Note

Some *O*-Stearoyl Derivatives of Chitosan Prepared *via* Its Schiff's Base Intermediates

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O-Substituted and *N*-unsubstituted derivatives of chitosan [(1→4)-linked 2-amino-2-deoxy-β-D-glucan] are useful as intermediates for the chemical modification of free amino groups and for polyelectrolyte complex formation with acidic polymers,^{1~3)} and also as a support polymer for immobilized enzymes.⁴⁾ Several *N*- and *O*-fatty acyl derivatives of chitosan have been prepared,^{5,6)} and lipid A in the *Salmonella* species⁷⁾ contains both *N*- and *O*-fatty acyl groups. However, no report has dealt with *O*-fatty acyl derivatives of chitosan having unsubstituted amino groups in the molecule.

We now report on some *O*-stearoyl derivatives of chitosan prepared *via* its Schiff's base intermediates.

In addition to the *N*-alkylidene and *N*-arylidene derivatives reported previously,^{8,9)} nine novel derivatives were prepared by the reactions of chitosan with *n*-butyl (1), *trans*-2-butenyl (2), 3-phenyl-2-propenyl (6), 1-naphthyl (8), 2-naphthyl (9), 2,6-dichlorobenz (3), 3,4-dichlo-

robenz (4), 2,4-dichlorobenz (5), and 4-methoxybenz (7)-aldehydes. All the products were obtained in gelled forms and isolated as dried, gelatinous products in yields of 82~94% on the basis of completely *N*-substituted derivatives. The products had IR absorptions at 1650~1640 cm⁻¹ (C=N) and the degree of substitution (DS) for the Schiff base was 0.8~1.0 on the basis of C/N ratio in the elemental analyses (Table I).

The *N*-arylidene derivatives swelled in pyridine at ~50°, but the *N*-alkylidene derivatives did not swell. *O*-Acylation was performed on the *N*-arylidene derivatives by the reaction of stearoyl chloride. The *O*-acyl products were isolated in yields of 62~89% (Table II) and were insoluble in the common organic solvents examined. The presence of *O*-stearoyl groups in the products was detected by strong IR absorptions at 2910 and 2850 (CH), 1740 cm⁻¹ (C=O) and by weak or almost no IR absorption at 3500~3400 cm⁻¹ (OH). The DS value for *O*-stearoyl groups in the products varied from 0.82 to 1.91 depending on the Schiff-base groups. Almost complete *O*-acylation was found in 9. A higher DS value for the *O*-stearoyl group can be obtained with the larger Schiff-base groups such as 9, because of its conformation of less steric hindrance of the free hydroxyl groups.⁶⁾

The Schiff-base groups were selectively removed by treatment with dilute hydrochloric acid in ethanol at room temperature, and essentially no release of *O*-stearoyl groups was found in this treatment. The *O*-stearoyl derivatives of chitosan were isolated as the hydrochloride salt in up to 92% yield (Table III).

EXPERIMENTAL

General. Chitosan was prepared from Flonac N (crab

TABLE I. DATA FOR SOME NEW *N*-ALKYLIDENE AND *N*-ARYLIDENE DERIVATIVES OF CHITOSAN

Alkylidene or arylidene group	Yield (%)	DS ^a	Formula ^b	Calcd. (%)				Found (%)			
				C	H	N	Cl	C	H	N	Cl
1	82	0.81	C ₆ H ₉ NO ₄ (C ₄ H ₆) _{0.81} (H ₂) _{0.19} ·0.80H ₂ O	50.91	7.91	6.46		51.16	7.63	6.44	
2	85	0.81	C ₆ H ₉ NO ₄ (C ₄ H ₆) _{0.81} (H ₂) _{0.91} ·0.80H ₂ O	50.96	7.33	6.43		50.93	7.35	6.46	
3	92	0.81	C ₆ H ₉ NO ₄ (C ₇ H ₄ Cl ₂) _{0.81} (H ₂) _{0.18} ·0.12H ₂ O	48.28	4.44	4.80	19.91	48.59	4.38	4.81	19.63
4	92	0.95	C ₆ H ₉ NO ₄ (C ₇ H ₄ Cl ₂) _{0.95} (H ₂) _{0.25} ·0.18H ₂ O	48.45	4.26	4.47	21.15	48.56	4.18	4.36	20.86
5	92	0.95	C ₆ H ₉ NO ₄ (C ₇ H ₄ Cl ₂) _{0.95} (H ₂) _{0.05} ·0.18H ₂ O	48.45	4.26	4.47	21.15	48.41	4.24	4.50	21.07
6	87	0.92	C ₆ H ₉ NO ₄ (C ₉ H ₈) _{0.92} (H ₂) _{0.08} ·0.67H ₂ O	61.64	6.47	5.04		61.56	6.50	5.05	
7	89	0.90	C ₆ H ₉ NO ₄ (C ₈ H ₈ O) _{0.90} (H ₂) _{0.10} ·0.55H ₂ O	57.16	6.36	5.05		57.11	6.30	5.04	
8	94	1.00	C ₁₇ H ₁₇ NO ₄ ·0.43H ₂ O	66.51	5.82	4.56		66.54	6.03	4.64	
9	94	1.00	C ₁₇ H ₁₇ NO ₄ ·0.43H ₂ O	66.51	5.82	4.56		66.57	5.83	4.59	

^a Degree of substitution; the value is based on elemental analyses, as shown in the formula.^b The repeating unit is shown.TABLE II. DATA FOR SOME STEAROYL DERIVATIVES OF *N*-ARYLIDENECHITOSANS

<i>N</i> -Aryliden group	Yield (%)	DS ^a for stearoyl group	Formula ^b	Calcd. (%)				Found (%)			
				C	H	N	Cl	C	H	N	Cl
3	n.d. ^c	1.22	C ₆ H ₇ NO ₄ (C ₇ H ₄ Cl ₂) _{0.56} (C ₁₈ H ₃₅ O) _{1.22} (H) _{1.66} ·0.57H ₂ O	65.52	9.44	2.40	6.79	65.39	9.81	2.42	6.74
4	69	1.05	C ₆ H ₇ NO ₄ (C ₇ H ₄ Cl ₂) _{0.80} (C ₁₈ H ₃₅ O) _{1.05} (H) _{1.35} ·1.43H ₂ O	61.85	8.71	2.37	9.58	61.90	8.61	2.42	9.61
5	n.d.	1.53	C ₆ H ₇ NO ₄ (C ₇ H ₄ Cl ₂) _{0.82} (C ₁₈ H ₃₅ O) _{1.53} (H) _{0.71} ·0.90H ₂ O	66.11	9.37	1.96	8.15	66.09	9.43	1.90	8.16
6	n.d.	1.67	C ₆ H ₇ NO ₄ (C ₉ H ₈) _{0.80} (C ₁₈ H ₃₅ O) _{1.67} (H) _{0.77} ·0.90H ₂ O	72.80	10.51	1.96		72.81	10.56	1.92	
7	89	0.84	C ₆ H ₇ NO ₄ (C ₈ H ₈ O) _{0.50} (C ₁₈ H ₃₅ O) _{0.84} (H) _{2.16} ·1.80H ₂ O	63.32	9.76	2.94		63.28	9.75	2.94	
8	71	1.46	C ₆ H ₇ NO ₄ (C ₁₁ H ₈) _{0.60} (C ₁₈ H ₃₅ O) _{1.46} (H) _{1.34} ·1.81H ₂ O	70.15	10.27	2.10		70.14	10.29	2.11	
9	62	2.00	C ₆ H ₇ NO ₄ (C ₁₁ H ₈) _{0.70} (C ₁₈ H ₃₅ O) _{2.00} (H) _{0.60} ·1.53H ₂ O	72.94	10.62	1.71		72.83	10.62	1.70	

^a Degree of substitution; the value is based on elemental analyses, as shown in the formula.^b The repeating unit is shown.^c Not determined.

TABLE III. DATA FOR SOME STEAROYLCHITOSAN HYDROCHLORIDE SALTS

Sample ^a	Yield (%)	DS ^c for stearoyl group	Formula ^b	Calcd. (%)			Found (%)		
				C	H	N	C	H	N
A	85	1.05	C ₆ H ₉ NO ₄ (C ₁₈ H ₃₅ O) _{1.05} (H) _{0.90} (HCl) _{0.90} ·1.99H ₂ O	58.68	10.20	2.75	58.91	10.01	2.75
B	92	1.53	C ₆ H ₉ NO ₄ (C ₁₈ H ₃₅ O) _{1.53} (H) _{0.47} (HCl) _{1.00} ·1.72H ₂ O	63.51	10.40	2.21	63.56	10.53	2.21
C	79	0.82	C ₆ H ₉ NO ₄ (C ₁₈ H ₃₅ O) _{0.82} (H) _{1.18} (HCl) _{0.93} ·1.48H ₂ O	56.90	9.38	3.20	56.95	9.37	3.10
D	83	1.91	C ₆ H ₉ NO ₄ (C ₁₈ H ₃₅ O) _{1.91} (H) _{0.01} (HCl) _{1.00} ·1.66H ₂ O	66.04	10.73	1.91	66.24	10.76	1.91

^a A, B, C and D were prepared from *N*-3,4-dichlorobenzylidene (4), *N*-2,4-dichlorobenzylidene (5), 4-methoxybenzylidene (7) and 2-naphthylmethylene (9) derivatives, respectively.

^b Degree of substitution; the value is based on elemental analyses, as shown in the formula.

^c The repeating unit is shown.

shell chitosan produced from Kyōwa Yushi Co. Ltd., Chiba) by retreatment in 40% NaOH solution containing NaBH₄ (0.1 g/100 ml) at 110° for 5 hr. The products contained almost no acetyl groups as examined by ¹H-NMR (D₂O-DCO₂D, 8:2, v/v), had $[\alpha]_D^{17} - 10^\circ$ (*c* 0.53, 2% acetic acid), and agreed within 0.3% error with the theoretical values of chitosan in the elemental analyses.

The other analytical methods have been cited previously.⁶⁾

N-Alkylidene- and *N*-arylidene-chitosans. These derivatives were prepared by our previously reported methods.^{8,9)}

N-Arylidene-*O*-stearoylchitosans. Each of the dried, powdered samples (104~318 mg, 0.001 mol) of *N*-arylidenechitosans was swollen in a mixture of pyridine (15 ml) and chloroform (10 ml) by standing overnight at 50° with mechanical stirring. Stearoyl chloride (4 mol/GlcN) was added dropwise to the swollen sample in an ice bath. After standing at room temperature for 5 hr, the mixture was stirred at 50° for 24 hr and then heated at 115° for 2 hr. The reaction mixture was poured into acetone (100 ml) with vigorous stirring to give precipitates. The precipitates were collected by filtration, washed with acetone and ether, and dried over P₂O₅ *in vacuo* at 100° for 5 hr.

O-Stearoylchitosans. Each 20 mg of the *O*-stearoyl derivatives of *N*-arylidenechitosan was suspended in ethanol (50 ml) containing 0.25 M HCl. The suspension was stirred at room temperature for 18 hr, and the precipitates were collected by centrifugation, washed with ethanol several times, and dried over P₂O₅ *in vacuo* at 100° for 5 hr.

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