

Drs. R. G. Hayter and R. D. Feltham for discussions and their interest in this work.

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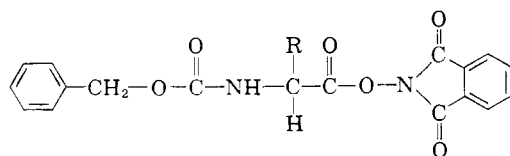
L. VASKA  
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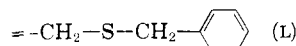
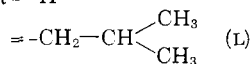
### A NOVEL ACTIVATED ESTER IN PEPTIDE SYNTHESIS

Sir:

Bodanszky and du Vigneaud<sup>1,2</sup> have introduced the *p*-nitrophenylesters of N-carbobenzoxycarboxylic acids for the synthesis of oligopeptides. The advantage of this method is obvious and has been discussed thoroughly by these authors. In extending this conception it seemed of interest to us to study activated esters of the general type (I) for their properties as N-acylating agents.

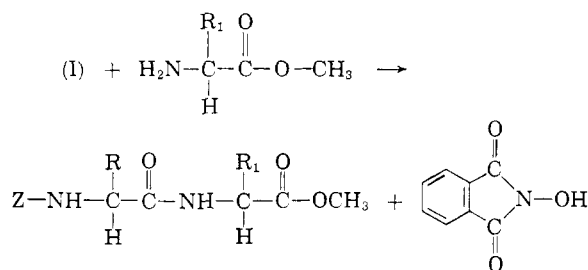


I, R = -H



and Z-L-prolyl-hydroxyphthalimide ester

These activated esters (I) are synthesized readily from N-carbobenzoxycarboxylic acids and N-hydroxyphthalimide<sup>3,6</sup> in the presence of dicyclohexylcarbodiimide.<sup>3</sup> They crystallize with ease from ethanol or carbon tetrachloride and have been obtained in yields of 40–80% of the theory depending on the amino acid employed. In the presence of another amino acid ester, the active esters (I) react within seconds, at 0° and in quantitative fashion with formation of a protected dipeptide according to the equation



The N-hydroxyphthalimide can be removed completely by shaking the reaction mixture with an aqueous solution of sodium bicarbonate in which the anion of the former is readily soluble with formation of a brilliant red coloration. With this method Z-L-leucyl-L-leucine-methyl ester and Z-glycyl-D,L-phenylalanine methyl ester have been synthesized in excellent yield and purity.

(1) M. Bodanszky, *Nature*, **163**, 1324 (1959).

(2) M. Bodanszky and V. du Vigneaud, *J. Am. Chem. Soc.*, **81**, 5688 (1959).

(3) D. F. Elliot and D. W. Russell, *Biochem. J.*, **66**, 49P (1957).

The tripeptide Z-glycyl-L-phenylalanylglycine methyl ester, synthesized by stepwise addition of glycine-ethyl ester to Z-L-phenylalanylhydroxyphthalimide ester, then decarbobenzoxylation and a new addition of Z-glycylhydroxyphthalimide ester, showed no trace of racemization in the test according to Anderson and Callahan.<sup>4</sup>

For the preparation of N-hydroxyphthalimide 1 mole of hydroxylamine hydrochloride and 2 moles of triethylamine are heated in 500 ml. of absolute ethanol until complete dissolution has occurred. To the hot solution 1 mole of N-carboethoxyphthalimide<sup>5,6</sup> is added at once with mechanical stirring. The solution changes to a deep red, owing to the formation of the triethylammonium salt of N-hydroxyphthalimide.

After immediate cooling to room temperature the solution is poured into 3 l. of acidified water. The product crystallizes spontaneously in form of fine, nearly colorless needles. After filtration, washing with water and drying over P<sub>2</sub>O<sub>5</sub> *in vacuo* the product is suitable for the preparation of activated esters; m.p. 230°; yield 70% (of theoretical).

The substance otherwise is identical with the previously described compound.<sup>7,8</sup> Details of this procedure will be published shortly.

**Acknowledgment.**—We are indebted to Professor F. Zilliken for valuable discussions.

(4) G. W. Anderson and F. M. Callahan, *J. Am. Chem. Soc.*, **80**, 2902 (1958).

(5) G. H. L. Nefkens, *Nature*, **185**, 309 (1960).

(6) G. H. L. Nefkens, G. I. Tesser and R. J. F. Nivard, *Rec. trav. chim.*, **79**, 688 (1960).

(7) L. Cohn, *Ann.*, **205**, 295 (1880).

(8) N. I. Putohkin, *J. Russ. Phys. Chem. Soc.*, **62**, 2203 (1930).

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### PEPTIDE SYNTHESIS VIA OXIDATION OF HYDRAZIDES

Sir:

Recently it was observed<sup>1</sup> that  $\gamma$ -glutamylhydrazide reacts with two equivalents of N-bromosuccinimide (NBS) to give pyrrolidonecarboxylic acid and nitrogen in quantitative yield. Accordingly, it appeared that this procedure under proper conditions might serve as a new method for peptide synthesis.

Z.Gly.NHNH<sub>2</sub> and Gly.OBz(NO<sub>2</sub>).HBr<sup>2</sup> were coupled instantaneously upon addition of two equivalents of NBS. The reaction was carried out in an ice-bath in the presence of three equivalents of triethylamine (TEA) using tetrahydrofuran (THF) as solvent. Z.Gly-Gly.OBz(NO<sub>2</sub>) was isolated after two minutes from the reaction mixture by addition of about 5 volumes of water, and recrystallized from ethanol-water, m.p. 99°, yield 86% (*Anal.* Calcd. for C<sub>13</sub>H<sub>15</sub>O<sub>7</sub>N<sub>3</sub>: C, 56.85; H, 4.77; N, 10.47. Found: C, 57.07; H, 4.95; N, 10.51). The over-all reaction may be summarized as shown

(1) P. M. Gallop, S. Seifter and C. Franzblau, unpublished results.

(2) Z, benzyloxycarbonyl; OBz(NO<sub>2</sub>), *p*-nitrobenzyl ester.