ing over calcium chloride, the solvent was removed and the residue distilled to give 31.6 g. (29%) of the second isomer of VIII, b.p. 125° at 0.4 mm., n^{25}_D 1.5632, which solidified after cooling and scratching, m.p. 35–36°.[13]

Anal. Calcd. for $C_{16}H_{15}N$: C, 86.85; H, 6.83; N, 6.33. Found: C, 86.82; H, 6.81; N, 6.39.

Benzhydrylation to Form IX.—This alkylation was carried out as described for the α-phenylethylation (procedure A) employing 0.25 mole each of potassium amide, phenylacetonitrile and benzhydryl chloride. After removing the ammonia and adding water, the ether layer was separated and the solvent distilled. The residual oil was taken up in methanol, and the solution cooled to precipitate 70.3 g. (99%) of α,β,β-triphenylpropionitrile (IX), m.p. 99.5–100.5°, reported m.p. 101.5–102°.[15]

The nitrile (32 g.) was refluxed for 48 hours with 11 g. of potassium hydroxide in 100 ml. of ethylene glycol monomethyl ether and 20 ml. of water to give 29.6 g. (87%) of α,β,β-triphenylpropionamide, m.p. 205–206°, reported m.p. 208–209°.[16]

Further Benzhydrylation of IX.—This alkylation was carried out in the usual manner employing 0.1 mole each of IX, potassium amide and benzhydryl chloride, except that the reaction time was increased to 2 hours. The crude product was recrystallized twice from ethanol to give 15 g. (33%) of α,α-dibenzhydrylphenylacetonitrile, m.p. 212–213°.

Anal. Calcd. for $C_{34}H_{27}N$: C, 90.81; H, 6.05; N, 3.12. Found: C, 90.97; H, 6.09; N, 2.99.

DURHAM, NORTH CAROLINA

COMMUNICATIONS TO THE EDITOR

POLYPEPTIDES. VI. POLY-α-L-GLUTAMIC ACID: PREPARATION AND HELIX-COIL CONVERSIONS[1]

Sir:

We have prepared high molecular weight poly-α-L-glutamic acid (PGA) from poly-γ-benzyl-α-L-glutamate[2] (PBG) without significant cleavage of peptide bonds by the treatment of the latter material with hydrogen bromide under anhydrous conditions. As shown by infrared dichroism measurements on solid samples this PGA is in the "α" or helical form[3] as the free acid, and changes to a random coil upon formation of the sodium salt, from which, however, the helical form can be regenerated.

It has been previously reported that attempts to remove the benzyl groups of PBG by catalytic reduction have been unsuccessful,[4] but that these groups could be removed by treatment with phosphonium iodide in glacial acetic acid.[4,2] Our inability to "reduce" PBG with phosphonium iodide in glacial acetic acid containing excess acetic anhydride or in acetic anhydride, led us to believe that the reaction is not a reduction, but rather an alkyl–oxygen ester cleavage brought about by hydrogen iodide.[5] The hydrogen iodide results from the reaction of phosphonium iodide and the water in glacial acetic acid.

Hydrogen bromide may be used to effect complete debenzylation of PBG in several solvents: *viz.*, anhydrous benzene, liquid sulfur dioxide, nitromethane and trifluoroacetic acid. A typical debenzylation in sulfur dioxide proceeds as follows: a 2% solution of PBG (molecular weight, 680,000; degree of polymerization, 3100) in sulfur dioxide is cooled to −60° in a pressure bottle and anhydrous hydrogen bromide bubbled through for ten minutes. The bottle is sealed, allowed to stand at room temperature for two hours, and then cooled again to −60°. The SO_2 is decanted from the precipitate, which is then washed thoroughly with ether and dried. Analysis for poly-L-glutamic acid: Calcd. for $(C_5H_7NO_3)_n$: C, 46.5; H, 5.4; N, 10.8. Found: C, 46.8; H, 5.9; N, 10.2; $[\eta]$ in 0.2 M Na_2SO_4 at pH 7.3 = 2.83. Molecular weight found from light scattering 135,000,[6] degree of polymerization, 1050. Alternatively, the polymerization of the γ-benzyl-N-carboxy-L-glutamate anhydride may be carried out in anhydrous benzene[3] and this benzene solution treated with hydrogen bromide without isolation of the PBG. After bubbling excess HBr through the solution at room temperature, within one hour a gelatinous precipitate forms which gradually becomes a soft white mass of PGA. Removal and washing is performed under anhydrous conditions to minimize peptide bond cleavage.

Oriented films of PGA from 4:1 dioxane:water solution may be obtained by unidirectional rubbing of such solutions on silver chloride plates. These films show infrared dichroism (Fig. 1) characteristic of that obtained with other high molecular weight helical polypeptides such as PBG,[7] namely, parallel dichroism[8] of the 3300 cm.$^{-1}$ band (NH stretch) and 1655 cm.$^{-1}$ band (C=O stretch, amide I) and perpendicular dichroism of the 1550 cm.$^{-1}$ (amide II) band. There is a also a strong, but weakly dichroic absorption at 1710 cm.$^{-1}$ (non-ionized carboxyl) but no band at 1575 cm.$^{-1}$ (ionized carboxyl).

Oriented films of the sodium salt of PGA are obtained from water solution (pH 7.3) but orient much less easily than the free acid. In this case the infrared dichroisms are less, opposite in sense

---

(1) Supported in part by the Office of the Surgeon General, Department of the Army.

(2) E. R. Blout, R. H. Karlson, P. Doty and B. Hargitay, THIS JOURNAL, **76**, 4492 (1954); E. R. Blout and R. H. Karlson, *ibid.*, **78**, in press (1956).

(3) L. Pauling and R. B. Corey, *Proc. Nat. Acad. Sci.*, **37**, 241 (1951).

(4) W. E. Hanby, S. G. Waley and J. Watson, *J. Chem. Soc.*, 3239 (1950).

(5) *Cf.* D. Ben-Ishai and A. Berger, *J. Org. Chem.*, **17**, 1564 (1952).

(6) We are indebted to Drs. P. M. Doty and A. Wada for the light scattering and viscosity measurements.

(7) E. J. Ambrose and A. Elliott, *Proc. Roy. Soc.* (*London*), **A205**, 47 (1951).

(8) The dichroism is parallel if the absorption is greatest when the electric vibration direction of the incident linearly polarized radiation is parallel to the orientation direction. The dichroism was measured using an improved method (A. S. Makas and W. A. Shurcliff, *J. Opt. Soc. Am.*, **45**, 998 (1955)) that avoids certain common errors.

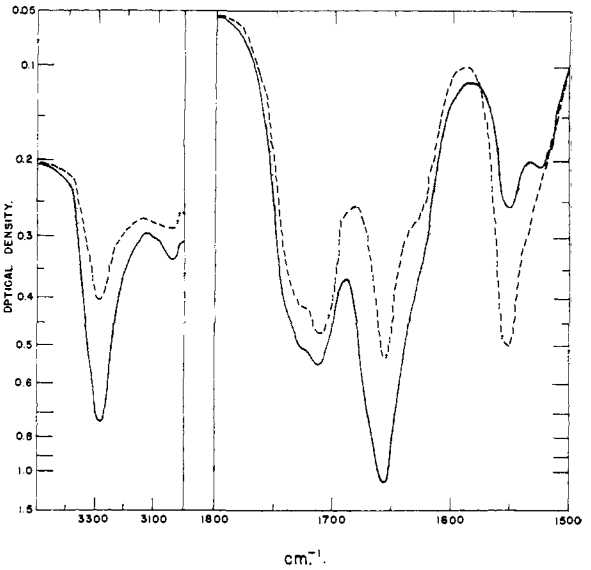

Fig. 1.—Infrared spectra of oriented film (on silver chloride plate) of poly-α-L-glutamic acid: —— electric vibration direction parallel to orientation direction; ---- electric vibration direction perpendicular to orientation direction.

to those obtained with the free acid, and correspond to those obtained with "β" polypeptides[7] or extended intermolecularly bonded configurations from random coils. The infrared spectra (Fig. 2) are characterized by perpendicular dichroism of the 3300 cm.$^{-1}$ and 1655 cm.$^{-1}$ bands. Particularly important is the appearance of a perpendicularly dichroic absorption band at 1628 cm.$^{-1}$ (C=O stretch, amide I). The 1550 cm.$^{-1}$ band is overshadowed by the 1575 cm.$^{-1}$ ionized carboxyl absorption band which is very strong.

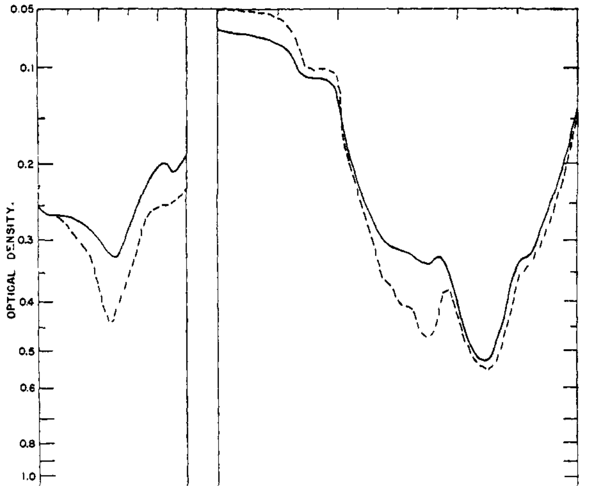

Fig. 2.—Infrared spectra of oriented film (on silver chloride plate) of sodium poly-α-L-glutamate: —— electric vibration direction parallel to orientation direction; ---- electric vibration direction perpendicular to orientation direction.

Solutions of the sodium salt of PGA upon acidification, dialysis, lyophilization and re-solution in dioxane:water give oriented films characteristic of helical configurations which correspond exactly in their infrared dichroic properties to that in Fig. 1. It is therefore concluded that *with PGA, helix → non-helix → helix transformations can be obtained by altering the ionization of the γ-carboxyl groups.*

It is interesting to speculate that similar transformations occur in solution. Some indication is offered by the large, reversible change in optical rotation of aqueous PGA solutions; $[\alpha]^{25}\mathrm{D}$ (0.2 *M* Na_2SO_4 solution) varies from −83° at *p*H > 7 to −8° at *p*H 4.5. These changes are comparable to those which occur with changes of configuration in PBG[9,10] and in proteins.[11]

---

(9) P. Doty, A. M. Holtzer, J. H. Bradbury and E. R. Blout, THIS JOURNAL, **76**, 4493 (1954).

(10) P. Doty and J. T. Yang, *ibid.*, **78**, 498 (1956).

(11) C. Cohen, *Nature*, **175**, 129 (1955).

CHEMICAL RESEARCH LABORATORY
POLAROID CORPORATION E. R. BLOUT
CAMBRIDGE 39, MASSACHUSETTS

THE CHILDREN'S CANCER RESEARCH FOUNDATION
THE CHILDREN'S MEDICAL CENTER M. IDELSON
BOSTON 15, MASSACHUSETTS

RECEIVED DECEMBER 12, 1955

POLYPEPTIDES. VII. POLY-γ-BENZYL-L-GLUTAMATE: THE HELIX-COIL TRANSITION IN SOLUTION[1]

Sir:

In earlier investigations[2] it has been shown that poly-γ-benzyl-L-glutamate exists in a randomly coiled configuration in dichloroacetic acid and in a rigid, rod-like helical configuration in a number of other organic solvents. Since the helix possesses a definite ordering of hydrogen bonds in one direction it would be anticipated that the conversion of this configuration into that of a random coil should have the characteristics of a first order transition. We have observed this transition both as a function of solvent composition and temperature and have found it to be rapidly reversible.

Solutions of equal concentration of poly-γ-benzyl-L-glutamate of high molecular weight[3,4] in ethylene dichloride and dichloroacetic acid were mixed in varying proportions and the intrinsic viscosity, the double refraction of flow and the optical rotation were determined. Except for a small change upon the first addition of the dichloroacetic acid solution to the ethylene dichloride solution[5] these properties remained essentially constant until a sharp change occurred in the vicinity of 76 volume per cent. dichloroacetic acid. At higher concentrations the properties were those of the pure dichloroacetic acid solution. The transition appeared to occur most sharply in terms of optical rotation, presumably because this quantity varied in direct proportion to the change

---

(1) This work was supported by the Office of Naval Research (N5ori-07654).

(2) (a) P. Doty, A. M. Holtzer, J. H. Bradbury and E. R. Blout, THIS JOURNAL, **76**, 4493 (1954); (b) P. Doty, J. H. Bradbury and A. M. Holtzer, *ibid.*, in press.

(3) E. R. Blout, R. H. Karlson, P. Doty and B. Hargitay, *ibid.*, **76**, 4492 (1954).

(4) E. R. Blout and R. H. Karlson, *ibid.*, in press.

(5) This small change at low dichloroacetic acid concentrations is thought to be associated with a change in the average orientation of the benzyl groups to the helix core.