,2-Dibromo spiro-derivative (10)—To a solution of **9** (0.024 g, 0.098 mmol) in CCl₄ was added Br₂ (0.012 mL, 2.5 equiv.). The resulting mixture was stirred in the dark for overnight at rt. Then the solvent and gaseous product was reduced under pressure. The yellow residue was dissolved in Et₂O and washed with Al₂O₃ and evaporated. The crude mixture was purified by silica gel column chromatography using 12% ethyl acetate in hexanes as an eluent to obtain the pure compound **10** as pale yellow solid (0.036 g, 90% yield). R_f = 0.67 (3:1 hexanes/EtOAc). mp 123–125 °C. ¹H NMR (500 MHz, CDCl₃): δ = 7.69 (dd, J = 7.7, 1.7 Hz, 2H), 7.47–7.45 (m, 3H), 5.75 (s, 1H), 3.82 (br s, 5H). ¹³C NMR (125 MHz, CDCl₃): δ = 176.6, 171.6, 155.5, 142.4, 130.8, 130.4, 128.9, 128.1, 127.0, 100.3, 85.5, 57.1, 47.2. IR (KBr): $\nu_{\max}$ = 3068, 3020, 2949, 2925, 2853, 1723, 1661, 1637, 1582, 1447, 1358, 1216, 756 cm⁻¹. HRMS (EI): m/z calcd for C₁₅H₁₁Br₂NO₃ (M+Na⁺): 433.8997; found 433.8996.

Tribromo spiro-derivative (11)—A solution of **10** (0.032 g, 0.080 mmol) and K₂CO₃ (0.032 g, 0.238 mmol) in 2 mL of dry dichloromethane was stirred in an ice-water bath.

Pyridinium tribromide (0.74 g, 0.232 mmol) was added slowly to the cold mixture. The reaction mixture was stirred for 2 h at 0 °C, and was allowed to slowly warm to rt overnight. The reaction mixture was stirred at rt until the disappearance of the starting materials, as evidenced by TLC. The reaction mixture was poured into a cold, saturated NH₄Cl solution (5 mL), extracted with ethyl acetate (3 × 10 mL), and washed with a saturated CuSO₄ solution (3 × 10 mL). The organic layer was dried with MgSO₄, filtered, and concentrated under reduced pressure. The crude products were purified by flash column chromatography over silica gel using the 6% ethyl acetate in hexanes as an eluent to obtain the pure compound **11** as pale yellow solid (0.028 g, 82% yield). R_f = 0.67 (3:1 hexanes/EtOAc). ¹H NMR (500 MHz, CDCl₃): δ = 7.69 (dd, J = 8.0, 1.6 Hz, 2H), 7.50–7.45 (m, 3H), 4.16 (s, 3H), 3.91 (d, J = 17.1 Hz, 1H), 3.75 (d, J = 17.1 Hz, 1H). ¹³C NMR (125 MHz, CDCl₃): δ = 172.1, 168.7, 156.2, 143.8, 131.0, 129.0, 128.1, 127.9, 127.1, 108.5, 85.3, 63.2, 46.8. IR (KBr): $\nu_{\max}$ = 2949, 2919, 2849, 1733, 1664, 1616, 1578, 1354, 1218, 1051, 946, 889, 763, 692 cm⁻¹. HRMS (EI): m/z calcd for C₁₅H₁₀Br₃NO₃ (M+Na⁺): 511.8103; found 511.8104.

Monobromo spiroisoxazoline (13)—To a previously cooled (–78 °C) solution of **4** (0.698 g, 2.73 mmol) in anhydrous THF (20 mL) was added slowly a solution of LHMDs (8.0 mL, 1M in THF). The resulting solution was allowed to warm to –10 °C over a period of 1h. After re-cooling to –78 °C, a solution of bromine (0.42 mL, 8.2 mmol) in CH₂Cl₂ (2 mL) was added slowly to the reaction mixture. The solution was allowed to warm to 0 °C over a period of 30 min. TLC observation showed the formation of a less polar bromo intermediate. Once this conversion was complete, DABCO (0.612 g, 5.46 mmol) was added to the reaction mixture, and the resulting mixture was refluxed for 3h. After the disappearance of the less polar material, as evidenced by TLC, the reaction mixture was diluted with EtOAc (50 mL) and washed with H₂O (10 mL), brine, dried over MgSO₄ and concentrated *in vacuo*. The crude mixture was purified by flash column over silica gel using 4:1 hexanes-ethyl acetate as an eluent to afford **13** (0.693 g, 76% yield) as a yellow solid. R_f = 0.65 (3:2 hexanes/EtOAc). mp 154–156 °C. ¹H NMR (500 MHz, CDCl₃): δ = 7.66–7.64 (m, 2H), 7.44–7.39 (m, 3H), 6.88 (s, 1H), 5.55 (s, 1H), 3.90 (s, 3H), 3.75 (d, J = 16.6 Hz, 1H), 3.39 (d, J = 16.6 Hz, 1H). ¹³C NMR (125 MHz, CDCl₃): δ = 194.7, 165.4, 155.2, 139.8, 130.6, 128.8, 128.5, 127.0, 117.8, 99.2, 84.5, 57.3, 45.7. IR (KBr): $\nu_{\max}$ = 3076, 3023, 2978, 2945, 1652, 1575, 1368, 1254, 1209, 1001, 759, 689 cm⁻¹. HRMS (EI): m/z calcd for C₁₅H₁₂BrNO₃ (M+Na⁺): 355.9892; found 355.9894.

Spiroisoxazoline (1)—To a solution of **13** (0.327 g, 0.976 mmol) in CH₂Cl₂ (10 mL) was added NBS (0.230 g, 1.95 mmol) and the resulting mixture was stirred for 24 h at rt under dark reaction conditions. Completion of the reaction was monitored by TLC analysis. The reaction mixture was then diluted with CH₂Cl₂ (10 mL) and washed with H₂O (50 mL), saturated Na₂S₂O₃ (2 × 50 mL), brine, dried over MgSO₄ and concentrated under reduced pressure. The crude product was purified by flash column over silica gel using 6% ethyl acetate in hexanes as an eluent to afford **1** (0.306 g, 76% yield) as a yellow solid. R_f = 0.73 (3:2 hexanes/EtOAc). mp 140–142 °C. ¹H NMR (500 MHz, CDCl₃): δ = 7.64–7.63 (m, 2H), 7.44–7.39 (m, 3H), 6.86 (s, 1H), 4.16 (s, 3H), 3.77 (d, J = 16.7 Hz, 1H), 3.44 (d, J = 16.7 Hz, 1H). ¹³C NMR (125 MHz, CDCl₃): δ = 189.9, 163.2, 155.4, 137.7, 130.8, 128.7, 128.2, 126.9, 119.5, 107.4, 85.2, 61.9, 45.0. IR (KBr): $\nu_{\max}$ = 3068, 2949, 2937, 2851, 1683, 1541,

1322, 894, 761, 692, 538 cm^{-1} . HRMS (EI): m/z calcd for $\text{C}_{15}\text{H}_{11}\text{Br}_2\text{NO}_3$ ($\text{M}+\text{Na}^+$): 433.8997; found 433.9001.

Spiroisoxazoline Core (14 and 15)—To a stirred solution of **1** (0.275 g, 0.67 mmol) in CH_2Cl_2 (10 mL) at room temperature was added a freshly prepared solution of $\text{Zn}(\text{BH}_4)_2$ (1.0 mL, 0.25 M in Et_2O).²³ After 10 min, H_2O (0.5 mL) was added, and the reaction mixture was stirred for a further 30 min. Following the addition of anhydrous MgSO_4 , the mixture was filtered and concentrated *in vacuo*. NMR of the crude product shows the formation of **14** and **15** in a 4:1 diastereomeric mixture. The crude residue was purified carefully by flash column chromatography over silica gel using 10% ethyl acetate in hexanes to obtain ($\pm$)**14** (0.178 g, 64% yield) and ($\pm$)**15** (0.045 g, 16% yield) as a colorless liquid.

Mixture of 14 and 15— ^1H NMR (500 MHz, CDCl_3): δ = 7.66–7.62 (m, 2.5H), 7.44–7.38 (m, 4.75H), 6.44 (s, 0.25H), 6.40 (s, 1H), 4.50 (s, 1H), 4.41 (s, 0.25H), 4.09 (d, J = 17.2 Hz, 1.25H), 3.78 (s, 0.75H), 3.77 (s, 3H), 3.11 (d, J = 17.2 Hz, 1.25H). ^{13}C NMR (125 MHz, CDCl_3): δ = 157.2, 157.1, 148.4, 148.1, 132.2, 131.6, 130.7, 130.5, 128.9, 128.8, 128.7 (2C), 126.8, 126.7, 120.8, 120.4, 112.93, 112.5, 89.4, 87.7, 75.2, 74.1, 60.13, 60.1, 44.1, 40.1.

Spiroisoxazoline Core (14)— R_f = 0.70 (1:1 hexanes/EtOAc). ^1H NMR (500 MHz, CDCl_3): δ = 7.67–7.65 (m, 2H), 7.43–7.39 (m, 3H), 6.41 (s, 1H), 4.51 (s, 1H), 4.09 (d, J = 17.2 Hz, 1H), 3.78 (s, 3H), 3.12 (d, J = 17.2 Hz, 1H). ^{13}C NMR (125 MHz, CDCl_3): δ = 157.1, 148.2, 132.2, 130.5, 128.9, 128.8, 126.8, 120.4, 112.4, 89.2, 74.2, 60.1, 40.1. IR (thin film): ν_{max} = 3384, 3056, 3007, 2934, 2839, 1577, 1447, 1359, 1309, 1217, 984, 905, 760, 691 cm^{-1} . HRMS (EI): m/z calcd for $\text{C}_{15}\text{H}_{13}\text{Br}_2\text{NO}_3$ ($\text{M}+\text{Na}^+$): 435.9154; found 435.9159.

Spiroisoxazoline Core (15)— R_f = 0.63 (1:1 hexanes/EtOAc). ^1H NMR (500 MHz, CDCl_3): δ = 7.65–7.63 (m, 2H), 7.45–7.40 (m, 3H), 6.45 (s, 1H), 4.42 (d, J = 8.0 Hz, 1H), 3.79 (s, 3H), 3.62 (d, J = 17.0 Hz, 1H), 3.40 (d, J = 17.0 Hz, 1H), 2.64 (d, J = 8.0 Hz, 1H–OH). ^{13}C NMR (125 MHz, CDCl_3): δ = 157.1, 148.4, 131.5, 130.7, 128.9, 128.5, 126.8, 120.9, 112.4, 87.7, 75.2, 60.1, 44.2. IR (thin film): ν_{max} = 3374, 3060, 3011, 2933, 2835, 1576, 1442, 1356, 1307, 1213, 980, 902, 759, 690 cm^{-1} . HRMS (EI): m/z calcd for $\text{C}_{15}\text{H}_{13}\text{Br}_2\text{NO}_3$ ($\text{M}+\text{Na}^+$): 435.9154; found 435.9157.

Mono bromo spiro-derivative (17)—To a previously cooled (-78°C) solution of **5** (0.660 g, 2.56 mmol) in anhydrous THF (15 mL) was added slowly a solution of LHMDs (6.5 mL, 1M in THF). The resulting solution was allowed to warm to -10°C over a period of 1h. After re-cooling to -78°C , a solution of bromine (0.33 mL, 6.4 mmol) in CH_2Cl_2 (2 mL) was added slowly to the reaction mixture. The solution was allowed to warm to 0°C over a period of 30 min. TLC observation showed the formation of a less polar bromo intermediate. Once this conversion was complete, DABCO (0.574 g, 5.08 mmol) was added to the reaction mixture, and the resulting mixture was refluxed for 3h. After the disappearance of the less polar material, as evidenced by TLC, the reaction mixture was diluted with EtOAc (50 mL) and washed with H_2O (10 mL), brine, dried over MgSO_4 and concentrated *in vacuo*. The crude mixture was purified by flash column over silica gel using 4:1 hexanes-ethyl acetate as an eluent to afford **17** (0.667 g, 78% yield) as a yellow solid. R_f

= 0.62 (1:1 hexanes/EtOAc). ^1H NMR (500 MHz, CDCl_3): δ = 7.65 (d, J = 6.35 Hz, 2H), 7.46–7.40 (m, 3H), 7.14 (s, 1H), 5.71 (s, 1H), 3.80 (s, 3H), 3.76 (d, J = 16.9 Hz, 1H), 3.51 (d, J = 16.9 Hz, 1H). ^{13}C NMR (125 MHz, CDCl_3): δ = 178.8, 171.1, 155.8, 141.5, 130.7, 128.9, 128.2, 126.9, 125.1, 101.2, 82.8, 56.6, 44.7. IR (KBr): ν_{max} = 3060, 2922, 2851, 1733, 1645, 1598, 1445, 1357, 1209, 901, 757, 689 cm^{-1} . HRMS (EI): m/z calcd for $\text{C}_{15}\text{H}_{12}\text{BrNO}_3$ ($\text{M}+\text{Na}^+$): 355.9892; found 355.9895.

Dibromo spiro-derivative (2)—A solution of **17** (0.300 g, 0.89 mmol) and K_2CO_3 (0.307 g, 2.22 mmol) in 10 mL of dry dichloromethane was stirred in an ice-water bath. Pyridinium tribromide (0.574 g, 1.79 mmol) was added slowly to the cold mixture. The reaction mixture was stirred for 2 h at 0 °C, and was allowed to slowly warm to rt overnight. The reaction mixture was stirred at rt until the disappearance of the starting materials, as evidenced by TLC. The reaction mixture was poured into a cold, saturated NH_4Cl solution (50 mL), extracted with ethyl acetate (3×100 mL), and washed with saturated CuSO_4 solution (3×50 mL). The organic layer was dried with MgSO_4 , filtered, and concentrated under reduced pressure. The crude products were purified by flash column chromatography over silica gel using 5% ethyl acetate in hexanes as an eluent to obtain the pure compound **2** as a white solid (0.294 g, 80% yield). R_f = 0.80 (3:2 hexanes/EtOAc). ^1H NMR (500 MHz, CDCl_3): δ = 7.67–7.65 (m, 2H), 7.48–7.57 (m, 3H), 7.21 (s, 1H), 4.16 (s, 3H), 3.85 (dd, J = 17.0 Hz, 1H), 3.49 (d, J = 17.0 Hz, 1H). ^{13}C NMR (125 MHz, CDCl_3): δ = 173.8, 167.8, 156.5, 142.4, 131.0, 129.0, 128.1, 126.9, 122.8, 108.8, 85.6, 62.5, 43.8. IR (KBr): ν_{max} = 3060, 2921, 2847, 1683, 1540, 1319, 1278, 1229, 951, 890, 763, 690 cm^{-1} . HRMS (EI): m/z calcd for $\text{C}_{15}\text{H}_{11}\text{Br}_2\text{NO}_3$ ($\text{M}+\text{Na}^+$): 433.8997; found 433.9001.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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Figure 1.
Spiroisoxazoline natural products.

Scheme 1.
Our synthetic strategy.

Scheme 2.
Retrosynthetic analysis.

Scheme 3.
Synthesis of the spiro-vinylogous acid of **3**.

Scheme 4.
1st Generation synthesis of spirodiene.

Scheme 5.1st Generation synthesis of bromo-spiroisoxazoline.

Scheme 6.
2nd Generation synthesis of bromo-spiroisoxazoline core.

Scheme 7.

2nd Generation synthesis of quinone-spiroisoxazoline core.

Table 1

Synthesis of methoxyenone.

Entry	Procedure	Ratio (5:4)	Yield (%)
1	10% HCl/MeOH	1:2	50
2	Conc. H ₂ SO ₄ /MeOH, reflux 12h	1:2	70
3	2.5 mol% <i>p</i> TSA/MeOH, reflux 6h	1:1.5	60
4	5 mol% TiCl ₄ /MeOH/Et ₃ N reflux 6h	1:2.5	97
5	CeCl ₃ ·7H ₂ O/MeOH, 6 min	1:2	40
6	NaH/THF, reflux 2h, then Me ₂ SO ₄	1:2	82
7	^t BuOK/THF, 10 min, then Me ₂ SO ₄	1:2.5	80