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Synthesis of 3-Acetamido-3-deoxy-(1→5)-α-D-ribofuranan by Ring-Opening Polymerization of 1,4-Anhydro-3-azido-α-D-ribopyranose Derivative

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ABSTRACT: A stereoregular 3-acetamido-3-deoxy-(1→5)-α-D-ribofuranan (3AAdRF) was synthesized by the synthetic route starting from selective ring-opening polymerization of 1,4-anhydro-3-azido-2-*O*-*t*-butyldimethylsilyl-3-deoxy-α-D-ribopyranose (A3ASR). A3ASR was polymerized by $BF_3 \cdot OEt_2$ catalyst at low temperature to give 3-azido-2-*O*-*t*-butyldimethylsilyl-(1→5)-α-D-ribofuranan (3AzSRF) with $\bar{M}_n$ of 1.4×10^4—1.9×10^4 and $[\alpha]_D$ of +249—+266°. 3AzSRF was reduced with $NaBH_4$ in a tetrahydrofuran (THF)–ethanol mixture to afford 3-amino-2-*O*-*t*-butyldimethylsilyl-3-deoxy-(1→5)-α-D-ribofuranan (3AmSRF). Acetylation of the amino group in 3AmSRF produced 3-acetamido-ribofuranan derivative (3AAdSRF), and then desilylation of the 3AAdSRF gave 3AAdRF with $\bar{M}_n$ ranging from 9.4×10^3 to 10.5×10^3 and $[\alpha]_D$ of +195°. The structure analysis was performed by ^{13}C and ^1H NMR spectroscopy, IR spectroscopy, and optical rotation measurements.

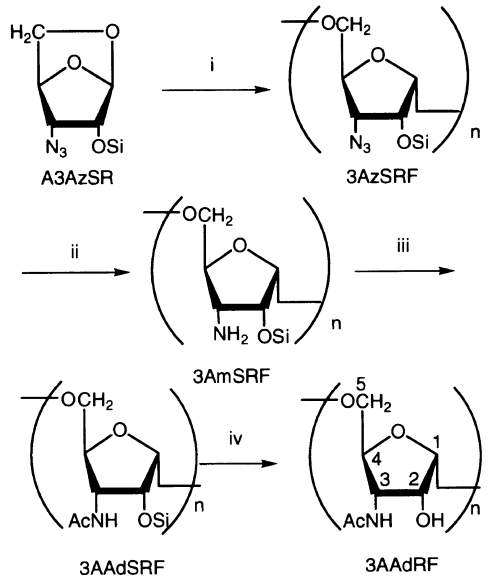
KEY WORDS 3-Acetamido-α-D-ribofuranan / Azido Polysaccharides / Azido Reduction / Ring-Opening Polymerization /

Ring-opening polymerization of anhydro sugars have provided a number of stereoregular polysaccharides such as 1,6-α-linked hexopyranans, 1,4-β-linked cellulose-type polysaccharides, and 1,5-α-linked glycofuranans having hydroxyl and acyl groups as the functional groups.[1-4] 1,6-α-Linked dextran-type polysaccharides possessing azido, amino, and sulfamide groups in addition to hydroxyl groups were also synthesized.[5] Polysaccharides containing sulfamide and sulfate groups exhibited high anticoagulant activities. Recently, a few new amino-containing polysaccharides, such as 1,5-α-linked amino-xylofuranans and -ribofuranans were synthesized by a synthetic route starting from the selective ring-opening polymerization of azido group-containing 1,4-anhydro sugars (=1,5-anhydro sugars).[6,7]

Since it was found that sulfated polysaccharides have an anti-HIV (human immunodeficiency virus) activity,[8] various sulfated polysaccharides with high anti-HIV activities were synthesized by the sulfation of natural and synthetic polysaccharides obtained by the ring-opening polymerization of 1,4-anhydro sugars.[9-13] Since curdlan sulfate had the most potent anti-HIV activity and low side-effects,[10] its Phase I/II test for an AIDS drug was carried out.[14]

Recently, biological activities of the acetamido group in polysaccharides have attracted much attention, since it was found that an acetamido group-containing natural polysaccharide chitin has a wide variety of biological activities, such as wound-healing effects to animals,[15] induction of neutrophil migration,[16] a drug-carrying ability with sustained release,[17] an activation for morphogenetic events in yeast,[18] and the induction of plant defense response.[19] Also, it is reported recently that many kinds of acetamido group-containing poly saccharides were isolated from *O*-antigenic chains, which were of biomedical property.[20,21]

To understand the relationship between the biological activity and the structure of acetamido-containing polysaccharides, it is necessary to study the synthesis of new kinds of acetamido-containing polysaccharides. In this field, Hatanaka and coworkers prepared a low molecular weight acetamido-containing oligomers.[22] The synthesis of an acetamido-containing polyxylose 3-acetamido-3-deoxy-(1→5)-α-D-xylofuranan was completed for the first time in our laboratory.[23]



Si = $Si(CH_3)_2C(CH_3)_3$ Ac = CH_3CO

i) $BF_3 \cdot O(C_2H_5)_2$ / CH_2Cl_2, ii) $NaBH_4$ / THF, EtOH,
iii) CH_3COCl / THF, iv) $(n\text{-}Bu)_4NF$ / THF.

Scheme 1. Synthesis of 3-acetamido-3-deoxy-(1→5)-α-D-ribofuranan (3AAdRF) by conversion of azido-ribofuranan 3AzSRF *via* both amino-ribofuranan 3AmSRF and acetamido-ribofuranan 3AAdSRF.

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In this paper, we report the synthesis of a new acetamido group-containing polyribose 3-acetamido-3-deoxy-(1→5)-α-D-ribofuranan.

The synthesis starts from selective ring-opening polymerization of 1,4-anhydro-3-azido-2-*O*-*t*-butyldimethylsilyl-3-deoxy-α-D-ribopyranose providing a stereoregular azido group-containing (1→5)-α-D-ribofuranan. The azido group in the polysaccharide was reduced into an amino group which was then acetylated to give acetamido and *t*-butyldimethylsilyl groups-containing polysaccharide. After removal of the OH-protective *t*-butyldimethylsilyl group, 3-acetamido-3-deoxy-(1→5)-α-D-ribofuranan was successfully synthesized. The structure analysis was performed by NMR and IR spectroscopies, and optical rotation measurements.

EXPERIMENTAL

Materials and Measurements

Lithium azide was prepared by the reaction of sodium azide with lithium sulfate according to the method in literature.[24] Solvents were distilled before use. 500 MHz ^{1}H and 125 MHz ^{13}C NMR spectra were taken on sample solutions in $CDCl_3$ or D_2O by use of a JEOL Lambda 500 spectrometer. The 2-dimensional H-H COSY-FG and C-H HMQC-FG measurements were used to assign the ^{1}H and ^{13}C absorptions. Tetramethylsilane (TMS) and sodium 2,2-dimethyl-2-silapentane-5-sulfonate (DSS) were used as internal standards for organic solvent and deuterium oxide, respectively. Specific rotations were measured in chloroform or aqueous solution at 20°C by use of a Perkin Elmer 240 polarimeter. Molecular weights of polymers were determined by gel permeation chromatography using $CHCl_3$ or phosphate buffer solution (pH = 7.0) as eluent and standard polystyrenes or pullulans as references, respectively.

Synthesis of 1,4-Anhydro-3-azido-2-O-t-butyldimethylsilyl-3-deoxy-α-D-ribopyranose (A3ASR)

1,4-Anhydro-α-xylopyranose (AX) was obtained by pyrolysis of xylose according to the method of Köll *et al.*[24] 1,4-Anhydro-2-*O*-*t*-butyldimethylsilyl-α-D-xylopyranose (A2SX) was prepared by reaction of AX (90.0 g, 0.68 mol) with *t*-butyldimethylsilyl chloride (103 g, 0.68 mol) in the presence of silver nitrate (78 g, 0.68 mol) at room temperature for 2 h. After work-up, A2SX (79.0 g, 0.32 mol) was obtained in 47.2% yield.

A2SX (19.7 g, 80.0 mmol) was allowed to react with trifluoromethanesulfonic anhydride (33.8 g, 108 mmol) in pyridine (160 mL) at 5°C for 1 h. After purification, 1,4-anhydro-2-*O*-*t*-butyldimethylsilyl-3-*O*-trifluoromethanesulfonyl-α-D-xylopyranose (A3TSX) (28.0 g, 74.0 mmol) was obtained in 92.5% yield.

1,4-Anhydro-3-azido-2-*O*-*t*-butyldimethylsilyl-3-deoxy-α-D-ribopyranose (A3ASR) was prepared by reaction of A3TSX (28.0 g, 74 mmol) with lithium azide (18.6 g, 370 mmol) in dry dimethylformamide (80 mL) at 85°C for 20 h. After work-up, purification of crude A3ASR was finally carried out by silica gel column chromatography using a hexane and ethyl acetate mixture (6 : 1 volume ratio) as an eluent. A3ASR (7.3 g, 1.97 mol) was obtained in 48.4% yield.

Selective Ring-Opening Polymerization of Azido-Containing Monomer A3ASR

Cationic ring-opening polymerization of A3ASR was carried out by using high vacuum technique.[26–28] A3ASR was polymerized with $BF_3 \cdot OEt_2$ as catalyst in methylene chloride at −10 and −20°C. Resulting polymer was purified by dissolution–reprecipitation with chloroform–methanol system for three times, and freeze-dried from benzene.

Reduction of Azido Group into Amino Group in Polyribose

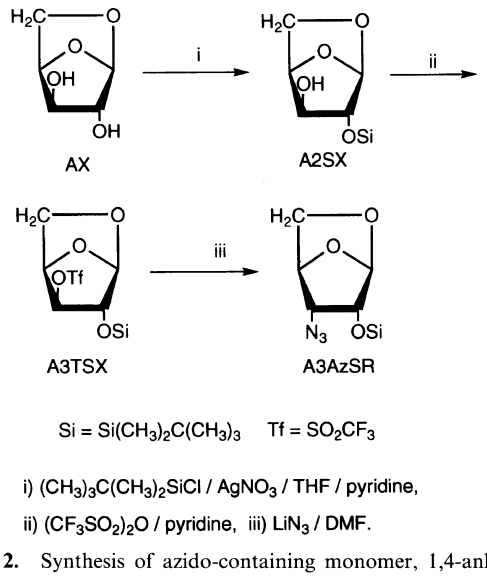
To 3-azido-2-*O*-*t*-butyldimethylsilyl-(1→5)-α-D-ribofuranan (3AzSRF) (0.20 g) solution in tetrahydrofuran (THF) (10 mL), sodium borohydride (0.28 g) solution in ethanol (5 mL) was added, followed by stirring at 40°C for 2 h. An amino polymer 3-amino-2-*O*-*t*-butyldimethylsilyl-3-deoxy-(1→5)-α-D-ribofuranan (3AmSRF) was recovered by pouring the reaction solution into water. The polymer was purified by dissolution–reprecipitation using THF–H_2O twice, and finally freeze-dried from benzene. The reduction was repeated again, so that no absorption of azido group was observed in IR spectrum of the polymer.

N-Acetylation of Amino Group in 3AmSRF

3AmSRF (200 mg) was acetylated with acetyl chloride (1 mL) in a mixture of THF–pyridine (5/10 mL) at 20°C under nitrogen atmosphere for 1 h. After work-up, 3-acetamido-2-*O*-*t*-butyldimethylsilyl-3-deoxy-(1→5)-α-D-ribofuranan (3AAdSRF) (140 mg, 59.7% yield) was obtained.

Desilylation of 3AAdSRF

t-Butyldimethylsilyl groups in 3AAdSRF (50 mg) were removed by reaction with tetra-*n*-butylammonium fluoride (1.8 mL) in THF (5 mL) at room temperature for 1 h. After desilylation, H_2O was added, and then THF was evaporated. The aqueous polymer solution was dialyzed with water for 3 d. 3-Acetamido-3-deoxy-(1→5)-α-D-ribofuranan (3AAdRF) (30 mg, 99.5% yield) was obtained by freeze-drying from water.



Scheme 2. Synthesis of azido-containing monomer, 1,4-anhydro-3-azido-2-*O*-*t*-butyldimethylsilyl-3-deoxy-α-D-ribopyranose (A3ASR) by (i) silylation, (ii) triflation, and (iii) azidation of anhydro xylose (AX).

Table I. ^{13}C Chemical shifts of 1,4-anhydro-α-D-xylopyranose (AX), 1,4-anhydro-2-*O*-*t*-butyldimethylsilyl-α-D-xylopyranose (A2SX), 1,4-anhydro-3-*O*-trifluoromethylsulfonyl-2-*O*-*t*-butyldimethylsilyl-α-D-xylopyranose (A3TSX), and 1,4-anhydro-3-azido-2-*O*-*t*-butyldimethylsilyl-3-deoxy-α-D-ribopyranose (A3ASR)[a]

Compoud designation	C1	C2	C3	C4	C5	$Si(CH_3)_2C(CH_3)_3$			F_3CSO_3		Solvent
						$C(CH_3)_3$	$C(CH_3)_3$	$(CH_3)_2$			
AX	106.513	81.327	79.838	79.797	64.604	—	—	—	—	—	D_2O
A2SX	104.761	80.948	79.476	76.992	61.873	25.763	18.022	−4.705	—	—	$CDCl_3$
A3TSX	104.136	78.554	90.638	75.091	62.638	25.557	17.915	−5.125	119.822	117.281	$CDCl_3$
A3ASR	102.614	77.979	63.534	79.541	65.838	25.680	18.211	−5.207	—	—	$CDCl_3$

[a] ^{13}C chemical shifts in ppm.

Table II. Selective ring-opening polymerization of 1,4-anhydro-3-azido-2-*O*-*t*-butyldimethylsilyl-3-deoxy-α-D-ribopyranose (A3ASR)

No.	Azido-monomer		$BF_3 \cdot OEt_2$	CH_2Cl_2	Temp	Time	Yield[a]		$\bar{M}_n$[b] $\times 10^4$	$[\alpha]_D^{20}$	1,5-α-Furanosidic[c]
		g	μL	mL	°C	h	g	%		deg.	unit, %
1	A3ASR	0.20	5	0.5	−20	22	0.16	80.5	1.93	+266	100
2	A3ASR	1.00	25	2.5	−10	20	0.62	62.3	1.70	+265	100
3	A3ASR	0.50	10	1.0	−20	24	0.27	54.0	1.39	+266	100
4	A3ASR	0.50	10	1.0	−20	24	0.39	77.2	1.66	+249	100

[a] Measured after purification. [b] Determined by GPC. [c] Determined by NMR spectroscopy.

RESULTS AND DISCUSSION

Selective Ring-Opening Polymerization of 1,4-Anhydro-3-azido-2-O-t-butyldimethylsilyl-3-deoxy-α-D-ribopyranose (A3ASR)

An azido-containing anhydro ribose 1,4-anhydro-3-azido-2-*O*-*t*-butyldimethylsilyl-3-deoxy-α-D-ribopyranose (A3ASR) was prepared by starting from 1,4-anhydro-xylose, according to the literature as shown in Scheme 2.[7,23]

1,4-Anhydro-2-*t*-butyldimethylsilyl(*t*-BDMS)-α-D-xylopyranose (A2SX) was prepared by partial-*t*-butyldimethylsilylation at 2-OH alone of (AX) in 47.2% yield. Subsequently, A2SX was allowed to react with trifluoromethanesulfonic anhydride to produce A3TSX in 92.5% yield. By reacting A3TSX with lithium azide, Walden inversion was caused at C3 carbon of the A3TSX to give a ribose-type monomer A3ASR. Structures of all compounds were determined by NMR and the chemical shifts were listed in Table I.

The azido-containing monomer, A3ASR had five sharp absorption peaks due to five carbons of ribopyranose at 102.614 (C1), 77.979 (C2), 63.534 (C3), 79.541 (C4), and 65.838 (C5) ppm. All NMR spectra indicated that the compounds were successfully prepared as designed.

The azido-containing monomer A3ASR was polymerized with a Lewis acid $BF_3 \cdot OEt_2$ as catalyst at low temperatures under high vacuum by use of a literature method.[7] Results of polymerization are summarized in Table II.

When A3ASR was polymerized with $BF_3 \cdot OEt_2$ as catalyst at −10°C and −20°C, polymers were obtained in 54—80% yields. These polymers had high number-average molecular weights of 1.39×10^4—1.93×10^4. The polymers obtained at −20°C exhibited high specific rotations from +249° to +266°, indicating that those are composed of an α structure. In ^{13}C NMR spectra

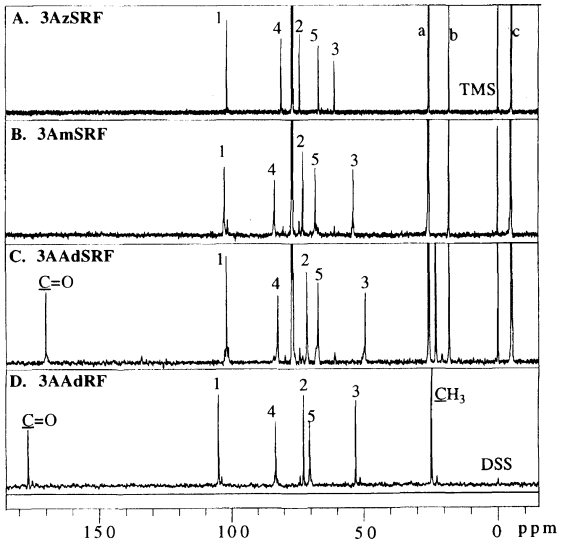


Figure 1. ^{13}C NMR Spectra of (A) 3-azido-2-*O*-*t*-butyldimethylsilyl-3-deoxy-(1→5)-α-D-ribofuranan ($CDCl_3$ as solvent), (B) 3-amino-2-*O*-*t*-butyldimethylsilyl-3-deoxy-(1→5)-α-D-ribofuranan ($CDCl_3$ as solvent), (C) 3-acetamido-2-*O*-*t*-butyldimethylsilyl-3-deoxy-(1→5)-α-D-ribofuranan ($CDCl_3$ as solvent), and (D) 3-acetamido-3-deoxy-(1→5)-α-D-ribofuranan (D_2O as solvent).

(Figure 1A), all absorptions due to five carbons constituting ribose residues appeared as five sharp peaks, representing that the polymers consisted of a completely stereoregular structure.[29]

Taking into account the high specific rotation and the stereoregular structure indicated by NMR, it was revealed that selective ring-opening polymerization occurred *via* 1,5-α-scission of the 1,4-anhydro sugar and the polymer was 3AzSRF.

^{13}C NMR absorptions of a stereoregular 3AzSRF were assigned using 2-dimensional H-H COSY-FG and C-H HMQC-FG spectra. The assignments and chemical shifts of 3AzSRF are shown in Table III. ^{13}C chemical shifts

Table III. ^{13}C chemical shifts of azido, amino, and acetamido groups-containing (1→5)-α-D-ribofuranans[a]

Polymer designation	C1	C2	C3	C4	C5	$Si(CH_3)_2C(CH_3)_3$			$CONHCH_3$		Solvent
						$C(CH_3)_3$	$C(CH_3)_3$	$(CH_3)_2$	C=O	CH_3	
3AzSRF[b]	101.89	74.22	61.91	81.23	67.06	25.66	18.07	−4.97	—	—	$CDCl_3$
3AmSRF[c]	102.70	73.30	54.02	83.70	68.20	25.64	18.15	−4.81	—	—	$CDCl_3$
3AAdSRF[d]	102.05	71.56	49.58	82.56	67.33	25.63	18.14	−4.89	170.17	23.16	$CDCl_3$
3AAdRF[e]	105.07	72.66	53.14	83.33	70.33	—	—	—	176.75	24.67	D_2O

[a] Chemical shift in ppm. [b] 3-Azido-2-*O*-*t*-butyldimethylsilyl-3-deoxy-(1→5)-α-D-ribofuranans. [c] 3-Amino-2-*O*-*t*-butyldimethylsilyl-3-deoxy-(1→5)-α-D-ribofuranan. [d] 3-Acetamido-2-*O*-*t*-butyldimethylsilyl-3-deoxy-(1→5)-α-D-ribofuranan. [d] 3-Acetamido-3-deoxy-(1→5)-α-D-ribofuranan.

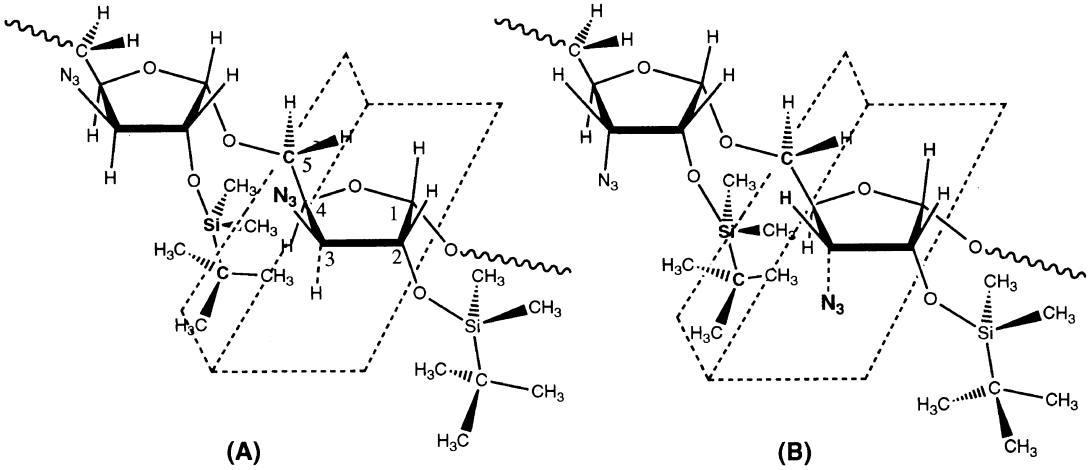


Figure 2. (A) Proposed structure of 3-azido-2-*t*-butyldimethylsilyl-3-deoxy-(1→5)-α-D-xylofuranan (3AzSXF). The azido group (N_3) exists between the *t*-butyldimethylsilyl group and O1–C5–C4 linkage. (B) Proposed structure of 3-azido-2-*t*-butyldimethylsilyl-3-deoxy-(1→5)-α-D-ribofuranan (3AzSRF). The azido group was under the plane of the ribose ring.

of the five carbons in azido group-containing (1→5)-α-ribofuranan appeared at 101.59 (C1), 74.22 (C2), 61.91 (C3), 81.23 (C4), and 67.06 (C5) ppm.

Reduction of the Azido Group into Amino Group in (1→5)-α-D-Ribofuranan (3AAdRF).

3AzSRF was reduced into 3AmSRF with sodium borohydride in a THF and ethanol mixture. The reduction of the azido group in 3AzSRF with sodium borohydride completed within 6 h in a THF–ethanol mixture at 40°C, producing 3AmSRF in 93.5% yield. The specific rotation of 3AmSRF was +123°. The reduction at higher temperature of 64°C gave the amino group-containing (1→5)-α-D-ribofuranan (3AmSRF) in 46.6—87.8% yield. This result indicates that much milder conditions were sufficient for the reduction of azido group in polyribose 3AzSRF than those for polyxylose 3AzSXF.[23] ^{13}C NMR spectra of the resulting polymer (3AmSRF) was measured in $CDCl_3$, as shown in Figure 1B. Since all carbon absorptions appeared as individual peaks, it was revealed that the polymer is 3AmSRF having the amino group at the C3 position. Assignments of the ^{13}C chemical shifts are shown in Table III.

Previously, we reported that reduction of a polyxylose homolog, *i.e.*, 3-azido-2-*O*-*t*-butyldimethylsilyl-3-deoxy-(1→5)-α-D-xylofuranan needed long time for completion.[23] Since the reduction of the polyribose derivative proceeded relatively smoothly, the reduction of the former polyxylose homolog was examined under similar conditions. The slow reduction of the azido-containing polyxylose was confirmed. The reason that the $NaBH_4$ reduction of the azido-containing polyxylose took longer time and higher temperature than that of polyribose might be due to a sterically hindered azido group in the polyxylose which is positioned between the *t*-butyldimethylsilyl group and O1–CH_2–C4 linkage (Figure 2A).

On the other hand, the azido group in the polyribose was under the plane of the ribose ring. Accordingly, the azido group can be easily attacked by reducing agent (Figure 2B).

N-Acetylation and Desilylation of 3-Amino-2-O-t-butyldimethylsilyl-3-deoxy-(1→5)-α-D-ribofuranan

The amino-ribofuranan 3AmSRF was acetylated with acetyl chloride in a THF and pyridine mixture at room temperature to give 3AAdSRF in 22.1—59.7% yields. The specific rotation of acetamido group-containing intermediate polysaccharide 3AzSRF was +125°.

^{13}C NMR spectrum and ^{13}C chemical shifts of 3AAdSRF are shown in Figure 1C and Table III, respectively. The ^{13}C NMR spectrum of 3AAdSRF showed a sharp single peak of C1 in 3AAdSRF at 102.05 ppm and the absorption peaks due to acetamido group at 170.17 ppm (C=O) and 23.16 ppm (CH_3), the three peaks of *t*-butyldimethylsilyl group appeared at 25.63, 18.14, and −4.89 ppm. The ^{13}C NMR spectrum of the 3AAdSRF indicates that complete acetylation of amino groups occurred.

Desilylation of 3AAdSRF was carried out with tetra-*n*-butylammonium fluoride in THF at room temperature

to give 3AAdRF in 58.1—99.5% yields. $\bar{M}_n$ ranged from 9.4×10^3 to 10.5×10^3 and $[\alpha]_D^{20}$ was in the range of $+194.9° — +194.7°$.

^{13}C NMR spectrum of 3AAdRF is shown in Figure 1D and the ^{13}C chemical shifts assigned by use of 2D NMR are exhibited in Table III. In the ^{13}C NMR spectrum, the C1 absorption appeared at 105.07 ppm as a single peak, as well as the carbonyl carbon ($CONHCH_3$) of acetamido group at 176.75 ppm and the carbon of methyl group ($CONHCH_3$) of acetamide at 24.67 ppm. On the other hand, the three absorption peaks of *t*-butyldimethylsilyl group ($Si(CH_3)_2C(CH_3)_3$) completely disappeared. Thus, it is revealed that the resulting polymer was 3AAdRF and complete desilylation took place without causing deacetylation.

Unlike acetamido group-containing natural polysaccharide chitin that was not soluble in water and organic solvents, the acetamido group-containing synthetic polyribose (3AAdRF) was soluble in water. Therefore, characterization of the acetamido group-containing polyribose was carried out in aqueous solution smoothly. The structure of the acetamido polyribose was determined by NMR on sample solutions in D_2O, the specific rotation was measured in aqueous solution and the molecular weight was measured by gel permeation chromatography (GPC) using phosphate buffer solution as an eluent. Thus, the water solubility is very important for chemical and biological study of acetamido group-containing synthetic polysaccharides.

In conclusion, a stereoregular acetamido group-containing ribofuranan, *i.e.*, 3-acetamido-3-deoxy-(1→5)-α-D-ribofuranan was synthesized for the first time by the four step reactions starting from the selective ring-opening polymerization of a 1,4-anhydro-azido-ribose derivative.

REFERENCES

1. C. Schuerch, "Adv. Carbohydr. Chem. Biochem.," Vol. 39, Academic Press, New York, N.Y., 1981, p 157.
2. T. Uryu, *Prog. Polym. Sci.*, **18**, 717 (1993).
3. H. Kamitakahara, F. Nakatsubo, and K. Murakami, *Macromolecules*, **27**, 5939 (1994).
4. H. Kamitakahara and F. Nakatsubo, *Macromolecules*, **29**, 51119 (1996).
5. K. Hatanaka, T. Yoshida, S. Miyahara, T. Sato, F. Ono, T. Uryu, and H. Kuzuhara, *J. Med. Chem.*, **30**, 810 (1987).
6. T. Yoshida, B. Kang, K. Hattori, H. Qing, and T. Uryu, *Macromolecules*, **29**, 3117 (1996).
7. B. W. Kang, K. Hattori, T. Yoshida, M. Hirai, Y. S. Choi, and T. Uryu, *Macromol. Chem. Phys.*, **198**, 1331 (1997).
8. R. Ueno and S. Kuno, *Lancet*, **ii**, 461 (1987).
9. O. Yoshida, H. Nakashima, T. Yoshida, Y. Kaneko, I. Yamamoto, K. Matsuzaki, T. Uryu, and N. Yamamoto, *Biochem. Pharm.*, **37**, 2887 (1988).
10. Y. Kaneko, O. Yoshida, R. Nakagawa, T. Yoshida, M. Date, S. Ogihara, S. Shioya, T. Matsuzawa, N. Nagashima, Y. Irie, T. Mimura, H. Shinkai, N. Yasuda, K. Matsuzaki, T. Uryu, and N. Yamamoto, *Biochem. Pharm.*, **39**, 793 (1990).
11. M. Witvrouw, D. Schols, G. Andrei, R. Snoeck, M. Hosoya, R. Pauwels, J. Balzarini, and E. De Clerck, *Antivir. Chem. Chemother.*, **2**, 171 (1991).
12. T. Yoshida, Y. Katayama, S. Inoue, and T. Uryu, *Macromolecules*, **25**, 4057 (1992).
13. T. Yoshida, C. Wu, L. Song, T. Uryu, Y. Kaneko, T. Mimura, H. Nakashima, and N. Yamamoto, *Macromolecules*, **27**, 4422 (1994).
14. M. Gordon, M. Guralnik, Y. Kaneko, T. Mimura, M. Baker, and W. Lang, *J. Med.*, **25**, 163 (1994).
15. Y. Shigemasa and S. Minami, "Biotechnology & Genetic Engineering Reviews," Vol. 13, M. P. Tombs, Ed., Intercept Ltd., Andover, 1995, p 383.
16. Y. Usami, Y. Okamoto, S. Minami, A. Matsuhashi, N. Kumazawa, S. Tanioka, and Y. Shigemasa, *J. Vet. Med. Sci.*, **56**, 1215 (1994).
17. S. Tokura, S. Baba, Y. Uraki, Y. Miura, N. Nishi, and O. Hasegawa, *Carbohydr. Polym.*, **13**, 273 (1990).
18. L. Popolo, D. Gilardelli, P. Bonfante, and M. Vai, *J. Bacteriology*, **179**, 463 (1997).
19. A. Savouré, C. Sallaud, J. El-Turk, J. Zuanazzi, P. Ratet, M. Schultze, A. Kondoṛosi, R. Esnault, and E. Kondorosi, *Plant J.*, **11**, 277 (1997) .
20. L. L. Leann and B. M. Perry, *Biochem Cell Biol.*, **75**, 315 (1997).
21. R. S. Haseley, O. Holst, and H. Brade, *Eur. J. Biochem.*, **244**, 761 (1997).
22. K. Komada, Y. Kobayashi, S. Nishimura, H. Kuzuhara, and J. Hatanaka, *J. Carbohydr. Chem.*, **14**, 481 (1995).
23. G. Borjihan and T. Uryu, *Macromolecules*, **31**, 5572 (1998).
24. H. B. Niels, *Acta Chem. Scand.*, **11**, 581 (1957).
35. P. Köll, S. Deyhim, and K. Heyns, *Chem. Ber.*, **106**, 3565 (1973).
36. J. Zachoval and C. Schuerch, *J. Am. Chem. Soc.*, **91**, 1165 (1969).
37. H. Ito and C. Schuerch, *J. Am. Chem. Soc.*, **101**, 5797 (1979).
38. K. Kobayashi and T. Kondo, *Macromolecules*, **21**, 6531 (1997).
29. T. Uryu, J. Yamanouchi, S. Hayashi, H. Tamaki, and K. Matsuzaki, *Macromolecules*, **16**, 320 (1983).