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Synthesis of procyanidins by stepwise- and self-condensation using 3,4-*cis*-4-acetoxy-3-*O*-acetyl-4-dehydro-5,7,3′,4′-tetra-*O*-benzyl-(+)-catechin and (−)-epicatechin as a key building monomer

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Abstract

3,4-*cis*-4-Acetoxy-3-*O*-acetyl-4-dehydro-5,7,3′,4′-tetra-*O*-benzyl-(+)-catechin (**1a**) or (−)-epicatechin (**1b**) reacted high regio- and stereo-selectively with 1.5 equiv of the 5,7,3′,4′-tetra-*O*-benzyloxyflavan-3-ol (**4a** or **4b**) in the presence of 1 equiv of TMSOTf to give the corresponding procyanidins. On the other hand, the self-condensation of **1a** in the presence of a catalytic amount of $B(C_6F_5)_3$ afforded wide-range procyanidins from dimer to 15-mer like a biomass.

Keywords: Procyanidin; Stepwise- and self-condensation; 4-Acetoxy-3-acetylcatechin; 4-Acetoxy-3-acetylepicatechin

Procyanidins (condensed tannins) are widely distributed in food such as fruits, beans, cocoa, and tea and show multiple biological activities such as antioxidant, anti-inflammatory, anti-atherosclerotic, and anti-allergy effects.[1–3] Thus, diets containing procyanidins are important for maintaining and improving health. However, naturally occurring procyanidins are composed of complex mixtures from dimer to about 15-mer with various linkages and stereochemical differences.[2–4] Furthermore, procyanidins are highly sensitive to air,[1] thus, isolation of each component is difficult. To investigate the biological activities and medicinal utilities of procyanidins, a synthetic supply of the desired procyanidin oligmers is required. For this purpose, development of controlled regio- and stereo-selective oligomerization is essential.

There are two strategies on the synthetic approach of procyanidin oligomers. One is stepwise-condensation, which is able to synthesize defined oligomers with an unambiguous stereo- and regiochemistry.[5] The other is self-condensation, which produces at once biomass-mimetic materials having various oligomers.[6] We designed a highly reactive and relatively stable monomer of 3,4-*cis*-4-acetoxy-3-*O*-acetyl-(+)-catechin and (−)-epicatechin as a donor. Here, we report the synthesis of various 4,8-linked procyanidins by using two kinds of Lewis acids, TMSOTf and $B(C_6F_5)_3$ as catalysts.

After Kawamoto's report,[5a,6] many condensation conditions for procyanidin oligomers using catechin derivatives and Lewis acids have been studied.[5,6] However, there were several synthetic problems, such as the reactive efficiency and the oligomerization control. From the previous reports,[5–7] the reactivity and the stereoselectivity might depend on the leaving group of the C4-position and the protecting group of C3–OH (Fig. 1). Recently, Suzuki et al. reported that the C4-acetoxy derivatives might be excellent candidates as building blocks for procyanidin synthesis.[7,8] Furthermore, the protecting group of C3–OH influences the stereoselectivity as well as the reactivity

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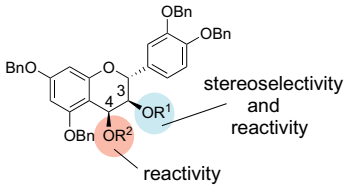


1a: R^1 = Ac, R^2 = Ac
2: R^1 = TBS, R^2 = Ac

Fig. 1.

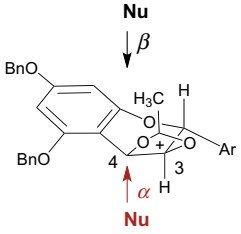


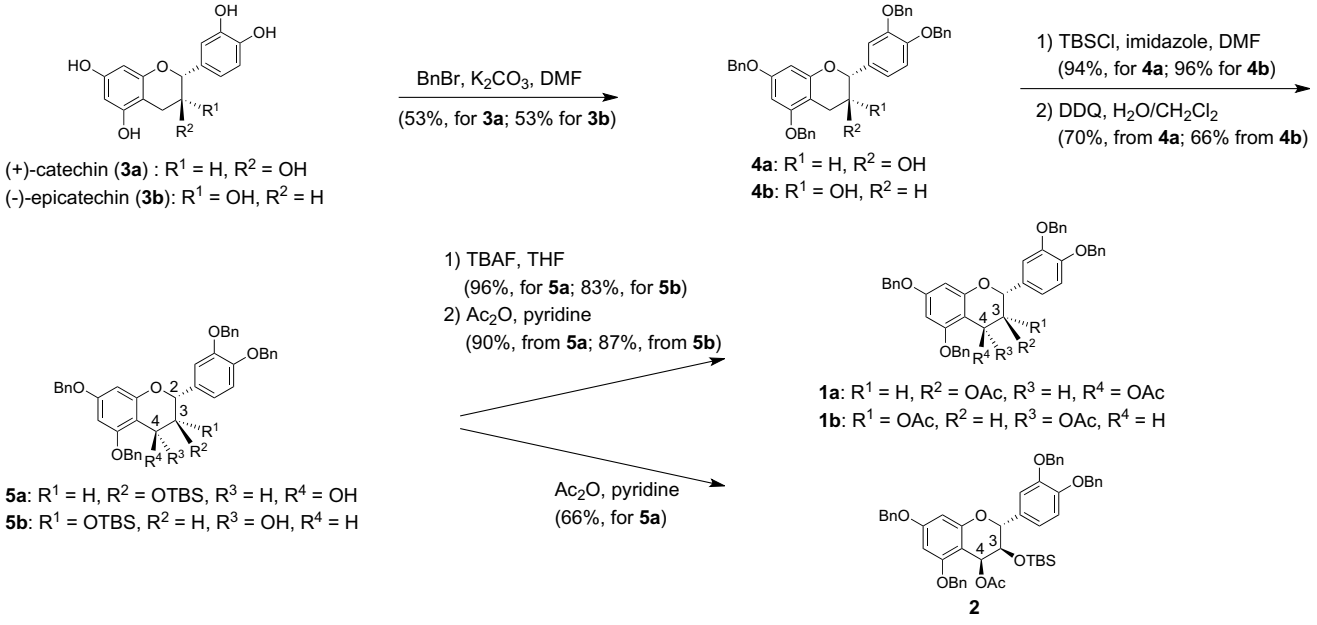
Fig. 2.

due to the steric factor and the neighboring participation-effect like the case of glycosylation (Fig. 2).[5–7] Considering these experimental observations we designed two monomers, 3-*O*-acetyl- and 3-*O*-TBS-, 4-acetoxy-perbenzylcatechin, **1a** and **2**.

5,7,3′,4′-Tetra-*O*-benzylcatechin (**4a**) was prepared from (+)-catechin (**3a**) (53%) according to Kawamoto's procedure.[6a,9] **4a** was then protected with TBSCl-imidazole (94%), and oxidized with DDQ in a suspension of CH_2Cl_2 and H_2O to give the 4-hydroxy **5a** in 70% yield (Scheme 1). The configuration of **5a** was determined to be 3,4-*cis* by NMR analysis.[10] **5a** was acetylated with Ac_2O in pyridine to give the 4-acetoxy-3-*O*-TBS derivative **2** in 66% yield. The TBS-group of **5a** was deprotected with TBAF (96%), then, the obtained product was acetylated with Ac_2O in pyridine to give the 3,4-*cis*-4-acetoxy-3-*O*-acetyl-4-dehydro-5,7,3′,4′-tetra-*O*-benzyl-(+)-catechin (**1a**)[11] in 90% yield (Scheme 1).

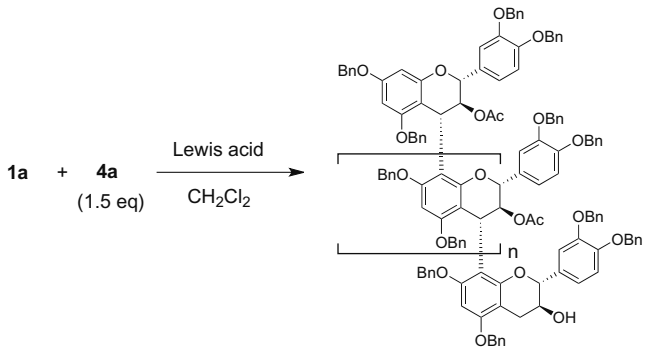
Condensation between **2** and **4a** was performed in the presence of $BF_3{\cdot}Et_2O$ to give only a trace amount of the partially desilylated dimers.[12] The low reactivity may be due to a large steric-hindrance of TBS at 3-OH of **2**. Instead of 3-*O*-TBS compound **2**, 3,4-*O*-diacetoxy **1a** was reacted with **4a** in the presence of $BF_3{\cdot}Et_2O$. Compound **1a** and **4a** smoothly condensed at 0 °C within 5 min to give oligomers, dimer **6** (57%), trimer **7** (20%), and tetramer **8** (3%) (Table 1, entry 1).[13] Concerning the configuration of 4,8-linkage of the oligomers, all of them were determined to be *trans* to 3*C*-OAc by $J_{3,4} = 9$–10 Hz.[13] Various Lewis acids were examined as promoters for the condensation reaction of **1a** and **4a** in CH_2Cl_2 (Table 1). Surprisingly, with only 1 equiv of TMSOTf at −78 °C for 5 min, the dimer was yielded up to 82% with high stereoselectivity (α/β 97/3)[14] (Table 1, entry 2).[15] The stereoselectivity and reactivity might arise from the neighboring participation-effect at 3-*O*-acetylate.[5f] After deacetylation of **6** (97%), careful hydrogenolysis by $H_2/Pd(OH)_2$ was carried out to give procyanidin B3 (**9**) in 91% yield (Scheme 2).[16]

For structural determination of procyanidins isolated from seed coats of red adzuki bean, *Vigna angularis*,[3] we synthesized a trimer by using **1b** and **4a** in the presence of TMSOTf. The 4-acetoxyepicatechin derivative **1b** was synthesized from (−)-epicatechin (**3b**) according to the same procedure as **1a** (Scheme 1).[10] The condensation between **1b** and **4a** in the presence of TMSOTf in CH_2Cl_2 for 5 min at −78 °C gave dimer **10** in 70% yield with a small



Scheme 1.

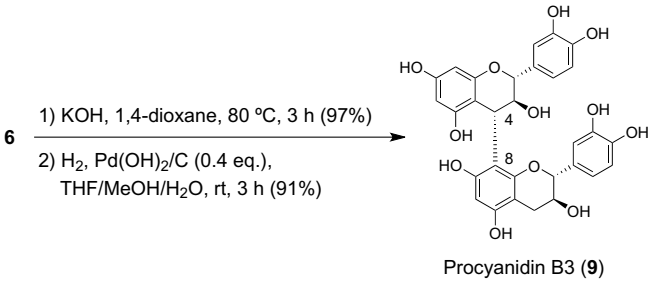
Table 1



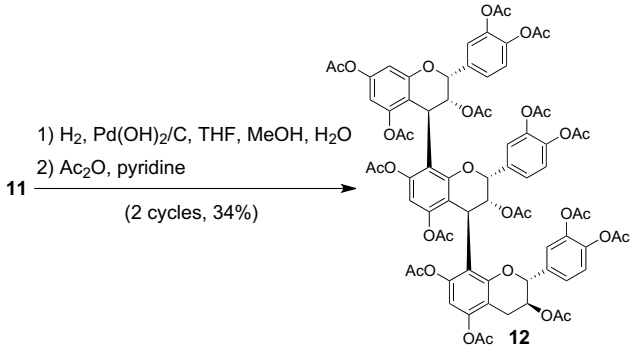
Entry	Lewis acid (equiv)	Temp (°C)	Time	Yield[a] (%) Dimer **6** ($n = 0$)	Trimer **7** ($n = 1$)	Tetramer **8** ($n = 3$)
1	$BF{\cdot}Et_2O$ (1)	0	5 min	57	20	3
2	TMSOTf (1)	−78	5 min	82[b]	10	Trace
3	TMSOTf (0.2)	−78	1 h	49	33	10
4	$Sc(OTf)_3$ (0.1)	0 to rt	1 h	49	20	4
5	$Sc(OTf)_3$ (0.1)	rt	30 min	61	22	4
6	$Yb(OTf)_3$ (1)	0 to rt	1 h	45	20	Trace
7	$In(OTf)_3$ (0.1)	0 to rt	5 h	44	19	2
8	$La(OTf)_3$ (1)	0 to rt	21 h	40	24	Trace
9	Cp_2HfCl_2 (1)	0 to rt	1 h	Trace	0	0
10	$B(C_6F_5)_3$ (0.1)	0	3.5 h	60	23	4

[a] A stereoselectivity was not determined without entry 2, dimer 6.
[b] A stereoselectivity ($\alpha/\beta = 97/3$) was determined by ^1H NMR after acetylation.

amount of higher molecular oligomers (Scheme 3). Further condensation of **1b** and **10** in the same condition was carried out to afford trimer **11** in 70% yield (Scheme 3). Finally, transformation of all protecting groups of **11** by



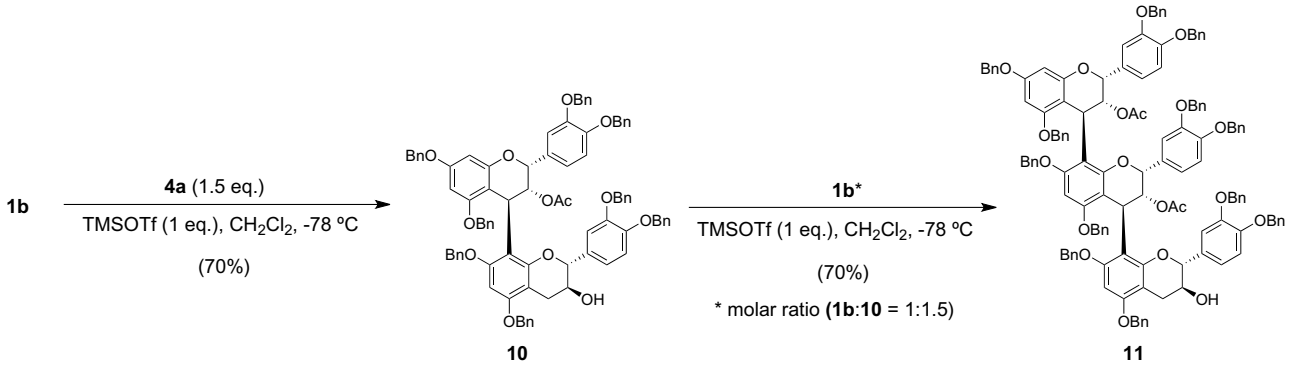
Scheme 2.



Scheme 4.

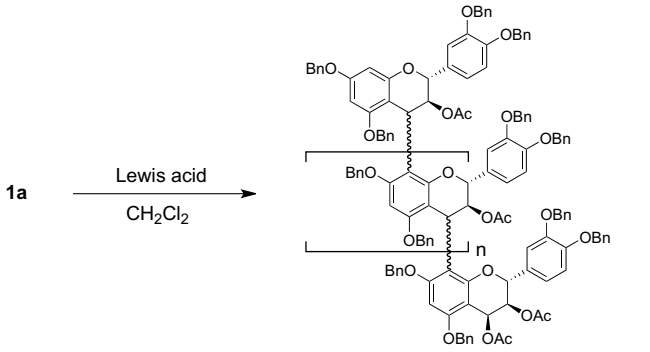
combination of hydrogenolysis and acetylation repeatedly gave acetate **12** (EC-(4β→8)-EC-(4 β→8)-C) (Scheme 4).[5j,17] By spectroscope (^1H NMR and ESI-Q-TOF MS) the properties of synthetic acetate (trimer) **12** were found to be identical to the acetate of the natural procyanidin isolated from red adzuki bean.[18] Thus, the trimer of azuki procyanidin is determined to be EC-(4β→8)-EC-(4β→8)-C.[19]

When $B(C_6F_5)_3$ or $Sc(OTf)_3$ was used as a promoter, the condensation reaction was proceeded with only a catalytic amount of Lewis acid (0.1 equiv) with higher trimer yield compared with that of TMSOTf (Table 1, entries 4 and 10). These findings prompted us to investigate the polymerization reaction of **1a** (Table 2). By using 10 mol % of $B(C_6F_5)_3$, **1a** was self-condensed to give a mixture of procyanidin oligomers for 5 h at 0 °C (93% yield).



Scheme 3.

Table 2



Entry	Lewis acid (mol %)	Temp (°C)	Time	Yield[a] (%)	Max DP[b]
1	TMSOTf (100)	−78	5 min	>89	12
2	$B(C_6F_5)_3$ (10)	0	5 h	>93	15 (7.4)[c]
3	$B(C_6F_5)_3$ (1)	rt	24 h	>85	11 (6.4)[c]
4	$Sc(OTf)_3$ (10)	0	5 h	>82	15

[a] The product was a mixture of procyanidin oligomers.
[b] Max DP was estimated by MALDI-TOF-MS using 2,5-dihydroxybenzoic acid as matrix.
[c] Parentheses indicate the DPn which was estimated by size-exclusion chromatography (SEC, polystyrene standards, $CHCl_3$).

The maximum degree of polymerization (max DP) detected by MALDI-TOF-MS was 15 and the number average degree of polymerization (DP_n) measured by SEC was 7.4 (Table 2, entry 2).[20] $Sc(OTf)_3$ also gave a similar result.

In summary, we designed and prepared the 4-acetoxy-3-acetylcatechin derivatives (**1a**,**b**) as a monomer unit for procyanidin synthesis. **1a**,**b** showed a high efficiency in both step-wise condensation with TMSOTf and self-condensation with $B(C_6F_5)_3$ to give procyanidin oligomers. Furthermore, the structure of a novel procyanidin-trimer from red adzuki bean, *Vigna angularis*, was clarified using the synthesized authentic samples. The synthesis of various procyanidin oligomers according to the above-mentioned method and the structural identification of polyphenols from natural source are in progress.

Acknowledgments

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Supplementary data

Copies of ^1H NMR spectra of **12** and the acetate of the natural sample from red adzuki bean. Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.tetlet.2008.02.173.

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8. The details of the substitution reaction of flavan skeleton were reported by Suzuki et al. They described that the stereochemistry of C4-position has no influence on the stereoselectivity and concluded that this reaction is S_N1-like reaction.
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10. Compound **5a** ($J_{2,3} = 9.3$ Hz, $J_{3,4} = 5.5$ Hz), **5b** ($J_{2,3} = 2.4$ Hz, $J_{3,4} = 1.4$ Hz).
11. Compound **1a** was already reported by Suzuki et al., but they carried out only a model study (one example) in the presence of $BF_3{\cdot}Et_2O$.[7b]
12. 1.5 equiv of **4a** to **2** was used. The reaction temperature was raised gradually from −78 °C to −10 °C. The products were decomposed at rt in the reaction mixture.
13. After acetylation, the structural identification was performed by various 1D and 2D (COSY, NOESY, HMQC, and HMBC) NMR experiments.

14. The stereoselectivity was determined using acetylated compound by ^{1}H NMR according to Saito's protocol.[5f,g]
15. Synthesis of procyanidin B using one equimolar amount of the nucleophile was recently reported, but the yield was not so good.[5k]
16. A long reaction time with a large amount of Pd catalyst lowered the yield because of cleavage of interflavan bonds.
17. To prevent cleavage of interflavan bonds, partial hydrogenolysis followed by acetylation was conducted repeatedly.
18. See Supplementary data.
19. This is the first report on isolation and structural determination of procyanidin-trimer found in red adzuki beans (*Vigna angularis*).[3]
20. Typical experimental procedure for the self-condensation: To a solution of **1a** (50 μmmol, 37.5 mg) in CH_2Cl_2 was added $B(C_5F_6)_3$ (5 μmmol, 2.6 mg) at 0 °C. After being stirred for 5 h, the reaction mixture was quenched by the addition of saturated aqueous $NaHCO_3$ and extracted with AcOEt. The combined extracts were dried over anhydrous $MgSO_4$ and the solvent was evaporated to yield condensed product (>93%, 35 mg).[6b]