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Total Synthesis of (±)-Jiadifenin, a Non-peptidyl Neurotrophic Modulator

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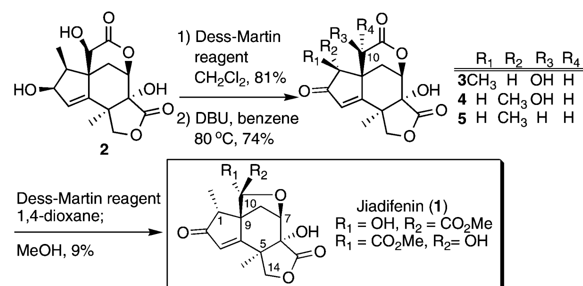
Fukuyama and colleagues recently reported the isolation of the majucin cage-like sesquiterpene jiadifenin (**1**) as an inseparable anomeric mixture in 0.001% yield from the methanol extract of the pericarps of *Illicium jiadifengpi*.[1] (2*S*)-Hydroxy-3,4-dehydroneomanjucin (**2**), also obtained from *I. jiadifengpi*, is a potential biosynthetic precursor of **1**. Indeed, Fukuyama and co-workers demonstrated that **2** could be converted by chemical means to jiadifenin (**1**) through the intermediacy of ketones **3** and **4**, themselves isolated from *Illicium majus*[2] (Scheme 1).

From a purely chemical perspective, the densely oxygenated and highly compact structure of jiadifenin (**1**) poses significant issues that invite propositions directed to its synthesis. Moreover, Fukuyama has shown that compound **1** promotes neurite outgrowth in the primary cultures of rat cortical neurons in concentrations as low as 0.1 μM.[1] Thus, the jiadifenins are appropriately classified as non-peptidyl neurotrophic factors.[3] Conceivably, the improved bioavailablity prospects of such compact non-peptidyl structures could be of value in the treatment of various neurodegenerative diseases.[3b] Recently, our laboratory has begun investigating this general field, starting with total syntheses as points of departure for broader explorations.[4] Herein, we report the first total synthesis of racemic jiadifenin (**1**), the establishment of a modality for its biological evaluation, and the discovery of somewhat more potent neurotrophic activity in fully synthetic compound **17** (vide infra).

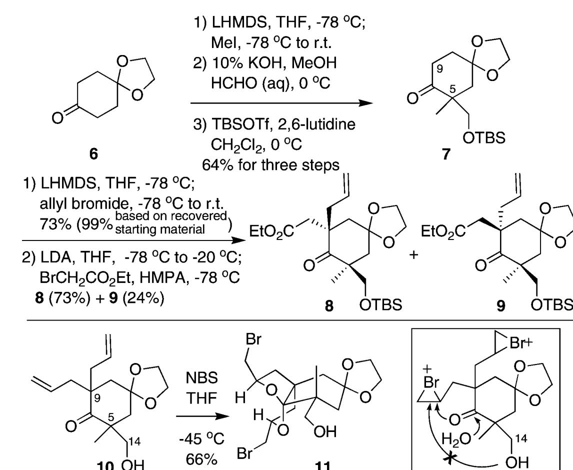
Given the reported convertibility of **4** to **1**, we envisioned a related oxidative ring contraction of a C(10) hydroxylated variant of **5** as the final stage of the synthesis of jiadifenin (Scheme 1). Preparation of **5** itself was to be accomplished through ring-closing operations of an α,α′-tetrasubstituted cyclohexanone, obtained from the commercially available 1,4-cyclohexanedione monoethylene ketal (**6**, Scheme 2).

Methylation of ketone **6**, followed by hydroxymethylation (under the thermodynamic conditions)[5] and protection of the resulting primary alcohol, produced **7** (Scheme 2). Anticipating difficulties in achieving full stereochemical control in the correlation of quaternary carbons 5 and 9, we explored desymmetrization of α,α′-diallycyclohexanone **10**. The hope was to distinguish the diastereotopically related allylic functions on the basis of their stereo relationships to the resident hydroxymethyl group at C(5). In the event, treatment of **10** as shown provided **11** in 66% yield and a mixture of other diastereomers in 24% yield.[6] Thus this formation of two tetrahydrofurans, via intervention of the formal "ketone hydrate", superseded participation of the primary alcohol at C(14) in the bromination event. While the exploitation of various internal structural implements in this series remains our long-term goal for stereocontrol, in the interim we turned to a program which starts with **7**. Sequential C(9) allylation and C(9) carboethoxymethylation afforded **8** and **9** in the yields and ratios shown.

Scheme 1



Scheme 2



Conversion of the ester moiety in **8** to a β-ketophosphonate, followed by intramolecular Horner–Wadsworth–Emmons reaction[7] and global deprotection, as shown, led to cyclopentenone **12** in 70% yield for three steps (Scheme 3). The use of a carbonate (see intermediate **12a**) as a one-carbon interpolation moiety served us well in obtaining diketolactone **13**.[8] Oxidation of the latter with *m*CPBA furnished the desired α-hydroxy product **13a** in 90% yield as a single isomer. Following stereoselective reduction, *trans*-diol **14** was in hand. The assigned relative configurations of the newly generated stereocenters were verified by single-crystal X-ray analysis of **14**.

Methylation of **14** via trianion formation, followed by a two-step oxidative cyclization, generated lactone **15** (Scheme 4). Stereoselective reduction of **15** under Luche conditions and C(10) hydroxyl incorporation via Davis's oxaziridine[9] afforded the α-hydroxy lactone **16**.[10] We hoped to accomplish simultaneous oxidation of the C(2) and C(10) hydroxyl groups, thereby prompting rearrangement of the α-ketolactone into the hydroxytetrahydrofurancarboxylate acetal moiety of jiadifenin (**1**). In practice, this

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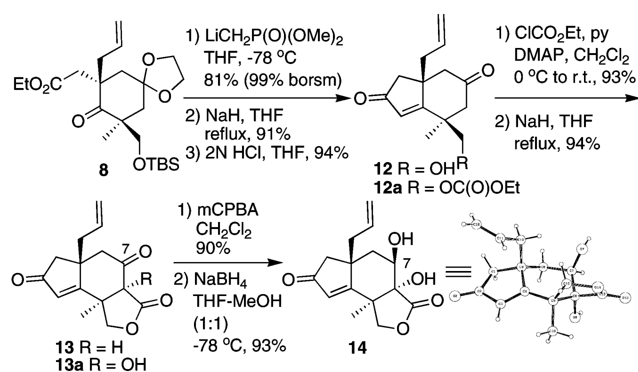
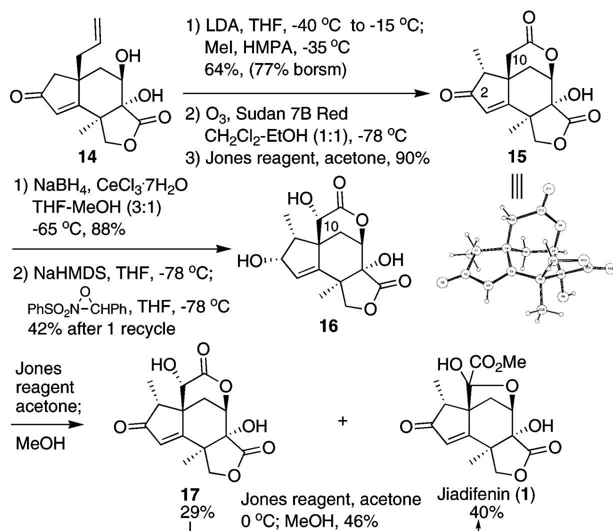
Scheme 3



Scheme 4



protocol led to isolation of a mixture of (1*R**,10*S**)-2-oxo-3,4-dehydroneomajucin (**17**) and jiadifenin (**1**). Following separation, **17** was further submitted to oxidative ring contraction to yield **1** in 46% yield after a prolonged reaction time. The overall consolidated yield for the conversion of **16** to jiadifenin was 53%. The spectroscopic data measured from fully synthetic **17** are in full accord with the published data of the compound, in tabular form.[2] Further confirmation came from the identity of the NMR spectra of synthetic jiadifenin (±)-**1** with spectra of natural jiadifenin, kindly provided by professor Fukuyama.[1] Thus, as a consequence of this interim, nonoptimized total synthesis (18 steps, current isolated yield 1.9%), jiadifenin, hitherto obtainable with only the greatest of difficulty, is now eminently available for biological investigation.

It was important to validate the claimed neurotrophic activity of fully synthetic **1**. This was accomplished by measuring the ability of **1** to stimulate NGF-mediated neurite outgrowth under the protocols provided below.[11] Thus, in our assay it is particularly clear that the effect of jiadifenin is that of upregulating the action of the NGF rather than functioning independently.[12] Remarkably, fully synthetic **17**, an intermediate en route to **1**, displays even stronger activity in this assay. Thus, neurite lengths enhanced by

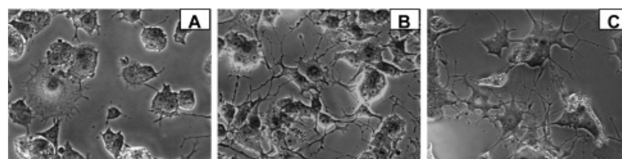


Figure 1. Images of neurons after treatment with (A) DMSO + NGF, (B) compound **17** in DMSO (0.3 μM) + NGF, and (C) compound **1** in DMSO (0.3 μM) + NGF.

17 and **1** were 184% ($P < 0.01$) and 162% ($P < 0.05$), respectively, relative to the DMSO−NGF control.

Given the encouraging results described above, research into this field continues in earnest in our laboratory at the level of chemical synthesis as well as biological follow-up.

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Supporting Information Available: Spectroscopic and analytical data for all intermediates, experimental procedures, and assay protocols. This material is available free of charge via the Internet at http://pubs.acs.org.

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(10) (a) Incorporation of the C(10) hydroxyl group was hampered by decomposition of both starting compound and product under the reaction conditions. Accordingly, it was necessary to cease the reaction well before the conclusion. We isolated the desired product **16** and the starting material in 26% and 73% yield, respectively. The recovered starting material was resubmitted to the reaction conditions to give **16** in 42% combined yield after one recycle. (b) A mixture of diastereomers was observed at C(10) of **16** (6:1 ratio, a major diastereomer as shown in **16**).
(11) Rat pheochromocytoma cells (PC12) were cultured in a 96-well collagen-coated plate in F-12K medium supplemented with 0.5% fetal calf serum and 50 ng/mL of NGF (2.5 S) with or without each compound at 0.3 μM in DMSO solution for 48 h. Fresh medium with the same supplements was placed on the cell for an additional 48 h. The cells were then fixed and examined by microscopy. From our PC-12 assay, the neurite outgrowth associated with neuronal differentiation was determined.
(12) In the absence of NGF, no neurite outgrowth was observed.

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