Hyperbranched 5,6-glucan as reducing sugar ball

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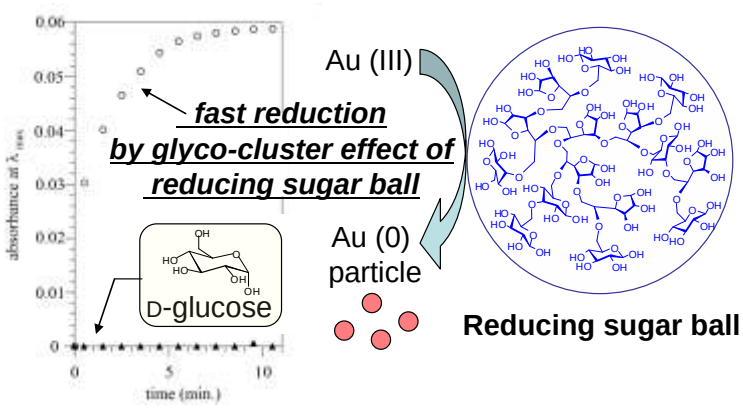
Graphical contents entry



A hyperbranched glycopolymer arranged with numerous reducing D-glucose units on the peripheries of the polymer, *i.e.*, "reducing sugar ball", possessed a higher reducing ability than D-glucose because of the glyco-cluster effect or the multivalent effect of the D-glucose units.

Abstract. The ring-opening polymerization of 5,6-anhydro-1,2-*O*-isopropylidene-α-D-glucofuranose (**1**) as a latent cyclic AB_2-type monomer was carried out using potassium *tert*-butoxide (*t*-BuOK) or boron trifluoride diethyletherate ($BF_3 \cdot OEt_2$) as an initiator in order to synthesize a novel hyperbranched glycopolymer. The anionic and cationic polymerizations proceeded via the proton-transfer reaction mechanism to produce the hyperbranched poly(5,6-anhydro-1,2-*O*-isopropylidene-α-D-glucofuranose) (**2**). In particular, the cationic polymerization with the slow-monomer-addition strategy is a facile method leading to the hyperbranched glycopolymers with high molecular weights and highly branched structures. The weight-average molecular weight ($M_{w,SEC\text{-}MALLS}$) values of **2** measured by multi-angle laser light scattering (MALLS) varied in the range from 7,400 to 122,400, which were significantly higher than the weight-average molecular weight ($M_{w,SEC}$) values determined by size exclusion chromatography (SEC). The intrinsic viscosities ([η]) of these polymers were very low in the range of 3.3-4.6 $mL \cdot g^{-1}$ and the Mark-Houwink-Sakurada exponents α were calculated to be 0.08-0.27. These results of the MALLS, SEC, and viscosity measurements suggested that these polymers exist in a compact spherical conformation in solution because of their highly branched structure. The synthesis of the hyperbranched 5,6-glucan (**3**) by hydrolysis of polymer **2** was also demonstrated. Polymer **3** is a novel water-soluble hyperbranched glycopolymer arranged with numerous reducing D-glucose units on the peripheries of the polymer, and has a higher reducing ability than D-glucose because of the glyco-cluster effect or the multivalent effect of the reducing D-glucose units. Therefore, polymer **3** should be called a "reducing sugar ball".

Introduction

The spherical architectures of highly branched macromolecules, such as dendrimers, star-shaped polymers, and hyperbranched polymers, have attracted much attention from the viewpoint of nanotechnology, because their numerous terminal units can be converted into various functional groups leading to novel nanomaterials.[1-32] Thus, various types of dendrimers, star polymers, and hyperbranched polymers have been synthesized and their properties were compared to the linear analogues. [23,33-46]

Spherical macromolecular architectures with their surfaces covered with sugars are of increasing interest due to their unique structures along with specific properties, such as the glycol-cluster or multivalent effect on the carbohydrate-protein interactions. For a spherical polymer with many terminal sugar residues, there are a few types of glycodendrimers, e.g., dendrimers prepared from the reaction of the dendrimer surface with mono- and disaccharides (sugar ball),[47-59] and dendrimers prepared through the convergent method using semi-dendritic branched oligosaccharides.[60-64] Although the synthesis of such glycodendrimer types was accomplished, dendrimers essentially consisting of sugar units, i.e., dendritic carbohydrate polymers, have not been prepared until now due to the complicated and time-consuming synthetic process involving numerous protection, deprotection, and purification procedures. Star-shaped glycopolymers were also prepared using the "core-first" method based on the RAFT polymerization[65,66] and the "arm-first" method based on the TEMPO-mediated radical polymerization.[67-70] Hyperbranched glycopolymers from the polymerization of AB_m-type monomers can be prepared more easily than glycodendrimers and star-shaped glycopolymers.[71-76] Recently, we proposed that the ring-opening multibranching polymerization of anhydro and dianhydro sugars as latent AB_m-type monomers should be a convenient method of synthesizing hyperbranched glycopolymers with a novel spherical macromolecular architecture,[77-83] for example, the hyperbranched polysaccharides from the cationic polymerization of 1,6-anhydro-β-D-hexopyranose, the hyperbranched poly(2,5-anhydro-D-glucitol) from 1,2:5,6-dianhydro-D-mannitol, and the hyperbranched polytetritols of 1,4-anhydrotetritol and 2,3-anhydrotetritol. For our synthetic strategy of hyperbranched glycopolymers, it is important to elucidate the effect of the structure of the sugar monomers on the ring-opening multibranching polymerization tendency and to clarify the relationship between the structure and properties of the hyperbranched glycopolymers.

In this article, we report the ring-opening multibranching polymerization of 5,6-anhydro-1,2-*O*-isopropylidene-α-D-glucofuranose (**1**) as a latent cyclic AB_2-type monomer using anionic or cationic initiators, as shown in Fig. 1, leading to the hyperbranched poly(5,6-anhydro-1,2-*O*-isopropylidene-α-D-glucofuranose) (**2**). In fact, Uryu et al. previously reported pioneering research on the ring-opening

polymerization of **1**,[84,85] though the polymer structure was insufficiently characterized as well as the solution properties. Therefore, the characteristics of the polymerization, the polymer structure, and the solution characteristics were discussed by the NMR measurements and the size exclusion chromatography (SEC) with a multi-angle laser light scattering (MALLS) detector and a viscosity detector. In addition, the synthesis of a hyperbranched 5,6-glucan (**3**) by hydrolysis of polymer **2** was demonstrated. Since polymer **3** is a novel water-soluble hyperbranched glycopolymer arranged with numerous reducing D-glucose units on the peripheries of the polymer, the reduction property of **3** is discussed from the viewpoint of the physical property as a "reducing sugar ball".

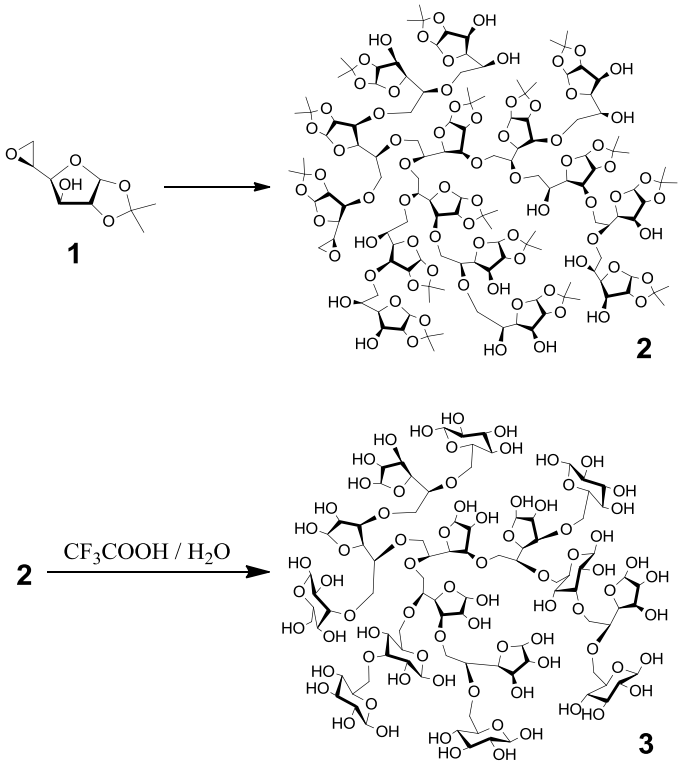


Fig. 1. Polymerization of 5,6-Anhydro-1,2-*O*-isopropylidene-α-D-glucofuranose (**1**) and synthesis of hyperbranched 5,6-glucan (**3**) by hydrolysis of polymer **2**.

Experimental

Materials. 5,6-Anhydro-1,2-*O*-isopropylidene-α-D-glucofuranose (**1**) was synthesized from 1,2-*O*-isopropylidene-α-D-glucofuranose using a procedure similar to that reported by Yus et al.[86] **1** was purified by column chromatography on silica gel with *n*-hexane/ethyl acetate (3/2, v/v, R_f = 0.15) and dried in a vacuum oven at 50 °C for 2 days. ^{1}H NMR ($CDCl_3$):δ (ppm) 1.32 and 1.48 (s, 6H, $2CH_3$), 2.88 and 3.00 (m, 2H, 2H-6), 3.11 (m, 1H, OH), 3.43 (m, 1H, H-5), 4.07 (m, 1H, H-4), 4.26 (m, 1H, H-3), 4.52 (m, 1H, H-2), 5.99 (d, 1H, H-1). ^{13}C NMR ($CDCl_3$):δ (ppm) 26.1 and 26.7 (2C, $2CH_3$), 45.9 (C-6), 50.2 (C-5), 75.1 (C-3), 79.2 (C-4), 85.0 (C-2), 104.9 (C-1), 111.8 (C). Potassium *tert*-butoxide (*t*-BuOK)(1.0 mol·L^{-1} in THF) was obtained from the Sigma-Aldrich Co., and used as received. Boron trifluoride diethyletherate (BF_3·OEt_2) was purchased from Kanto Chemical Co., Ltd. (Tokyo, Japan) and distilled over CaH_2 under reduced pressure. Silica gel 60N (Kanto Chemical Co., Ltd.; spherical shape; particle size, 40-50 μm; neutral) was used for the column chromatography. Methyl iodide (>99.5%), silver oxide (>99.0%), an ammonia aqueous solution (28.0 - 30.0 %), tetrachloroauric (III) acid ($HAuCl_4$) tetrahydrate (>99.0%), trifluoroacetic acid (>99.0%), dry dichloromethane (dry CH_2Cl_2) (>99.5%; water content, <0.001%), dry tetrahydrofuran (THF) (>99.5%; water content, <0.001%), dry acetonitrile (CH_3CN) (>99.5%; water content, <0.001%), methanol (>99.5%), acetone (>99.0%), *n*-hexane (>95.0%), ethyl acetate (>99.5%), dimethylformamide (DMF) (>99.5%), and chloroform ($CHCl_3$) (>99.0%) were obtained from Kanto Chemical Co., Ltd., and used without further purification.

Measurements. The spectra of ^{1}H (400 MHz), ^{13}C NMR (100 MHz), distortionless enhancement by polarization transfer (DEPT), and ^{13}C-^{1}H correlation spectroscopy (COSY) were recorded using a JEOL JNM-A400II instrument. The molecular weight values of polymers **2**, **4** and **6** were determined by size exclusion chromatography (SEC) in THF (1.0 mL·min^{-1}) at 40 °C using an Agilent 1100 series instrumentation equipped with two Shodex KF-804L columns (linear, 8.0 mm × 300 mm; exclusion limit, 4 × 10^5; bead size, 7 μm), a DAWN 8 multi-angle laser light scattering (MALLS) detector (Wyatt Technology, Santa Barbara, CA), a Viscostar viscosity detector (Wyatt Technology), and an Optilab rEX

refractive index detector (Wyatt Technology). The absolute molecular weights ($M_{w,SEC\text{-}MALLS}$), polydispersity ($M_{w,SEC\text{-}MALLS}/M_{n,SEC\text{-}MALLS}$), intrinsic viscosity ([η]), refractive index change ($dn \cdot dc^{-1}$), and Mark-Houwink-Sakurada constants, α and K, were estimated by ASTRA 5.1.6.0 software (Wyatt Technology). The relative molecular weight ($M_{w,SEC}$) and the polydispersity ($M_{w,SEC}$ / $M_{n,SEC}$) in THF were calculated on the basis of a polystyrene calibration. The molecular weight values of polymer **3** were determined by size exclusion chromatography (SEC) in a 0.2 $mol \cdot L^{-1}$ $NaNO_3$ aqueous solution (1.0 $mL \cdot min^{-1}$) at 40 °C using an Agilent 1100 series instrumentation equipped with two Tosoh TSKgel $GMPW_{XL}$ columns (linear, 7.8 mm × 300 mm; exclusion limit, 5×10^7; bead size, 13 μm), a DAWN 8 multi-angle laser light scattering (MALLS) detector (Wyatt Technology), a Viscostar viscosity detector (Wyatt Technology), and an Optilab rEX refractive index detector (Wyatt Technology). The $M_{w,SEC\text{-}MALLS}$, $M_{w,SEC\text{-}MALLS}/M_{n,SEC\text{-}MALLS}$, [η], $dn \cdot dc^{-1}$, and Mark-Houwink-Sakurada constants, α and K, were estimated by ASTRA 5.1.6.0 software (Wyatt Technology). Preparative SEC for the chloroform-soluble polymers was performed in chloroform (3.5 $mL \cdot min^{-1}$) at 23 °C using a JAI LC-9201 (Japan Analytical Industry Co., Ltd., Japan) equipped with a JAI JAIGEL-3H column (20 mm x 600 mm; exclusion limit, 7×10^4) and a JAI RI-50s refractive index detector. The preparative SEC for the water-soluble polymers was performed in water (14 $mL \cdot min^{-1}$) at 23 °C using a JAI LC-928 (Japan Analytical Industry Co., Ltd., Japan) equipped with a JAI JAIGEL-W253-40 column (40 mm x 500 mm; exclusion limit, 5×10^4) and a JAI RI-50s refractive index detector. The elemental analysis was performed using a MICRO CORDER JM10 and Dionex DX-500 (Instrumental Analysis Division, Management Center, Creative Research Instaction Sousei, Hokkaido University). The measurement of the optical rotations was performed in $CHCl_3$ or H_2O using a Jasco DIP-1000 digital polarimeter (λ = 589 nm). The FI-MASS spectroscopy measurement was performed using a JEOL JMS-SX102A mass spectrometer (GC-MS & NMR Laboratory, Graduate School of Agriculture, Hokkaido University). The UV-vis spectra were measured at 70 °C with a 10-mm path length using a JASCO V-550 spectrometer with a deuterium lamp as the light source for the UV range (190-350 nm) and a halogen lamp for the visible range (330-900 nm). The dynamic light scattering (DLS) measurement of the obtained Au particles was performed at 25 °C using

an Otsuka Electronics FDLS-3000 light scattering spectrophotometer equipped with a solid state laser (λ = 532 nm, scattering angle = 90°). Before the measurement, the samples were filtered using 0.45 μm PTFE membrane filters to eliminate any dust particles. The data analysis was carried out using the histogram methods including the Marquadt analysis.

Anionic Polymerization of 1. All the anionic polymerizations of **1** were carried out using a storage flask with a needle valve under an argon atmosphere. A typical polymerization procedure (entry 2 in Table 1) is as follows: 1.0 mol·L^{-1} *t*-BuOK in a THF solution (0.50 mL, 0.50 mmol, [**1**]/[*t*-BuOK] = 20) was added to a solution of the monomer **1** (2.02 g, 10.0 mmol) in dry THF (4.50 mL, [**1**] = 2.0 mol·L^{-1}) at 50 °C using a microsyringe. After 600 h, the reaction mixture was poured into a large amount of methanol, then the solution was neutralized with dry ice. After evaporating the solvent, the residue was purified by reprecipitation from methanol-water to yield the crude polymer. The precipitate was redissolved in $CHCl_3$ (15 mL) and applied to the preparative SEC to remove the unreacted monomer **1**. The concentrated polymer solution was dried *in vacuo* to give a white solid polymer **2** in a 68.4 % yield (1.38 g); $M_{w,SEC}$ (THF) = 1,400; $M_{w,SEC}$ / $M_{n,SEC}$ (THF) = 1.28; $M_{w,SEC\text{-}MALLS}$ (THF) = 7,400; $M_{w,\ SEC\text{-}MALLS}$ / $M_{n,\ SEC\text{-}MALLS}$ (THF) = 1.37; dn·dc^{-1} (THF) = 0.080 mL·g^{-1}; $[\alpha]_D$ = -23.1° (*c* 1.00, $CHCl_3$, 24 °C). ^{1}H NMR (CD_2Cl_2):δ 5.87 (br.s, H-1), 4.80 -3.20 (m, including peaks at 4.59, 4.48, 4.26, 4.01, 3.73, 3.60, and 3.47 ppm), 3.06 (br.s), 2.93-2.70 (m, epoxy group), 1.44 (s, =C(CH_3)$_2$) and 1.28 ppm (s, =C(CH_3)$_2$). ^{13}C NMR (CD_2Cl_2):δ 112.14 (=*C*(CH$_3$)$_2$), 105.74 (C-1), 105.44 (C-1), 85.55, 84.20, 82.31, 80.46, 74.65, 73.45, 68.60, 48.49 (epoxy group), 46.98 (epoxy group), 26.96 (=C(*C*H$_3$)$_2$), and 26.35 ppm (=C(*C*H$_3$)$_2$). It is hard to estimate the degree of branching value because the NMR peaks due to each branch structure were broadened and overlapped.

Cationic Polymerization of 1. All procedures were performed using a Schlenk flask under an argon atmosphere. A typical procedure for the cationic polymerization (entry 4 in Table 1) is as follows: BF_3·OEt_2 (8.0 μL, 62.5 μmol, [**1**]/[BF_3·OEt_2] = 40) was added to a solution of **1** (0.505 g, 2.50 mmol) in dry CH_2Cl_2 (5.0 mL, [**1**] = 0.5 mol·L^{-1}) at 10 °C using a microsyringe. After 24 h, the polymerization

was terminated by adding methanol containing a small portion of an aqueous ammonia solution. After evaporating the solvent, the residue was purified by reprecipitation from methanol-water to yield the crude polymer. The precipitate was redissolved in $CHCl_3$ (4 mL) and applied to the preparative SEC to remove the unreacted monomer **1**. The concentrated polymer solution was dried *in vacuo* to give a white solid polymer **2** in a 68.5 % yield (346 mg); $M_{w,SEC}$ (THF) = 4,100; $M_{w,SEC}/M_{n,SEC}$ (THF) = 1.29; $M_{w,SEC\text{-}MALLS}$ (THF) = 15,000; $M_{w,\ SEC\text{-}MALLS}/M_{n,\ SEC\text{-}MALLS}$ (THF) = 1.45; $dn\cdot dc^{-1}$ (THF) = 0.079 $mL\cdot g^{-1}$; $[\alpha]_D$ = -50.6° (*c* 1.00, $CHCl_3$, 24 °C). The ^1H and ^{13}C NMR spectra of **2** (entry 4) are almost similar to those of entry 7.

Cationic Polymerization of 1 using Slow-monomer-addition Strategy. A typical procedure for the cationic polymerization (entry 7 in Table 1) using the slow-monomer-addition strategy[87] is as follows: $BF_3\cdot OEt_2$ (32.0 μL, 250 μmol, [**1**]/[$BF_3\cdot OEt_2$] = 40) was added to a solution of **1** (0.404 g, 2.00 mmol) in dry CH_2Cl_2 (4.0 mL, [**1**] = 0.5 $mol\cdot L^{-1}$) at 10 °C using a microsyringe. A dry CH_2Cl_2 (16.0 mL, [**1**] = 0.5 $mol\cdot L^{-1}$) solution of **1** (1.62 g, 8.00 mmol) in a microsyringe was then slowly added to the mixture at 10 °C using a dosing pump. The drip rate was 15.0 $\mu L\cdot min^{-1}$, i.e., the addition time of **1** was 17.8 h. After 24 h, the polymerization was terminated by adding 400 mL of methanol containing a small portion of an aqueous ammonia solution. After evaporating the solvent, the residue was purified by reprecipitation from methanol-water to yield the crude polymer. The precipitate was redissolved in $CHCl_3$ (15 mL) and applied to the preparative SEC to remove the unreacted monomer **1**. The concentrated polymer solution was dried *in vacuo* to give a white solid polymer **2** in a 66.1 % yield (1.34 g); $M_{w,SEC}$ (THF) = 8,600; $M_{w,SEC}/M_{n,SEC}$ (THF) = 1.26; $M_{w,SEC\text{-}MALLS}$ (THF) = 122,400; $M_{w,\ SEC\text{-}MALLS}/M_{n,\ SEC\text{-}MALLS}$ (THF) = 1.70; $dn\cdot dc^{-1}$ (THF) = 0.074 $mL\cdot g^{-1}$; $[\alpha]_D$ = -22.2° (*c* 1.00, $CHCl_3$, 25 °C). ^1H NMR (CD_2Cl_2):δ 5.87 (br.s, H-1), 5.20 -2.40 (br, including peaks at 4.47, 4.28, 4.18, 3.99, 2.91(epoxy group), 2.81 (epoxy group), 2.66 and 2.63 ppm), 1.44 (s, $=C(CH_3)_2$), 1.28 ppm (s, $=C(CH_3)_2$), and 0.89 ppm. ^{13}C NMR (CD_2Cl_2):δ 111.95 ($=C(CH_3)_2$), 106.83 (C-1), 105.40 (C-1), 101.49, 86.00-63.00 (m, including peaks at 85.62, 84.24, 82.16, 79.80, 75.35, 72.03, 69.39, 68.38), 27.05

(=C(CH_3)$_2$), and 26.42 ppm (=C(CH_3)$_2$). It is hard to estimate the degree of branching value because the NMR peaks due to each branch structure were broadened and overlapped.

Methylation of polymer 2. Silver oxide (0.50 g, 2.20 mmol) and methyl iodide (3.00 mL, 48.6 mmol) were added to polymer **2** (78.2 mg, 0.39 unit-mmol, entry 2 in Table 1) in 5 mL of dry acetonitrile. After the mixture was refluxed for 21 h, the insoluble parts were removed by filtration and the solvent was evaporated. The residue was redissolved in $CHCl_3$ (1 mL) and applied to the preparative SEC to prepare the resulting polymer. The concentrated polymer solution was dried *in vacuo* to give a white solid polymer **4** in an 80.1 % yield (67.0 mg); The degree of methylation, which was estimated from the ^{1}H NMR measurement, was ca. 100 %. $M_{w,SEC\text{-}MALLS}$ (THF) = 4,200; $M_{w,SEC\text{-}MALLS}/M_{n,SECMALLS}$ (THF) = 1.49; $dn \cdot dc^{-1}$ (THF) = 0.076 mL·g^{-1}; [η] = 4.7 mL·g^{-1}; The Mark-Houwink-Sakurada exponents, α and K, are 0.22 and 0.90 mL·g^{-1}, respectively. ^{1}H NMR (CD_2Cl_2):δ 5.83 (br.s, H-1), 4.55 (br.s), 4.30-3.20 (m, including peaks at 4.11, 3.93, 3.89, 3.77, 3.61, 3.60, 3.48 (OCH_3), 3.46 (OCH_3) and 3.44 ppm (OCH_3)), 3.00-2.70 (m, including br.s at 2.91, 2.86, and 2.76 ppm, epoxy group) 1.45 (s, =C(CH_3)$_2$), and 1.29 ppm (s, =C(CH_3)$_2$). ^{13}C NMR (CD_2Cl_2):δ 111.60 (=*C*(CH_3)$_2$), 105.06 (C-1), 83.60-80.50 (m, including peaks at 83.51, 82.05, and 81.28 ppm), 78.50, 76.90-75.00 (m, including peak at 76.75 ppm), 74.00-67.00 (m, including peaks at 72.00, 69.97, and 67.50 ppm), 59.50-57.00 (m, including peaks at 58.43 and 57.64 ppm, O*C*H$_3$), 48.15, 46.49, 29.68, 26.77 (=C(*C*H$_3$)$_2$), and 26.28 ppm (=C(*C*H$_3$)$_2$).

Synthesis of 5,6-anhydro-1,2-*O*-isopropylidene-3-*O*-methyl-α-D-glucofuranose (5). Silver oxide (4.60 g, 19.8 mmol) and methyl iodide (1.20 mL, 19.8 mmol) were added to **1** (1.95 g, 9.64 mmol) in 5 mL of dry acetonitrile. After the mixture was refluxed for 20 h, the insoluble parts were removed by filtration and the solvent was evaporated. The residue was purified by column chromatography on silica gel with *n*-hexane/ethyl acetate (2/1, v/v; R_f = 0.31) to afford a colorless liquid **5**. Yield: 1.92 g (92.1%). ^{1}H NMR ($CDCl_3$):δ 5.88 (d, *J* = 3.2 Hz, 1H, H-1), 4.58 (m, 1H, H-2), 3.82 (m, 1H, H-4), 3.68 (m, 1H, H-3), (s, 3H, OCH_3), 3.46, 3.20 (m, 1H, H-5), 2.88 (m, 1H, H-6), 2.74 (m, 1H, H-6), 1.43 (s, 3H, C(CH_3)$_2$) and 1.29 ppm (s, 3H, C(CH_3)$_2$). ^{13}C NMR ($CDCl_3$):δ 111.9 (*C*(CH_3)$_2$), 105.3 (C-1), 84.4 (C-2),

82.0 (C-3), 81.6 (C-4), 58.4 (OCH_3), 48.1 (C-5), 46.9 (C-6), 26.9 ($C(CH_3)_2$) and 26.3 ppm ($C(CH_3)_2$). Anal. Calcd for $C_{10}H_{16}O_5$ (216.23): C, 55.55; H, 7.46. Found: C, 55.24; H, 7.31. FI-MS (*m*/*z*): 216 (M^+).

Anionic Polymerization of 5. The anionic polymerizations of **5** were carried out using a storage flask with a needle valve under an argon atmosphere. 1.0 mol·L^{-1} *t*-BuOK in a THF solution (0.30 mL, 0.30 mmol, [**1**]/[*t*-BuOK] = 20) was added to a solution of **5** (1.30 g, 6.00 mmol) in dry THF (2.70 mL, [**1**] = 2.0 mol·L^{-1}) at 60 °C using a microsyringe. After 180 h, the reaction mixture was poured into a large amount of methanol, and the solution was neutralized with dry ice. After evaporating the solvent, the residue was purified by reprecipitation from chloroform-*n*-hexane to yield the crude polymer. The precipitate was redissolved in $CHCl_3$ (9 mL) and applied to the preparative SEC to remove the unreacted monomer. The concentrated polymer solution was dried *in vacuo* to give a white solid polymer, poly(5,6-anhydro-1,2-*O*-isopropylidene-3-*O*-methyl-α-D-glucofuranose) (**6**), in a 65.9 % yield (857 mg); $M_{w,SEC\text{-}MALLS}$ (THF) = 11,100; $M_{w,SEC\text{-}MALLS}/M_{n,SEC\text{-}MALLS}$ (THF) = 1.26; d*n*·d*c*$^{-1}$ (THF) = 0.062 mL·g^{-1}; [η] = 6.8 mL·g^{-1}; Mark-Houwink-Sakurada exponents α and K are 0.50 and 0.06 mL·g^{-1}, respectively. $[\alpha]_D$ = -51.0° (*c* 1.00, $CHCl_3$, 25 °C). ^{1}H NMR ($CDCl_3$):δ 5.93 (d, *J* = 3.2 Hz, H-1), 5.83, 4.54, 4.49 (d, *J* = 3.7 Hz, H-2), 4.27 (d, *J* = 8.7 Hz, H-6), 4.01 (br.s, H-3 and H-4), 3.87, 3.76 (br.s, H-5), 3.53 (br.s, H-6) 3.46 (s, 3H, O*CH*$_3$), 1.45 (s, C(*CH*$_3$)$_2$), 1.27 (s, C(*CH*$_3$)$_2$) and 1.17 ppm. ^{13}C NMR ($CDCl_3$):δ 111.03, 110.95 (*C*(CH_3)$_2$), 105.01 (C-1), 82.95 (C-4), 81.76 (C-2), 79.10 (C-3), 75.65 (C-5), 74.37 (C-6), 72.63, 57.61 (OCH_3), 27.58, 26.76 ($C(CH_3)_2$) and 26.30 ($C(CH_3)_2$).

Hydrolysis of polymer 2. Polymer **2** (0.60 g, 2.97 unit-mmol, entry 7) was dissolved in 5 mL of a 85/15 (v/v) trifluoroacetic acid/water mixture. The solution was magnetically stirred at room temperature for 18 hr, and diluted with 40 mL of water. The solution was neutralized using an aqueous ammonia solution, then dialyzed in water for several days using a Spectra/Por® CE (cellulose Ester) membrane (MWCO: 1000 Dalton, SPECTRUM). After freeze-drying from water, the residue was redissolved in water (5 mL) and applied to the preparative SEC to purify the resulting polymer. After freeze-drying from water, the hyperbranched 5,6-glucan (**3**) was isolated as a white powder. The yield was 62.7 % (335 mg, entry 9). $M_{w,SEC\text{-}MALLS}$ (0.2 mol·L^{-1} $NaNO_3$ aq.) = 74,400; $M_{w,\ SEC\text{-}MALLS}/M_{n,\ SEC\text{-}}$

$_{MALLS}$ (0.2 mol·L^{-1} $NaNO_3$ aq.) = 2.74; dn·dc^{-1} (0.2 mol·L^{-1} $NaNO_3$ aq.) = 0.135 mL·g^{-1}; $[\alpha]_D$ = +17.3° (*c* 1.00, H_2O, 25 °C). ^{1}H NMR (entry 9, D_2O including acetone as internal reference):δ 6.05 (br.s), 5.49 (br.s, α-franosyl unit, H-1), 5.23 (br.s, β-franosyl and α-pyranosyl units, H-1), and 5.00-3.20 ppm (m, including peaks at 4.64 (br.s, β- pyranosyl unit, H-1), 4.28, 4.12, 3.96, 3.71, 3.56, 3.48, 3.26), 1.44, and 1.23 ppm. ^{13}C NMR (entry 9, D_2O including acetone as internal reference):δ 102.94 (β-franosyl unit, C-1), 97.30 (α-franosyl unit, C-1), 96.58 (β- pyranosyl unit, C-1), 92.74 (α- pyranosyl unit, C-1), and 88.00-62.00 ppm (m, including signals at 80.92, 77.83, 76.28, 75.48, 74.68, 73.33, 72.04, and 70.45 ppm). For entry 8: $[\alpha]_D$ +53.6° (*c* 1.00, H_2O, 25 °C). ^{1}H NMR (entry 8, D_2O including acetone as internal reference):δ 6.01 (br.s), 5.48 (br.s, α-franosyl unit, H-1), 5.22 (br.s, β-franosyl and α-pyranosyl units, H-1), 4.65 (br.s, β- pyranosyl unit, H-1) and 4.50 – 3.20 ppm (m, including peaks at 4.33, 4.28, 4.12, 4.08, 3.99, 3.60, 3.52, 3.49, 3.47, 3.44, 3.40, 3.34, 3.33, 3.25), 1.52, and 1.36 ppm. ^{13}C NMR (entry 8, D_2O including acetone as internal reference):δ 103.24 (C-1), 102.80 (β-franosyl unit, C-1), 97.31 (α-franosyl unit, C-1), 96.56 (β- pyranosyl unit,C-1), 92.72 (α- pyranosyl unit, C-1), 85.62, 82.97, 80.89, 80.38, and 79.00-68.00 ppm (m, including signals at 77.65, 76.43, 76.25, 76.05, 75.68, 75.49, 74.62, 74.24, 74.15, 73.27, 72.21, 72.01, 71.68, 71.60, 71.37, 71.03, 70.56, and 69.90 ppm).

Formation of Gold Nanoparticle. A typical procedure for the formation of the gold particles is as follows (run 1 in Fig. 15): Polymer **3** (37.5 mg, 0.231 unit-mmol, entry 8) was first dissolved in deionized (Mill-Q) water (5 mL). The polymer solution (46 unit-mmol·L^{-1}, 1.8 mL) was filtered, then kept at 70 °C for 30 minutes in the UV-cell equipped with a stirring bar. To the polymer solution, the tetrachloroauric acid ($HAuCl_4$) aqueous solution (0.01 mol·L^{-1}, 6.67 μL) was added. The UV-vis spectra of the polymer–metal solution were measured in the wavelength range of 450-600 nm at 1 min intervals with the polymer solution as a reference. The obtained absorbance data were corrected by removing the absorption of the $HAuCl_4$ aqueous solution.

Results and discussion

Polymerization. The ring-opening polymerizations of 5,6-anhydro-1,2-*O*-isopropylidene-α-D-glucofuranose (**1**) were carried out using potassium *tert*-butoxide (*t*-BuOK) as the anionic initiator or boron trifluoride diethyletherate ($BF_3 \cdot OEt_2$) as the cationic initiator. The typical results are summarized in Table 1. When *t*-BuOK was used in dry tetrahydrofuran (THF) as the solvent, the reaction mixtures immediately changed from colorless to dark brown as the reaction progressed, and the polymerization homogeneously proceeded until the end of the polymerization, while the cationic polymerization using $BF_3 \cdot OEt_2$ in dry methylene chloride (CH_2Cl_2) homogeneously proceeded without any color change. The polymers obtained using *t*-BuOK or $BF_3 \cdot OEt_2$ were a light-yellow solid or a white solid, respectively, which were soluble in chloroform, THF, DMF and methanol, but insoluble in water. The polymer yields for the anionic polymerization (entries 1-3) were in the range of 52.9 to 69.5 % and the increase in conversion was very slow, e.g., 52.9 % at 60 °C for 160 hr (entry 1), 68.4 % at 50 °C for 600 hr (entry 2) and 69.5 % at 50 °C for 770 hr (entry 3). The weight-average molecular weights ($M_{w,SEC}$) measured by size exclusion chromatography (SEC) of the resulting polymers ranged from approximately 1,400 to 2,000. In contrast, the polymer yield for the cationic polymerization (entry 4) was 68.5% for 24 hr and the $M_{w,SEC}$ was 4,100, indicating that the cationic polymerization is much better than the anionic one.[88]

In general, highly branched polymers are known to have spherical conformations in a solution, and the $M_{w,SEC}$s of these polymers were often claimed to be too low because the hydrodynamic volumes of these polymers are smaller than the corresponding linear polymers used for the calibration. Therefore, the absolute weight-average molecular weight ($M_{w,SEC\text{-}MALLS}$) of the resulting polymers was also measured using SEC equipped with a multiangle laser light scattering instrument (MALLS), and was compared with the $M_{w,SEC}$ values in Table 1. The $M_{w,SEC\text{-}MALLS}$ values for entries 1-4 ranged from 7,400 to 15,000, which were ca. 1.6-5.5 those of the $M_{w,SEC}$ values. These results suggest that the resulting polymer has a compact form in solution because of its branched structure.

From the viscosity measurement in THF using SEC equipped with the viscosity detector, the intrinsic viscosities ([η]) of the resulting polymers were observed. The solution viscosities of the polymers obtained using *t*-BuOK were very low in the range from 4.0 to 4.6 $mL \cdot g^{-1}$. The $M_{w,SEC\text{-}MALLS}$ dependence

of [η] for the polymer (entry 2) is shown in Fig. 2. On the basis of these results, the exponents (α) of the Mark-Houwink-Sakurada equations ([η] = KM^{α}) of the polymers obtained using *t*-BuOK were determined to be 0.25-0.27. In general, α = 0.5 suggests that the polymer behaves as dense spheres, α = 0.6-0.8 for a flexible chain and α > 1 for an elongated rod, whereas it is well-known that the α value was less than 0.5 for the various hyperbranched polymers. Our experimental α values were noticeably low, which are ascribed to the compact spherical structure in solution. Therefore, the SEC, MALLS, and viscosity measurement results suggested that the polymer obtained using *t*-BuOK was a branched spherical molecule, i.e., a hyperbranched poly(5,6-anhydro-1,2-*O*-isopropylidene-α-D-glucofuranose) (**2**).

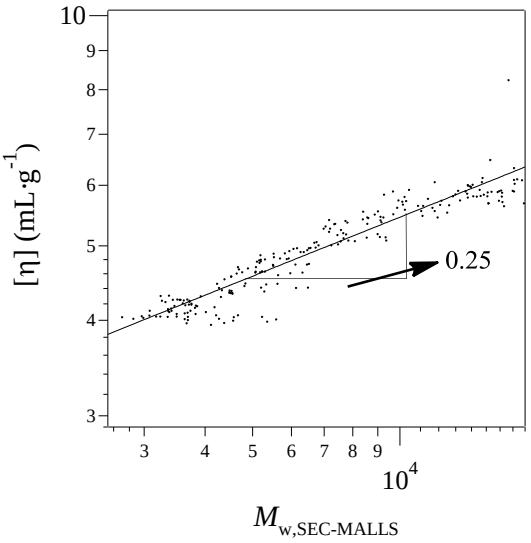


Fig. 2. Double logarithmic plots of [η] vs $M_{w,\ SEC\text{-}MALLS}$ for polymer **2** in THF (entry 2)

The solution viscosity of the polymer (entry 4) obtained using $BF_3{\cdot}OEt_2$ was 3.4 mL·g^{-1}, which was lower than those of the polymers obtained using *t*-BuOK, regardless of the higher molecular weight polymer. In addition, the α value was 0.11, which was less than those of the polymers obtained using *t*-BuOK. These results indicate that the polymer obtained using $BF_3{\cdot}OEt_2$ should have a more compact

spherical structure in solution than the polymer obtained using *t*-BuOK, i.e., the polymer obtained using $BF_3 \cdot OEt_2$ should have a higher degree of branching value than the polymer obtained using *t*-BuOK.

As the efficient preparation method of hyperbranched polymer with both a high molecular weight and a high branching value degree, the slow-monomer-addition strategy[87] was used for the cationic polymerization of **1**. According to the established time of monomer addition, 16 mL of the 0.5 $mol \cdot L^{-1}$ monomer solution was slowly added to the mixture of 4 mL of monomer solution and $BF_3 \cdot OEt_2$ at 10 °C using a dosing pump. These results are summarized as entries 5-7 in Table 1. The slow-monomer-addition strategy results in a very low monomer concentration actually present in the reaction mixture. Thus, the exclusive reaction of the monomer with the growing polyfunctional macromolecules occurs, resulting in a high molecular weight and a high degree of branching. Based on a longer addition time, i.e., a decrease in the drop rate, the $M_{w,SEC\text{-}MALLS}$ values dramatically increase up to 122,400 (entry 7). Interestingly, regardless of the higher molecular weight polymer, the [η] values were in the range from 3.3 to 3.8 $mL \cdot g^{-1}$, which was similar to the value for entry 4. The α values of the Mark-Houwink-Sakurada equations were determined to be very low in range of 0.08 to 0.09, suggesting that the resulting polymers have a highly branched spherical structure. In addition, the polydispersities ($M_{w,SEC}/M_{n,SEC}$) were found to be in the relatively narrow range of 1.26-1.33. These results indicate that the slow-monomer-addition strategy is a facile method for preparing the hyperbranched polymer **2** with a controlled molecular weight and highly branched structure.

Polymer Structure. In order to investigate the polymer structure, the ^1H and ^{13}C NMR measurements were carried out in CD_2Cl_2. Parts a and b of Fig. 3 show the ^1H NMR spectra of the polymers obtained using *t*-BuOK (entry 2) and $BF_3 \cdot OEt_2$ (entry 7), respectively. Though the unreacted monomer was removed using the preparative SEC, in both spectra, the peaks due to the epoxy groups slightly exist in the range of 2.6-3.0 ppm as the starting polymer end, which is a characteristic of a hyperbranched polymer. From a comparison of both spectra, the spectrum of the polymer obtained using *t*-BuOK is obviously different from that of the polymer using $BF_3 \cdot OEt_2$, recognizing that the structure of the former is more regular than that of the latter because of distinct splitting of the peaks and narrow peak width for

the anionically obtained polymer. This trend is also recognized by the comparison of the ^{13}C NMR spectra in Fig. 4. Ring-opening of the monosubstituted epoxide occurs in two ways, i.e., by α- or β-scission. In general, the polymerization using an anionic catalyst predominantly cleaves the CH_2-O bond (β-scission) via an SN_2 displacement to form the regular head-to-tail linkage. Therefore, the difference in the spectra is mainly caused by the stereoregularity of the C-5 carbon in the polymer. Though the anionically obtained polymer has higher stereoregularity, the ^{13}C NMR spectrum contains many peaks due to the C-6 methylene carbons, as shown in Fig. 4c, indicating that the polymer obviously contains some branched structures as irregular structure.

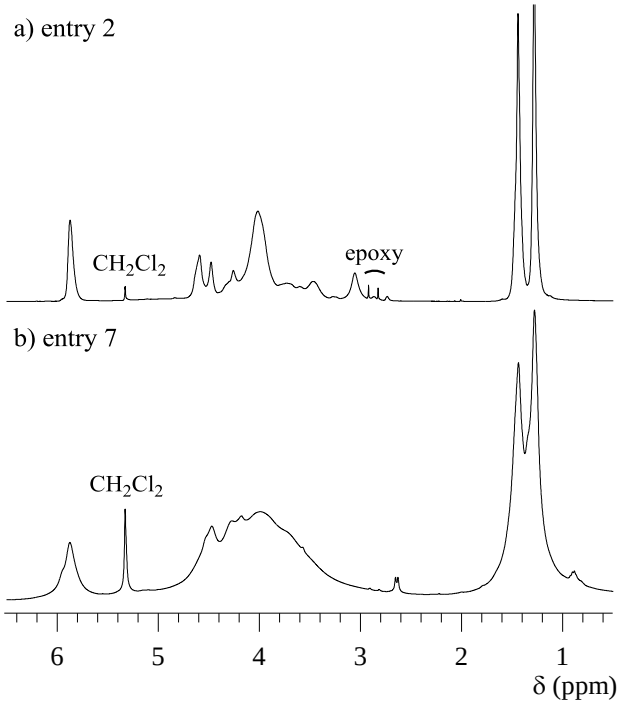


Fig. 3. ^1H NMR spectra of polymer **2** in CD_2Cl_2 (TMS as internal standard).
a) entry 2 and b) entry 7.

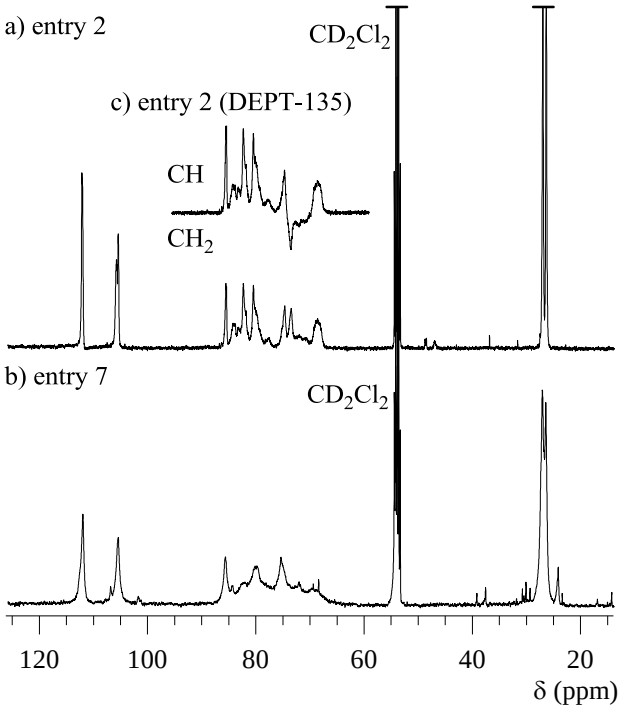


Fig. 4. ^{13}C NMR spectra of polymer **2** in CD_2Cl_2 (TMS as internal standard). a) entry 2 and b) entry 7, and DEPT-135 spectrum of entry 2.

In order to investigate the branching structure in polymer **2**, the ^{1}H and ^{13}C NMR spectra of the methylated hyperbranched polymer (**4**) were compared with those of the linear analog, poly(5,6-anhydro-1,2-*O*-isopropylidene-3-*O*-methyl-α-D-glucofuranose) (**6**). The polymer **4** was prepared by the methylation of the anionically obtained polymer **2** (entry 2) using CH_3I and Ag_2O in dry CH_3CN, as shown in Fig. 5. On the other hand, the linear polymer **6** was obtained by the anionic polymerization of 5,6-anhydro-1,2-*O*-isopropylidene-3-*O*-methyl-α-D-glucofuranose (**5**) using *t*-BuOK, as shown in Fig 6. Parts a and b of Fig. 7 show the ^{1}H NMR spectra of **4** and **6**, respectively. The ^{1}H NMR spectrum of **4** consists of some broad peaks, which is obviously different from that of **6**. In particular, the methoxy peaks of **4** were split three or more, indicating that there are some methoxy groups in different surroundings, as shown in Fig. S1 (see Supplementary Information). A comparison of the ^{13}C NMR spectra in Fig. S2 (see Supplementary Information) also shows the obvious difference. The ^{13}C NMR

spectrum of **6** mainly consists of 10 sharp peaks corresponding to the repeating unit, while the spectrum of **4** includes some broad peaks which are due to the irregular structure. In addition, the Mark-Houwink-Sakurada exponent α of **4** was 0.22, whereas the α value of **6** was 0.50. The NMR and viscosity results strongly support the fact that polymer **4** obviously had some branched structure as an irregular structure, i.e., the polymerization of **1** proceeded through the ring-opening multibranching mechanism to produce the hyperbranched poly(5,6-anhydro-1,2-*O*-isopropylidene-α-D-glucofuranose) **2**.

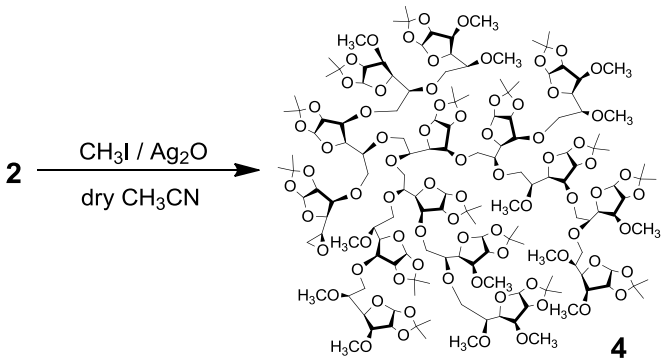


Fig. 5. Methylation of polymer **2**.

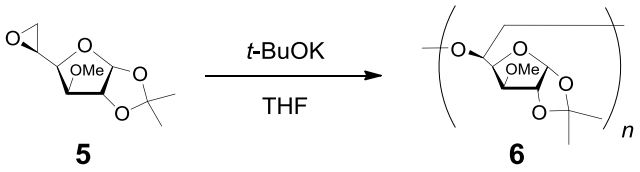


Fig. 6. Anionic polymerization of 5,6-anhydro-1,2-*O*-isopropylidene-3-*O*-methyl-α-D-glucofuranose (**5**).

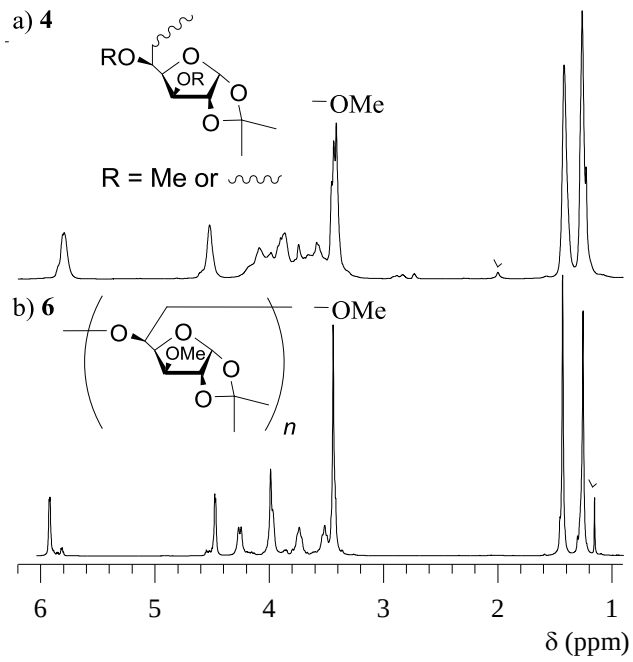


Fig. 7. ^{1}H NMR spectra of a) polymer **4** and b) polymer **6** in $CDCl_3$ (TMS as internal standard).

Polymerization mechanism. Fig. 8 and 9 represent the proposed anionic and cationic polymerization mechanisms, respectively. During the anionic polymerization, the ring-opening occurred through only β-scission to afford the linear unit with the regular head-to-tail linkage. The branches of the polymers should be produced by the proton transfer from the hydroxy group at C-3 to the other alkoxide and the following ring opening of **1** by the alkoxide at C-3. On the other hand, for the cationic polymerization, the protonated **1** reacted with the epoxy group of a second monomer, and two kinds of linkages through the α- or β-scission were formed as a linear structure. The branched structure should be produced by proton transfer from the hydroxyl groups in the polymer chains to the oxygen atom of the epoxy group. These reactions mentioned above simultaneously occurred in the polymerization systems and consequently produced the hyperbranched polymer **2**.

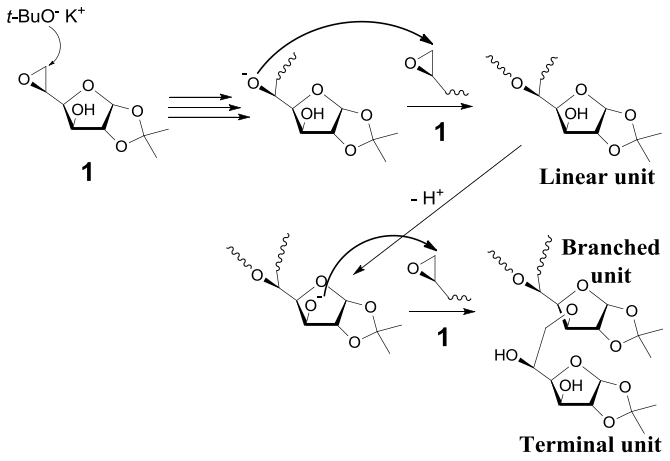


Fig. 8. Proposed mechanism for the anionic polymerization of **1**.

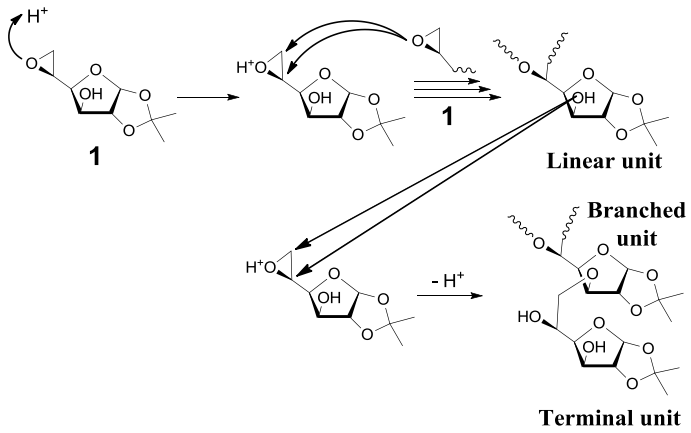


Fig. 9. Proposed mechanism for the cationic polymerization of **1**.

Hydrolysis of 2. The hydrolysis of polymer **2** was carried out using a mixture of trifluoroacetic acid and water (85/15, v/v) to produce the hyperbranched poly(5,6-anhydro-α-D-glucofuranose) (**3**), e.g., the hyperbranched 5,6-glucan. The obtained polymers **3** were white powders which were soluble in water, and insoluble in chloroform, THF, DMF and methanol, which remarkably differed from the solubility of **2**. Fig 10 and 11 show the ^{1}H and ^{13}C NMR spectra of **3** (entries 8 and 9) obtained from **2** (entries 2 and 7), respectively. Based on these spectra, it is concluded that the isopropylidene groups were mostly hydrolyzed.

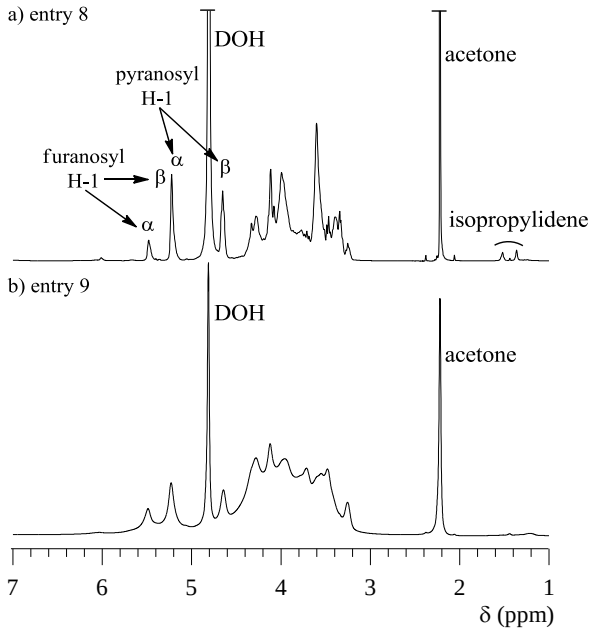


Fig. 10. ^{1}H NMR spectra of polymer **3** in D_2O (acetone as internal standard). a) entry 8 and b) entry 9.

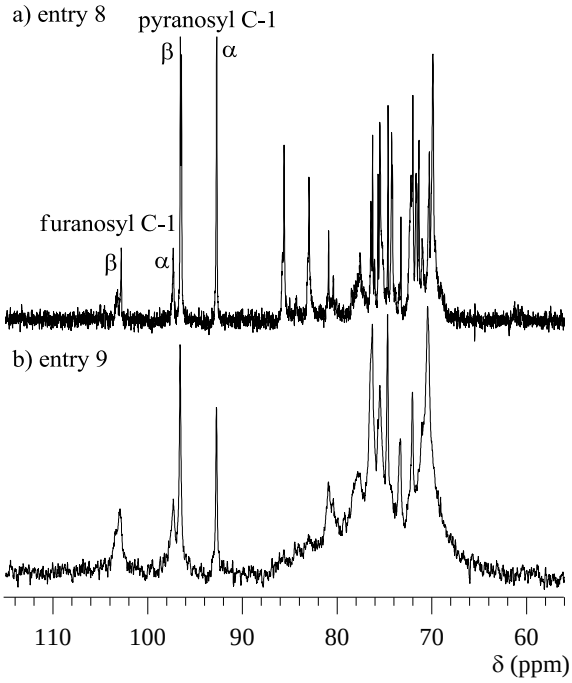


Fig. 11. ^{13}C NMR spectra of polymer **3** in D_2O (acetone as internal standard). a) entry 8 and b) entry 9.

The physical data of the obtained **3** are listed in Table 2. The $M_{w,SEC\text{-}MALLS}$ values for entries 8 and 9 are 3,740 to 74,400, respectively, which were lower than the calculated M_w values. This indicates that some linkage may break under the hydrolysis conditions. However, the exponents α of the Mark-Houwink-Sakurada equations were determined to be 0.36 for entry 8 and 0.26 for entry 9, as shown in Fig. 12, indicating that the obtained polymer still have the branched structure.

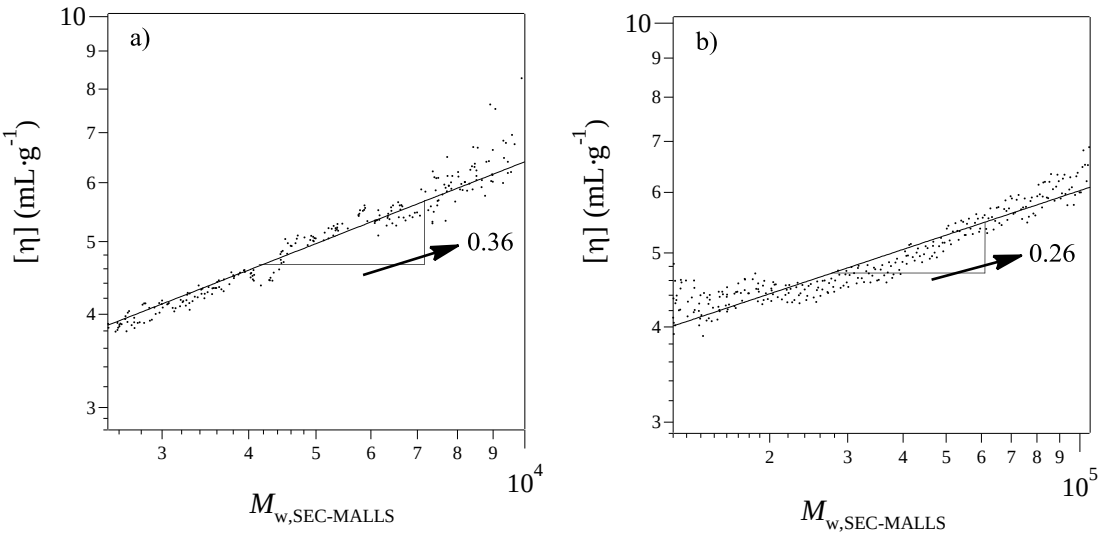


Fig. 12. Double logarithmic plots of [η] vs $M_{w,\ SEC\text{-}MALLS}$ for polymer **3** in 0.2 mol L^{-1} $NaNO_3$ aqueous solution. a) entry 7 and b) entry 8.

The ^1H and ^{13}C NMR spectra of **3** consisted of very complicated peaks, which were constructed by the difference sequences of the D-glucose units. In the ^{13}C NMR spectra, four distinct signals exist in the C-1 region. The big signals at 92.74 and 96.58 ppm in Fig 11a, which are very similar to the chemical shifts of the carbons for α- and β-D-glucopyranoses,[89] should be assigned to the carbons of the α- and β-D-glucopyranose units in **3**, respectively. In addition, the small signals at 97.30 and 102.94 ppm should be assigned to the carbons of the α- and β-D-glucofranose units in **3**, respectively, since these signals are very similar to the chemical shifts of 3,5,6-tri-*O*-methyl α- and β-D-glucofranoses.[90] This result

indicates that after hydrolysis, most of the D-glucofranose units in **2** changes to the D-glucopyranose unit in **3**. As shown in Fig 13, the D-glucofranose units with a hydroxyl group at C-5 could isomerize to the D-glucopyranose units, e.g., the 6-linked terminal and 3,6-linked linear D-glucofranose units isomerize to the corresponding D-glucopyranose units, while the 5,6-linked linear and 3,5,6-linked dendritic units do not change. Since the intensities of the D-glucopyranose units in Fig 11 are stronger than those of D-glucofranose, polymer **3** includes numerous 6-linked terminal and 3,6-linked linear D-glucopyranose units, which is a characteristic of a hyperbranched polymer.

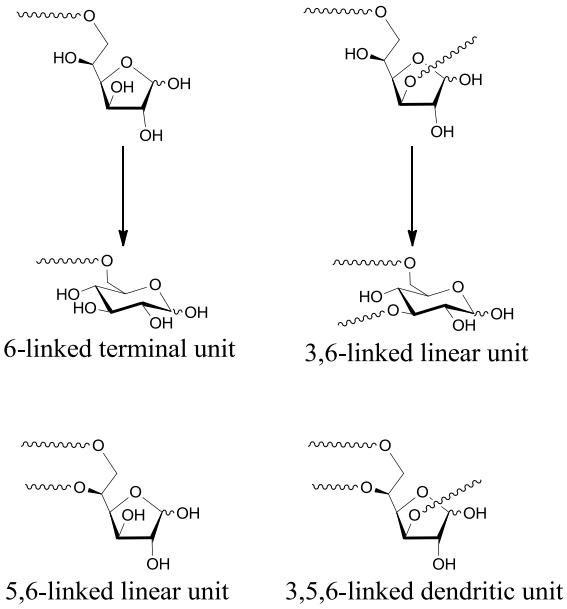


Fig 13. Possible isomerizations of polymer **3** to produce the D-glucopyranose units.

Formation of gold nanoparticle using 3. As mentioned in the previous section, polymer **3** is a novel hyperbranched glycopolymer arranged with numerous reducing D-glucose units on the peripheries of the hyperbranched polymer, whereas almost all of the glycodendrimers, star-shaped glycopolymers, and hyperbranched glycopolymers consist only of nonreducing sugar ends. Therefore, polymer **3** is a particularly interesting material, which should be called a "reducing sugar ball". Polymer **3** probably has a higher reducing ability than D-glucose because of the glyco-cluster effect or the multivalent effect of

the reducing D-glucose units. Such a hyperbranched glycopolymer with numerous reducing sugar units is also of considerable interest in a wide variety of fields such as biochemical and medicinal applications.

To estimate the reducing ability of **3**, the formation of gold particles, which is derived from the reduction of $HAuCl_4$, is demonstrated in the aqueous solution of **3**. Fig 14 shows the UV-vis spectra after 150-min mixing of **3** and $HAuCl_4$ aqueous solutions at 70 °C (runs 1-3). As a comparative experiment, the UV-vis spectrum of the mixture of D-glucose and $HAuCl_4$ aqueous solutions (run 4) is also shown in Fig 14.[91] After mixing of the **3** and $HAuCl_4$ aqueous solutions, the color of the system changed from colorless to light red with time, as shown in Fig. S3 (see Supplementary Information). The UV-vis spectra of the **3**-$HAuCl_4$ system have a λ_{max} that ranged from 512 nm to 514 nm, which due to the formation of the gold particles. From previous studies,[92] the average gold particle diameter can be estimated from the λ_{max}, i.e., gold particles of ca. 8 - 12 nm were obtained by the mixing of **3** and the $HAuCl_4$ aqueous solutions. Thus, the average hydrodynamic diameter of the resulting Au particles (run 2 in Fig. 14), which was measured by a dynamic light scattering (DLS) technique, was ca. 11.3 nm, as shown in Fig. S4 (see Supplementary Information).

To obtain the relationship between the formation of the gold particles and the reaction time, the increasing trend in the UV-vis absorbance at λ_{max} is presented in Fig. 15. For all the **3**-$HAuCl_4$ systems (runs 1-3), the UV-vis absorbance sharply increases without an induction time and finally became a constant value within 10 min.[93,94] On the other hand, the D-glucose-$HAuCl_4$ system (run 4) shows a long induction time, then the absorbance sharply increases as an autocatalytic system. A similar autocatalytic tendency with a long induction time was also reported using the sugar-persubstituted poly(amidoamine) dendrimer, i.e., a sugar ball with numerous nonreducing sugar units.[95] The results indicate that **3** has a high reducing ability that originated from the high density of the reducing sugars in **3**, i.e., a glyco-cluster effect or multivalent effect of the reducing sugars. Thus, the reaction rates within 10 min for the **3**-$HAuCl_4$ systems were much faster than that for the D-glucose-$HAuCl_4$ system, as shown in Fig 15b. This indicates that "reducing sugar ball" **3** effectively act as a sugar based reductant.

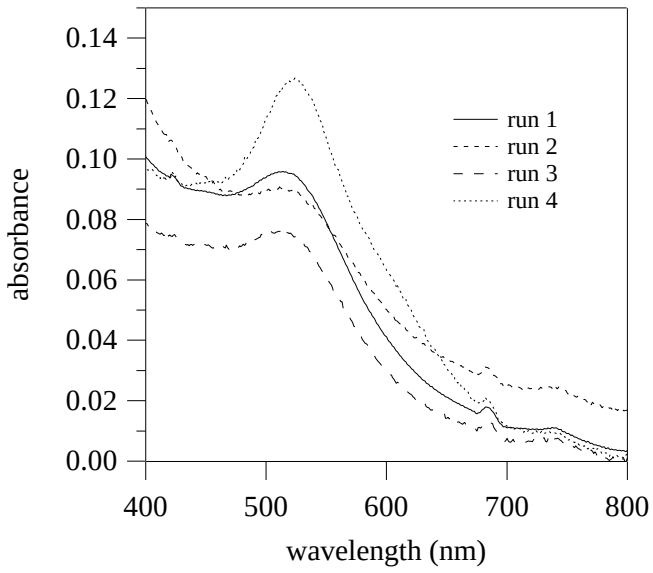


Fig. 14. UV-vis spectra after 150 min mixing of polymer **3** (or D-glucose) and $HAuCl_4$ at 70 ^oC. Run 1; polymer **3** (46 unit-mmol L^{-1} of entry 8, 1.8 mL) and $HAuCl_4$ (0.01 mol L^{-1}, 6.67 μL) aqueous solutions, run 2; polymer **3** (25 unit-mmol L^{-1} of entry 8, 1.8 mL) and $HAuCl_4$ (0.01 mol L^{-1}, 6.67 μL) aqueous solutions, run 3; polymer **3** (46 unit-mmol L^{-1} of entry 9, 1.8 mL) and $HAuCl_4$ (0.01 mol L^{-1}, 6.67 μL) aqueous solutions, and run 4; D-glucose (46 unit-mmol L^{-1}, 1.8 mL) and $HAuCl_4$ (0.01 mol L^{-1}, 6.67 μL) aqueous solutions. The maximal wavelengths (λ_{max}) are 512 nm for run 1, 514 nm for run 2, 514 nm for run 3 and 526 nm for run 4.

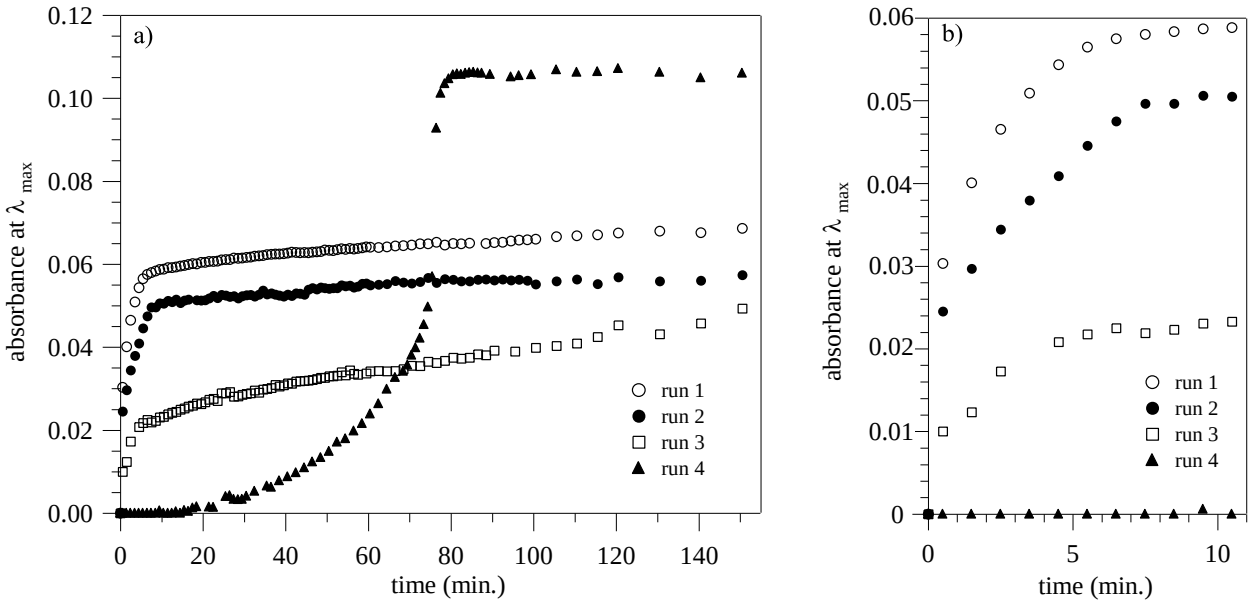


Fig. 15. Change in the UV absorption at λ_{max} of mixtures of polymer **3** (or D-glucose) and $HAuCl_4$ as a function of reaction time at 70 °C. Run 1 (open circle); polymer **3** (46 unit-mmol L^{-1} of entry 8, 1.8 mL) and $HAuCl_4$ (0.01 mol L^{-1}, 6.67 μL) aqueous solutions at 512 nm, run 2 (closed circle); polymer **3** (25 unit-mmol L^{-1} of entry 8, 1.8 mL) and $HAuCl_4$ (0.01 mol L^{-1}, 6.67 μL) aqueous solutions 514 nm, run 3 (open square); polymer **3** (46 unit-mmol L^{-1} of entry 9, 1.8 mL) and $HAuCl_4$ (0.01 mol L^{-1}, 6.67 μL) aqueous solutions 514 nm, and run 4 (closed triangle); D-glucose (46 unit-mmol L^{-1}, 1.8 mL) and $HAuCl_4$ (0.01 mol L^{-1}, 6.67 μL) aqueous solutions 526 nm. a) Overall figure and b) the expanded figure in the range of 0 to 10 min. The obtained absorbance data were corrected by removing the absorption of the $HAuCl_4$ aqueous solution.

Conclusions. In this study, we performed the anionic or cationic ring-opening multibranching polymerization of 5,6-anhydro-1,2-*O*-isopropylidene-α-D-glucofuranose (**1**) as a latent cyclic AB_2-type monomer. The polymerizations proceeded through the proton-transfer reaction mechanism to produce hyperbranched poly(5,6-anhydro-1,2-*O*-isopropylidene-α-D-glucofuranose) (**2**). In particular, the cationic polymerization with the slow-monomer-addition strategy is a facile method leading to the polymers with a high molecular weight and highly-branched structure. The weight-average molecular weight ($M_{w,SEC\text{-}MALLS}$) values of **2** measured by multi-angle laser light scattering (MALLS) varied in the range of 7,400 to 122,400, which were significantly higher than the weight-average molecular weight ($M_{w,SEC}$) values by size exclusion chromatography (SEC). The intrinsic viscosities ([η]) of these

polymers were very low in the range of 3.3-4.6 $mL \cdot g^{-1}$ and the Mark-Houwink-Sakurada exponent α was calculated to be 0.08-0.28. These results of the MALLS, SEC, and viscosity measurements suggested that these polymers exist in a compact spherical conformation in solution because of their highly-branched structure. A water-soluble hyperbranched 5,6-glucan (**3**) was also synthesized by the hydrolysis of polymer **2**. Polymer **3** is a novel hyperbranched glycopolymer arranged with numerous reducing D-glucose units on the peripheries of the polymer, and has a high reducing ability derived from the glycol-cluster effect or the multivalent effect of the reducing D-glucose units, i.e., the "reducing sugar ball" **3** effectively acts as a sugar-based reductant. Such a hyperbranched glycopolymer with numerous reducing sugar units can be expected to be useful in a wide variety of fields such as biochemical and medicinal applications; for example, a biocompatible reductant, antioxidant, and a part of the carrier for a drug delivery system, etc.

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Supplementary Information. Possible repeating units of polymer **4**; ^{13}C NMR spectra of polymer **4** and polymer **6** in $CDCl_3$; Demonstration pictures of the formation of gold particles using polymer **3** (entry 8); Dynamic light scattering (DLS) measurements of Au particles; C-H COSY spectrum of polymer **3** (entry 8) in D_2O. These materials are available free of charge via the Internet.

Table 1. The polymerization results of **1**

entry	Cat.	Temp °C	Polymerization time hr	addition time hr	yield %	$M_{w,SEC}$ (M_w/M_n) [c]	$M_{w,SEC-MALLS}$ (M_w/M_n) [d]	α [d]	[η] [d] mL·g^{-1}	K [d] mL·g^{-1}
1	*t*-BuOK[a]	60	160	-	52.9	2,000 (1.26)	7,700 (1.22)	0.27	4.6	0.40
2	*t*-BuOK[a]	50	600	-	68.4	1,400 (1.28)	7,400 (1.37)	0.25	4.0	0.48
3	*t*-BuOK[a]	50	770	-	69.5	1,600 (1.34)	8,900 (1.33)	0.27	4.3	0.38
4	$BF_3{\cdot}OEt_2$[b]	10	24	-	68.5	4,100 (1.29)	15,000 (1.45)	0.11	3.4	1.16
5	$BF_3{\cdot}OEt_2$[b]	10	24	4.4	73.4	5,600 (1.32)	37,000 (1.19)	0.09	3.3	1.22
6	$BF_3{\cdot}OEt_2$[b]	10	24	13.3	75.5	6,500 (1.33)	81,300 (1.23)	0.09	3.4	1.26
7	$BF_3{\cdot}OEt_2$[b]	10	24	17.8	66.1	8,600 (1.26)	122,400 (1.70)	0.08	3.8	1.42

[a] Conditions: Ar atmosphere; [M]/[I] = 20; solvent, THF; [M] = 2.0 mol·L^{-1}. [b] Condition : Ar atmosphere; [M]/[I] = 40; solvent, CH_2Cl_2; [M] = 0.5 mol·L^{-1}. [c] Determined by SEC using polystyrene standards in THF. [d] The weight average molecular weight $M_{w,SEC-MALLS}$, polydispersity $M_{w,SEC-MALLS}/M_{n,SEC-MALLS}$, intrinsic viscosity [η], and Mark-Houwink-Sakurada constants, α and K ([η] = K$M_{w,SEC-MALLS}^{\alpha}$), were determined by SEC in THF equipped with MALLS and viscosity detectors.

Table 2. The results of hydrolysis of **2** to prepare **3**

entry	Starting Material **2**	$M_{w,SEC\text{-}MALLS}$ of **3** [a] ($M_{w,SEC\text{-}MALLS}$/ $M_{n,SEC\text{-}MALLS}$)	α [a]	[η] [a] mL·g^{-1}	K [a] mL·g^{-1}
8	entry 2	3,740 (1.26)	0.36	4.4	0.22
9	entry 7	74,400 (2.74)	0.26	5.0	0.29

[a] The weight average molecular weight $M_{w,SEC\text{-}MALLS}$, polydispersity $M_{w,SEC\text{-}MALLS}/M_{n,SEC\text{-}MALLS}$, intrinsic viscosity [η], and Mark-Houwink-Sakurada constants, α and K ([η] = K$M_{w,SEC\text{-}MALLS}^{\alpha}$), were determined by SEC in 0.2 mol·L^{-1} $NaNO_3$ aqueous solution equipped with MALLS and viscosity detectors.

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88 A similar polymerization tendency was reported by Uryu et al.[84,85]

89 The chemical shifts of the carbons for α- and β-D-glucopyranoses in D_2O are 93.18 and 97.00 ppm, respectively.

90 G. Just and D. Crosilla, *Can. J. Chem*.,1980, **58**, 2349. The chemical shifts of the protons for 3,5,6-tri-*O*-methyl α- and β-D-glucofranoses in $CDCl_3$ are 5.40 and 5.10 ppm, respectively. Based on the ^{1}H NMR data of 3,5,6-tri-*O*-methyl α- and β-D-glucofranoses and CH-COSY spectrum of polymer **3** (entry 8) in Fig. S5 (see Supplementary Information), the signals at 97.30 and 102.94 ppm in Fig. 11a are assigned to the carbons of the α- and β-D-glucofranose units in **3**, respectively.

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93 In run 3 using the high molecular weight polymer **3** (entry 9), the intensity at λ_{max} is consistently rather weaker compared to run 1 using the low molecular weight sample (entry 8), probably because the sugar units on the peripheries of polymer **3** at the same unit-mol concentration is less for the high molecular weight polymer than the low molecular weight polymer, i.e., the inner sugar units of the high molecular weight polymer cannot effectively be utilized as a reductant because of the sugar units in the enclosed space.

94 The final intensity at λ_{max} for the D-glucose-$HAuCl_4$ system was greater than those for the **3**-$HAuCl_4$ system at the same mol concentration. The main factor is attributed to the links between the sugar units in **3**, which cannot act as a reductant.

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Supplementary Material (ESI) for Polymer Chemistry

Supplementary Information for:

Hyperbranched 5,6-Glucan as Reducing Sugar ball

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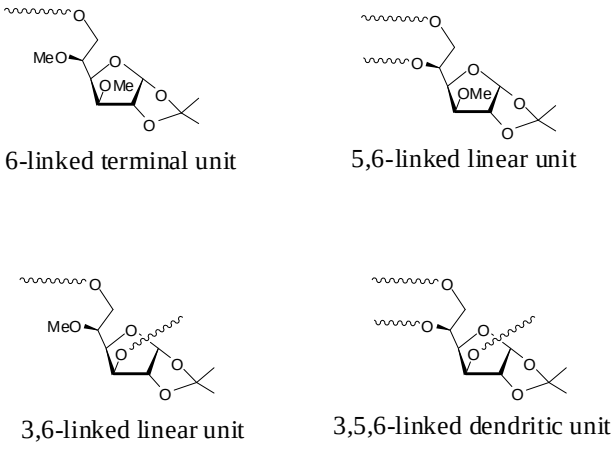

Fig. S1. Possible repeating units in polymer **4**.

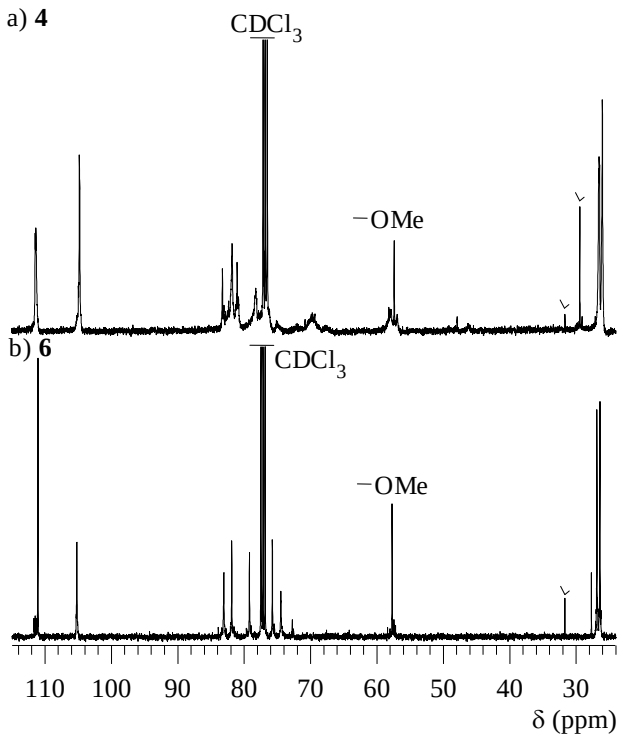

Fig. S2. ^{13}C NMR spectra of a) polymer **4** and b) polymer **6** in $CDCl_3$ (TMS as internal standard).

Fig. S3. Demonstration (run 1 in Fig 14) of the formation of gold particles using polymer **3** (entry 8). The left vial contains polymer **3** (46 unit-mmol·L^{-1}, 1.8 mL) and $HAuCl_4$ aqueous solution (0.01 mol·L^{-1},

6.67μL) at 70 °C (after 150-min mixing), while the right vial is the control experiment in the absence of polymer **3**.

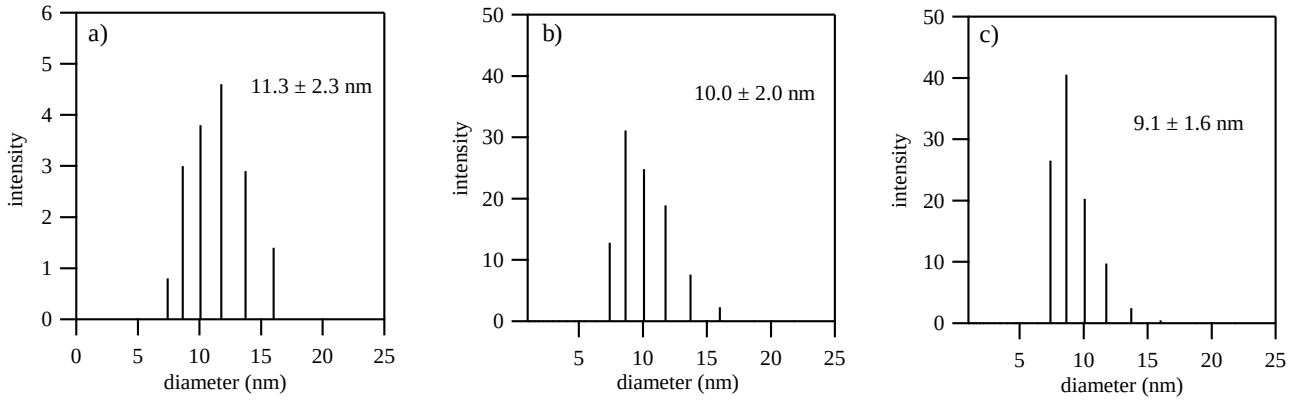

Fig. S4. Particle size distribution and average hydrodynamic diameter estimated by dynamic light scattering (DLS) measurements of run 2 in Fig 14. a) scatter average hydrodynamic diameter, b) weight average hydrodynamic diameter, and c) number average hydrodynamic diameter calculated using the histogram methods including the Marquadt analysis.

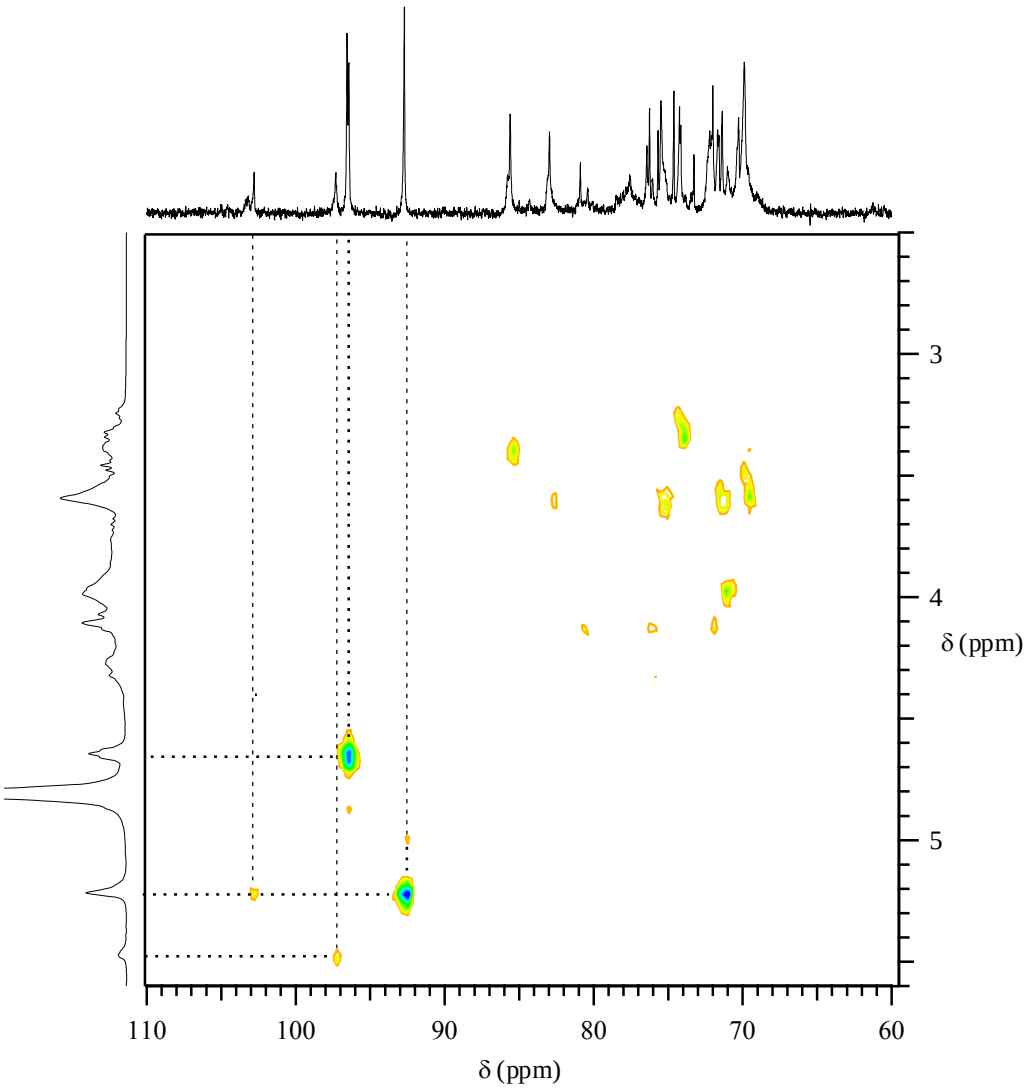

Fig. S5. C-H COSY spectrum of polymer **3** (entry 8) in D_2O (acetone as internal standard).