Circular Dichroism Studies on Poly-L-cyclohexylalanine in the Far-ultraviolet Region

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THE preparation of cyclohexyl-L-alanine and of its homopolymers has been already described,[1] but not the far-ultraviolet optical rotatory properties of poly-L-cyclohexyl alanine as this polymer is barely soluble in organic solvents transparent in the u.v. spectral region.

It has recently been found that many polypeptides can easily be dissolved in concentrated strong acids like sulphuric acid[2] and methanesulphonic acid.[3] In these media the polypeptide chain is stable enough to permit measurements to be made.

Poly-L-cyclohexylalanine, prepared by polymerization of the corresponding *N*-carboxy-anhydride in dimethylformamide and using n-butylamine as the initiator (the monomer to initiator molar ratio was 35), was dissolved in pure methanesulphonic acid. The circular dichroism (c.d.) spectrum of the polymer in this solvent exhibits the typical shape of polypeptides in the random coil conformation,[4] with a weak positive band located at 220 nm. ($\Delta\epsilon = 0{\cdot}44$) and a strong negative band located at 196 nm. ($\Delta\epsilon = -\ 3{\cdot}8$). The molar ellipticity value of the negative band is lower than that observed for random polypeptides like poly-L-glutamic acid and poly-L-lysine in aqueous solutions. This behaviour is typical of polypeptides dissolved in concentrated sulphuric acid and methanesulphonic acid.[3] Addition of water to the acidic solution induces a sharp conformational transition in the polymer. The c.d. spectrum in 89.1% (w/w) methanesulphonic acid shows a marked negative shoulder at 225 nm. (curve 1 of the Figure): the solution was perfectly clear and the c.d. spectrum did not change with time. In 59·1% methanesulphonic acid the c.d. spectrum is characteristic of a polypeptide having the conformation of a right-handed α-helix, with negative dichroic bands at 225 and 210 nm., and a positive band at 196 nm. (curve 2a in the Figure). In this case the interpretation of the c.d. spectrum in terms of conformation is unequivocal, since in the polymer there are no optically active side-chain chromophores whose contributions to the optical activity could overlap those from the peptide transitions. The intensities of the c.d. bands in the spectrum recorded in 59·1% methanesulphonic acid immediately after the preparation of the solution (which was slightly opalescent) are about 50% lower than those expected for the pure α-helical form.[4] Furthermore it was observed that the c.d. diminishes and the turbidity increases as the solution ages (curve 2b in the Figure).

Two hypotheses can be invoked to explain the low intensities of the c.d. bands in 59·1% methanesulphonic acid. First, the polymer may not be completely in the helical form at this solvent composition. Unfortunately, it was not possible to increase the water content above 40% since extensive polymer precipitation occurs.

Second, if very small precipitated polymer particles are present in solution, the intensity of the dichroic bands may be reduced to a large extent (50% or more), even when the polymer is completely in the helical form. According to Urry and his co-workers, this modification in the c.d. pattern of incompletely homogeneous solutions of helical polymers arises both from absorption flattening and from scattering distortion.[4c]

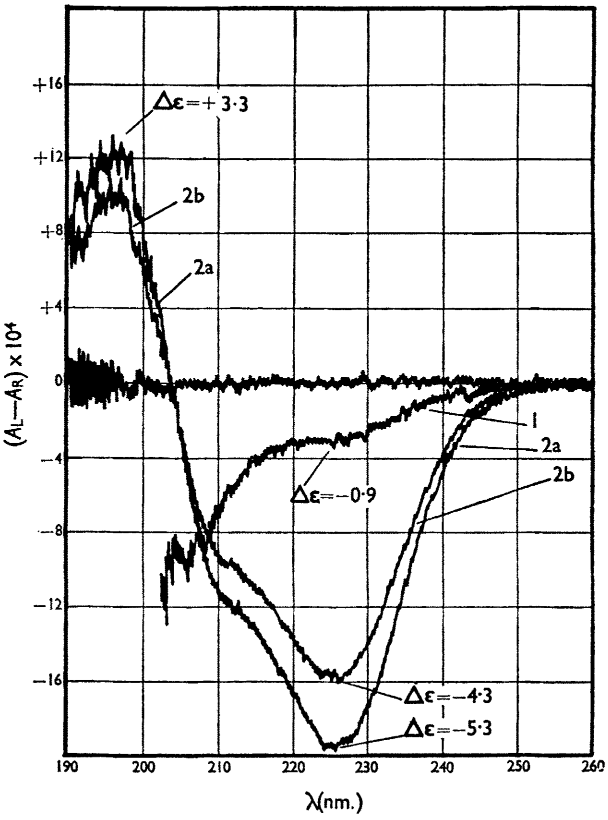

FIGURE. *Original c.d. spectra of poly-*L*-cyclohexylalanine (recorded on a Roussel–Jouan Mod. II Dichrograph) in methanesulphonic–water mixtures*: Curve 1: 89·1% $MeSO_3H$ (*polymer concentration* 0·510 *g./l.*). Curves 2: 59·1% $MeSO_3H$ (*polymer concentration* 0·560 *g./l.*). *Spectrum recorded immediately after the preparation of the solution* (2a) *and 2 hr. after the preparation of the solution* (2b).

The Δε *values reported in the Figure have been calculated according to the usual equation. In all measurements a* 0·1 *cm. cell was used.*

The observed time-dependence of the c.d. spectrum strongly supports the second hypothesis, *i.e.*, poly-L-cyclohexylalanine in 59·1% methanesulphonic is completely in the right-handed α-helical form, the observed low intensities of the c.d. bands being due to the presence of very small precipitated polymer particles in the system.

More detailed and complete results including c.d. and u.v. measurements on poly-L-cyclohexylalanine in solvent mixtures of various compositions and on a block copolymer [DL-glutamic acid]$_m$[L-cyclohexylalanine]$_n$ in water will be reported elsewhere.

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