Biodegradable Polymer Made of Styrene and *N*-Benzyl-4-vinylpyridinium Chloride

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ABSTRACT: A copolymer of styrene with *N*-benzyl-4-vinylpyridinium chloride (BVP), poly(styrene-*co*-N-benzyl-4-vinylpyridinium chloride) (PST-*co*-BVP), was degradable by activated sludge in soil when the oligo-styrene portion was sufficiently small. The degradation of the equimolar copolymer followed first-order kinetics when the polymer sample was 1.0 or 0.5 g/kg and gave a half-life of 5.6 days. The degradation of PST-*co*-BVP with a reduced BVP content did not follow first-order kinetics under the aforementioned conditions but appeared to follow the kinetics when the amount of the polymer sample was sufficiently small. Under the ultimate conditions, the half-life of PST-*co*-BVP that contained 10.6 mol % BVP was estimated to be 12.5 days, and the half-life of PST-*co*-BVP that contained 5 mol % BVP was expected to be 30–40 days. The incorporation of 5 mol % BVP appeared sufficient for making PST-*co*-BVP substantially biodegradable if we did not expect exceptionally rapid degradation. PST-*co*-BVP was different from conventional polystyrene but possessed biodegradability. Random scission of the main chain much predominated over uniform scission from the end of the polymer chain in the biodegradation of PST-*co*-BVP. The cleavage of the main chain at BVP appeared predominant over that of oligo-styrene. © 2006 Wiley Periodicals, Inc. J Appl Polym Sci 100: 554–559, 2006

Key words: biodegradable; polystyrene; functionalization of polymers

INTRODUCTION

Biodegradable polymers have received increasing attention.[1–6] The development of these polymers, however, has mainly been focused on their biodegradability, and the polymers do not always possess necessary characteristics for the wide use of synthetic polymers. Therefore, it is favorable if general synthetic polymers are made substantially biodegradable by partial modification of the chemical structure.

The incorporation of a biodegradable unit into the main chain is effective because oligomers are biodegradable, unlike high-molecular-weight polymers.[7–11] The connection of oligomers with biodegradable chemical structures forms biodegradable polymers. Matsumura et al.[6] made poly(carboxylic acid) biodegradable by the incorporation of vinyl alcohol into the main chain. Previously, we reported the digestion of crosslinked poly(*N*-benzyl-4-vinylpyridinium chloride) by activated sludge,[12] and made poly(methyl methacrylate),[12] poly(vinyl acetate),[13] and polyacrylonitrile[14] biodegradable by the incorporation of *N*-benzyl-4-vinylpyridinium chloride (BVP) into the main chain.

In this work, we attempted to develop biodegradable polymers made of styrene (ST), and we investigated biodegradability of copolymers of ST with BVP [poly-(styrene-*co*-N-benzyl-4-vinylpyridinium chloride) (PST-*co*-BVP)]. Although the dimer of ST is biodegradable,[11] the biodegradation of polystyrene (PST) is extremely difficult, and we investigated the biodegradability of PST-*co*-BVP systematically with six types of copolymers in molar ratios of 1:1–9:1.

EXPERIMENTAL

Materials

We washed ST with a 5% aqueous sodium hydroxide solution, and this was followed by drying with calcium hydride. We purified 4-vinylpyridine (VP) by distillation under a reduced pressure. We used other chemicals without further purification. We obtained activated sludge from Sewage Works of Hikone City, Japan, immediately before biodegradation, and we washed it three times with sterilized physiological saline. We obtained soil from the shore of Lake Biwa and washed it with water with a Soxhlet extraction apparatus for 48 h. After it was dried, we washed the soil with ethyl acetate with a Soxhlet extraction apparatus for 15 h. After it was dried, we sterilized the soil by autoclaving at 121°C for 180 min. After 2 days, we repeated the autoclaving and dried it to a constant

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weight before use. Artificial sewage was prepared according to a literature recipe[15] and used to assist with the biodegradation. Peptone (6.0 g), meat extract (4.0 g), urea (1.0 g), sodium chloride (0.30 g), potassium chloride (0.14 g), calcium chloride (0.14 g), magnesium sulfate (0.10 g), and sodium hydrogen phosphate (1.0 g) were dissolved in 1.0 L of deionized water, and the pH was adjusted to 8.5. The chemical oxygen demand (COD) of this undiluted solution was about 10,000 mg/L. We diluted this solution with an appropriate amount of deionized water to prepare artificial sewage of the prescribed COD concentration.

Copolymer of ST with BVP (PST-*co*-BVP)

The free-radical copolymerization of ST with VP was performed in a 500-mL, three-necked flask equipped with a mechanical stirrer, a reflux condenser, and a gas inlet with 2,2'-azobisisobutyronitrile as an initiator in toluene at 80°C for 24 h. After the reaction, we removed volatile materials by evaporation with a rotary evaporator and dried them under a reduced pressure. We dissolved the residue that contained the copolymer of ST with VP [poly(styrene-*co*-4-vinylpyridine) (PST-*co*-VP)] into a mixed solvent of tetrahydrofuran and ethanol (in a weight ratio of 1:1). We added an equimolar amount of benzyl chloride to VP that was contained in PST-*co*-VP, and we allowed them to react at 70°C for 24 h. After the reaction, we removed volatile materials by evaporation with a rotary evaporator and added hexane to the residue to isolate PST-*co*-BVP. We dried the isolated polymer to a constant weight under a reduced pressure at room temperature. We ascertained the chemical structure of PST-*co*-BVP by elemental analyses performed at the Elementary Analyses Center of Kyoto University. The intrinsic viscosity of PST-*co*-BVP was determined at 25°C in ethanol or dimethylformamide (DMF) that contained 10 g/L $MgCl_2 \cdot 6H_2O$. We performed gel permeation chromatography (GPC) with a Shimadzu LC-10AD high-performance-liquid-chromatography system with an SPD-10A ultraviolet–visible detector, a CTO-10A column oven, and a Chromatopac C-R7A-plus data processor (Shimadzu Corp., Kyoto, Japan). Shodex GF310HQ and GF510HQ (Showa Denko, Tokyo, Japan) were used as columns, and we used DMF that contained 10 g/L $MgCl_2 \cdot 6H_2O$ as an eluate.

Degradation of PST-*co*-BVP by a treatment with activated sludge in soil

We performed the degradation of PST-*co*-BVP by a treatment with activated sludge in soil. A prescribed amount of the polymer was dissolved in a mixed solvent of ethanol and tetrahydrofuran (in a weight ratio of 1:1) and mixed with the washed soil, and it was dried to a constant weight under a reduced pres-

sure at room temperature to remove the solvents from the test soil.

We mixed a prescribed amount of the washed activated sludge and artificial sewage with the test soil that contained the polymer sample. We added the artificial sewage, 0.2 g in COD per gram of the polymer sample, to assist with the biodegradation. The total amounts of water and activated sludge were settled to 200 mL/kg and 14.2 g/kg (in wet weight), respectively. We allowed the mixture to stand at room temperature. After a prescribed time, we recovered the remaining polymer by extraction with a mixture of ethanol and tetrahydrofuran in a weight ratio of 1:1 for 48 h with a Soxhlet extraction apparatus. We removed fine soil particles contained in the extractive by centrifugation at 2000g for 20 min. We repeated this centrifugation three times and removed solvents by evaporation with a rotary evaporator. We added ethyl acetate to the residue to isolate the recovered polymer sample, and we dried the isolated polymer to a constant weight under a reduced pressure at room temperature. Before these experiments in biodegradation, we performed a series of experiments to ascertain the reliability of the recovery procedure.

RESULTS AND DISCUSSION

Because the biodegradation of PST is difficult, we suspected that the biodegradability of PST-*co*-BVP mainly depended on the nature of the oligo-ST portion contained in PST-*co*-BVP, and we investigated the degradation of PST-*co*-BVP systematically, starting from the equimolar copolymer.

We performed the degradation of PST-*co*-BVP by a treatment with activated sludge in soil at room temperature. After the treatment, we recovered the residual polymer by extraction, which was followed by the removal of the solvents and precipitation of the recovered polymer by the addition of ethyl acetate to the residue. We dried the recovered polymer to a constant weight under a reduced pressure. Low-molecular-weight oligomers and other organic materials soluble in ethyl acetate were not included in the residual weight. Figures 1 and 2 show the time course of the weight reduction during the treatment. Here, the residual weight is the total weight of the polymeric materials recovered after the treatment. Figures 3 and 4 show the time course of $t \ln 2/\ln[a/(a - x)]$ during the treatment. Here, t is the treatment time (days), a is the initial amount of the polymer (g/kg), and x is the amount of the polymer that disappears before t (g/kg).

Biodegradation of an equimolar copolymer of ST with BVP (PST-*co*-BVP$_1$)

An equimolar copolymer of ST with BVP (PST-*co*-BVP$_1$) contained 43.1 mol % ST, 49.1 mol % BVP, and
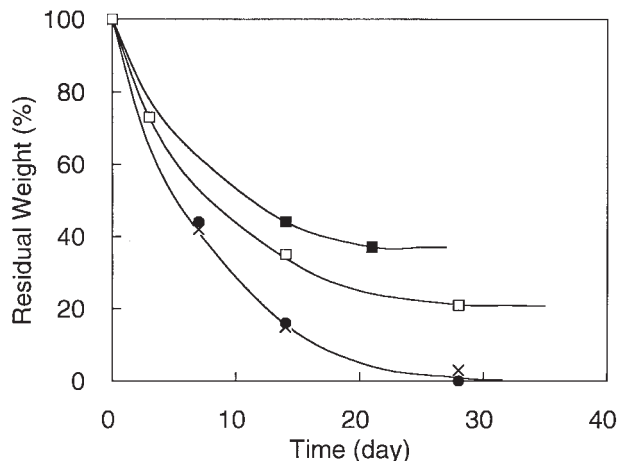
Figure 1 Time course of the weight reduction of PST-*co*-BVP during the treatment with activated sludge in soil at room temperature (the amount of the polymer sample used for the treatment is given in parentheses): (×) PST-*co*-BVP$_1$ (0.5 g/kg), (●) PST-*co*-BVP$_1$ (1.0 g/kg), (□) PST-*co*-BVP$_2$ (0.5 g/kg), and (■) PST-*co*-BVP$_2$ (1.0 g/kg).

7.8 mol % VP. The intrinsic viscosity of PST-*co*-BVP$_1$, determined in ethanol containing 10 g/L of MgSO$_4$ · 6H$_2$O at 35°C, was 0.26 dL/g.

The closed circles and cross marks in Figure 1 show the time course of the weight reduction of PST-*co*-BVP$_1$ during the treatment with activated sludge in soil. Here, the cross marks and closed circles show the results obtained with 0.5 and 1.0 g/kg PST-*co*-BVP$_1$, respectively. The time courses were very close.

The closed circles and cross marks in Figure 3 show the time course of $t \ln 2/\ln[a/(a - x)]$ during the treatment of PST-*co*-BVP$_1$. The cross marks and closed circles show the results obtained with 0.5 and 1.0 g/kg
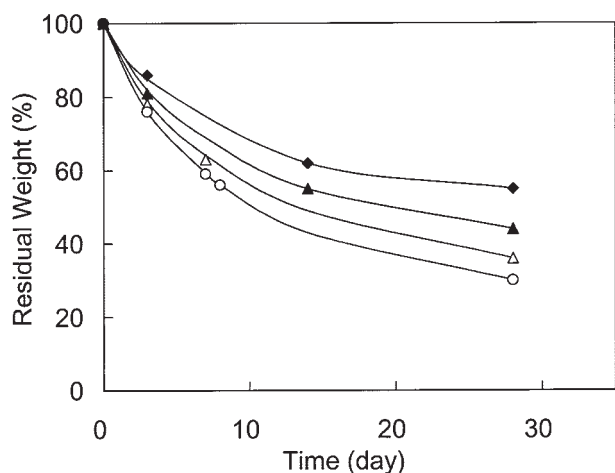
Figure 2 Time course of the weight reduction of PST-*co*-BVP during the treatment with activated sludge in soil at room temperature (the amount of the polymer sample used for the treatment was 0.5 g/kg): (○) PST-*co*-BVP$_3$, (△) PST-*co*-BVP$_4$, (▲) PST-*co*-BVP$_5$, and (◆) PST-*co*-BVP$_8$.
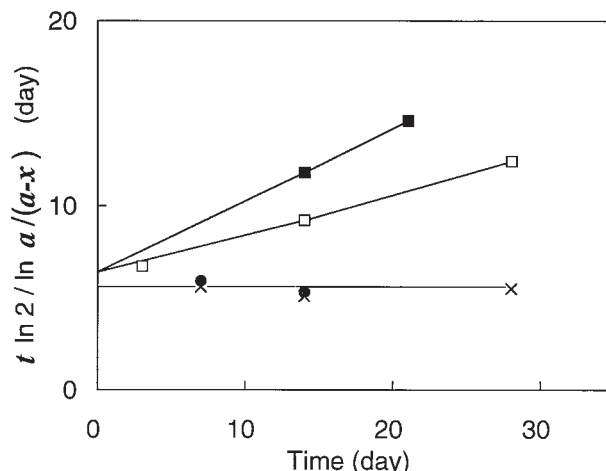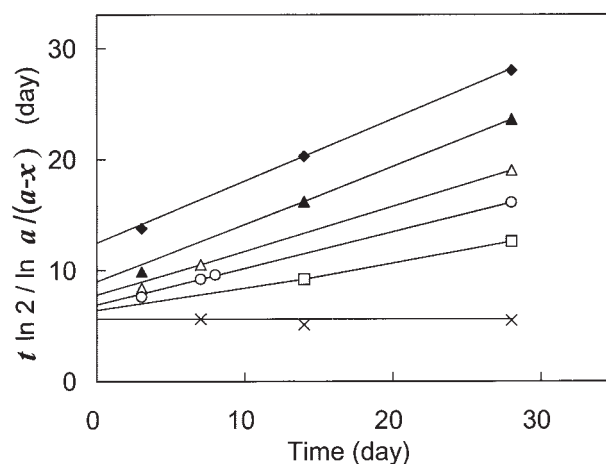

Figure 3 Time course of $t \ln 2/\ln[a/(a - x)]$ during the treatment of PST-*co*-BVP with activated sludge in soil at room temperature (the amount of the polymer sample used for the treatment is given in parentheses): (×) PST-*co*-BVP$_1$ (0.5 g/kg), (●) PST-*co*-BVP$_1$ (1.0 g/kg), (□) PST-*co*-BVP$_2$ (0.5 g/kg), and (■) PST-*co*-BVP$_2$ (1.0 g/kg).

PST-*co*-BVP$_1$, respectively. These time courses were very close and did not depend on t.

These experimental results indicated that the biodegradation of PST-*co*-BVP$_1$ followed first-order kinetics under the conditions. The addition of 1.0 g/kg PST-*co*-BVP$_1$ appeared not too much and was within the ability of activated sludge used for the treatment. The half-life of PST-*co*-BVP$_1$ was 5.6 days.

Biodegradation of a copolymer of ST with BVP in a molar ratio of 2:1 (PST-*co*-BVP$_2$)

A copolymer of ST with BVP in a molar ratio of 2:1 (PST-*co*-BVP$_2$) contained 63.0 mol % ST, 35.5 mol %

Figure 4 Time course of $t \ln 2/\ln[a/(a - x)]$ during the treatment with activated sludge in soil at room temperature (the mount of the polymer sample used for the treatment was 0.5 g/kg): (×) PST-*co*-BVP$_1$, (□) PST-*co*-BVP$_2$, (○) PST-*co*-BVP$_3$, (△) PST-*co*-BVP$_4$, (▲) PST-*co*-BVP$_5$, and (◆) PST-*co*-BVP$_8$.

BVP, and 1.5 mol % VP. The molar ratio of ST to BVP was about 1.8; that is, the average number of ST units in the oligo-ST portion was 1.8. The weight-average molecular weight of PST-*co*-BVP$_2$ determined by GPC analysis was 116,000.

We performed the degradation of PST-*co*-BVP$_2$ in the same manner. The open and closed squares of Figure 1 show the time course of the weight reduction during the treatment; the concentrations of PST-*co*-BVP$_2$ were 0.5 and 1.0 g/kg, respectively. Weight reduction was faster when a smaller amount of PST-*co*-BVP$_2$ was used (open squares); this made a sharp contrast with PST-*co*-BVP$_1$, for which the weight reduction did not depend on the amount of the polymer, as shown by the cross marks and closed circles in Figure 1. This marked difference between PST-*co*-BVP$_1$ and PST-*co*-BVP$_2$ may have derived from the more difficult biodegradation of PST-*co*-BVP$_2$ than PST-*co*-BVP$_1$. The addition of 0.5 g/kg PST-*co*-BVP$_2$ appeared too much with respect to the degradation ability of activated sludge.

The open squares and closed squares of Figure 3 show the time course of $t \ln 2 / \ln[a/(a - x)]$ for PST-*co*-BVP$_2$; the concentrations of the polymer were 0.5 and 1.0 g/kg, respectively. Obviously, the biodegradation of PST-*co*-BVP$_2$ did not follow first-order kinetics under the conditions. This result indicates that the addition of 0.5 g/kg PST-*co*-BVP$_3$ was still overloading with respect to the ability of the activated sludge contained in the test soil. However, the slope of the linear relationship shown in Figure 3 was lower in the case of open squares, that is, with a lower amount of PST-*co*-BVP$_2$. This result suggests that the linear relationship shown in Figure 3 will become independent of the treatment time when the amount of the polymer sample is sufficiently small. In addition, the linear relationship shown by open squares coincides with that shown by closed squares at the treatment time of zero. This result suggests that the linear relationship shown by Figure 3 may give 6.4 days as the half-life under the ultimate conditions when a sufficiently small amount of PST-*co*-BVP$_3$ is used. On the basis of this assumption, we evaluated the half-life of PST-*co*-BVP$_2$ to 6.4 days when the amount of PST-*co*-BVP$_2$ was sufficiently small and did not exceed the degradation ability of microorganisms contained in the activated sludge in test soil.

Biodegradation of a copolymer of ST with BVP in a molar ratio of 3 : 1 (PST-*co*-BVP$_3$)

A copolymer of ST with BVP in a molar ratio of 3 : 1 (PST-*co*-BVP$_3$) contained 73.5 mol % ST, 26.5 mol % BVP, and 0.0 mol % VP. The molar ratio of ST to BVP was about 2.8; that is, the average number of ST in the oligo-ST portion was 2.8. The weight-average molec-

ular weight of the copolymer determined by GPC analysis was 105,000.

Because we anticipated poorer biodegradability for PST-*co*-BVP$_3$ than for PST-*co*-BVP$_2$, we performed the biodegradation of PST-*co*-BVP$_3$ with a 0.5 g/kg polymer sample. The open circles of Figures 2 and 4 summarize the experimental results. The open circles of Figure 4 show an increase in $t \ln 2 / \ln[a/(a - x)]$ with t, and the half-life of PST-*co*-BVP$_3$ was evaluated to 6.9 days by the extension of the linear relation to time zero. This half-life is reasonable under the ultimate conditions in which the amount of the polymer is sufficiently small with respect to the degradation ability of the microorganisms in the soil.

Biodegradation of a copolymer of ST with BVP in a molar ratio of 4 : 1 (PST-*co*-BVP$_4$)

A copolymer of ST with BVP in a molar ratio of 4:1 (PST-*co*-BVP$_4$) contained 80.2 mol % ST, 19.4 mol % BVP, and 0.4 mol % VP. A molar ratio of ST to BVP was about 4.2; that is, the average number of ST units in the oligo-ST portion was 4.2. The weight-average molecular weight of PST-*co*-BVP$_4$ determined by GPC analysis was 79,000.

We performed the degradation of PST-*co*-BVP$_4$ with a 0.5 g/kg polymer sample. The open triangles of Figures 2 and 4 summarize the results. The open triangles of Figure 4 show an increase in $t \ln 2 / \ln[a/(a - x)]$ with t, and the half-life of PST-*co*-BVP$_4$ was evaluated to be 7.8 days by the extension of the linear relation to time zero. This half-life is reasonable under the ultimate conditions when the amount of the polymer sample is sufficiently small.

Biodegradation of PST-*co*-BVP containing a smaller amount of BVP (PST-*co*-BVP$_5$ and PST-*co*-BVP$_8$)

A copolymer of ST with BVP in a molar ratio of 5 : 1 (PST-*co*-BVP$_5$) contained 84.0 mol % ST, 15.7 mol % BVP, and 0.4 mol % VP. The molar ratio of ST to BVP was about 5.4; that is, the average number of ST units in the oligo-ST portion was 5.4. The weight-average molecular weight of PST-*co*-BVP$_5$ determined by GPC analysis was 73,900. A copolymer of ST with BVP in a molar ratio of 8 : 1 (PST-*co*-BVP$_8$) contained 88.0 mol % ST, 10.6 mol % BVP, and 1.5 mol %VP. The molar ratio of ST to BVP was about 8.3; that is, the average number of ST units in the oligo-ST portion was 8.3. The weight-average molecular weight of PST-*co*-BVP$_8$ determined by GPC analysis was 264,000.

The closed triangles and closed rhombuses of Figures 2 and 4 summarize the experimental results for PST-*co*-BVP$_5$ and PST-*co*-BVP$_8$, respectively. The half-lives of PST-*co*-BVP$_5$ and PST-*co*-BVP$_8$ were 9.0 and 12.5 days, respectively, by the extension of the linear
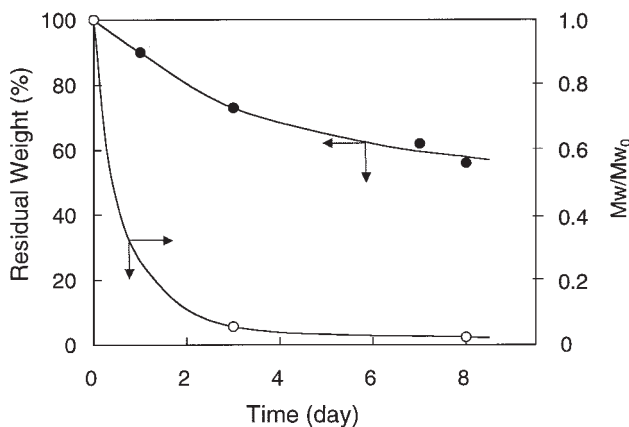
Figure 5   Time course of (●) the residual weight and (○) the rate of reduction of the molecular weight of the recovered polymer during the treatment of PST-*co*-BVP$_3$ with activated sludge in soil at room temperature. M_w is the molecular weight of the recovered polymer; M_{w0} is the molecular weight of the polymer before the biological treatment. The amount of the polymer sample used for the treatment was 0.5 g/kg.

relation of the closed triangles and closed rhombuses of Figure 4 to time zero, respectively. These half-live are reasonable under the ultimate conditions when the amount of the polymer sample is within the degradation ability of the microorganisms contained in the test soil.

Mode of chain scission of PST-*co*-BVP$_3$ during biodegradation

We investigated the mode of chain scission during the treatment of PST-*co*-BVP with activated sludge in detail with PST-*co*-BVP$_3$ as an example. Because recovered PST-*co*-BVP did not exhibit a perceivable change in the elementary composition, we estimated the rate of chain shortening of the recovered polymer simply on the basis of the rate of the reduction of the molecular weight, that is, the division of the molecular weight of the recovered polymer by that before the treatment.

When the first scission occurs at the center of the polymer chain, the molecular weight of the recovered polymer may dramatically decrease to half, but the reduction of the gravimetric weight may be negligible at this stage. Similarly, when random scission inside the polymer chain predominates, the reduction of the molecular weight of the recovered polymer may predominate much over the reduction of the gravimetric weight. On the other hand, when the first scission occurs at the end of the polymer chain, the reduction of the gravimetric weight and the reduction of the molecular weight of the recovered polymer may be negligible at this stage. Similarly, in the case of uniformly successive scission from the end of the polymer

chain, the reduction of the molecular weight of the recovered polymer and the reduction of the gravimetric weight may occur in parallel.

We summarize the experimental results in Figure 5. The concentration of the polymer sample was 0.5 g/kg. We show the time course of the residual weight during the treatment by closed circles. The open circles show the time course of the rate of reduction of the molecular weight of the recovered polymer. Obviously, the reduction of the molecular weight much predominated over the reduction of the gravimetric weight. This result clearly indicates that cleavage predominates at the BVP portion over the trimer portion. The biodegradation of the trimer of ST appears to be more difficult than that of BVP, although the dimer of ST is biodegradable.[11]

Influence of the size of the oligo-ST portion on the half-life of PST-*co*-BVP

Figure 6 shows the relation between the half-life of PST-*co*-BVP and the average number of ST units contained in the oligo-ST portion. Except for PST-*co*-BVP$_1$, we estimated the half-lives by the extension of the treatment time to zero in linear relations shown in Figures 3 and 4. The half-lives were significant under ultimate conditions when the amount of the polymer sample was sufficiently small and did not exceed the ability of the microorganisms in the soil. However, Figure 6 suggests that the half-life under ultimate conditions could be in the range of 30–40 days when PST-*co*-BVP contained 5 mol % BVP; that is, the oligo-ST portion contained 20 ST units. The incorporation of 5 mol % BVP into the main chain would be sufficient to make PST substantially biodegradable if we do not expect exceptionally rapid degradation in the natural environment, although such biodegradation was real-
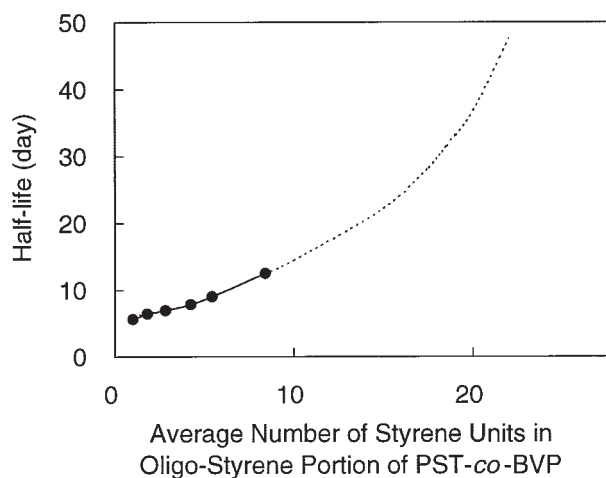
Figure 6   Relation between the half-life and average number of ST units in the oligo-ST portion of PST-*co*-BVP.
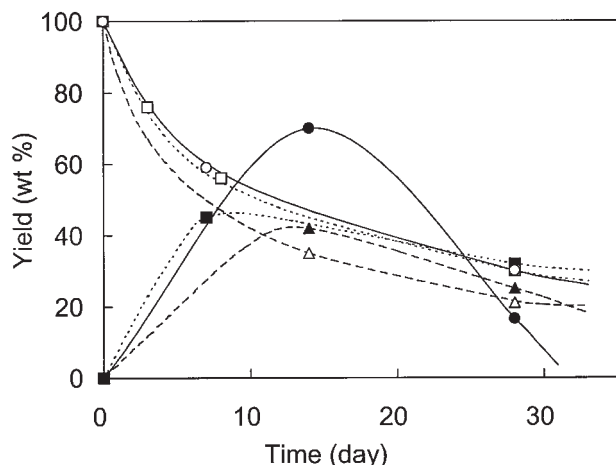
Figure 7 Time course of the yields of (□,○,△) the residual polymer and (■,●,▲) low-molecular-weight products that were soluble in ethyl acetate during the treatment of (□,■) PST-*co*-BVP$_2$, (○,●) PST-*co*-BVP$_3$, and (△,▲) PST-*co*-BVP$_4$ with activated sludge in soil. The amount of the polymer sample used for the treatment was 0.5 g/kg.

ized when a very low amount of the polymer sample was used.

Intermediate materials produced during the treatment of PST-*co*-BVP with activated sludge in soil and isolated by extraction with ethyl acetate

After the treatment of PST-*co*-BVP with activated sludge in soil, we recovered the residual polymer by extraction with a mixture of ethanol and tetrahydrofuran in a weight ratio of 1 : 1, as described in the Experimental section. After the removal of soil particles from the extracts by centrifugation, we removed the solvents by evaporation with a rotary evaporator and added ethyl acetate to the residue to precipitate and isolate the recovered polymer. However, we found intermediate materials in the layer of ethyl acetate, and we tried to isolate and identify them. The time course of the total amount of the intermediate products is shown in Figure 7 for PST-*co*-BVP$_2$, PST-*co*-BVP$_3$, and PST-*co*-BVP$_4$ together with the time course of the residual weight of PST-*co*-BVP$_2$, PST-*co*-BVP$_3$, and PST-*co*-BVP$_4$.

We made every effort to identify the recovered intermediates but did not succeed at this stage. The ultraviolet spectrum of the recovery showed a maximum absorbance at 275 nm. Column chromatography with silica gel as the stationary phase and ethyl acetate as the eluate revealed that the recovery was a compli-

cated mixture of several components. Unfortunately, we could not separate and identify these individual compounds definitively at this stage, although elementary analyses, Fourier transform infrared spectroscopy, and ^{1}H- and ^{13}C-NMR spectroscopy of the mixture suggested the presence of a carboxyl group in the intermediate products, in addition to the presence of a phenyl group and BVP. Chemical analyses of the individual components may provide important information on the mechanism of the biodegradation of PST-*co*-BVP by activated sludge. The determination of the chemical structure of these intermediate products is now under consideration.

For the degradation of poly(methyl methacrylate-*co*-N-benzyl-4-vinylpyridinium chloride) and poly(acrylonitrile-*co*-N-benzyl-4-vinylpyridinium chloride) by the treatment with activated sludge in soil, we did not obtain such intermediate materials. Therefore, the recovery of intermediate products during the treatment of PST-*co*-BVP with activated sludge reflects the difficult biodegradation of PST and oligo-ST.

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