Synthesis and Biological Activities of Polymers Containing *exo*-3,6-Epoxy-1,2,3,6-Tetrahydrophthalic Glycinylimide

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SYNOPSIS

The monomer, *exo*-3,6-epoxy-1,2,3,6-tetrahydrophthalic glycinyl imide(ETGI), was prepared by the Diels-Alder reaction of *N*-glycinylmaleimide and furan. Poly(ETGI), poly(ETGI-*co*-methacrylic acid)[poly(ETGI-*co*-MA)] and poly(ETGI-*co*-vinylacetate)[poly(ETGI-*co*-VAc)] were synthesized by photoinitiated homopolymerization of ETGI or copolymerizations of ETGI with MA and VAc. Synthesized ETGI, poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) were characterized by IR and ^{1}H-NMR spectroscopies, elemental analysis, and gel permeation chromatography. The *in vitro* cytotoxicities of ETGI, poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) were evaluated using K-562 human leukemia cells and HeLa cells. *In vitro* cytotoxicity of monomer and polymers at a concentration of 1.0 mg/mL against K-562 human leukemia cells increased in the following order: poly(ETGI-*co*-MA) > poly(ETGI-*co*-VAc) > poly(ETGI) > ETGI. The cytotoxicities of copolymers against HeLa cells are less cytotoxic than ETGI at a dosage of 0.02, 1.0, and 5.0 mg/mL. The copolymers were very effective at any dosage tested. The *in vivo* antitumor activities of ETGI, poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) were also evaluated against mice bearing sarcoma 180. *In vivo* antitumor activity of monomer and polymers at a dosage of 80 mg/kg increased in the following order: ETGI > poly(ETGI-*co*-VAc) > poly(ETGI-*co*-MA) > poly(ETGI) > 5-fluorouracil (5-FU). ETGI and polymers containing ETGI showed higher antitumor activity than 5-FU at any dosage tested. © 1996 John Wiley & Sons, Inc.

INTRODUCTION

The 1 : 2 regulary alternating copolymer of divinyl ether with maleic anhydride (DIVEMA) has been extensively studied for its structure and broad biological activities. The structural feature for the hydrolyzed form of DIVEMA contains the carboxylic group as a hydrophilic part and sugar moieties such as pyran or furan ring as a hydrophobic part. DIVEMA has been shown to possess good antitumor, antiviral, antibacterial, and antifungal activities[1,2] and interferon-inducing ability in animal test. However, it has also several toxic side effects such as enlarged livers and spleens, etc. Afterward, many attempts have been made to obtain a polymeric drug like DIVEMA with reduced side effects.[3-8] Breslow et al.[9] prepared DIVEMA with narrow molecular weight distribution by the photopolymerization technique in solvent with or without a photoinitiator.[10-12] Several studies have been made also in this laboratory to develop a polymeric antitumor agent.[13-19]

The aim of this study is to obtain new biologically active polymer from *exo*-3,6-epoxy-1,2,3,6-tetrahydrophthalic glycinyl imide (ETGI). Polymers containing ETGI were expected to show considerably high biological activities because they have amino acid moiety in repeating unit and their anionic character after hydrolysis is similar to that of DIVEMA.

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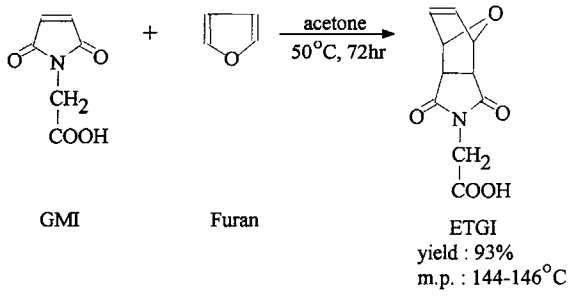

Figure 1 Synthesis procedure of ETGI.

In this work, the monomer ETGI was synthesizedby the Diels-Alder reaction of glycinylmalemide (GMI) and furan. Poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) were prepared by the homopolymerization of ETGI and copolymerizations of ETGI and the corresponding monomers. The structures of ETGI, poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc)were identified by IR and ^{1}H-NMR spectroscopies Average molecular weights of synthesized poly-mers were determined by gel permeation chromathography.

In vitro cytotoxicities of the prepared ETGI and polymers were evaluated with K-562 human leukemia cells and HeLa cells as a target cell. *In vivo* antitumor activities of the prepared ETGI and polymers against sarcoma 180 also were investigated using tumor bearing Balb/C mice.

EXPERIMENTAL

Materials

Maleic anhydride (Junsei Chem.) was recrystallized from chloroform. Glycine (Junsei Chem.) was recrystallized from distilled water. Furan, methacrylic acid (MA), and vinyl acetate (VAc) were purified by conventional methods.

For *in vitro* test, K-562 human leukemia cell line and HeLa cell line were used as a target tumor cell. For *in vivo* test, Balb/C mice and sarcoma 180 cell line were purchased from the Center of Genetic Engineering (Korea Institute of Science and Technology).

Instruments

IR spectra were obtained on a Jasco FTIR-5300 spectrophotometer using KBr disc. ^{1}H-NMR spectra were recorded on a FT-300 MHz Bruker A-3000 spectrophotometer. Average molecular weights were determined by gel permeation chromatography (GPC; Water, Water-410).

Elemental analysis was performed by Carlo Erba Instruments model EA1108 elemental analyzer.

Synthesis of ETGI

The synthesis of ETGI is shown in Figure 1. To use in synthesis of ETGI, GMI was prepared by the reaction of maleic anhydride and glycine in acetic acid at room temperature for 3 h according to the literature procedure.[20-22] A solution of 3.9 mL (0.054 mol) of furan in 10 mL of acetone and a solution of 5 g (0.032 moL) of GMI in 50 mL of acetone were mixed in a three-necked flask equipped with magnetic stirrer and nitrogen inlet and the mixed solution was stirred at 50°C for 72 h. The contents were precipitated in petroleum ether. The white precipitate was filtered and recrystallized from chloroform (yield, 6.7 g, 93%). The melting point of ETGI was 145°C. The prepared ETGI was identified by IR and ^{1}H-NMR spectra. Elemental analysis calculated for $C_{10}H_9NO_5$: C = 53.82%; H = 4.06%; N = 6.28%; found, C = 53.12%; H = 4.36%; N = 6.19%.

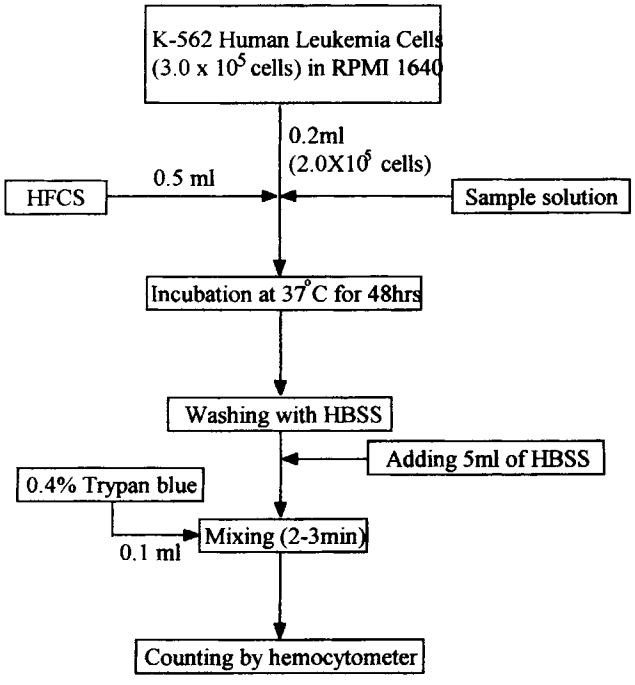

Figure 2 Procedure to determine the cytotoxicity by the dye exclusion method.

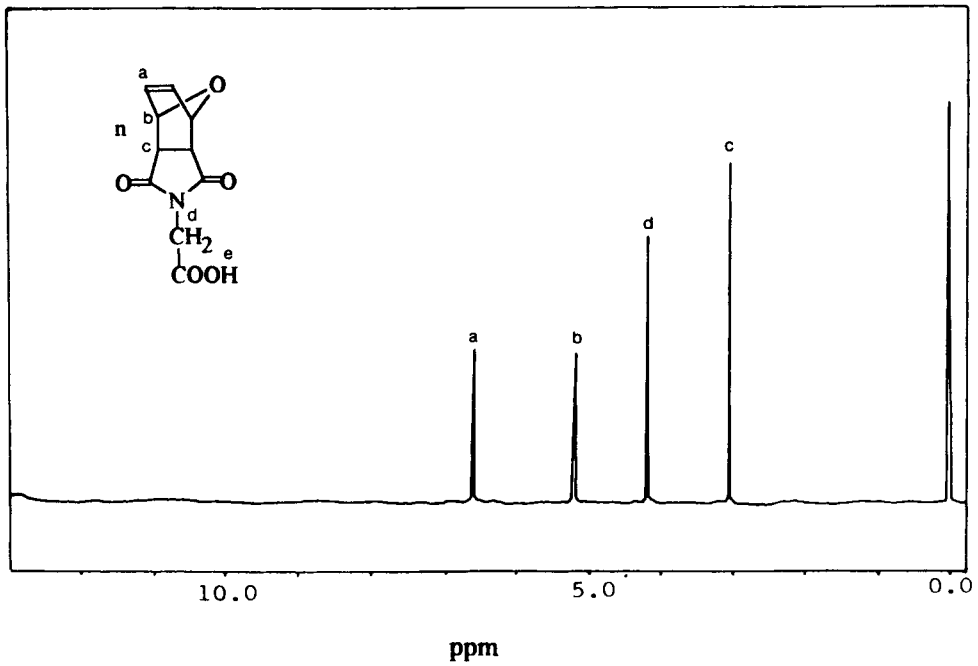

Figure 3 ^{1}H-NMR spectrum of ETGI (300 MHz, DMSO-d6).

Synthesis of Polymers

Poly(ETGI)

ETGI (1.12 g, 0.005 mol) and dimethoxybenzoin (DMB) (0.051 g, 0.1 mmol) were dissolved in 10 mL of the mixed solvent of 2-butanone and acetone (v/v), introduced into a dry quartz polymerization tube. The solution was degassed twice by purging with purified N_2 gas. The tube was sealed and placed in a photochemical chamber re-

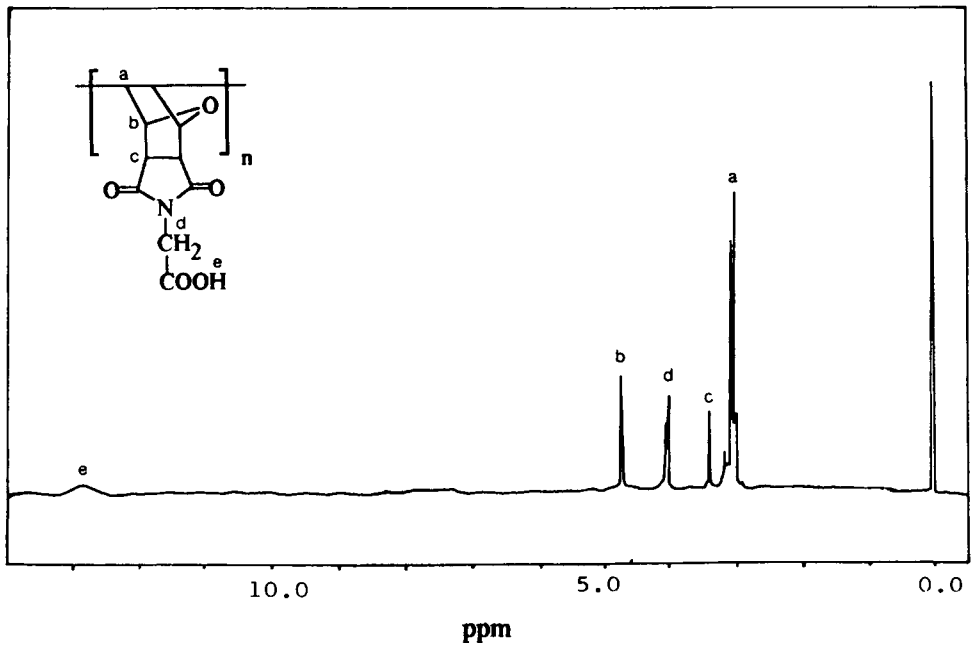

Figure 4 ^{1}H-NMR spectrum of poly(ETGI) (300 MHz, DMSO-d6).

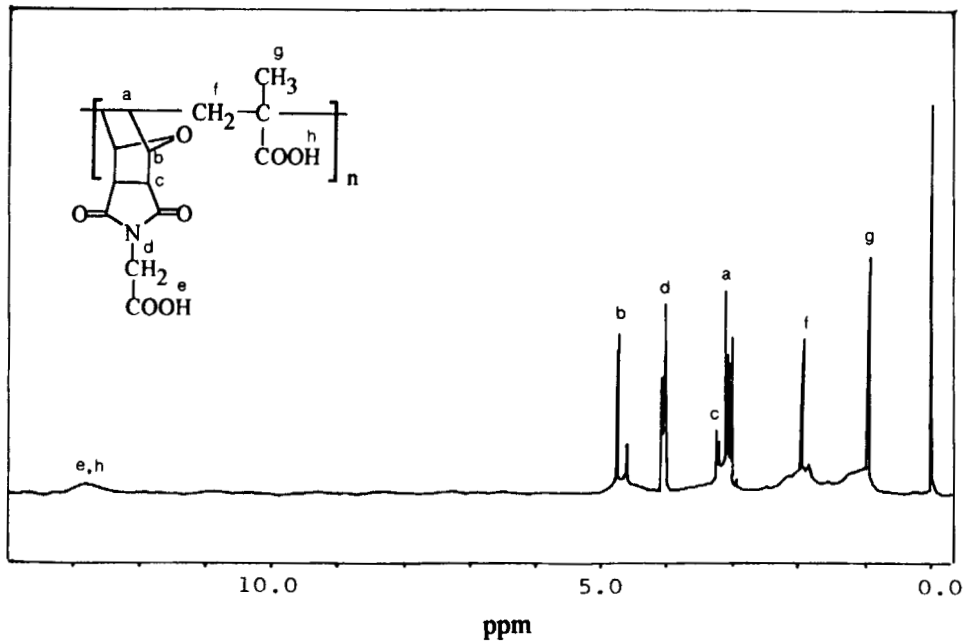

Figure 5 ^{1}H-NMR spectrum of poly(ETGI-*co*-MA) (300 MHz, DMSO-d6).

actor using UV lamp (λ_{max} = 313 nm) at 25°C for 72 h. The contents were poured into four times diethylether on the basis of the mixed solvent that was used in polymerization and followed by three subsequent precipitations from the mixed solvent into diethylether to remove the unpolymerized

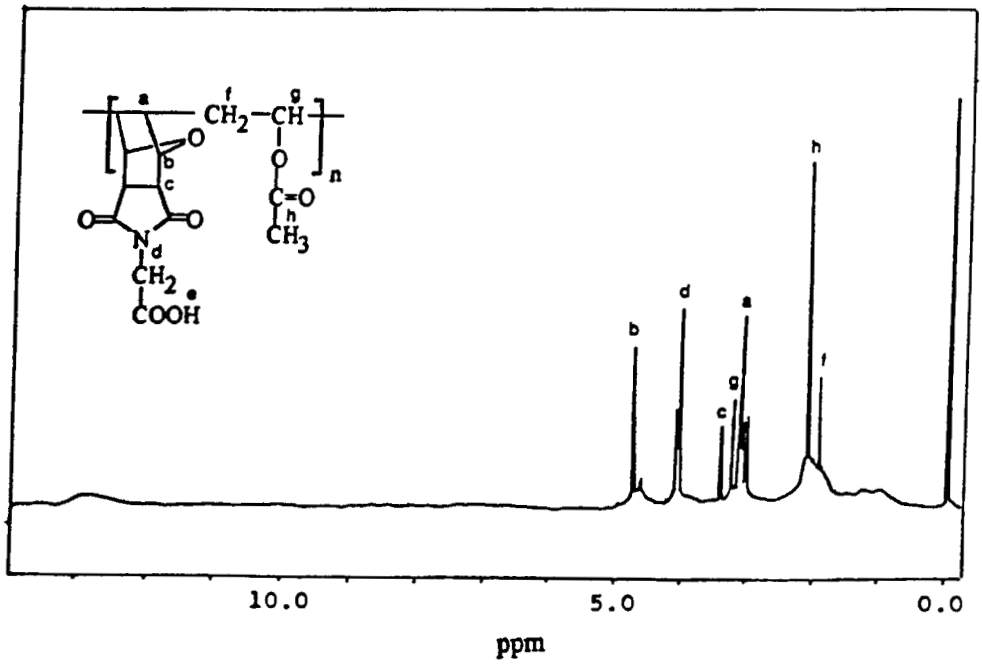

Figure 6 ^{1}H-NMR spectrum of poly(ETGI-*co*-VAc) (300 MHz, DMSO-d6).

Table I Solubility of ETGI and Polymers Containing ETGI

Solvent	ETGI	Poly(ETGI)	Poly(ETGI-*co*-MA)	Poly(ETGI-*co*-VAc)
Dimethylsulfoxide	○	○	○	○
N,N-Dimethylformamide	○	○	○	○
Water	○	○	○	○
Methanol	△	△	△	△
Acetone	○	△	○	○
2-Butanone	△	×	×	×
Chloroform	△	×	×	×
Tetrahydrofuran	○	×	×	×
1,4-Dioxane	○	×	×	×
Diethylether	×	×	×	×
Toluene	×	×	×	×
n-Hexane	×	×	×	×

○, good soluble; △, partially soluble; ×, insoluble.

monomer. Precipitated poly(ETGI) was collected by filtration and dried under vacuum until reached to a constant weight (yield, 23.3%).

Poly(ETGI-co-MA)

ETGI (1.12 g, 0.005 mol), MA (0.22 g, 0.0025 mol), and DMB (0.077 g, 0.15 mmol) were dissolved in 10 mL of the mixed solvent of 2-butanone and acetone (v/v), charged into a dry quartz polymerization tube. Poly(ETGI-*co*-MA) was obtained by using a procedure similar to that applied in the homopolymerization of ETGI (yield, 31.8%). Elemental analysis: found, C = 61.34%; H = 5.98%; N = 5.32%.

Poly(ETGI-co-VAc)

ETGI (1.12 g, 0.005 mol), VAc (0.43 g, 0.005 mol), and DMB (0.103 g, 0.2 mmol) were dissolved in 10 mL of the mixed solvent of 2-butanone and acetone (v/v), charged into a dry quartz polymerization tube. Poly(ETGI-*co*-VAc) was obtained by the same procedures as poly(ETGI-*co*-MA) (yield, 26.5%). Elemental analysis: found, C = 58.51%; H = 5.29%; N = 5.37%.

Measurement of Average Molecular Weight

Number (M_n) and weight average molecular weights (M_w) of poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) were determined by GPC equipped with μ-microstyragel column using dimethylformamide as an eluent at 80°C. Monodisperse polystyrene standard samples were used for molecular weight calibration. The concentration of polymers was 0.1% or less.

Analysis of Copolymers

The contents of ETGI moiety in poly(ETGI-*co*-MA) and poly(ETGI-*co*-VAc) were calculated from C, N, H data using Carlo Erba instruments model EA1108 elemental analyzer.

Assay of Biological Activity

Cytotoxicity of ETGI and Its Polymers

The procedure to investigate the *in vitro* cytotoxicity is as follows (Fig. 2). The assay was carried out in 96-well flat-bottom tissue culture plates. Target cells were K-562 human leukemia cells and HeLa cells.

Table II Content and Average Molecular Weights of Polymers Containing ETGI

	Content (%)			Average Molecular Weight		
Polymer	ETGI	MA	VAc	$\bar{M}_n$	$\bar{M}_w$	$\bar{M}_w/\bar{M}_n$
Poly(ETGI)	100.0	—	—	15,200	24,100	1.58
Poly(ETGI-*co*-MA)	53.7	46.3	—	13,300	19,800	1.49
Poly(ETGI-*co*-VAc)	59.6	—	40.4	15,900	27,100	1.70

Table III ***In Vitro*** **Cytotoxicities of ETGI and Polymers Containing ETGI Against K-562 Human Leukemia Cells**

Concentration (mg/mL)	Cytotoxicity (%)			
	ETGI	Poly(ETGI)	Poly(ETGI-*co*-MA)	Poly(ETGI-*co*-VAc)
5.0	75.1	73.5	89.8	94.9
1.0	69.5	72.9	82.5	81.0
0.1	67.8	65.5	77.7	78.0
0.02	38.4	31.1	61.6	62.9

The cells were suspended at 3×10^5 cells/mL in a culture medium containing heat inactivated fetal calf serum. The target cells (0.7 mL) and a polymer solution (4.3 mL) were added to each well of the plate. The assay was run in sextuplet. The plates were incubated at 37°C in a 5% CO_2 incubator for 24 h and then the cell suspension was centrifuged at 600 *g* at room temperature for 5 min. After centrifugation, the cells precipitated were washed twice with Hanks' balanced salt solution (HBSS), and then were added 5 mL of HBSS. Viability of the cells was determined with trypan blue by dye exclusion method. The percent cytotoxicity was calculated by following equation:

$$\text{Cytotoxicity (\%)} = \frac{\text{Number of Control Cells} - \text{Number of Sample Cells}}{\text{Number of Control Cells}} \times 100$$

Antitumor Activity of ETGI and Polymers Containing ETGI

To evaluate the antitumor activity of ETGI and polymers containing ETGI, mice bearing sarcoma 180 tumor cells were used. Balb/C mice ($n = 10$) were first intraperitonearly (i.p.) implanted with sarcoma 180 cells (2×10^5). The animals were then treated with a saline of sample at days 1–4. Three different dosages were tested: 0.8, 80, and 800 mg/kg. For comparison, antitumor activities of free 5-fluouracil (5-FU) also were tested by the same method. A control group was divided into two groups. One group was treated with sarcoma 180 cells along with the same volume of saline and the other group was treated with only sarcoma 180 cells. The ratio (T/C) of survival times of the polymer-treated (T) to that of control groups (C) was used as an index of the antitumor activity. Each group consisted of 10 animals.

RESULTS AND DISCUSSION

Identification of ETGI and Polymers

The structures synthesized monomer and polymers were confirmed by IR and ^{1}H-NMR spectroscopies. The IR spectrum of ETGI shows characteristic absorption peaks at 3450 cm^{-1} (— COOH), 1745 and 1690 cm^{-1} (— C=O), 1630 cm^{-1} (— CH=CH —), 1465 cm^{-1} (— CH_2 —), 1210 cm^{-1} (— C — O — C —). In Figure 3, ^{1}H-NMR spectrum of ETGI showed olefinic protons at 6.5 ppm, methine protons of cyclic ether ring at 5.2 ppm, methylene and methine protons of imide ring at 4.0, 3.4 ppm, and a proton of carboxylic acid at 12.8 ppm.

The absorption peak assignable to the C=C bond of monomeric ETGI disappeared at 1630 cm^{-1} in IR spectrum of poly(ETGI). In Figure 4, the absorption peak assignable to the olefinic protons of

Table IV ***In Vitro*** **Cytotoxicities of ETGI and Polymers Containing ETGI Against HeLa Cells**

Concentration (mg/mL)	Cytotoxicity (%)			
	ETGI	Poly(ETGI)	Poly(ETGI-*co*-MA)	Poly(ETGI-*co*-VAc)
5.0	83.6	79.5	76.6	72.9
1.0	80.4	69.4	77.5	71.0
0.1	71.6	63.5	65.8	64.2
0.02	71.0	58.6	62.8	64.2

Table V *In Vivo* Antitumor Activities of ETGI and Polymers Containing ETGI Against Sarcoma 180

Samples	Dose (mg/kg)	Survival Times (day)[a]	*T/C* (%)
Control	—	14.7 ± 2.3	100
	Saline	15.7 ± 0.5	100
5-FU	800.0	5.9 ± 0.3	39
	80.0	21.3 ± 1.3	140
	0.8	20.3 ± 1.8	134
ETGI	800.0	58.7 ± 12.8	276
	80.0	50.4 ± 20.4	237
	0.8	26.7 ± 13.0	125
Poly(ETGI)	800.0	23.7 ± 1.4	161
	80.0	24.3 ± 1.6	165
	0.8	21.7 ± 2.1	148
Poly(ETGI-*co*-MA)	800.0	24.5 ± 11.5	162
	80.0	25.3 ± 2.3	172
	0.8	24.8 ± 3.2	169
Poly(ETGI-*co*-VAc)	800.0	25.5 ± 1.9	174
	80.0	26.7 ± 1.9	182
	0.8	26.2 ± 1.6	178

[a] Values are means ± SE.

monomeric ETGI was not observed at 6.5 ppm. Methine protons of polymer backbone were observed at 2.9 ppm. The peaks due to methine protons of cyclic ether ring, methylene and methine protons of imide ring, and a proton of carboxylic acid are the same as those of monomeric ETGI.

The ^{1}H-NMR spectrum of poly(ETGI-*co*-MA) is shown in Figure 5. The absorption peaks due to protons of ETGI moiety in poly(ETGI-*co*-MA) were assigned to the same as those of poly(ETGI). Methylene protons, methyl protons, and a proton of carboxylic acid of MA moiety in poly(ETGI-*co*-MA) were observed at 1.9, 0.9, and 12.6 ppm, respectively. The peaks assigned to the olefinic proton of ETGI and MA moiety at 6.5 and 7.1 ppm disappeared in Figure 5.

Figure 6 is the ^{1}H-NMR spectrum of poly(ETGI-*co*-VAc). The absorption peaks due to protons of ETGI moiety in poly(ETGI-*co*-VAc) were assigned to the same as those of poly(ETGI). The peaks at 2.1, 1.9, and 3.4 ppm were assigned to methyl, methylene, and methine protons of VAc unit, respectively. The peaks due to vinyl protons in ETGI and VAc were not observed in Figure 6.

Solubility, Average Molecular Weights, and Composition of Polymers

Solubilities of ETGI, poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) are listed in Table I. ETGI and poly(ETGI-*co*-MA) are good soluble in polar solvents such as water, DMF. Poly(ETGI) and poly(ETGI-*co*-VAc) are soluble in water at elevated temperature (50°C).

The average molecular weights of poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) measured by GPC were listed in Table II. Number average molecular weight of synthesized poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) were 15,200, 13,300 and 15,900, respectively.

The contents of ETGI moiety in poly(ETGI-*co*-MA) and poly(ETGI-*co*-VAc) calculated from C, N, H data by elemental analyses were listed in Table II. ETGI contents in poly(ETGI-*co*-MA) and poly(ETGI-*co*-VAc) are 53.7% and 59.6%, respectively.

In Vitro *Cytotoxicity of ETGI and Polymers Containing ETGI*

K-562 human leukemia cells and HeLa cells were treated with ETGI, poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) at various concentrations of the samples (0.02–5 mg/mL). Table III and IV showed the effect of sample concentration on the *in vitro* cytotoxicity against the target cells. As known in Table III, *in vitro* cytotoxicity of monomer and polymers at concentration of 1.0 mg/mL increased in the following order: poly(ETGI-*co*-MA) > poly(ETGI-*co*-VAc) > poly(ETGI) > ETGI. Poly(ETGI-*co*-MA) and poly(ETGI-*co*-VAc) showed more cytotoxic against K-562 cells than ETGI at concentration range from 0.02 to 5 mg/mL. Poly(ETGI) showed a similar cytotoxicity against K-562 cells. In Table IV, poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) showed less cytotoxic against HeLa cells than ETGI at concentration range tested (0.02–5 mg/mL).

In Vivo *Antitumor Activity of ETGI and Polymers Containing ETGI*

The results of *in vivo* antitumor activities of ETGI, poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) against sarcoma 180 are listed in Table V. In Table V, the antitumor activity of 5-FU is also shown for comparison. The ratio T/C was used as an index of the antitumor activity:

$$T/C\ (\%) = \frac{\text{Survival Time of Treated Mice (T)}}{\text{Survival Time of Control Group Mice (C)}} \times 100$$

In Table V, the *in vivo* antitumor activity of monomer and polymers at a dosage of 80 mg/kg against Balb/C mice bearing sarcoma 180 tumor cells increased in the following order: ETGI > poly(ETGI-*co*-VAc) > poly(ETGI-*co*-MA) > poly(ETGI) > 5-FU. The lifespan of mice treated with 5-FU is longer than that of the control group at low dosages but that is remarkably short at a high dosage (800 mg/kg). 5-FU has enhanced survival at low dosages (0.8 and 80 mg/kg) but has not proven to be effective at high dosage (800 mg/kg) because of its side toxicity.

The survival times of mice treated with poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) were longer than those of treated with 5-FU at dosage range from 0.8 to 800 mg/kg. The T/C values of mice treated with ETGI at concentrations of 80 and 800 mg/kg were 237 and 276%, respectively. The antitumor activity of ETGI was higher than that of 5-FU at higher dosages (80 and 800 mg/kg). The antitumor activity of poly(ETGI-*co*-MA) was similar to that of poly(ETGI-*co*-VAc) at all dosages tested.

CONCLUSIONS

The monomer ETGI was prepared from *N*-glycinylmaleimide and furan and polymers such as poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) were prepared by the photohomopolymerization of ETGI or photocopolymerizations of ETGI with MA and VAC in 2-butanone using DMB at 25°C. The contents of ETGI unit in poly(ETGI-*co*-MA) and poly(ETGI-*co*-VAc) were 53.7% and 59.6%, respectively. The average molecular weights and polydispersities of copolymers determined by GPC were as follows:

Poly(ETGI): $\bar{M}_n$ = 15,200, $\bar{M}_w$ = 24,000, $\bar{M}_w/\bar{M}_n$ = 1.58

Poly(ETGI-*co*-MA): $\bar{M}_n$ = 13,300, $\bar{M}_w$ = 19,800, $\bar{M}_w/\bar{M}_n$ = 1.49

Poly(ETGI-*co*-VAc): $\bar{M}_n$ = 15,900, $\bar{M}_w$ = 27,100, $\bar{M}_w/\bar{M}_n$ = 1.70

The *in vitro* cytotoxicity of monomer and polymers at concentration of 1.0 mg/mL against K-562 human leukemia cells increased in the following order: poly(ETGI-*co*-MA) > poly(ETGI-*co*-VAc) > poly(ETGI) > ETGI. Poly(ETGI-*co*-MA) and poly(ETGI-*co*-VAc) showed more cytotoxic against K-562 cells than ETGI at concentration range tested from 0.02 to 5 mg/mL. Poly(ETGI) showed a similar cytotoxicity against K-562 leukemia cells. Poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) showed less cytotoxic against HeLa cells than monomeric ETGI at a concentration range tested (0.02–5 mg/mL).

The *in vivo* antitumor activity of monomer and polymers at a dosage of 80 mg/kg against Balb/C mice bearing sarcoma 180 tumor cells increased in the following order: ETGI > poly(ETGI-*co*-VAc) > poly(ETGI-*co*-MA) > poly(ETGI) > 5-FU. The antitumor activities of poly(ETGI), poly(ETGI-*co*-MA), and poly(ETGI-*co*-VAc) against sarcoma 180 were higher than that of 5-FU at all dosages tested (0.8–800 mg/Kg). The T/C values of mice treated with ETGI at the concentrations of 80 and 800 mg/kg were 237 and 276%, respectively. The antitumor activity of ETGI was higher than that of 5-FU at higher dosages (80 and 800 mg/kg).

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REFERENCES

1. G. Butler, *J. Polym. Sci.*, **48,** 279 (1960).
2. G. Butler and A. Zampini, *J. Macromol. Sci. Chem.*, **A11,** 491 (1977).
3. R. M. Ottenbrite, E. Goodell, and A. Munson, *Polymer,* **18,** 461, (1977).
4. R. M. Ottenbrite, K. Kuus, and K. Kaplan, *Polym. Prepr. Am. Chem. Soc., Div. Polym. Chem.*, **24,** 25 (1983).
5. R. M. Ottenbrite, *J. Macromol. Sci. Chem.*, **A22,** 819 (1985).
6. F. W. Bassett Jr., and O. Vogel, *J. Polym. Sci.: Chem. Ed.*, **21,** 891 (1983).
7. T. Sato, Junzo Sunamoto, and R. M. Ottenbrite, *J. Bioactive Comp. Polym.*, **1,** 448 (1986).
8. M. S. Melzer, *Eur. J. Cancer,* **16,** 15 (1980).
9. D. S. Breslow, E. I. Edwards, and N. R. Newberg, *Nature,* **346,** 160 (1973).
10. A. S. Sartoelli and D. G. Johns, *Handbook of Experimental Pharmacology,* Vol. 38, Parts 1 and 2, Springer, Berlin, 1974/1975.
11. P. Ehrlich, in *Readings in Pharmacology,* L. Shuster, Ed., Little, Brown, Boston, 1962, p. 231.
12. H. Ringsdorf, *J. Polym. Sci.: Symp.*, **51,** 135 (1975).
13. N. J. Lee, S. H. Oh, C. S. Ha, J. K. Lee, and W. J. Cho, *J. Kor. Ind. Eng. Chem.*, **1,** 2, 190 (1990).
14. N. J. Lee, C. S. Ha, and W. J. Cho, *Polymer (Korea),* **15,** 2, 211 (1991).

15. N. J. Lee, I. S. Kim, C. S. Ha, and W. J. Cho, *J. Kor. Ind. Eng. Chem.*, **2,** 156 (1991).
16. M. S. Shim, N. J. Lee, C. S. Ha, and W. J. Cho, *Polymer (Korea)*, **15,** 4, 489 (1991).
17. W. M. Choi, I. S. Kim, C. S. Ha, W. J. Cho, and N. J. Lee, *J. Bioactiv. Compat. Polym.*, **7,** 1, 39 (1992).
18. N. J. Lee, C. S. Ha, and W. J. Cho, *J. Macromol. Sci.-Pure Appl. Chem.*, **A29**(2), 161 (1992).
19. G. T. Gam, J. G. Jeong, N. J. Lee, Y. W. Lee, C. S. Ha, and W. J. Cho, *J. Appl. Polymer Sci.*, to appear.
20. T. Oishi, Y. Otsubo, and M. Fujimoto, *Polym. J.*, **24,** 527 (1992).
21. T. Oishi, A. Karmori, and M. Fujimoto, *J. M. S.-Pure Appl. Chem.*, **231,** A29 (1992).
22. D. H. Rich, P. D. Gesellchen, A. Tong, A. Cheung, and C. K. Buckner, *J. Med. Chem.*, **18,** 1004 (1975).

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