

## The Use of Polyhexamethylenecarbodi-imide, an Insoluble Condensing Agent, in Peptide Synthesis

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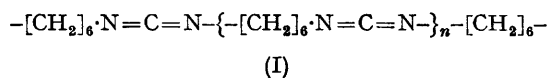
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RECENTLY peptide and polypeptide synthesis on a polymer support has been developed with striking success. In Merrifield "solid-phase peptide synthesis" the growing polypeptide chain is bound to an insoluble polymer while the *N*-blocked amino-acid is in solution.<sup>1</sup> A "reverse Merrifield" method has been developed lately in which the active ester of the *N*-blocked amino-acid is bound to the insoluble polymer, the free peptide ester being in solution.<sup>2,3</sup>

The carbodi-imides are well-known condensing agents in peptide synthesis.<sup>4</sup> Various polymers containing the carbodi-imide functional group in their backbone chain have been known for some time as polymers with film- and fibre-forming capabilities.<sup>5</sup>

We report the use of an insoluble polycarbodi-imide as a condensing agent in peptide synthesis. The condensing agent is bound to an insoluble carrier while the acylamino-acid, as well as the growing peptide chain, is in solution. Among various polycarbodi-imides which have been

tested, best results were obtained using polyhexamethylenecarbodi-imide (I).



The polymer (I) was obtained by the catalytic decarboxylation of 1,6-di-isocyanate hexane using 3-methyl-1-phenyl-3-phospholene 1-oxide as a catalyst,<sup>6,7</sup> and dry *N*-methyl-2-pyrrolidone as a solvent. The resulting polymer was treated with ethanol to block terminal isocyanate groups present, filtered off, ground, and fractionated by extraction with boiling methylene chloride in order to remove any low-molecular-weight compounds. The product was then treated with acetyl *N*-hydroxysuccinimide<sup>8</sup> which acetylates any free amino-group which might have been formed during the polymerization.

Z-Gly-Gly-OEt was obtained by suspending 20 mmole of compound (I) in methylene chloride containing Z-Gly-OH (2.5 mmole), HCl-Gly-OEt

(2.5 mmole), and  $E_3N$  (2.5 mmole), with stirring for 12 hr. The polymer was removed by filtration and washed with methylene chloride; the organic solvent was then removed *in vacuo*. The residue was dissolved in wet ethyl acetate and washed with 1*N*-HCl, 5%  $NaHCO_3$ , and water. On evaporation, a crystalline product remained, yield 92%, m.p. 79–80° (reported<sup>9</sup> m.p. 82–83°). Analogously Pht-Gly-L-Glu-(OBz)<sub>2</sub> was obtained from Pht-Gly-OH and HCl-L-Glu-(OBz)<sub>2</sub>; yield 89%, m.p. 92–94°,  $[\alpha]_D^{25} - 16.8$  [*c* 2.0 (EtOH)] {reported<sup>10</sup> m.p. 90–93°,  $[\alpha]_D^{23} - 17.1$  [*c* 2.0 (EtOH)]}, NS-di-Z-L-Cys-Gly-OBz was obtained from NS-di-Z-L-Cys-OH and HCl-Gly-OBz;

yield 93%, m.p. 116–118°,  $[\alpha]_D^{25} - 44.8$  [*c* 2.0 (dimethylformamide)] {reported<sup>11</sup> m.p. 118–119°,  $[\alpha]_D^{25} - 45.5$  [*c* 2.0 (DMF)]}, and acetyl *N*-hydroxysuccinimide from acetic acid and *N*-hydroxysuccinimide; yield 82%; m.p. 129–130° (reported<sup>8</sup> m.p. 130°).

Work is under way to use this reagent in the synthesis of various low- and high-molecular-weight peptides as well as in the synthesis of nucleotides and polynucleotides.

All compounds reported in this Communication gave satisfactory nitrogen analyses.

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