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# Novel Chemical Synthesis of Ginkgolic Acid (13:0) and Evaluation of Its Tyrosinase Inhibitory Activity

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**ABSTRACT:** A novel efficient synthesis of ginkgolic acid (13:0) from abundant 2,6-dihydroxybenzoic acid was successfully developed through a state-of-the-art palladium-catalyzed cross-coupling reaction and catalytic hydrogenation with an overall yield of 34% in five steps. The identity of the synthesized ginkgolic acid (13:0) was confirmed by nuclear magnetic resonance, mass spectrometry, infrared, and high-performance liquid chromatography. The reaction sequence of this method can be readily extended to the synthesis of other ginkgolic acids. The synthesized ginkgolic acid (13:0) exhibited promising anti-tyrosinase activity ($IC_{50}$ = 2.8 mg/mL) that was not correlated to antioxidant activity as probed by 1,1-diphenyl-2-picrylhydrazyl, 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid), ferric reducing ability of plasma, and oxygen radical absorbance capacity assays. The synthetic strategy developed in this work will significantly facilitate biological studies of ginkgolic acids that have great potential applications in food and pharmaceuticals.

**KEYWORDS:** *Ginkgolic acid, synthesis, tyrosinase inbibitory activity, cross-coupling reaction, antioxidant activity*

## INTRODUCTION

Ginkgolic acids (GAs) are natural salicylic acid derivatives of long alkyl or alkenyl substituents at the *ortho* position to the carboxyl group. GAs exist in various plant materials, including cashew nuts,[1,2] nutmeg,[3] liverwort,[4] brown alga,[5] and the leaves, sarcotesta, and nuts of *Ginkgo biloba* L.[6−8] Many beneficially effects of GAs have been found, such as antivirus,[8] antifungi,[9] antibacterial,[10] antitumor,[11] and tyrosinase inbibitory activities.[12,13] The structures of five typical GAs were illustrated in Figure 1. The long side chains usually contain 13−17 carbon atoms and 0−2 double bonds.

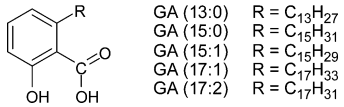


**Figure 1.** Five typical GAs.

Despite the wide spectrum of biological activities of GAs, their natural availability is very limited. As a result, many synthetic studies have been carried out to develop an efficient way to obtain GAs for biological activity investigation. Tyman et al.[14,15] applied a sequential alkylation and carboxylation of 2-fluoroanisole with alkylithium and $CO_2$, followed by demethylation with $BCl_3$, and afforded GA (15:0) in a low yield. Then, the strategy of chain elongation of 6-methylsalicylate was practiced and produced a higher yield of GA (15:0). Fürstner et al. concisely prepared GA (15:0) using the Suzuki cross-coupling reaction and directly constructed the side chain.[16] Satoh et al. successfully synthesized GA (15:0)[17,18] and GA (15:2)[19] using an annelation reaction of isoxazoles with ethyl acetoacetate and a Wittig reaction.

As a member of GAs, GA (13:0) has been recognized as an anti-inflammatory principle component in metabolites of brown alga,[5] and its strong molluscicidal activity has also been uncovered.[20] The natural abundance of GA (13:0) is particularly low. However, the synthetic investigations on GA (13:0) were scare and unsatisfactory. A reaction sequence of 2-alkyne-1-palmitate addition with 1-methoxy-1,4-cyclohexadiene, followed by demethylation and hydrolysis, was developed to produce GA (13:0),[21] regardless of complicated operations and harsh conditions involved. Therefore, to facilitate biological studies of GA (13:0) that has great potential applications in food and pharmaceuticals, an efficient synthetic method for GA (13:0) has to be developed.

In this study, a new strategy was applied to synthesize GA (13:0), employing state-of-the-art catalytic cross-coupling reactions to build the long side chain from abundant 2,6-dihydroxybenzoic acid. Then, tyrosinase inbibitory activity of the synthesized GA (13:0) was evaluated, and the mechanism was also briefly probed. Herein, the details of this work were described.

## MATERIALS AND METHODS

**Chemicals.** 9-Borabicyclo-[3.3.1]-nonane dimer (9-BBN) and 1-tridecene were purchased from Sigma-Aldrich and TCI (Japan), respectively, and used as obtained. Ethylene glycol dimethyl ether, thionyl chloride ($SOCl_2$), 4-dimethylaminopyridine (DMAP), 2,6-dihydroxybenzoic acid, $PdCl_2$(dppf), Pd/C, trifluoromethanesulfonic anhydride, pyridine, sodium methylate, and KOH were purchased from Aladdin-Reagent (China) and used as obtained. GA (13:0) standard was purchased from Shanghai Ronghe Co., Ltd. (China).

**Chemical Synthesis of GA (13:0).** *Synthesis of Trifluoromethanesulfonic Acid 2,2-Dimethyl-4-oxo-4H-1,3-benzodioxin-5-yl Ester (**3**).* Acetone (14.8 mL, 208 mmol) and $SOCl_2$ (3.74 mL, 52 mmol) were added to a solution of 2,6-dihydroxybenzoic acid (**1**) (3.08 g, 20 mmol) and DMAP (122.2 mg, 1 mmol) in 1,2-dimethoxyethane (DME) (15

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mL). The mixture was allowed to be stirred with a magnetic stirring bar in a round-bottomed flask at 0 °C for 1 h and at room temperature for 24 h. Saturated aqueous $NaHCO_3$ solution was added to the mixture, and the aqueous solution was extracted with ethyl acetate (50 mL × 3). The combined organic solution was washed with saturated aqueous NaCl solution and dried over anhydrous $MgSO_4$, and the solution was concentrated using a rotary evaporator at 40 °C under reduced pressure. The residue was purified by silica gel column chromatography using an isocratic elution with hexane/ethyl acetate (3:1, v/v) to afford compound **2**.

Anhydrous pyridine (1.2 mL, 15 mmol) and trifluolomethanesulfonic anhydride (0.82 mL, 5 mmol) were successively added to a solution of compound **2** (0.68 g, 3.5 mmol) in anhydrous dichloromethane (5 mL), and the mixture was allowed to be stirred at 0 °C for 1 h followed by 3 h at room temperature. The reaction mixture was extracted with ethyl acetate (50 mL × 3), and the combined organic solution was washed with saturated aqueous NaCl solution and dried over anhydrous $MgSO_4$. The solvent was removed under reduced pressure, and the residue was purified by silica gel column chromatography using an isocratic elution with hexane/ethyl acetate (3:1, v/v) to afford the triflate **3**.

*Synthesis of 2,2-Dimethyl-5-tridec-1-enyl-benzo[1,3]dioxin-4-one (**4′**).* A solution of 1-tridecene (124 mg, 0.68 mmol) in *N*,*N*-dimethylformamide (DMF) was stirred with $K_2CO_3$ (94.8 mg, 0.68 mmol) at ambient temperature for 30 min prior to the addition of the triflate **3** (200 mg, 0.62 mmol) and $PdCl_2$(dppf) (16.9 mg, 0.02 mmol) under a nitrogen atmosphere. The reaction mixture was heated for 12 h at 75 °C, and the solvent was evaporated. The residue was partitioned between $Et_2O$ and water (30 mL each), and the organic layer was separated and dried ($Na_2SO_4$). The solvent was removed under reduced pressure, and the crude product is purified by silica gel column chromatography with hexane/ethyl acetate (20:1, v/v) as the eluent, thus affording a product whose structure could be confirmed by mass spectrometry (MS) and proton nuclear magnetic resonance ($^1$H NMR).

*Preparation of 5-Tridecyl-2,2-dimethylbenzo[1,3]dioxin-4-one (**4**).* Reduction of the double bond in the long unsaturated chain substituent of compound **4′** was conducted to afford compound **4**. A solution of compound **4′** (111.5 mg, 0.31 mmol) in EtOAc in a round-bottomed flask was added with Pd/C (5%) (99 mg, 0.047 mmol) under a nitrogen atmosphere, followed by exclusion of $N_2$ through a three-way valve using vacuum pump and injection of $H_2$ with a hydrogen balloon. After 12 h of reaction at ambient temperature, $H_2$ in the round-bottomed flask was excluded. The mixture was filtered to remove Pd−C, and the organic phase was washed with water. The organic layer was separated and dried ($Na_2SO_4$), and the solvent was removed under reduced pressure to afford compound **4**.

**Preparation of GA (13:0) (5).** The hydrolysis of compound **4** was conducted to furnish the GA (13:0) **5**. A solution of compound **4** (17 mg, 0.048 mmol), in aqueous KOH (50%, 0.2 mL) and dimethyl sulfoxide (DMSO) (0.5 mL) was heated at 80 °C for 1 h. The reaction mixture was cooled to ambient temperature and diluted with water (20 mL), followed by the acidification with HCl (1 mol/L). The acidification was terminated when pH of the mixture reached 2. This mixture was then extracted with ethyl acetate (50 mL), and the organic layer was separated and dried over $Na_2SO_4$. The solvent was removed under reduced pressure, and the crude product was subjected to column chromatography on silica gel using an isocratic elusion with hexane/ethyl acetate/HAc (70:29:1, v/v/v) to afford compound **5**.

**NMR Analysis of Compounds 3 and 5.** $^1$H NMR analysis of the above purified compounds **3**, **4′**, and **5** was carried out to confirm their structure. NMR spectra of compounds **3**, **4′**, and **5** were recorded on a Bruker 500 MHz NMR spectrometer at room temperature in $CDCl_3$, with the solvent residual peak as the internal reference. Chemical shifts were expressed in $\delta$ values.

**MS Analysis of Compounds 4′, 4, 5, and 5′.** The purity of compound **5** was confirmed by high-performance liquid chromatography (HPLC), and the identities of compounds **4′**, **4**, and **5** were confirmed by a Thermo Finnigan LTQ-DECA-XP-MAX linear ion trap mass spectrometer (Thermo Fisher Scientific, Waltham, MA). Thermo Finnigan Xcalibur software (version 2.1) was used for data acquisition and processing. The analysis was monitored by an electrospray ionization (ESI) interface with a detector voltage of 1.5 kV, from $m/z$ 150 to 800 in the mass analyzer, and with an even time of 1.0 s. The ESI parameters were as follows: source voltage, 2.50 kV; sheath gas flow rate, 50 Arb; aux/sweep gas flow rate, 10 arb; capillary voltage, −41.00 V; and capillary temperature, 330 °C. ESI was operated in both negative-ion (NI) and positive-ion (PI) modes.

**Fourier Transform Infrared (FTIR) Spectroscopy Analysis of Compound 5.** A FTIR spectrum of the purified compound **5** was recorded by a FTIR spectrophotometer (Tensor 27, Bruker, Germany) using a KBr disk containing 1% finely ground samples.

**HPLC Analysis of Compound 5.** HPLC analysis of compound **5** was performed using an Agilent Technologies Series 1200 system (Agilent, Santa Clara, CA) equipped with a G1322A automatic degasser, a G1311A quaternary pump, a G1367 auto sampler, and an ultraviolet−visible (UV−vis) diode array detector (DAD). A C18 column (250 mm length, 4.6 mm inner diameter, and 5 $\mu$m particle size, Merck Germany, Ltd., Germany) operated at 30 °C was used, and the sample was eluted using an isocratic solvent system containg methanol/1% HAc (90:10), at a flow rate of 1.0 mL/min. A total of 20 $\mu$L of sample was injected into the system and detected at 310 nm by UV detection.

**Tyrosinase Inhibitory Activity Assay.** The mushroom tyrosinase was obtained from Worthington Biochemical Corporation, Freehold, NJ. L-3,4-Dihydroxyphenylalanine (L-DOPA) and kojic acid were purchased from Sigma-Aldrich (St. Louis, MO). The tyrosinase assay was performed with modification, as reported by Zhang et al.[22] A total of 80 $\mu$L of 0.1 M phosphate-buffered saline (PBS) at pH 6.8, 20 $\mu$L of mushroom tyrosinase diluted in the phosphate buffer (985 units/mL), and various concentrations of different test samples dissolved in 50 $\mu$L of 90% methanol were inserted into 96-well plates for 5 min of pre-incubation at 30 °C. A total of 50 $\mu$L of L-DOPA was then added to start the enzymatic reaction, and absorbance at 492 nm was measured by a Thermo Scientific Multiskan GO UV/vis microplate spectrophotometer (Thermo Scientific, Waltham, MA) to observe dopachrome formation for 20 min. All experiments were carried out at least in triplicate. The percentage of inhibition was calculated as follows:

$$\text{percentage of inhibition} = [(A - B) - (C - D)]/(A - B) \times 100$$

where $A$ is OD at 492 nm with tyrosinase but without the test sample, $B$ is OD at 492 nm without the test sample and tyrosinase, $C$ is OD at 492 nm with the test sample and tyrosinase, and $D$ is OD at 492 nm with the test sample but without tyrosinase. Kojic acid was tested as a positive control.

## RESULTS AND DISCUSSION

**Synthesis of Trifluoromethanesulfonic Acid 2,2-Dimethyl-4-oxo-4*H*-1,3-benzodioxin-5-yl Ester (3).** Preparation of compound **2** commenced with ketalization of 2,6-dihydroxybenzoic acid (**1**) according to a procedure described by Hadfield et al.[23] The silica gel thin-layer chromatography (TLC) using a mixed solvent system of hexanes and ethyl acetate (2:1) was applied to monitor the progress of the reaction. Visualization of TLC with a UV irradiation at 254 nm revealed that the addition amount of acetone was critical for the conversion rate of the reaction. Initially, acetone was added at the molar ratio of 2.6:1 to 2,6-dihydroxybenzoic acid, and the reaction did not proceeded well. After several attempts, the addition amount of acetone was finally optimized to a molar ratio of 10:1 to 2,6-dihydroxybenzoic acid and a good yield of 67% was obtained (Figure 2a).

The remaining hydroxyl group of compound **2** was readily converted to the triflate **3** as a colorless solid in a good yield of 73% by treatment with trifluoromethanesulfonic anhydride and pyridine in anhydrous dichloromethane following the procedure established by Uchiyama et al.[24] (Figure 2a). The structure of synthesized compound **3** was confirmed by $^1$H NMR spectros-


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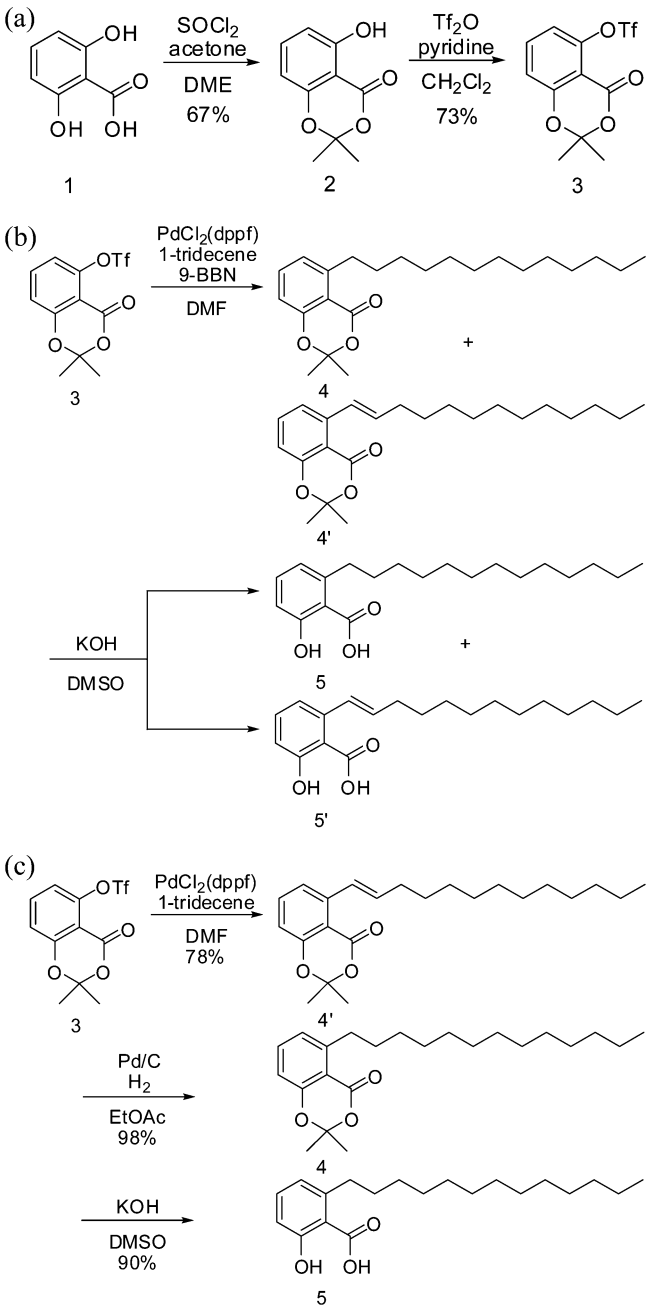


**Figure 2.** (a) Synthesis of trifluoromethanesulfonic acid 2,2-dimethyl-4-oxo-4*H*-1,3-benzodioxin-5-yl ester (**3**). (b) Synthesis of GA (13:0) (**5**) with 9-BBN. (c) Synthesis of GA (13:0) without 9-BBN.

copy after purification by flash column chromatography. $^1$H NMR (500 MHz, $CDCl_3$) $\delta$: 7.62 (t, $J$ = 8 Hz, 1H; aromatic proton), 7.07 (d, $J$ = 8 Hz, 1H; aromatic proton), 7.02 (d, $J$ = 8 Hz, 1H; aromatic proton), 1.78 (s, 6 H; 2 × C$\underline{H}_3$). The $^1$H NMR data of synthesized compound **3** were in agreement with those reported by Uchiyama et al.[24]

**Synthesis of GA (13:0) with 9-BBN.** A direct preparation of 5-tridecyl-2,2-dimethylbenzo-[1,3]-dioxin-4-one (**4**) through a Suzuki cross-coupling reaction was attempted according to Fürstner's method.[16] 1-Tridecene borane was hydroborated with 9-BBN and generated 1-tridecane borane *in situ*. The resultant alkylborane was treated with $K_2CO_3$ to form the activated boron complex that then participated in the palladium-catalyzed Suzuki cross-coupling reaction as the nucleophile and coupled with the triflate **3**. To optimize the Suzuki cross-coupling reaction, numerous reaction conditions were tested.

As a start, anhydrous tetrahydrofuran (THF) was chosen as the solvent, NaOMe was applied to provide alkaline circumstance, and a mixture of $PdCl_2$ and dppf was used as the catalyst. The reaction was carried out open to air, and the system was not hermetically sealed. When the reaction was conducted at 65 °C, the solvent tended to be evaporated, which made the reaction difficult to proceed. To change the situation, the reaction temperature was changed to 60 °C and the reaction system was hermetically sealed with other conditions unchanged. The progress of the reaction was monitored by TLC, and no obvious product was observed even after 12 h. Then, air was excluded from the reaction system by nitrogen before heated, and the reaction was carried out under a nitrogen atmosphere during heating. Unfortunately, although a new product was observed on TLC, the conversion rate of the triflate **3** was very poor even, with excessive 1-tridecene and 9-BBN and a prolonged reaction time.

Because of the poor conversion of the triflate starting material, we questioned the efficacy of the complexation between $PdCl_2$ and dppf. Therefore, the commercially available catalyst complex $PdCl_2$(dppf) was directly employed instead of generation *in situ* from $PdCl_2$ and dppf. To our delight, most of the triflate starting material was converted to new products, as monitored by TLC. After extraction with ethyl acetate and washing with water, the crude product was purified by silica gel column chromatography. The purified product was subjected to hydrolysis by the treatment of KOH in DMSO at 80 °C for 2 h, and then the reaction mixture was acidified with HCl and extracted with ethyl acetate. However, $^1$H NMR analysis revealed that the new product was a complex mixture of unidentified compounds, and no desired product was observed.

As a result, we doubted that THF was presumably not the appropriate solvent for controlling the pathway of the Suzuki cross-coupling reaction. Screening of the solvents found that DMF was the suitable solvent for the reaction. Further optimization of the reaction condition exhibited that $K_2CO_3$ was preferred to $NaOCH_3$ to provide a milder reaction condition. Under the optimized condition, the desired product was obtained in low yield because of the severe side reaction. Subsequently, this product was hydrolyzed with KOH in DMSO at 80 °C for 2 h and furnished the final product, GA (13:0) (**5**) (Figure 2b). Therefore, GA (13:0) can be synthesized in this route but with a low overall yield, and the performance of this method has to be improved.

Through careful examination of the side reaction during the cross-coupling reaction, a major byproduct with a molecular weight of 318, which was 2 less than that of compound **5**, was observed (Figure 2b). Thus, we speculated that there was a double bond in the long chain substituent of the byproduct that was finally identified as (*E*)-2-hydroxy-6-(tridec-1-enyl)benzoic acid (**5′**) by ESI−MS (Figure 3a). The molecular ion $[M - H]^-$ at $m/z$ 317 and $[M - COOH]^-$ at $m/z$ 273 in the NI mode ESI−MS spectrum strongly supported the structure of compound **5′**. The byproduct was reasoned to be generated directly from the coupling of 1-tridecene and the triflate without the assistance of 9-BBN. This interesting finding immediately led us to modification of the reaction sequence without 9-BBN to improve the overall efficiency.

**Modified Reaction Sequence for Synthesizing GA (13:0) without 9-BBN.** On the basis of the above investigations, the reaction sequence was modified to the direct coupling of 1-tridecene and the triflate without 9-BBN, followed by catalytic


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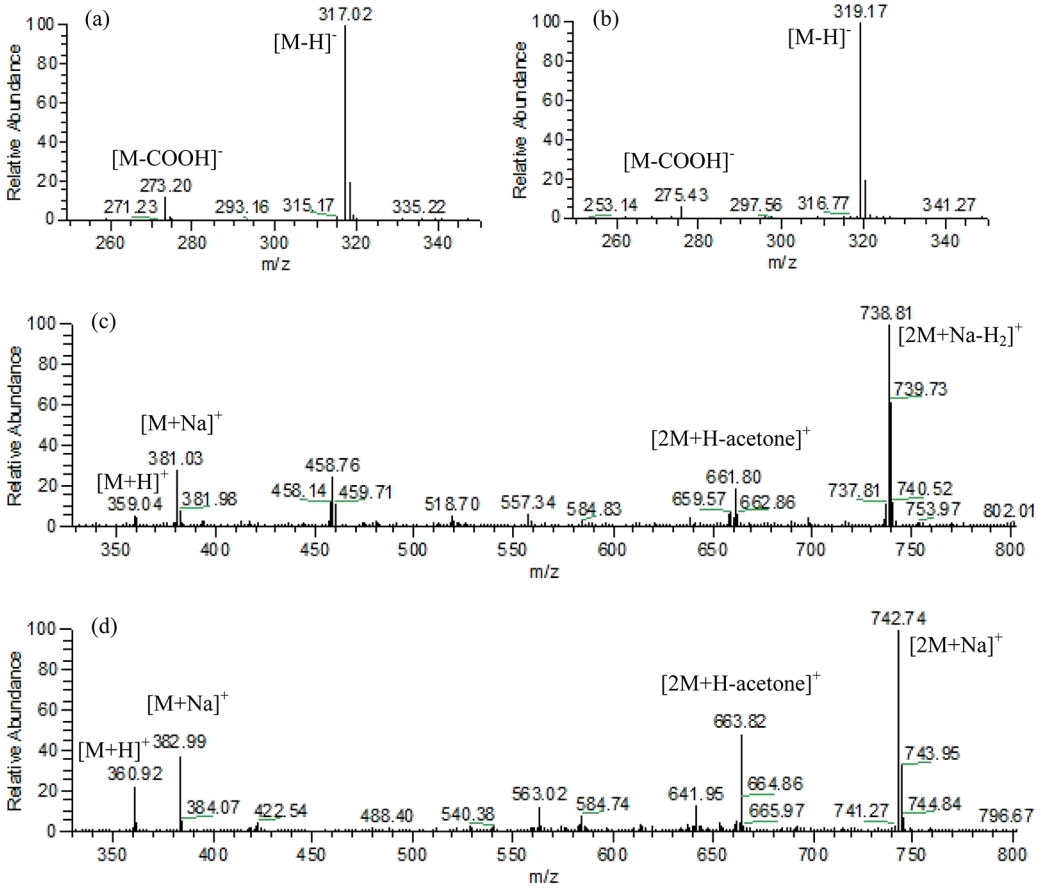


**Figure 3.** (a) ESI−MS spectrum of (*E*)-2-hydroxy-6-(tridec-1-enyl)benzoic acid (**5′**). (b) ESI−MS spectrum of the synthesized GA (13:0). (c) ESI−MS spectrum of compound **4′**. (d) ESI−MS spectrum of compound **4**.

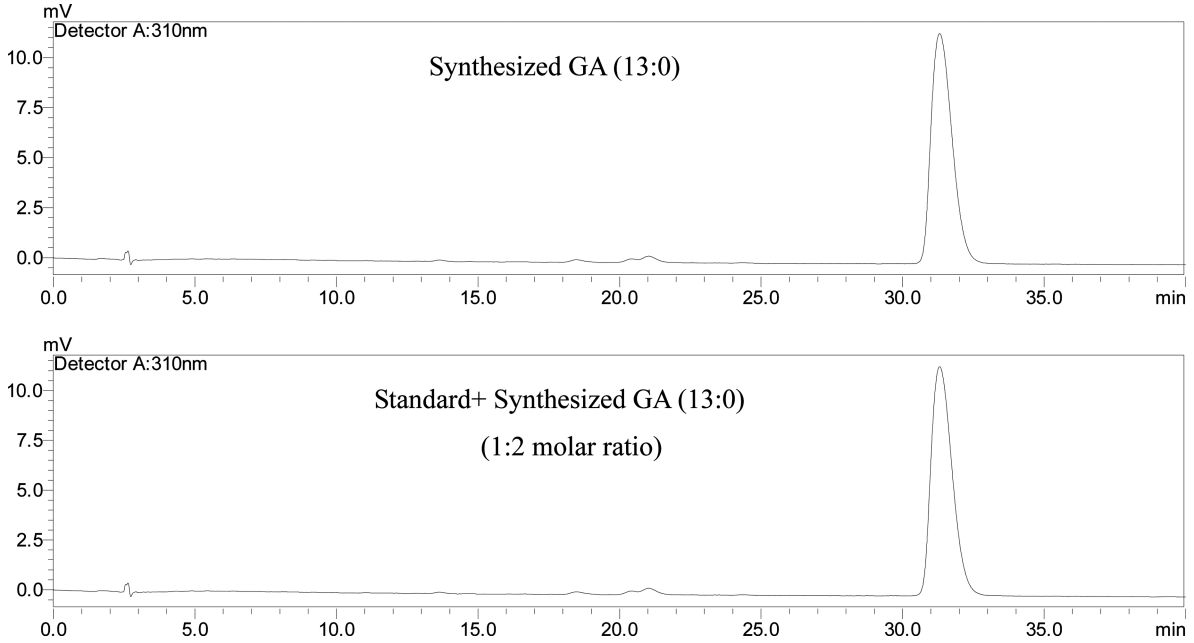


**Figure 4.** HPLC analysis of the synthesized GA (13:0).

hydrogenation. As expected, the coupling of 1-tridecene and the triflate proceeded smoothly to give compound **4′** by the treatment of $K_2CO_3$ and catalytic $PdCl_2$(dppf) under $N_2$ in anhydrous DMF (Figure 2c). The reaction was completed in 12 h. After extraction, chromatography purification afforded compound **4′** in a good yield of 78%. The structure of compound **4′** was confirmed by the molecular ion $[M + H]^+$ at $m/z$ 359, $[M + Na]^+$ at $m/z$ 381, $[2M + Na - H_2]^+$ at $m/z$ 739, and $[2M + H - acetone]^+$ fragment at $m/z$ 662 in the PI mode ESI−MS spectrum (Figure 3c).


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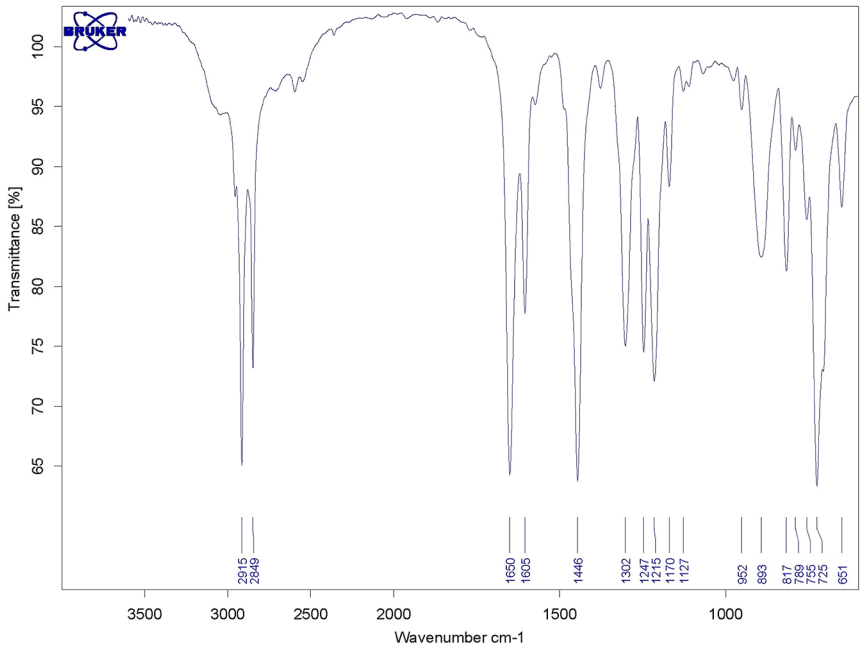


**Figure 5.** IR spectrum of the synthesized GA (13:0).

At first, catalytic hydrogen transfer with triethylsilane was tried to hydrogenate the double bond of compound **4′**. However, this method was not applicable to compound **4′** and only produced a low yield of the desired compound **4**. Then, reduction of the double bond was exercised with Pd/C-catalyzed hydrogenation with hydrogen gas. Desired compound **4** was readily obtained in an excellent yield of 98% (Figure 2c). The molecular ion $[M + H]^+$ at $m/z$ 361, $[M + Na]^+$ at $m/z$ 383, $[2M + Na]^+$ at $m/z$ 743, and $[2M + H - \text{acetone}]^+$ fragment at $m/z$ 664 in the PI mode ESI−MS spectrum strongly supported the structure of compound **4** (Figure 3d). Finally, hydrolysis of compound **4** with KOH furnished the target compound, GA (13:0), in an excellent yield of 90% (Figure 2c). It has to be noted that the order of hydrogenation reduction and hydrolysis is critical. Once hydrogenation reduction was carried out after hydrolysis of compound **4′**, a significantly lower yield was afforded, presumably because of the influence of the hydroxyl and carboxyl groups on catalytic transformation during the reduction reaction.

**Structure Analysis of GA (13:0) (5).** To confirm the identity of the synthesized GA (13:0), it was compared to the commercially available standard by HPLC analysis. As shown in Figure 4, the synthesized GA (13:0) had the same retention time as that of the standard and a mixture of the synthesized GA (13:0) and the standard (2:1 molar ratio) produced only one sharp single peak, demonstrating the same identity of the synthesized GA (13:0) and the standard.

The structure of the synthesized GA (13:0) was further confirmed by $^1$H NMR, ESI−MS, and infrared (IR). $^1$H NMR (400 MHz, $CDCl_3$) $\delta$: 11.27 (br, 1H; COO$\underline{H}$), 7.35 (t, $J$ = 8 Hz, 1H; aromatic proton), 6.87 (d, $J$ = 8 Hz, 1H; aromatic proton), 6.77 (d, $J$ = 8 Hz, 1H; aromatic proton), 2.97 (m, 2H; Ar−C$\underline{H}_2$), 1.27 (br, 22H; 11 × C$\underline{H}_2$), 0.89 (t, $J$ = 7 Hz, 3H; C$\underline{H}_3$). The $^1$H NMR data of the synthesized GA (13:0) were in agreement with those reported by Kazlauskas et al.[5] The molecular ion $[M - H]^-$ at $m/z$ 319 and $[M - COOH]^-$ fragment at $m/z$ 275 in the NI mode ESI−MS spectrum strongly supported the structure of GA (13:0) (Figure 3b). IR ($cm^{-1}$): 2915 (aliphatic C−H stretching vibration), 2849 (aliphatic C−H strecting vibration), 1650 (carboxyl C═O stretching vibration), 1605 (aromatic C═C stretching vibration), 1446 (aliphatic C−H bending vibration), 1127−1302 (C−O stretching vibration), and 651−952 (phenyl ring vibration) (Figure 5). These aborption peaks in IR indicated the presence of a phenyl ring, hydroxyl group, carboxyl group, and aliphatic chain.

Therefore, it was confirmed that GA (13:0) was successfully synthesized in this work. Overall, the synthetic sequence is efficient and straightforward with operational simplicity. The strategy developed in this study can be readily extended to synthesize other GAs.

**Tyrosinase Inhibitory Activity Assay.** Tyrosinase inhibitory activity assay of the synthesized GA (13:0) was performed employing kojic acid as the positive control. It was demonstrated that the synthesized GA (13:0) had a dose-dependent inhibitory effect on mushroom tyrosinase (Figure 6). The concentration

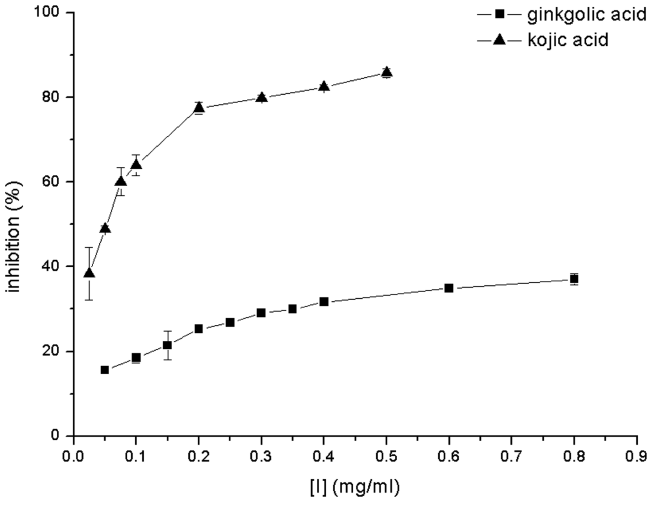


**Figure 6.** Tyrosinase inhibitory activity assay of the synthesized GA (13:0).


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inhibited by 50% tyrosinase activity ($IC_{50}$) was estimated to be 2.8 mg/mL, and the $IC_{50}$ value of the positive control kojic acid was 0.047 mg/mL. Although the tyrosinase inhibitory activity cannot compete with kojic acid, the promising activity encouraged further studies on GAs to improve the activity.

To elucidate the mechanism of tyrosinase inhibitory activity of the synthesized GA (13:0), its antioxidant activities were determined. The synthesized GA (13:0) exhibited very limited 1,1-diphenyl-2-picrylhydrazyl (DPPH) and 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) radical scavenging activities and almost no activities in ferric reducing ability of plasma (FRAP) and oxygen radical absorbance capacity (ORAC) assays. These results suggested that tyrosinase inhibitory activity of the synthesized GA (13:0) was not correlated to the antioxidant activity, which is consistent with the study by Kubo et al. that demonstrated that inhibition of mushroom tyrosinase was affected by specifically binding to the active site of tyrosinase on a competitive basis.[13]

In conclusion, an efficient synthesis of GA (13:0) from abundant 2,6-dihydroxybenzoic acid was successfully established with a palladium-catalyzed cross-coupling reaction of 1-tridecene and catalytic hydrogenation. The identity of the synthesized GA (13:0) was confirmed by NMR, MS, IR, and HPLC. The reaction sequence of this method is concise and can be readily extended to the synthesis of other GAs. Development of this method will greatly facilitate biological studies of GAs that have great potential applications in food and pharmaceuticals. The synthesized GA (13:0) exhibited promising anti-tyrosinase activity but no antioxidant activity, as probed by DPPH, ABTS, FRAP, and ORAC assays, suggesting that no correlation exists between anti-tyrosinase activity and antioxidant activity. A further study on the synthesis and biological activity of GAs is undergoing and will be reported in due course.

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### Notes

The authors declare no competing financial interest.

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