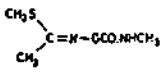
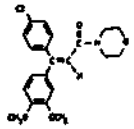
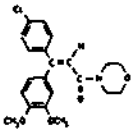
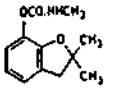
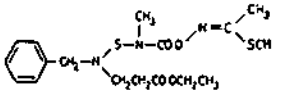
Table 1. The Structures of Peak in the Chromatograms

Chromatogram	Peaks	Commercial name	Chemical name	Structure
Figure 3	A	Methomyl	S-methyl N-(methyl-carbamoyloxy) thioacetimidate	
	B	Dimethomorph E	4-[3-(4-chlorophenyl)-3-(3,4-dimethoxyphenyl)acryloyl] morpholine	
	C	Dimethomorph Z	4-[3-(4-chlorophenyl)-3-(3,4-dimethoxyphenyl)acryloyl] morpholine	
Figure 4	A	Carbofuran	2,3-dihydro-2,2-dimethylbenzofuran -7-yl methylcarbamate	
	B	Alanycarb	Ethyl(Z)-N-benzyl-N-[[methyl(1-methyl thioethylideneamino-oxycarbonyl)amino]thio]-β-alaninate	

shown in Table 1.

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Chemoenzymatic Synthesis of (3R,5R)-3,6-diamino-5-hydroxyhexanoic Acid, the Amino Acid Moiety of (+)-Negamycin

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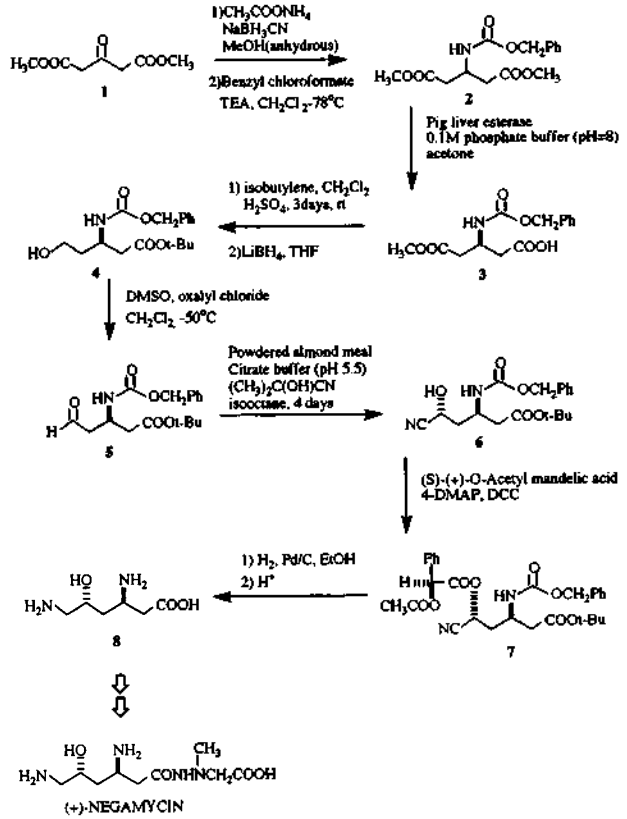
(+)-Negamycin was isolated from *Streptomyces purpeofuscus* in 1970.[1] It inhibits the growth of Gram-negative and Gram-positive bacteria.[1] (+)-Negamycin shows the misleading of genetic code and the inhibition of protein synthesis.[2] This compound contains two interesting structural features including lysine and hydrazide units with two asymmetric centers which served as an attractive target molecule for the stereoselective synthesis. Modification of the unnatural amino acid moiety of (+)-negamycin showed no antibacterial activity. This result showed that δ-hydroxy-β-amino lysine is a key moiety for the antibiotic activity. This antibiotic has been synthesized in both racemic[3] and optically active forms.[4] Naturally chiral starting materials were used for the total synthesis of (+)-negamycin.

Since the abilities of enzymes as chiral catalysts have been recognized for many years, we planned to control the chiral centers of the amino acid moiety of (+)-negamycin with enzymes. However, the stereochemistry of C(3) of (3R,



Scheme 1

5R)-3,6-diamino-5-hydroxyhexanoic acid has been controlled successfully with pig liver esterase.[4a,6] Here, we report the control of stereocenter of C(5) streochemistry of 3, 6-diamino-5-hydroxyhexanoic acid of (+)-negamycin by using mandelonitrile lyase (EC 4.2.1.0).

A chemoenzymatic procedure was taken as our synthetic strategy as shown in Scheme 1. By using Borch's method[7] the dimethyl β-oxoglutarate was reduced with CH_3COONH_4/ $NaBH_3CN$/ dry MeOH to give dimethyl β-aminoglutarate. The amino group was protected with the benzyloxycarbonyl group in 85% yield. Porcine liver esterase hydrolyzed **2** and (3R)-monomethyl ester **3** was formed in 90% yield.[6] The chiral half ester **3** with S configuration was treated with isobutylene-H_2SO_4 to produce *tert*-butyl ester. When $LiBH_4$ was used to reduce the N-benzyloxycarbonyl methyl t-butyl β-aminoglutarate, the tert-butyl ester group was stable during the methyl ester reduction. The alcohol **4** was treated with oxalyl chloride/DMSO and Et_3N at -50 °C to give aldehyde **5** in 55% yield.

At this point, we introduced the second chiral center at the C(5) with mandelonitrile lyase (EC 4.2.1.0). A crude extract from ground almond in an aqueous buffer or ground almond meal itself have been used as a catalyst to produce optically active (R)-cyanohydrin.[8] Powdered almond meal as a cheap catalyst has been used for the synthesis of optically acitve aliphatic cyanohydrins with acetone cyanohydrin as a transcyanation agent. The reaction was carred out with aldehyde **5** (0.3 mmol), acetone cyanohydrin(0.46 mmol), defatted almond meal (0.2 g) as a catalyst in citrate buffer (pH=5.5, 2.5 mL) and isooctane (20 mL) at room temperature for 4 days. This method allowed us to obtain

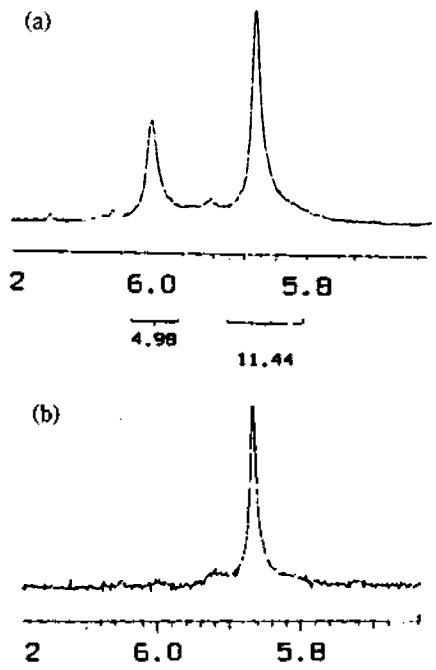


Figure 1. ^1H NMR (200 MHz) spectra (methine region) of the (S)-(+)-O-Acetylmandelic ester of 6 obtained by mandelonirile lyase. (a) before chromatographic purification. (b) after chromatographic purification.

the (R) enriched cyanohydrin **6** in 65% yield. In order to assign the absolute configuration and determine chiral purity, the cyanohydrin **6** was converted into (S)-(+)-O-acetylmandelate ester with (S)-(+)-O-actyl mandelic acid, DCC and 4-DMAP.[9] The procedure gave a 42% yield. The ^1H NMR spectrum of (S)-(+)-O-acetylmandelate ester **7** showed that the product was a mixture of (R)- and (S)-cyanohydrin in the ratio of 7 : 3 (Figure 1). One of the possible explanations for the low optical purity of the cyanohydrin is longer reaction time. During the longer reaction time, the chemical reaction leads to the formation of racemate.[8a] However, the (R)-cyanohydrin ester **7** was easily purified with column chromatography (silicalgel, 1.0 cm × 60 cm, R_f =0.5, EtOAc : *n*-Hexane=3 : 7) (Figure 1). Hydrogenolysis (H_2, 10% Pd-C, EtOH, HCl) of **7** afforded **8**. The *N*-protected group and mandelate ester were cleavaged in the acidic condition of the hydrogenolysis. The crude product was purified by anion exchange resin column chromatography [2.2 × 20 cm, Bio-Rad (AG 1-8) 200-400 mesh, formate form, 2 N formic acid] to give (3R,5R)-3,6-diamino-5-hydroxyhexanoic acid in 42% yield.

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10. ^{1}H NMR (200 MHz, $CDCl_3$) δ **3**: 2.73 (m, 4H), 3.67 (s, 3H), 4.4 (m, 1H), 5.9 (s, 2H), 5.62 (bd, *J*=9.1 Hz, 1H), 7.35 (s, 5H). **4**: 1.41 (s, 9H), 2.5 (dq, *J*=5.1 Hz, 15.7 Hz, 2H), 3.2 (bs, 1H), 3.62 (m, 2H), 3.95 (m, 1H), 5.1 (s, 2H), 5.7 (bd, *J*=9.1 Hz, 1H), 7.35 (s, 5H). **5**: 1.4 (s, 9H), 2.5 (d, *J*=5.86 Hz, 2H), 2.76 (t, *J*=5.86 Hz, 2H), 4.4 (m, 1H), 5.1 (s, 2H), 5.9 (bd, *J*=8.0 Hz, 1H), 7.3 (s, 5H), 9.72 (s, 1H). **6**: 1.45 (s, 9H), 2.0 (m, 2H), 2.55 (dq, *J*=4.9 Hz, 16.3 Hz, 2H), 4.19 (m, 1H), 4.5 (m, 1H), 4.67 (bs, 1H), 5.12 (s, 2H), 5.95 (bd, *J*=8.4 Hz, 1H) 7.35 (s, 5H). 7: 1.4 (s, 9H), 2.0 (m, 2H), 2.2(s, 3H), 2.7 (m, 2H), 4.2 (m, 1H), 5.12 (s, 2H), 5.46 (t, *J*= 7.8 Hz, 1H), 5.85 (s, 1H), 7.3-7.45 (m, 10H). ^{1}H NMR (500 MHz, D_2O) δ **8**: 1.8 (m, 1H), 2.1 (m, 1H), 2.7 (m, 2H), 2.87 (dd *J*=4.51 Hz, 17.95 Hz, 1H), 3.2 (d, *J*= 12.83 Hz, 1H), 3.37 (dd, *J*=4.04 Hz, 12.80 Hz, 1H), 4.1 (m, 1H). ES-MS (negative) **8**: m/z 161 $[M-H]^-$.

Itaconate Copolymer Bearing the Second-Order Nonlinear Optical Chromophores in Both Side Chains

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The nonlinear optical (NLO) properties of polymeric materials have been highlighted as a subject of numerous investigations for application in electro-optic (EO) and photonic devices over a decade.[1-3] Particularly, side chain polymeric structures drew much interest owing to their ease of synthesis and processibility. We also could find a lot of new features about the nonlinear optical active copolymers recently.[1-3] Resulting from our synthetic strategy, we could optimize our polymeric structure to maximize the second-order NLO effect employing a new monomer unit such as itaconate. Itaconic ester is a very promising monomer that can contain two NLO chromophores in one repeating unit. It was found that the itaconate is capable of building the special copolymer with commonly used comonomers such as methylmethacrylate, styrene, etc.[4] Assuming that the noncentrosymmetry could be induced under poling process practically, the second-order nonlinear optical coefficient was known to be directly proportional to the concentration of the active chromophore. In this respect, we conclude our choice of itaconate as the most favorable NLO monomer. The purpose of this work is how much improvement of NLO effect we can achieve with the two fold increase of the chromophore density in the copolymer. Therefore, we introduced our used chromophore of nitrostilbene in these structures by way of direct esterification and Mitsunobu reaction easily. Even though we employed several kinds of comonomers, we only reported here about the synthesis and NLO properties of the itaconate copolymers with methyl methacrylate.

In this study, we prepared itaconate monomers bearing second-order NLO-active dyes as shown in Scheme 1. 2-Methylene-succinic acid bis-[2-(methyl-{4-[2-(4-nitrophenyl)-vinyl]-phenyl}-amino)-ethyl] ester (monomer I) was synthesized by Mitsunobu reaction using diisopropylazodicarboxylate (DIAD) and triphenylphosphine in THF (49.9% yield).[5] We also obtained 2-methylene-succinic acid bis-(2-{4-[2-(4-nitrophenyl)-vinyl]-phenoxy}-hexyl) ester (monomer II) by simple direct esterification using a Dean-stark apparatus under refluxing the reaction mixture in toluene with H_2SO_4/p-toluensulfonic acid as acid catalysts (81.7% yield).[6]

We carried out radical copolymerization of methylmethacrylate (MMA) and itaconate monomer in freshly distilled N-methyl-2-pyrrolidinone (NMP) (Figure 1). A mixture of itaconate monomer I (1.46 g, 2.11 mmole), MMA