Total Synthesis of Robustaflavone, a Potential Anti-Hepatitis B Agent

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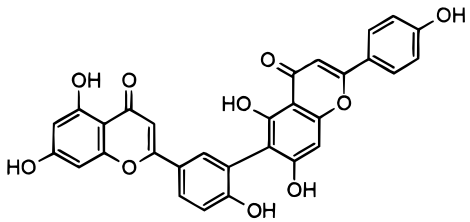
Robustaflavone, a naturally occurring compound, is an inhibitor of hepatitis B virus replication in vitro. Robustaflavone is a biflavanoid composed of two units of apigenin (5,7,4′-trihydroxyflavone) joined via a biaryl linkage between the 6-position of one unit and the 3′-position of the other (I6,II3′-biapigenin). The natural material was isolated from the seed-kernels of *Rhus succedanea*. To provide ready access to sufficient quantities of material for continued biological studies, as well as to provide a general route for the preparation of structural analogues, a total synthesis of robustaflavone was pursued. The total synthesis was approached by constructing apigenin ethers containing functionalities at the 6- and 3′-positions which could be cross-coupled using transition metal catalysis. Key steps of the synthesis included development of a regioselective iodination of an apigenin derivative at the 6-position. Also key was the formation of an apigenin 3′-boronate using a palladium-catalyzed exchange of the corresponding 3′-iodide with a diboron reagent. Finally, identification of appropriate reaction conditions for Suzuki coupling to form the sterically congested 6−3‴ biaryl bond of robustaflavone provided access to the desired biflavanoid system. This work represents the first total synthesis of robustaflavone.

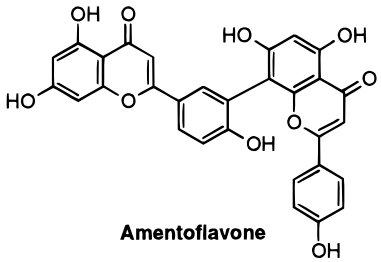
Introduction

We recently reported that robustaflavone, a naturally occurring biflavanoid, is a potent nonnucleoside inhibitor of hepatitis B virus (HBV) replication.[1] HBV is one of the most serious health problems in the world today and is listed as the ninth leading cause of death by the World Health Organization.[2] Approximately 300 million persons are chronically infected with HBV worldwide, with over one million of those in the United States. The Centers for Disease Control estimates that over 300000 new cases of acute HBV infection occurs in the United States each year, resulting in 4000 deaths due to cirrhosis and 1000 due to hepatocellular carcinoma.[3] The highest incidence of HBV infection occurs in the Far East and sub-Saharan Africa, where approximately 20% of the population are chronically infected.[4] Infection can be prevented through the use of several extremely effective recombinant vaccines.[5] Despite the availability of these vaccines, HBV infection remains the most significant viral pathogen infecting man, particularly in underdeveloped countries.

Robustaflavone was first[6] isolated in 1973, as its hexa-*O*-methyl ether, from leaf extracts of *Agathis robusta*, and later[7] in larger quantities from the seed-kernels of *Rhus succedanea*. Robustaflavone is a biflavanoid composed of two apigenin (5,7,4′-trihydroxyflavone) units connected via a biaryl linkage between their 6- and 3′-positions. No synthesis of robustaflavone has been reported. A recent synthesis of a related biflavanoid, amentoflavone (I8,II3′-biapigenin), which consists of two apigenin units connected via a biaryl linkage between respective 8- and 3′-positions, was achieved using Suzuki coupling[8] of an apigenin 8-boronic acid derivative with an appropriate 3′-iodoapigenin analogue.[9] We adopted this general methodology in our approach to the total synthesis of robustaflavone.



Robustaflavone



Amentoflavone

Chemistry

The total synthesis of robustaflavone was approached via construction of two apigenin derivatives, one substi-

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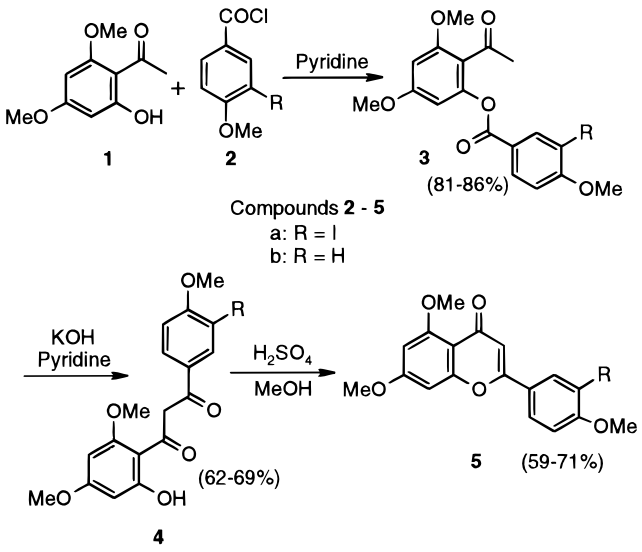
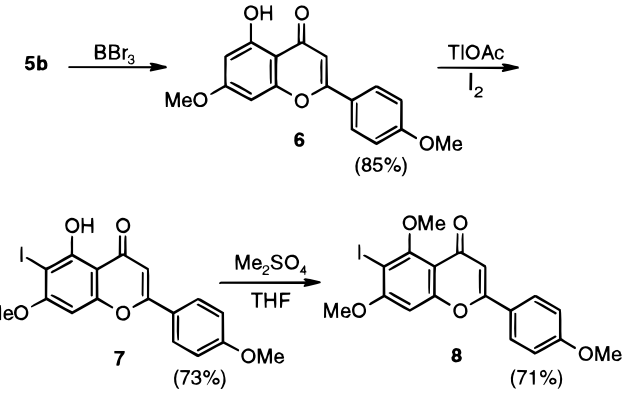
Scheme 1



Scheme 2



tuted in the 6-position and one substituted in the 3′-position, with groups that could be coupled using transition metal-catalyzed cross-coupling methodology. The synthesis of apigenin derivatives substituted in the 3′-position is straightforward (Scheme 1), involving esterification of phloroacetophenone dimethyl ether (**1**) with 3-substituted *p*-anisoyl chlorides, such as 3-iodo-*p*-anisoyl chloride (**2a**). Baker[10]−Venkataraman[11] rearrangement of the resulting ester (**3a**) to the *β*-diketone **4a** was achieved by heating in pyridine at 100 °C in the presence of powdered KOH. Cyclization of the diketone **4a** under acidic conditions provided 3′-iodoapigenin trimethyl ether (**5a**).[12] This route was also utilized to prepare apigenin trimethyl ether (**5b**), starting with **1** and *p*-anisoyl chloride.

The preparation of apigenin derivatives substituted in the 6-position presented a more difficult challenge, as direct electrophilic substitution of apigenin ethers occurs preferentially in the 8-position. An extensive search of the chemical literature yielded only two examples of 6-halogenated apigenin derivatives; the first, described in 1939, reported 6-bromoapigenin trimethyl ether as an intermediate in a total synthesis of apigenin.[13] However, it was later determined that the position of the ring bromination had been incorrectly assigned in the original report, and that the actual intermediate was 8-bromoapigenin trimethyl ether.[14] A second route described the iodination of apigenin 7,4′-dimethyl ether with iodine in a solution of iodic acid and ethanol, which provided a mixture of the 6-iodo and 8-iodo derivatives, in a 1:4 ratio.[15] The desired 6-iodinated derivative was reportedly purified by fractional recrystallization, but in very poor yield.

The desired 6-iodinated species was prepared in excellent yield with almost exclusive regioselectivity by exploiting the *ortho*-directing capabilities of thallium(I) salts in the iodination of phenols[16] (Scheme 2). Selective demethylation of apigenin trimethyl ether (**5b**) in the 5-position was accomplished with 1.1 equiv of boron tribromide, to afford apigenin 7,4′-dimethyl ether (**6**). Iodination of **6** with 1.0 equiv of I_2 in the presence of 1.2 equiv of thallium(I) acetate in CH_2Cl_2 provided 6-iodoapigenin 7,4′-dimethyl ether (**7**), in excellent yield, containing only trace amounts (less than 1% determined by ^1H NMR) of the 8-iodinated species. Methylation of **7** with dimethyl sulfate afforded the desired 6-iodoapigenin trimethyl ether (**8**).

Formation of biaryl systems is efficiently achieved via the palladium-catalyzed cross-coupling of aryl halides and aryl boronic acids (Suzuki coupling)[8] or aryl halides and aryl stannanes (Stille coupling),[17] either of which could be applied to the synthesis of robustaflavone from the iodinated species **5a** or **8**. Conceivably, the iodide of either of these derivatives could be converted to a boronic acid or a stannane and then cross-coupled with the other iodide to afford robustaflavone hexamethyl ether.

In a related example, derivatives of the biflavanoid amentoflavone were synthesized using the palladium-catalyzed cross-coupling of apigenin ethers bearing a boronic acid in the 8-position with apigenin ethers having an iodide in the 3′-position, in very good yields.[9] The apigenin-8-boronic acids were synthesized from the 8-iodinated derivatives via halogen−lithium exchange, followed by quenching with trimethyl borate and aqueous workup. Attempted conversion of the 3′-iodide **5a** via halogen−lithium exchange followed by trimethyl borate quench were unsuccessful in our hands, as for others.[9] Additionally, our attempt to prepare the corresponding apigenin 6-boronic acid derivative from **8** using this technique were also unsuccessful, in contrast to the reported[9] simple conversion of the 8-iodinated isomer to its corresponding boronic acid.

Milder general methods for the preparation of both stannanes[18] and boronic acids[19] have been described, using the palladium-catalyzed exchange of aryl halides with nucleophilic distannane and diboron reagents, respectively. These methods were applied to compound **5a** to afford both the corresponding stannane derivative **9a** and boronate **9b**, illustrated in Scheme 3. Treatment

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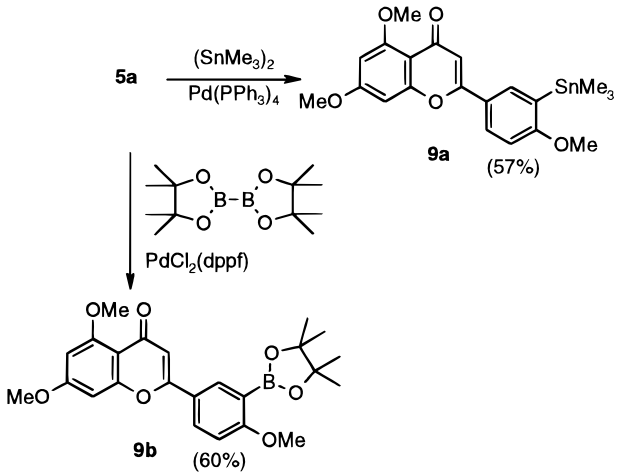
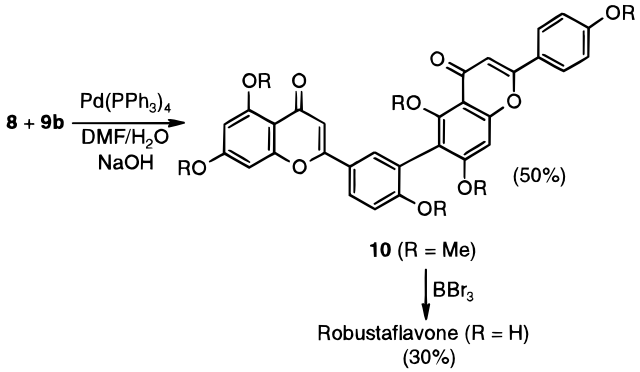
Scheme 3



Scheme 4



of **5a** with commercially available hexamethylditin in the presence of catalytic $(Ph_3P)_4Pd$ in refluxing toluene afforded the trimethylstannane **9a**. Similarly, treatment of **5a** with bis(pinacolato)diboron in the presence of catalytic $PdCl_2$(dppf) and K_2CO_3 in DMF at 85 °C provided boronate ester **9b** in 64% yield. Bis(pinacolato)-diboron was prepared via treatment of tetrakis(trimethyl-amino)diboron[20] with pinacol as previously described[21] and recently became commercially available.

Attempted Stille coupling of **9a** with iodide **8** in a variety of solvents (DMF, toluene, dioxane, THF) using several palladium catalysts [$(Ph_3P)_4Pd$, $Pd(OAc)_2$, $PdCl_2$, $(Ph_3P)_2PdCl_2$] failed to provide any significant formation of robustaflavone hexamethyl ether (**10**, data not shown). When using toluene as solvent, a very small amount of **10** could be detected by TLC, by comparison with an authentic standard prepared by methylation of natural robustaflavone with dimethyl sulfate,[7] but the major product formed in the reaction was apigenin trimethyl ether (**5b**). It is commonly accepted that transmetalation from tin to palladium represents the rate-limiting step in Stille couplings.[17] Thus, because iodide **8** is particularly reactive toward oxidative addition, accelerated by the presence of the two electron-donating *o*-methoxyl groups, reduction of the aryl iodide apparently occurred much faster than transmetalation. However, it is unclear whether **5b** was formed exclusively via reduction of the iodide **8**, or also by proteodesilylation of **9a**. Following the reaction progress by TLC appeared to indicate that the concentration of **8** decreased over time, while that of **9a** was unchanged, suggesting that reduction of **8** was the likely source of **5b**.

In contrast to Stille coupling, transmetalation from boron to palladium in Suzuki couplings is rapid, and oxidative addition is generally the rate-limiting step.[8] We thus turned to coupling of boronate **9b** with aryl iodide **8**. Using standard conditions described[19] for coupling of pinacol boronate esters with aryl halides ($PdCl_2$(dppf), DMF, KOAc, 80 °C), we observed that, again, the major product formed was apigenin trimethyl ether (**5b**). Though we anticipated that coupling between **9b** and **8** may be sluggish, due to the steric congestion of the biaryl bond being formed, we were comforted to learn that others[22] had successfully prepared biaryl systems containing similar degrees of steric crowding utilizing Suzuki methodology.

Evaluation of a variety of reaction conditions, conducted by changing the solvent, palladium catalyst, and base, identified conditions that afforded the desired robustaflavone hexamethyl ether (**10**). Conducting the reaction in DMF containing 10% H_2O, 4.0 equiv of NaOH, and 10 mol % $Pd(PPh_3)_4$ as catalyst afforded **10** in 30% yield, with only small amounts of **5b** formed as a byproduct, and trace amounts of unreacted iodide **8** (Scheme 4). Utilization of NaOH as base resulted in greatly accelerated reaction rates relative to those observed when using K_2CO_3, and the Suzuki coupling was generally complete within 1 h. Increasing the equivalence of **9b** to 1.2 increased the yield of **10** to 35%, and further increasing the stoichiometry of **9b** to 2.0 equiv increased the yield of **10** to 50%. The desired material, robustaflavone hexamethyl ether (**10**), was readily purified using silica gel column chromatography.

Deprotection of **10** was initially attempted using standard mineral acid conditions, such as HBr and HI. In all cases, the use of mineral acids resulted in Wessely–Moser rearrangement,[23] and amentoflavone was the major product isolated. Similar results were obtained using aqueous (HBr and HI) or anhydrous (HBr in HOAc) conditions.

Complete demethylation of **10** was achieved by treatment with 12 equiv of BBr_3 in refluxing $CHCl_3$. Attempts to deprotect using lesser amounts of BBr_3, or at lower temperatures, did not achieve complete demethylation, and the resulting products contained significant quantities of partially demethylated materials. Following demethylation, crude robustaflavone was obtained in 88.9% yield. Column chromatography through silica gel (toluene/pyridine/formic acid, 20:10:1) afforded robustaflavone in 30% yield.

Summary

Robustaflavone represents an important lead compound in the search for potential anti-hepatitis B agents. Our preliminary in vitro evaluations determined that robustaflavone possesses anti-hepatitis B activity com-

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parable to several nucleoside analogues currently in clinical trials[1] as well as acts synergistically with these agents.[24]

We have described the first total synthesis of robustaflavone. The key steps included a thallium-assisted regioselective iodination of apigenin 7,4′-dimethyl ether in the 6-position, allowing efficient access to 6-iodoapigenin trimethyl ether (**8**). This step was critical, because an efficient route to 6-halogenated flavones has not been described previously. Another key step was conversion of 3′-iodoapigenin trimethyl ether (**5b**) to its corresponding 3′-pinacol boronate derivative **9b**, via the palladium-catalyzed cross-coupling of **5b** with bis(pinacolato)-diboron. The corresponding 3′-stannane **9a** failed to couple with iodide **8** under Stille conditions, and attempts to convert either of iodides **5b** or **8** to their corresponding boronic acids using standard methods (halogen−lithium exchange/trialkylborate quench) failed. Identification of reaction conditions which allowed Suzuki coupling between boronate **9b** with iodide **8** furnished the critical 6−3‴ biaryl linkage, to afford the desired robustaflavone skeleton in the form of its hexamethyl ether (**10**). Finally, demethylation under nonaqueous conditions using BBr_3 provided access to synthetic robustaflavone, which was identical in all respects to the natural product. This total synthesis will allow access to needed quantities of robustaflavone for further biological studies as well as provide an efficient method for the synthesis of structural analogues.

Experimental Section

General Experimental. Column chromatography was conducted with EM Science silica gel 60 (70−230 mesh) with indicated eluents. Analytical thin-layer chromatography was performed with silica gel 60 F_{254} precoated glass-backed plates (250 μm) with indicated eluents. Bis(pinacolato)diboron was prepared from tetrakis(dimethylamino)diboron[20] as previously described.[21] All other reagents and solvents were purchased from commercial sources and used without further purification.

2,4-Dimethoxy-6-hydroxyacetophenone (1). To a solution of phloroacetophenone hydrate (20.5 g, 110 mmol) and K_2CO_3 (22.1 g, 160 mmol) in 150 mL of acetone was added dimethyl sulfate (27.7 g, 220 mmol) slowly over 30 min with mechanical stirring. The solution was heated at reflux overnight and then poured into 500 mL of H_2O, which produced a white solid. The material was collected on a Büchner funnel, rinsed with 1 L of H_2O and air-dried. Recrystallization from 80 mL of 90% MeOH afforded white needles (16.1 g, 74.4%): mp 79−80 °C (lit.[12] mp 82−83 °C); ^{1}H NMR ($CDCl_3$) δ 2.61 (s, 3 H), 3.82 (s, 3 H), 3.86 (s, 3 H), 5.92 (d, 1 H, J = 2.4 Hz), 6.06 (d, 1 H, J = 2.4 Hz), 14.03 (s, 1 H).

3-Iodo-*p*-anisic Acid. To a mechanically stirred suspension of *p*-anisic acid (59.90 g, 394 mmol) and iodine (100.0 g, 394 mmol) in a mixture of 325 mL of glacial acetic acid and 60 g of concd H_2SO_4, heated to 45 °C with a water bath, was added dropwise a solution of 40 g of concd HNO_3 in 60 mL of HOAc at such a rate that the temperature was maintained between 40 and 50 °C (ca. 90 min). After addition, the mixture was stirred for 30 min at 50 °C and then diluted with 400 mL of H_2O, which produced a pink solid. The material was collected on a Büchner funnel, rinsed with 1 L of 10% $Na_2S_2O_4$ and 1 L of H_2O, and then air-dried. Recrystallization from 700 mL of pyridine/MeOH (1:1) provided colorless plates. The crystals were collected on a Büchner funnel, rinsed with 500 mL of MeOH, and then allowed to air-dry. After drying under high vacuum overnight, 71.4 g (65.2%) of the desired product was obtained; mp 243−244 °C (lit.[12] mp 238 °C); ^{1}H NMR (DMSO-d_6) δ 3.91 (s, 3 H), 7.00 (d, 1 H, J = 8.7 Hz), 7.95 (dd, 1 H, J = 8.7, 1.8 Hz), 8.27 (d, 1 H, J = 1.8 Hz), 12.89 (br s, 1 H).

3-Iodo-*p*-anisoyl Chloride (2a). To a suspension of PCl_5 (33.0 g, 158 mmol) in 30 mL of $CHCl_3$ was added, with magnetic stirring, 3-iodo-*p*-anisic acid (40.0 g, 144 mmol) in small portions. The solution was heated at reflux for 1 h under a gentle nitrogen sweep, during which time the solution became homogeneous. The solvent was evaporated under reduced pressure, and then the residue was distilled under high vacuum. The fraction distilling at 152−156 °C (5 mmHg; lit.[12] bp 183−185 °C, 12−13 mmHg) was collected, which rapidly solidified as a pink solid. Recrystallization from 200 mL of hexane/CH_2Cl_2 afforded the desired product as white needles (36.4 g, 85.5%): mp 57−58 °C; ^{1}H NMR ($CDCl_3$) δ 3.99 (s, 3 H), 6.88 (d, 1 H, J = 8.7 Hz), 8.13 (dd, 1 H, J = 8.7, 2.4 Hz), 8.52 (d, 1 H, J = 2.4 Hz); EI-MS m/z 296 (M^+, 42), 261 (100).

4,6-Dimethoxy-2-(3′-iodo-4′-methoxybenzoyloxy)acetophenone (3a). To a solution of **1** (5.00 g, 25.5 mmol) in 15 mL of pyridine was added **2a** (9.08 g, 30.6 mmol), and the solution was heated to 100 °C in an oil bath with magnetic stirring for 10 min. The solution was cooled to room temperature and then diluted with 20 mL of MeOH. The solution was cooled in an ice bath and, following scratching with a glass rod, colorless needles formed. The crystals were collected on a Büchner funnel, rinsed with cold MeOH and air-dried to provide 10.1 g (86.4%) of the desired product (mp 146 °C), which was used without further purification. An analytical sample was obtained via recrystallization from acetone: mp 156−157 °C (lit.[12] mp 158 °C; ^{1}H NMR ($CDCl_3$) δ 2.47 (s, 3 H), 3.83 (s, 3 H), 3.87 (s, 3 H), 3.96 (s, 3 H), 6.34 (d, 1 H, J = 2.1 Hz), 6.41 (d, 1 H, J = 2.1 Hz), 6.87 (d, 1 H, J = 8.7 Hz), 8.12 (dd, 1 H, J = 8.7, 2.1 Hz), 8.55 (d, 1 H, J = 2.1 Hz); EI-MS m/z 456 (M^+, 41), 261 (100).

1-(3′-Iodo-4′-methoxyphenyl)-3-(2″-hydroxy-3″,6″-dimethoxyphenyl)-1,3-propanedione (4a). To a suspension of **3a** (9.12 g, 20.0 mmol) in 20 mL of pyridine was added 2.80 g (50 mmol) of powdered KOH, and the solution was heated to 100 °C with magnetic stirring in an oil bath for 10 min. The solution was cooled to room temperature and treated with 10 mL of HOAc, which produced a yellow paste. Addition of 20 mL of MeOH and cooling in an ice bath afforded a yellow powder, which was collected on a Büchner funnel, rinsed with 100 mL of MeOH, and air-dried to provide 5.68 g (62.3%) of the desired product: mp 170−171 °C (lit.[12] mp 168 °C); ^{1}H NMR ($CDCl_3$) δ 3.51 (s, 3 H), 3.82 (s, 3 H), 3.97 (s, 3 H), 4.48 (s, 2 H), 5.85 (d, 1 H, J = 2.1 Hz), 6.09 (d, 1 H, J = 2.1 Hz), 6.89 (d, 1 H, J = 8.7 Hz), 7.96 (dd, 1 H, J = 8.7, 2.1 Hz), 8.40 (d, 1 H, J = 2.1 Hz), 13.68 (s, 1 H); EI-MS m/z 456 (M^+, 70), 425 (40), 261 (83), 181 (100).

3′-Iodo-5,7,4′-trimethoxyflavone (5a). To a magnetically stirred solution of **4a** (5.00 g, 11.0 mmol) in 60 mL of HOAc at 100 °C (water bath) was added 10 mL of 20% H_2SO_4/HOAc. After stirring at 100 °C for 10 min, the solution was poured into 250 mL of H_2O, which produced a white precipitate. The solid was collected on a Büchner funnel, rinsed with 500 mL of H_2O, and air-dried. Recrystallization from 150 mL of dioxane produced white needles (2.83 g, 58.9%). Evaporation of the mother liquor and recrystallization afforded an additional 1.35 g (total yield 4.18 g, 87.0%): mp 209−210 °C (lit.[12] mp 223 °C); ^{1}H NMR (DMSO-d_6) δ 3.83 (s, 3 H), 3.91 (s, 3 H), 3.92 (s, 3 H), 6.50 (d, 1 H, J = 2.1 Hz), 6.73 (s, 1 H), 6.91 (d, 1 H, J = 2.1 Hz), 7.14 (d, 1 H, J = 8.7 Hz), 8.06 (dd, 1 H, J = 8.7, 2.4 Hz), 8.41 (d, 1 H, J = 2.4 Hz); EI-MS m/z 438 (M^+, 100), 407 (16). Anal. Calcd for $C_{18}H_{15}IO_5$: C, 49.29; H, 3.42. Found: C, 48.98; H, 3.39.

4,6-Dimethoxy-2-(4′-methoxybenzoyloxy)acetophenone (3b). To a solution of **1** (20.0 g, 102 mmol) in 60 mL of pyridine was added 20.9 g (122 mmol) *p*-anisoyl chloride (**2b**), and the solution was heated at 100 °C for 10 min with magnetic stirring in an oil bath. The solution was cooled to room temperature and then diluted first with 50 mL of EtOH followed by 50 mL of H_2O. The mixture was cooled to 0 °C in an ice bath which, after scratching with a glass rod, produced shiny white plates. The crystals were collected on a Büchner

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funnel, rinsed with cold 50% EtOH, and air-dried to afford 27.4 g (81.4%) of desired product: mp 97−98 °C (lit.[25] mp 115−116 °C); ^{1}H NMR ($CDCl_3$) δ 2.46 (s, 3 H), 3.82 (s, 3 H), 3.86 (s, 3 H), 3.88 (s, 3 H), 6.37 (d, 1 H, J_{AB} = 2.1 Hz), 6.39 (d, 1 H, J_{AB} = 2.1 Hz), 6.96 (d, 2 H, J = 8.9 Hz), 8.09 (d, 2 H, J = 8.9 Hz); EI-MS m/z 330 (M^+, 23), 135 (100). Anal. Calcd for $C_{18}H_{18}O_6$: C, 65.45; H, 5.49. Found: C, 65.55; H, 5.57.

1-(4′-Methoxyphenyl)-3-(2″-hydroxy-3″,6″-dimethoxyphenyl)-1,3-propanedione (4b). To a solution of **3b** (30.0 g, 90.8 mmol) in 120 mL of pyridine was added 12.7 g (153 mmol) powdered KOH, and the solution was heated to 100 °C in an oil bath with magnetic stirring for 10 min. The mixture was cooled to room temperature and then treated with 50 mL of glacial acetic acid, which produced a thick yellow paste. The mixture was diluted with 100 mL of EtOH, which afforded a homogeneous solution, followed by 100 mL of H_2O. After being cooled to 0 °C, the product crystallized as yellow prisms. The crystals were collected on a Büchner funnel, rinsed with cold 50% EtOH, and air-dried to provide 20.72 g (69.1%) of desired product: mp 132−133 °C (lit.[25] mp 147−149 °C); ^{1}H NMR ($CDCl_3$) δ 3.48 (s, 3 H), 3.81 (s, 3 H), 3.89 (s, 3 H), 4.51 (s, 2 H), 5.83 (d, 1 H, J = 2.4 Hz), 6.09 (s, 1 H, J = 2.4 Hz), 6.97 (d, 2 H, J = 9.1 Hz), 7.94 (d, 2 H, J = 9.1 Hz), 13.74 (s, 1 H); FTIR (KBr) 3071, 1724, 1584, 1182, 1138 cm^{-1}; EI-MS m/z 330 (M^+, 46), 135 (100). Anal. Calcd for $C_{18}H_{18}O_6$: C, 65.45; H, 5.49. Found: C, 65.38; H, 5.54. The ^{1}H NMR spectra also indicated the presence of enol tautomers.

5,7,4′-Trimethoxyflavone (apigenin trimethyl ether, 5b). A suspension of **4b** (18.6 g, 56.4 mmol) in 200 mL of glacial acetic acid was heated to 100 °C with magnetic stirring in an oil bath. To this suspension was added 40 mL of 20% H_2SO_4 in acetic acid, and the mixture was stirred at 100 °C for 10 min. The mixture was poured into 1 L of H_2O, which produced a pale-yellow gelatinous solid. The solid was collected on a Büchner funnel, allowed to partially dry by drawing air through the funnel, and then partitioned between 600 mL each of $CHCl_3$ and H_2O. The organic layer was separated and washed with 600 mL each of 5% $NaHCO_3$ and saturated brine, dried over magnesium sulfate, filtered, and evaporated to afford a light yellow solid. Recrystallization from 300 mL of acetone provided white needles (12.45 g, 70.8%): mp 159−160 °C (lit.[26] mp 156 °C); ^{1}H NMR (DMSO-d_6) δ 3.83 (s, 3 H), 3.85 (s, 3 H), 3.90 (s, 3 H), 6.50 (d, 1 H, J = 2.1 Hz), 6.67 (s, 1 H), 6.84 (d, 1 H, J = 2.1 Hz), 7.09 (d, 2 H, J = 8.9 Hz), 7.99 (d, 2 H, J = 8.9 Hz); FTIR (KBr) 1644, 1348, 1256, 1121, 831 cm^{-1}; EI-MS m/z 312 (M^+, 100). Anal. Calcd for $C_{18}H_{16}O_5$: C, 69.22; H, 5.16. Found: C, 69.35; H, 5.32.

7,4′-Dimethoxy-5-hydroxyflavone (6). To a solution of **5b** (10.0 g, 32.1 mmol) in 200 mL of anhydrous CH_2Cl_2 was added a 1 M solution of BBr_3 (35.3 mL, 35.3 mmol) dropwise over 15 min at room temperature with magnetic stirring. A thick yellow precipitate formed rapidly during the addition. After stirring 5 h, the reaction was quenched by adding 200 mL of EtOH, and the solvent was then evaporated in vacuo. The yellow residue was triturated with 300 mL of boiling 50% EtOH. After cooling to room temperature, the yellow solid was collected on a Büchner funnel, rinsed with 500 mL of 50% EtOH, and air-dried. Recrystallization from 1.5 L of EtOH provided 8.16 g (85.4%) of the title product as fine, pale-yellow needles: mp 176−177 °C (lit.[27] mp 174−175 °C); ^{1}H NMR ($CDCl_3$) δ 3.88 (s, 3 H), 3.89 (s, 3 H), 6.36 (d, 1 H, J = 2.1 Hz), 6.48 (d, 1 H, J = 2.1 Hz), 6.57 (s, 1 H), 7.01 (d, 2 H, J_{AB} = 9.0 Hz), 7.84 (d, 2 H, J_{AB} = 9.0 Hz), 12.81 (s, 1 H); EI-MS m/z 298 (M^+, 100). Anal. Calcd for $C_{17}H_{14}O_5$: C, 68.45; H, 4.73. Found: C, 68.25; H, 4.77.

7,4′-Dimethoxy-5-hydroxy-6-iodoflavone (7). To a solution of **6** (2.98 g, 10.0 mmol) in 300 mL of CH_2Cl_2 was added thallium(I) acetate (3.16 g, 10.2 mmol). With magnetic stirring, a solution of iodine (2.54 g, 10.0 mmol) in 200 mL of CH_2Cl_2 was added dropwise over 1 h. During the addition, a fine suspension of thallium salts precipitated. The solution was stirred at room temperature overnight and then filtered through a bed of Celite to remove the precipitated salts. The filtrate was extracted sequentially with 500 mL each of 5% $NaHCO_3$, 10% $Na_2S_2O_4$, and saturated brine and then dried over magnesium sulfate and filtered. Evaporation in vacuo provided an orange solid. Recrystallization from 300 mL of $CHCl_3$/EtOH (1:2) afforded fine yellow needles (3.10 g, 73.1%); mp 227−228 °C (lit.[15] mp 205−207 °C); ^{1}H NMR ($CDCl_3$) δ 3.90 (s, 3 H), 4.00 (s, 3 H), 6.54 (s, 1 H), 6.64 (s, 1 H), 7.02 (d, 2 H, J = 9.0 Hz), 7.85 (d, 2 H, J = 9.0 Hz), 13.84 (s, 1 H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 181.7, 164.4, 163.5, 162.9, 161.3, 158.3, 128.2, 123.3, 114.6, 104.4, 104.3, 90.5, 90.4, 56.8, 55.5; EI-MS m/z 424 (M^+, 100). Anal. Calcd for $C_{17}H_{13}IO_5$: C, 48.14; H, 3.09. Found: C, 48.30; H, 3.11.

6-Iodo-5,7,4′-trimethoxyflavone (8). To a solution of **7** (3.00 g, 7.08 mmol) and K_2CO_3 (1.47 g, 10.6 mmol) in 150 mL of THF was added dimethyl sulfate (1.07 g, 8.49 mmol), and the solution was heated at reflux overnight. The solvent was evaporated and the residue partitioned between 50 mL each of $CHCl_3$ and H_2O. The organic phase was separated and washed with saturated brine, dried over magnesium sulfate, filtered, and evaporated to provide a yellow solid. Recrystallization from $CHCl_3$/EtOH (1:2, 150 mL) afforded pale-yellow needles (1.49 g). Column chromatography of the evaporated mother liquor (100 g of silica gel, 2% MeOH/CH_2Cl_2) afforded an additional 720 mg of desired product (total yield 2.21 g, 71.3%); mp 202−204 °C (lit.[15] mp 191−194 °C); ^{1}H NMR ($CDCl_3$) δ 3.89 (s, 3 H), 3.94 (s, 3 H), 4.01 (s, 3 H), 6.62 (s, 1 H), 6.78 (s, 1 H), 7.01 (d, 2 H, J = 9.1 Hz), 7.83 (d, 2 H, J = 9.1 Hz); EI-MS m/z 438 (M^+, 65), 311 (100). Anal. Calcd for $C_{18}H_{15}IO_5$: C, 49.34; H, 3.45. Found: C, 48.58; H, 3.21.

5,7,4′-Trimethoxy-3′-(trimethylstannyl)flavone (9a). To a solution of **5a** (438 mg, 1.00 mmol) in 30 mL of toluene were added $Pd(Ph_3P)_4$ (150 mg, 0.13 mmol) and hexamethylditin (655 mg, 2.00 mmol), and the solution was heated to reflux under N_2 for 16 h. The solution was filtered and the solvent evaporated in vacuo. The residue was dissolved in 75 mL of $CHCl_3$, washed with 75 mL of saturated brine, dried over magnesium sulfate, filtered, and evaporated to provide a white crystalline solid. The solid was triturated with 10 mL of EtOH, collected on a Buchner funnel, rinsed with fresh EtOH, and air-dried (270 mg, 56.8%). An analytical sample was obtained via recrystallization from EtOH, which produced fine colorless needles: mp 154−155 °C; ^{1}H NMR (DMSO-d_6) δ 0.31 (s, 9 H), 3.83 (s, 3 H), 3.85 (s, 3 H), 3.91 (s, 3 H), 6.51 (d, 1 H, J = 2.4 Hz), 6.65 (s, 1 H), 6.82 (d, 1 H, J = 2.4 Hz), 7.08 (d, 1 H, J = 8.7 Hz), 7.86 (d, 1 H, J = 2.4 Hz), 8.01 (dd, 1 H, J = 8.7, 2.4 Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 177.9, 166.5, 164.0, 161.4, 161.0, 160.1, 134.3, 131.7, 128.6, 124.1, 109.1, 107.7, 96.0, 92.9, 56.4, 55.7, 55.5; EI-MS m/z 476 (M^+, 64), 461 (100). Anal. Calcd for $C_{21}H_{24}O_5Sn$: C, 53.09; H, 5.09. Found: C, 52.74; H, 5.01.

Pinacol 5,7,4′-trimethoxyflavone-3′-boronate (9b). A solution of **5a** (2.00 g, 4.56 mmol), bis(pinacolato)diboron (1.50 g, 5.93 mmol), KOAc (1.79 g, 18.24 mmol), and $PdCl_2$(dppf) (372 mg, 0.456 mmol) in 60 mL of DMF was stirred at 80 °C overnight. The reaction mixture was filtered, diluted with 200 mL of EtOAc, and then washed with H_2O (3×) and brine. After drying over sodium sulfate, the solvent was evaporated. The product was chromatographed over silica gel (EtOAc/MeOH, 96:4) to afford 1.21 g (60.5%) of the desired product as a gray solid. The material could be recrystallized from CH_2Cl_2/EtOAc, as pale gray needles: mp 218−220 °C; ^{1}H NMR ($CDCl_3$) δ 1.39 (s, 12 H), 3.91 (s, 3 H), 3.92 (s, 3 H), 3.96 (s, 3 H), 6.38 (d, 1 H, J = 2.2 Hz), 6.60 (d, 1 H, J = 2.2 Hz), 6.66 (s, 1 H), 6.96 (d, 1 H, J = 8.9 Hz), 7.96 (dd, 1 H, J = 8.9, 2.4 Hz), 8.18 (d, 1 H, J = 2.4 Hz); ^{13}C NMR ($CDCl_3$) δ 177.9, 166.6, 164.0, 161.0, 160.9, 160.0, 134.9, 130.4, 123.3, 114.4, 110.6, 109.3, 107.8, 96.0, 93.0, 83.9, 56.4, 55.9, 55.7, 24.7; FTIR (KBr) 1645, 1602, 1330, 1148 cm^{-1}; APCI-MS m/z 439 (MH^+, 100). Anal. Calcd for $C_{24}H_{27}BO_7 \cdot {}^1/_2H_2O$: C, 64.45; H, 6.31. Found: C, 64.17; H, 6.05.

Robustaflavone Hexamethyl Ether (10). To a solution of **8** (25.0 mg, 0.057 mmol) and **9a** (50.0 mg, 0.114 mmol) in DMF/H_2O (9:1), which had been deoxygenated for 15 min by bubbling N_2, were added NaOH (9.1 mg, 0.23 mmol) and Pd-

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$(PPh_3)_4$ (6.6 mg, 0.0057 mmol), and the reaction was stirred at 80 °C for 2 h. The solution was diluted with CH_2Cl_2 and then extracted with H_2O and saturated brine. The organic layer was dried over magnesium sulfate, filtered, and evaporated in vacuo. Column chromatography (silica gel, CH_2Cl_2/MeOH, 96:4) afforded the desired product **10** (17.7 mg, 50%), identical with an authentic sample prepared via methylation of robustaflavone:[7] mp 296−297 °C (lit.[7] mp 303−305 °C) ^{1}H NMR ($CDCl_3$) δ 3.62 (s, 3 H), 3.85 (s, 3 H), 3.87 (s, 3 H), 3.91 (s, 6 H), 3.96 (s, 3 H), 6.37 (d, 1 H, J = 2.1 Hz), 6.60 (d, 1 H, J = 2.4 Hz), 6.69 (s, 1 H), 6.74 (s, 1 H), 6.89 (s, 1 H), 7.04 (d, 2 H, J = 9.0 Hz), 7.10 (d, 1 H, J = 8.7 Hz), 7.81 (d, 1 H, J = 2.4 Hz), 7.89 (d, 2 H, J = 9.0 Hz), 7.89 (dt, 1 H, J = 2.1, 8.7 Hz); HR-EIMS m/z 623.1917 (MH^+, requires 623.1917). Anal. Calcd for $C_{36}H_{30}O_{10}\cdot 0.2CH_2Cl_2$: C, 68.34; H, 4.81. Found: C, 68.28; H, 5.19. (Note: dichloromethane was extremely difficult to remove from this material, consistent with our observations of the hexamethyl ether prepared from natural robustaflavone).

Robustaflavone. To a solution of **10** (75.0 mg, 0.12 mmol) in 10 mL of dry $CHCl_3$ was added BBr_3 (1.0 M in CH_2Cl_2, 1.45 mL, 1.45 mmol), and the resulting yellow slurry was stirred at reflux overnight. The reaction mixture was cooled to room temperature, quenched by the careful addition of MeOH, and evaporated in vacuo. The resulting orange solid was triturated with MeOH, the solvent again evaporated in vacuo, and the solid partitioned between EtOAc and 1 M NaOH. The organic layer was discarded, and the aqueous layer was extracted with EtOAc. After cooling to 0 °C, the aqueous layer was carefully acidified to pH 3.0 by the dropwise addition of 3 M HCl. The resulting yellow precipitate was collected by vacuum filtration, rinsed with water, and air-dried (38.7 mg). The crude material was chromatographed through silica gel, eluting with a mixture of toluene/pyridine/formic acid (20:10:1). Appropriate fractions were combined and evaporated to afford 19.4 mg (30.0%) robustaflavone. An analytical sample was obtained via recrystallization from pyridine/H_2O (1:1): mp 370−372 °C, dec (lit.[1] mp 350−352 °C). Spectral data of synthetic robustaflavone was identical to that recently reported[1] for the natural product isolated from *Rhus succedanea*. HR−FABMS m/z 538.0912 (M^+, requires 538.0900). Anal. Calcd for $C_{30}H_{18}O_{10}\cdot H_2O$: C, 64.75; H, 3.60. Found: C, 64.95; H, 3.91.

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