ORGANIC LETTERS

2006
Vol. 8, No. 24
5441−5443

Total Synthesis of (*S*)-Equol

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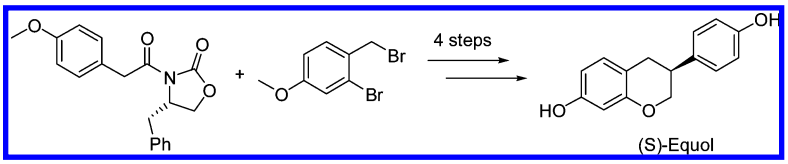
Received August 19, 2006

ABSTRACT



The first enantioselective total synthesis of (*S*)-equol is reported. The described route relies on an Evans alkylation to form the stereocenter and an intramolecular Buchwald etherification to generate the chroman ring. Key features of this method include its brevity, its scalability, and the low cost of starting materials.

Soy isoflavanoids such as daidzein and genistein (Figure 1) have recently attracted increased attention for their estrogenic activity, and their potential for use in menopausal hormone replacement therapy[1] and the treatment of breast cancer[2] has been suggested. However, daidzein can be metabolized in vivo to give (*S*)-equol (**1a**),[3] which has higher estrogenic activity than daidzein,[4] and may actually increase the proliferation of breast cancer cells.[5]

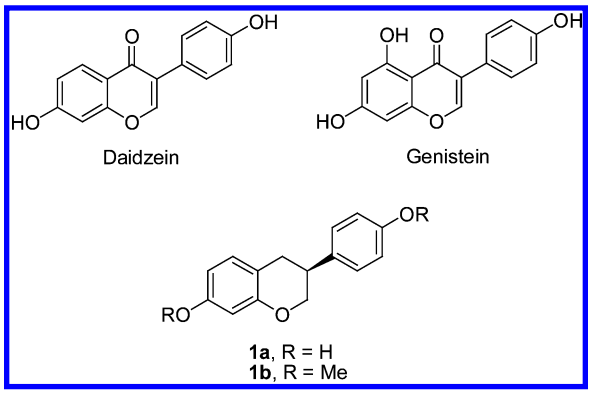


Figure 1. Examples of soy isoflavanoids and their metabolic products.

Equol possesses a single stereogenic center at the C3 position of the chroman ring. As anticipated, the (*R*)- and (*S*)-enantiomers have differing biological activities, with the naturally occurring (*S*)-enantiomer possessing greater activity toward estrogen receptor β (ERβ) and the (*R*)-enantiomer possessing greater activity toward estrogen receptor α (ERα).[6] Thus, to elucidate the precise biological effects of these metabolites, it is necessary to conduct studies using enantiopure material. (*S*)-Equol has been obtained previously via synthesis of the racemate from daidzein, followed by separation using chiral HPLC[6] and by bacterial metabolism of dihydrodaidzein.[7] Also, Ferreira and co-workers have

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10.1021/ol0620444 CCC: $33.50
Published on Web 11/02/2006

demonstrated the enantioselective synthesis of dimethoxy analogue **1b**;[8] however, the synthesis provided only small quantities of material and the nontrivial cleavage of the methyl ethers was not attempted. Here, we report a brief and cost-effective total synthesis of (*S*)-equol that can be conducted on a large scale to produce over 10 g of the final product from a single batch.

Retrosynthetic analysis (Figure 2) revealed that the chroman skeleton could be formed via intramolecular palladium-

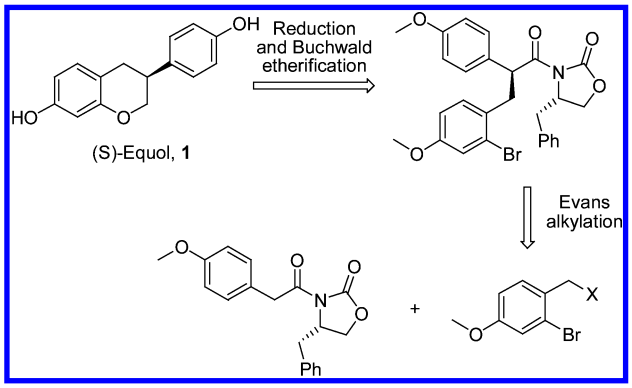
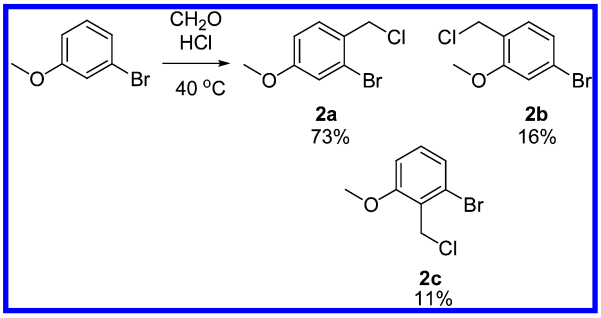


Figure 2. Retrosynthetic analysis of (*S*)-equol.

catalyzed Buchwald etherification, which is known to proceed smoothly even when deactivating electron-rich substituents are present on the aryl bromide.[9] To obtain the required chiral alcohol precursor, we envisioned an Evans alkylation[10] using 2-bromo-4-methoxybenzyl halide and a chiral *N*-acyloxazolidinone derived from 4-methoxyphenylacetic acid.

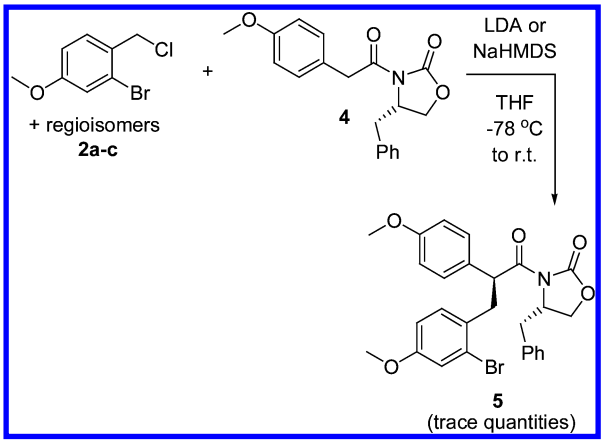
The synthesis of 2-bromo-4-methoxybenzyl bromide is known but requires five synthetic steps from *p*-nitrotoluene.[11] Alternatively, Lythgoe and co-workers reported the synthesis of 2-bromo-4-methoxybenzyl chloride (**2**) in one step via the chloromethylation of 3-bromoanisole.[12] Following the literature procedure, we reacted 3-bromoanisole with formaldehyde and hydrogen chloride gas, and the product we obtained after fractional distillation was characterized by a narrow boiling point in the gas chromatograph; however, the ^{1}H NMR spectrum revealed three different products, leading us to believe that we had in fact formed three regioisomers.[13] By analogy to the ^{1}H NMR shifts of similar chemical structures, we were able to assign the product distribution for the three isomers (**2a**−**c**) as shown in Scheme 1.[14]

Scheme 1. Chloromethylation of 3-Bromoanisole



Although we were hesitant to carry through material of such low purity, this option was still far more appealing than attempting the five-step synthesis of the benzyl bromide, provided that we would be able to remove products arising from the unwanted regioisomers in a subsequent synthetic step. We first attempted the reaction of benzyl chlorides **2a**−**c** with chiral *N*-acyloxazolidinone **4** using LDA as the base but observed very poor recovery of both the product and the starting material **4** (Scheme 2). Changing the base

Scheme 2. Initial Attempts of Evans Alkylation



from LDA to NaHMDS improved the yield slightly, but we were still lacking a method suitable for large-scale use. In the previous synthesis of dimethoxy equol, Ferreira and co-workers reported low (13−30%) yields when using an oxazolidinone as their chiral auxiliary and hypothesized that the good leaving group ability of the oxazolidinone results in ketene formation in the presence of base.[7] They instead used an ephedrine-derived imidazolidinone auxiliary, but due to the higher cost and increased number of synthetic steps required, we were motivated to find reaction conditions that would be conducive to use of the oxazolidinone.

We found that increasing the reactivity of the electrophile by converting the benzyl chlorides **2a**−**c** to benzyl bromides

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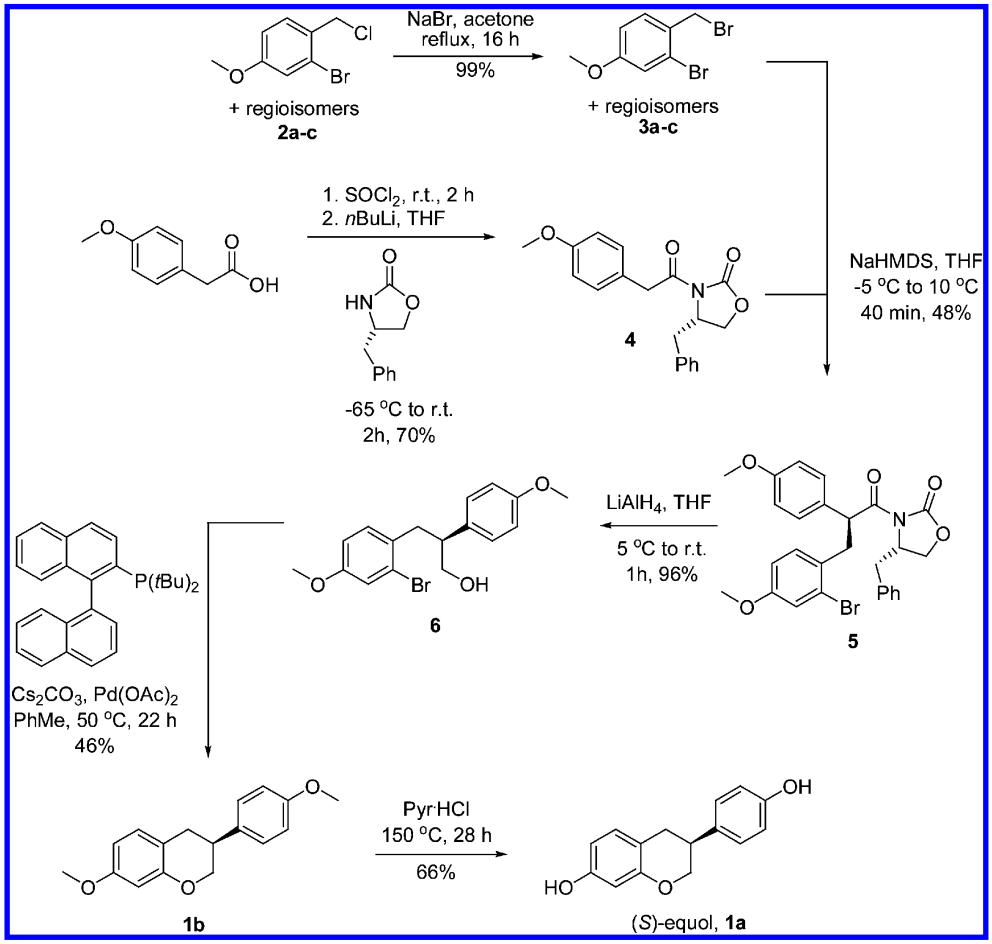
(13) The original publication dates back to 1956, prior to the availability of ^{1}H NMR. Thus, it is unclear whether the initially reported synthesis also yielded a mixture of three regioisomers that were unknowingly separated in a later step.

(14) The product distribution was calculated from the integrations of the benzyl protons of each of the regioisomers in the ^{1}H NMR spectrum. The chemical shifts of these protons in $CDCl_3$ are: d 4.84 (**2c**), 4.68 (**2a**), and 4.59 (**2b**).

Scheme 3. Synthesis of (*S*)-Equol



3a−**c** via Finkelstein reaction,[15] along with premixing the electrophile with **4** prior to addition of the base, enabled the reaction to proceed much more cleanly (Scheme 3). The resulting yield for the alkylation reaction is admittedly modest, yet acceptable considering that the theoretical yield of the correct isomer is only 73%. The presence of multiple regioisomers in the crude product mixture prohibited analysis of the diastereoselectivity of the reaction, but after recrystallization of **5**, we observed no evidence of unwanted regioisomers or diastereomers in the ^{1}H NMR spectrum.

To complete the synthesis, we first cleaved the chiral auxiliary using $LiAlH_4$, providing **6** as the desired substrate for the Buchwald etherification reaction. Reaction of **6** with $Pd(OAc)_2$ in the presence of di-*tert*-butyl-binaphthylphosphine yielded dimethoxy equol **1b**. As previously mentioned, cleavage of the methyl ether protecting groups is not trivial, as overly forcing conditions will result in opening of the chroman ring. Previous work had shown, however, that pyridine hydrochloride provides the desired balance of reactivity,[16] and upon reaction with **1b**, we indeed were able to obtain (*S*)-equol **1a**.

From a single reaction batch, we were able to obtain over 10 g of (*S*)-equol in an overall yield of 9.8%. Chiral LC/MS analysis revealed a chemical purity of >99% and an enantiomeric ratio of >99.9:0.1, with no detection of the (*R*)-enantiomer.[17] The enantioselectivity of this route is higher than that obtained in the previous partial synthesis,[7] and to the best of our knowledge, the quantity of material produced is greater than the current world's supply of isolated (*S*)-equol.

In summary, this report describes the first total synthesis of (*S*)-equol, utilizing a route that is brief, cost-effective, and scalable. Future work will focus on optimizing yields and selectivity through screening of both reagents and reaction conditions.

Acknowledgment. The authors kindly thank Prof. John A. Katzenellenbogen of the University of Illinois, Urbana−Champaign, for his advice toward developing the synthetic route.

Supporting Information Available: Details of all experimental procedures, characterization data, and NMR spectra for all new compounds. This material is available free of charge via the Internet at http://pubs.acs.org.

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(17) The sample was analyzed using the procedures described in ref 6.