Communications to the Editor

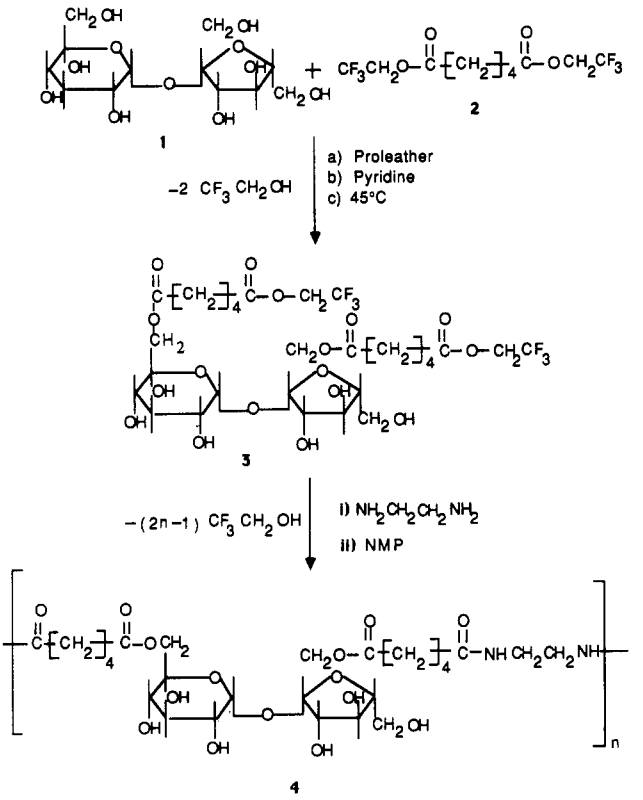
Chemoenzymatic Synthesis of Novel Sucrose-Containing Polymers

Typical chemical catalysts provide the chemoselectivity required for the preparation of high molecular weight poly(esters, amides, ols, acrylates, etc.) while enhancing reaction rates. There is an increasing need, however, to impart additional selectivity (e.g., regio- or stereoselectivity) in the synthesis of polymers.[1] This is particularly important in the synthesis of optically active polymers or the synthesis of linear polymers from monomers with functionality greater than 2. Recent advances in enzymatic catalysis in nonaqueous media have shown that enzymes are useful catalysts for the synthesis of polyesters and phenolic resins.[2,3] Unlike conventional chemical catalysts, enzymes can synthesize polymers with exquisite selectivity. For example, in previous work we have shown that an alkaline protease from a *Bacillus* sp. catalyzes the polycondensation of sucrose and an adipic acid derivative, which results in an alternating linear polyester containing sucrose in the backbone.[4] The high degree of regioselectivity provided by the enzyme enabled sucrose (with eight free hydroxyl groups) to react as if it were a diol. In this manner, no cross-linking was observed.[5]

The major drawback with enzymic polymer syntheses is the nearly universally slow rates of catalysis—the aforementioned sucrose polyester synthesis is complete only after 3 weeks. It occurred to us that a far more efficient approach would be to use enzymes only for the highly selective step(s) in polymer synthesis (such as monomer preparation) and to employ conventional chemical catalysts for the bulk polymer synthesis. In this work, we describe the chemoenzymatic synthesis of two novel polymers containing sucrose. The first is a polyesteramide with sucrose contained in the polymer backbone, while the second is a polyacrylate with sucrose as a side group. In both instances, the high degree of regioselectivity afforded by enzymatic catalysis is used to modify the sucrose before chemical polymerization is performed.

Poly(sucrose adipamide) Synthesis. Poly(sucrose adipamide) synthesis was carried out as shown in Scheme I. For the present case, 0.86 g (0.1 M) of sucrose was dissolved in 25 mL of pyridine containing 3.1 g (0.4 M) of bis(2,2,2-trifluoroethyl) adipate (**2**). Excess **2** was used to improve the yield of diester (relative to monoester). Higher sucrose concentrations could not be used due to the limited solubility of sucrose in pyridine; however, this reaction cannot be carried out in aqueous solutions as ester hydrolysis would be the predominant reaction. The enzyme used for this synthesis was Proleather, an alkaline protease from a *Bacillus* sp. Our previous work indicates that under these conditions this enzyme catalyzes the synthesis of sucrose 1′,6-bis(trifluoroethyl adipate).[4] The reaction was initiated by the addition of 15 mg/mL of Proleather,[6] and the suspension[7] was magnetically stirred under nitrogen at 150 rpm for 5 days at 45 °C. The reaction was terminated by filtering off the enzyme and evaporating the pyridine and unreacted **2**. The sucrose 1′,6-bis(trifluoroethyl adipate) (**3**) was purified by using silica gel chromatography with an eluent consisting of ethyl acetate/methanol/water (18:1.25:1). The diester **3** was obtained in 20% yield. No triester was formed.

Scheme I



Polymerization was carried out by adding 15 mg (0.125 M) of ethylenediamine to 0.19 g (0.125 M) of **3** in 2 mL of *N*-methylpyrrolidone (NMP). The solution was stirred at 35 °C for 24 h. Results of gel permeation (GPC) and thin-layer (TLC) chromatographies[8] indicated conversion of **3** was quantitative. A substantial byproduct (ca. 50%) was found to be sucrose monoadipate (the ester linkage at either the C-6 or C-1′ position), presumably formed by the reaction of ethylenediamine with the internal ester linkage between the sucrose and the adipate derivative. The resulting poly(sucrose adipamide) was recovered by evaporating NMP under vacuum at 50 °C. The product was washed with acetone and dried under vacuum at 45 °C. The polymer **4** was obtained in 48% recovered yield, 75 mg, and was a semicrystalline solid, mp = 218–225 °C; $[\alpha]^{25}_D$ = 16.8° (*c* 1, DMF), M_n = 4800, M_w = 8100. Anal. Calcd for $C_{26}H_{42}O_{15}N_2$ (per repeat unit): C, 50.2; H, 6.8; O, 38.6; N, 4.5. Found: C, 48.9; H, 6.8; O, 33.1; N, 6.6. The slight decrease in the ratio of O/N may be due to the formation of trace amounts of poly(ethyleneadipamide). The polymer **4** was insoluble in water but was soluble in a variety of polar organic solvents including pyridine, DMF, NMP, dimethyl sulfoxide, dimethylacetamide, methanol, and ethanol. Structural analysis of **4** by infrared spectroscopy[9] was consistent with incorporation of sucrose into the polymer backbone as shown in Scheme I.

Poly(sucrose acrylate) Synthesis. Polyacrylate synthesis with sucrose was performed as shown in Scheme II. Specifically, 3.42 g (0.1 M) of sucrose was dissolved in 100

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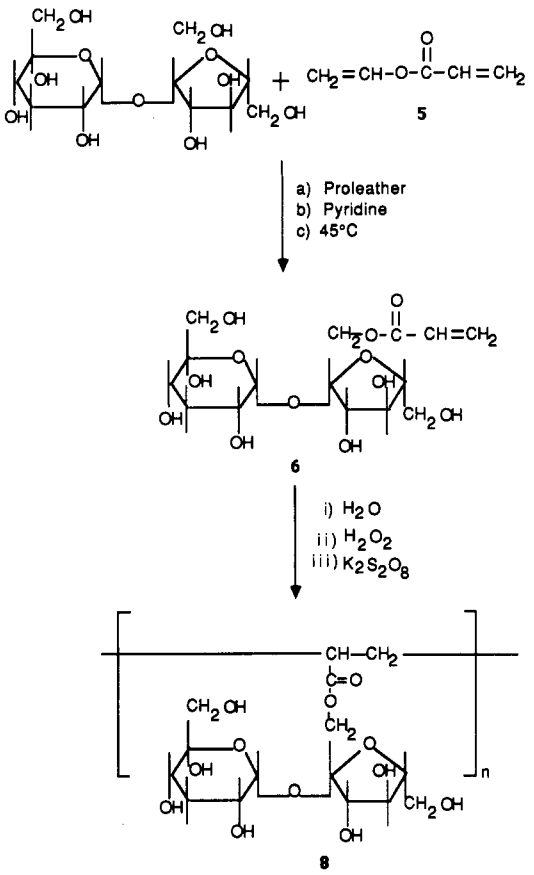
Scheme II



mL of pyridine containing 5.88 g (0.6 M) of vinyl acrylate (**5**). Hydroquinone (0.5% w/v) was added to inhibit polymerization of **5** during the sucrose acrylate synthesis. The reaction was initiated by addition of 15 mg/mL of Proleather, and the suspension was magnetically stirred under nitrogen at 150 rpm for 5 days at 45 °C, leading to ca. 60% conversion of sucrose. The reactions were terminated by filtering off the enzyme and evaporating the pyridine and unreacted **5**, and the product was purified and separated by silica gel chromatography with an eluent consisting of ethyl acetate/methanol/water (18:1.25:1). The sucrose monoester **6** was obtained in 28% yield, 1.10 g.[10] The ester was an amorphous solid, mp = 78 °C; $[\alpha]^{25}_D$ = 50.4° (*c* 1, H_2O).

Poly(sucrose acrylate) synthesis was carried out by dissolving 0.1 g (0.25 M) of **6** in 1 mL of H_2O, and the solution was sparged with N_2 for 10 min. Potassium persulfate (0.15%) and 0.2% hydrogen peroxide were added, and the solution was stirred at 25 °C for 24 h. The resulting poly(sucrose 1'-acrylate) was recovered by precipitation with acetone, filtered, and dried under vacuum at 45 °C. The polymer **8** was obtained in 80% recovered yield (80 mg) and was characterized as an amorphous solid, $[\alpha]^{25}_D$ = 38.3° (*c* 0.67, H_2O), M_n = 57 000, M_w = 91 000.[8] Anal. Calcd for $C_{15}H_{23}O_{12}$ (per repeat unit): C, 45.5; H, 6.1; O, 48.5. Found: C, 43.2; H, 5.9; O, 47.0. The polymer was soluble in a variety of polar organic solvents including water, DMF, and NMP. The structure of **8** is shown in Scheme II and is consistent with the IR results.[11]

In summary, we have demonstrated the chemoenzymatic synthesis of sucrose-containing polymers. The sugar resides either in the backbone of a poly(sucrose adipamide) or as a side group of a polyacrylate. We are currently investigating a number of potential applications of these novel polymers including water-absorbent materials, biocompatible polymers, and hydrogels. Chemoenzymatic synthesis represents a significant advantage over either purely chemical approaches (enhanced regioselectivity) or purely enzymatic routes (greater speed and, in the case of poly(sucrose acrylate), substantially higher molecular weights). This work represents another advance in the use of combined enzymatic and chemical syntheses in nonaqueous media.[12]

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References and Notes

(1) Chen, S. H.; Tsai, M. L. *Macromolecules* **1990**, *23*, 5055–5058. Wulff, G.; Dhal, P. K. *Macromolecules* **1990**, *23*, 4525–4527. Black, W. A. P.; Dewar, E. T.; Rutherford, D. *J. Chem. Soc.* **1963**, 4433–4439.
(2) Wallace, J. S.; Morrow, C. J. *J. Polym. Sci., Part A: Polym. Chem.* **1989**, *27*, 2553–2567. Margolin, A. L.; Crenne, J.-Y.; Klibanov, A. M. *Tetrahedron Lett.* **1987**, *28*, 1607–1610.
(3) Dordick, J. S.; Marletta, M. A.; Klibanov, A. M. *Biotechnol. Bioeng.* **1987**, *30*, 31–36. Ryu, K.; Stafford, D. R.; Dordick, J. S. *ACS Symp. Ser.* **1989**, *389*, 141–157.
(4) Patil, D. R.; Rethwisch, D. G.; Dordick, J. S. *Biotechnol. Bioeng.* **1991**, *37*, 639–646.
(5) Viswanathan, T.; Toland, A.; Liu, R.-Q. *J. Polym. Sci., Part C: Polym. Lett.* **1990**, *28*, 95–100.
(6) Proleather is an alkaline protease from a *Bacillus* sp. and was obtained as a crude powder from the Amano International Enzyme Co. (Troy, VA).
(7) Proleather, as with nearly all enzymes, is insoluble in pyridine.
(8) For poly(sucrose adipamide) characterization, GPC was performed with a Zorbax PSM-bimodal-S column (Du Pont; MW range from 500 to 1 × 10^6) with DMF (1 mL/min) as eluent. TLC was performed on silica plates with *n*-butanol/ethanol/water (5:3:2) as mobile phase. For poly(sucrose acrylate) characterization, GPC was performed with an Ultrahydrogel linear column (Waters Associates; MW range from 500 to 5 × 10^6) with 0.1 M $NaNO_3$ as eluent.
(9) IR results were as follows: –NH (3086, 3303, 1640 cm^{-1}); –OH (broad at 3400 cm^{-1}); C=O (1742 cm^{-1}); C–N (1437 cm^{-1}).
(10) Identification of this compound was performed with ^{13}C NMR in DMSO: δ 165.6 (C=O), 130.7 (H_2C=), 129.1 (HC=), 102.1 (C-2'), 91.5 (C-1), 82.7 (C-5'), 76.7 (C-3'), 74.1 (C-4'), 72.7 (C-3), 72.5 (C-5), 71.5 (C-2), 69.9 (C-4), 64.0 (C-1'), 62.1 (C-6'), 60.0 (C-6). The only shifts observed from that of sucrose are a downfield shift in C-1' (for sucrose, δ 62.1) and an upfield shift in C-2' (for sucrose δ 104.0). This indicates the acylation of only the C-1' hydroxyl.
(11) IR results were as follows: for sucrose 1'-acrylate, C=C (1646 cm^{-1}), C=O (1728 cm^{-1}), –OH (3416 cm^{-1}); for polymer, –C=O (1730 cm^{-1}), –OH (3399 cm^{-1}), disappearance of C=C absorption at 1646 cm^{-1}.
(12) Parida, S.; Dordick, J. S. *J. Am. Chem. Soc.* **1991**, *113*, 2253–2259. Riva, S.; Klibanov, A. M. *J. Am. Chem. Soc.* **1988**, *110*, 3291–3295.

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