Hence the amination of enolates derived from **4** with dibenzyl or di-*tert*-butyl azodiformates provides an efficient route for conversion of carboxylic acid derivatives to both chiral α-hydrazino acids (**1**) and α-amino acids (**2**).

Supplementary Material Available: Experimental descriptions, analytical procedures, and spectral data for new compounds (12 pages). Ordering information is given on any current masthead page.

Photoinduced Reversible Conformational Transition of Polypeptide Solid Membranes

T. Kinoshita, M. Sato, A. Takizawa,* and Y. Tsujita

Department of Materials Science & Technology
Nagoya Institute of Technology
Gokiso-cho, Showa-ku, Nagoya 466, Japan

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The photocontrol of physical and chemical properties of polymer gels and membranes with photochromic residues has been the subject of numerous recent investigations.[1-4] Photoinduced conformational changes of polypeptides in solution have been observed for poly(L-aspartates)[5-9] and poly(L-glutamates),[10-14] with azobenzene derivatives in their side chains. No report has appeared, however, on photoinduced α-helix to coil transition of polypeptide solid membrane. We report here on the photocontrol of the secondary structure of polypeptide solid membranes composed of poly(L-glutamic acid) (PGA) containing pararosaniline (rose) groups in the polymer side chains based on a cooperative effect between photodissociation of the pararosaniline moiety and

Scheme I

the induced acid dissociation of the L-glutamic acid group in the membrane.

PGA polymers with incorporated pararosaniline groups (rose-PGA) were synthesized by the condensation reaction of PGA with pararosaniline in the presence of *N*-hydroxybenzotriazole (HOBt) and 1-ethyl-3-(3-(dimethylamino)propyl)carbodiimide
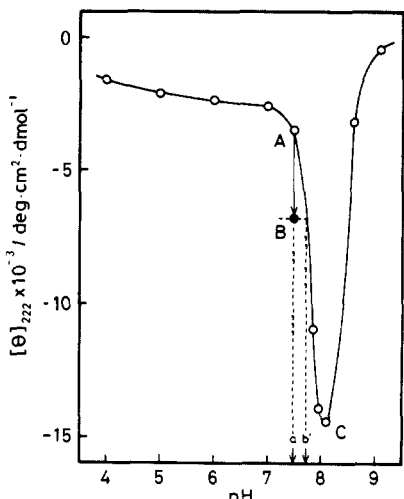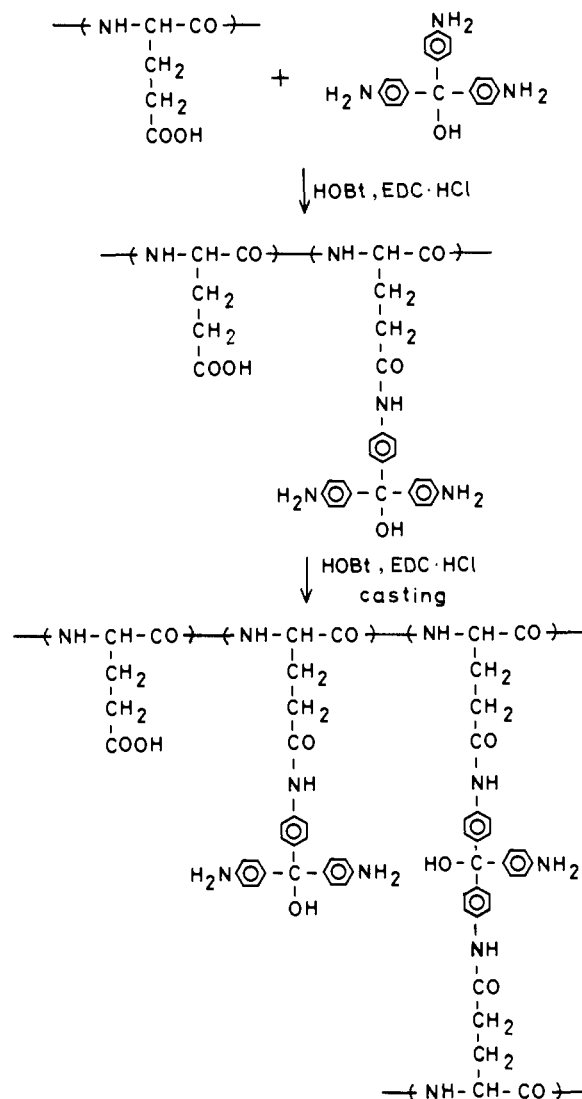
Figure 1. pH dependence of minimum ellipticity, $[\theta]_{222}$, of a dark-adapted membrane of poly(L-glutamic acid) containing 10.5 mol % pararosaniline groups in aqueous solution at 25 °C.

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Scheme II

rose-PGA : random coil

α-helix
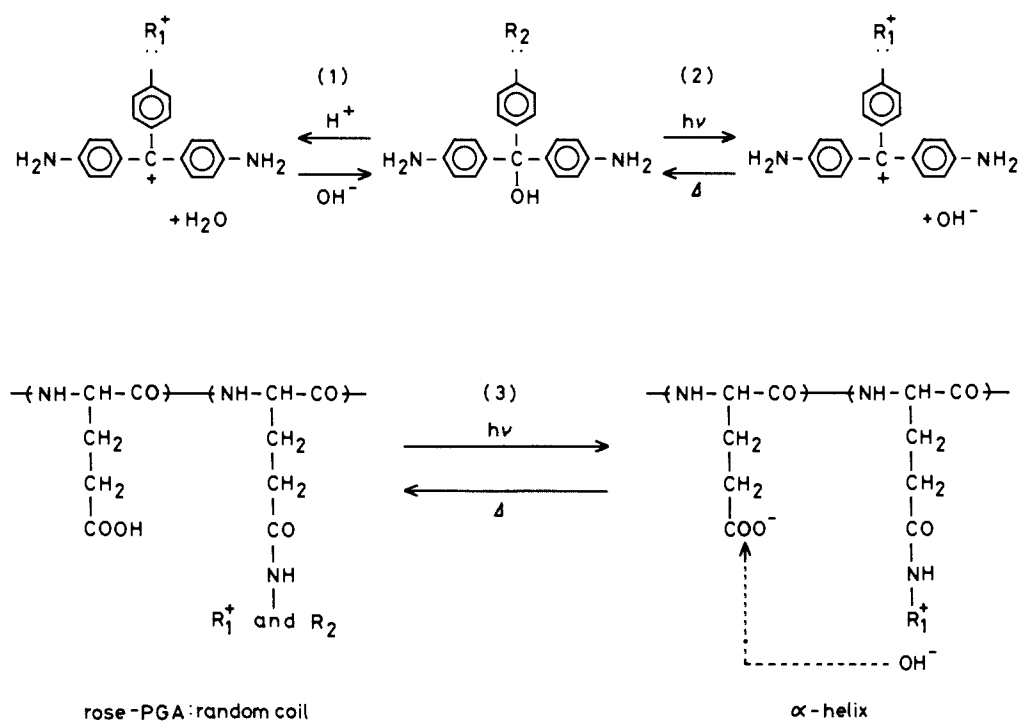
hydrochloride (EDC·HCl) in dimethylformamide (DMF) solution at room temperature for 24 h (Scheme I). L-Glutamic acid residues, 10.5 mol %, were converted to the pararosaniline derivatives.[15] A rose-PGA membrane of ca. 3-μm thickness was prepared by casting a DMF solution of the rose-PGA containing HOBt and EDC·HCl at 40 °C and allowing the solvent to evaporate (Scheme I). The membrane obtained was no longer soluble in DMF and in aqueous solution of NaOH, indicating that only a fraction of rose moieties with amino groups may also act as a cross-linking agent in the concentrated solution of rose-PGA during the casting process.

Circular dichroism (CD) measurements were made with a JASCO J-40 apparatus at 25 °C to elucidate the pH-induced conformational transition of the dark-adapted rose-PGA membrane. The helix stability at various pHs was represented by a pH dependence of the molar ellipticity at 222 nm, suggesting loss of α-helix structure at low and high pH regions and formation of the helix near neutrality (Figure 1). This transition behavior arises from the difference in the state of ionization of the side chains in the rose-PGA; i.e., the L-glutamic acid side chain is negatively ionized at high pH, whereas the rose moiety is positively ionized at low pH[16] (eq 1 in Scheme II). The repulsion forces among these charged groups of the same sign at low and high pHs, therefore, will disturb the formation of α-helix structure of the membrane. On the other hand, at the transition region these charged groups will be neutralized by the oppositely charged groups, which results in the formation of helix structure in the rose-PGA membrane. The pK_a of the rose dye was shown to be ca. 7.6;[16] however, the transition region of the rose-PGA membrane seems to be shifted to higher pH value, since the pK_a of the L-glutamic acid group, which reduces the electrostatic repulsion between cationic rose moieties in acid pH region, was shown to be ca. 5.0 on pure poly(L-glutamic acid).[17] It is well-known that the hydrophobic environment around PGA effectively reduces the degree of dissociation of the side chain carboxyl groups, which results in a large shift of the pH of the helix to coil transition of the PGA toward higher values.[13,18] Therefore, the transition
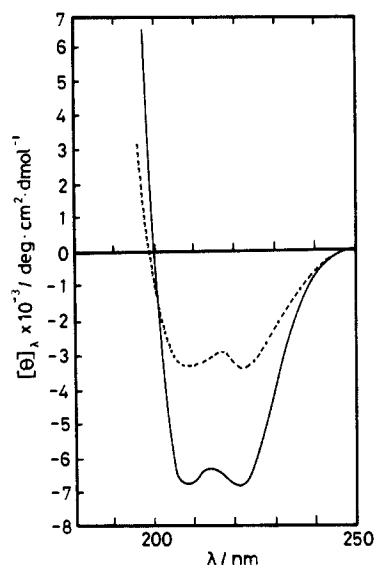
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Figure 2. CD spectra of a membrane of poly(L-glutamic acid) containing 10.5 mol % pararosaniline groups, before (---) and after (—) irradiation, in aqueous solution at 25 °C. The irradiation was carried out at pH 7.5.

behavior of the rose-PGA membrane in Figure 1 can be explained in terms of the hydrophobic environment in the membrane provided by triarylmethane groups of the rose side chains around L-glutamic acid moieties; i.e., the triarylmethane groups with low dielectric constant may also produce a shifting of the pK_a of the -COOH side chain toward higher values.

The light irradiation of the rose-PGA membrane was carried out at pH 7.5 with a high-pressure mercury arc lamp (Ushio 500 W), using a glass filter (Toshiba D-33s). The irradiation produced a strong increase of the CD bands at 222 and 208 nm (Figure 2). The relative increase of the helix content can be estimated to be ca. 100%. During the irradiation, the molar ellipticity of the membrane at 222 nm monotonously increased from -3.40 × 10³ to -6.84 × 10³ deg·cm²·dmol⁻¹, which corresponds to the increase in [θ]₂₂₂ value from point A to B in Figure 1 without

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passing through the minimum point C. It may say, therefore, that this photoirradiation is apparently equivalent to the replacement of the aqueous solution of pH 7.5 (a in Figure 1) by that of pH 7.8 (b′) in the dark. Triarylmethane leucohydroxide derivatives are known to dissociate into ion pairs under ultraviolet light irradiation with production of a hydroxide ion[19] (eq 2 in Scheme II). Changes in the UV–visible absorption spectrum of the rose-PGA membrane effected by light irradiation could be correlated with the photodissociation of the rose moiety, yielding a hydroxide ion and the corresponding dye. This suggests that the pH value in the membrane can be controlled by light irradiation via the photodissociation of the rose side chains. In addition, Figure 1 shows that the conformations of the rose-PGA membrane at pH 7.5 are very sensitive to small increase in pH. These results imply that the photoinduced α-helix formation can be explained in terms of the cooperative effect between the photodissociation of the rose moiety with production of a hydroxide ion and the induced acid dissociation of L-glutamic acid moieties accompanied by the increase in pH in the membrane phase on UV irradiation (eq 3 in Scheme II). One additional important aspect should be noted; i.e., after removal of the light, CD bands at 222 and 208 nm gradually decreased again and returned to the initial value after 100 min in the dark at 25 °C, thus confirming the reversibility of the change. It was also found that the photoinduced conformational transitions of the rose-PGA membrane depend on the rose group content and pH value of the aqueous solution at which irradiation is carried out. The study of the detailed photoresponsiveness of the rose-PGA membrane is in progress.

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First Two-Step 1,3-Dipolar Cycloadditions: Nonstereospecificity[†]

Rolf Huisgen,* Grzegorz Mloston, and Elke Langhals

*Institut für Organische Chemie der
Universität München, D-8000 München 2, Germany*

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All mechanistic criteria point to the concertedness of 1,3-dipolar cycloadditions.[1] An alleged violation of stereospecificity for an azomethine imine[2] is an artifact.[3] We report on the first nonstereospecific (3 + 2) cycloadditions.

Sustmann's PMO model of concerted cycloadditions[4] envisions *two* cases in which the stepwise mechanism might compete with the concerted one. Two similar HO–LU interaction energies correspond to a minimum of rate, and a *diradical* pathway is expected, if stabilizing substituents are present.

A second limiting case is foreseen when the interaction HO-(1,3-dipole)–LU(dipolarophile) is strongly dominant in the transition state. The higher the difference in π-MO energies of reactants, the lower would be the energy contribution by the second HO–LU interaction; in the extreme it should no longer outweigh the additional entropy requirements of the concerted process. A

unilateral electron flow should result, establishing *one* bond between the reactants, i.e., generation of a *zwitterionic intermediate*.[5]

Sulfur and carbon possess the same electronegativity on the Pauling scale. Absence of the more electronegative O or N atoms brings *thiocarbonyl ylides*[6] close to the high MO energies of the allyl anion, electronic prototype of 1,3-dipoles. An aliphatic thiocarbonyl ylide and an ethylene derivative bearing four electron-attracting substituents offer a reactant pair with extremely different MO energies.

Nitrogen evolution from the spiro-1,3,4-thiadiazoline **1a**[7,8] in THF furnished 2,2,4,4-tetramethylcyclobutan-1-one-3-thione *S*-methylide (**2a**) which combined in situ with dimethyl 2,3-dicyanofumarate to give the *cis,trans-isomeric* cycloadducts **5a** and **6a**;[9] the yield of 94% and the 48:52 ratio were based on ¹H NMR
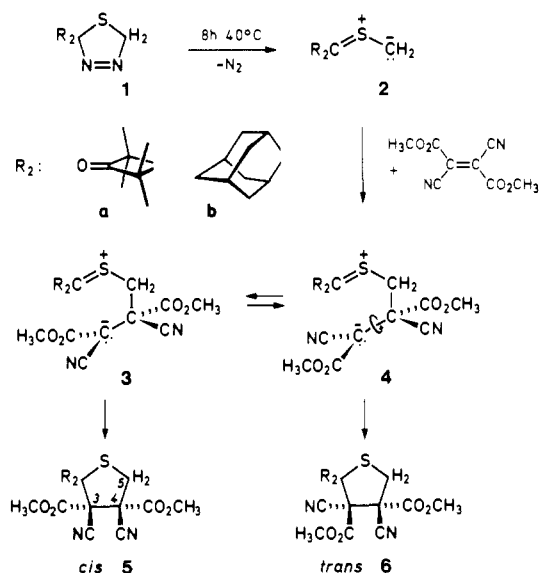
analysis. The separated adducts are stable under the reaction conditions indicating kinetic control. At no stage did the solution of 2,3-dicyanofumaric ester[10] show sign of the cis isomer. We consider rotation of the *zwitterionic intermediates* **3a** and **4a** responsible for the nonstereospecific course.

The cis,trans ratio **5a**/**6a** increases with solvent polarity. **5a** (%): CCl₄ 37, toluene 36, CS₂ 40, THF 48, CH₂Cl₂ 47, acetone 60, CH₃CN 62. The small growth is ascribed either to increasing approach to rotational equilibrium, in accord with the steric course of (2 + 2) cycloadditions via zwitterions,[11] or to solvent dependence of the rotamer equilibrium.

The thiadiazoline **1b** eliminates N₂ with a similar half-life as **1a** (33 min vs. 25 min, xylene, 45 °C), and adamantanethione *S*-methylide (**2b**) is an active 1,3-dipole.[12] Its in situ reaction with dimethyl dicyanofumarate in THF provided the cis,trans cycloadducts **5b** and **6b**, 90%, 41:59 (57:43 in the more polar acetonitrile). Each adduct, isolated crystalline, results from kinetic control.

The cis,trans assignment in the **a** and **b** series was based on solvent dependence of the isomer ratio, higher solubility of cis vs. trans,[13] X-ray analysis of a related cis adduct, and cis,trans in-

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† Dedicated to Professor David Ginsburg on the occasion of his 65th birthday.

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