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PREPARATION OF N-(2-BENZOYL-1-METHYLVINYL)AMINO ACID DICYCLOHEXYLAMMONIUM SALTS FOR PEPTIDE SYNTHESIS

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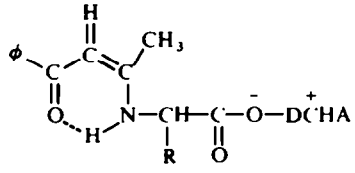
Abstract—An improved method for synthesizing highly acid labile N-(2-Benzoyl-1-methylvinyl)amino acids is reported. The derivatives were prepared as dicyclohexylamine salts thus overcoming the limitations experienced with potassium salts.

DANE *et al.*[1] successfully used the 2-benzoyl-1-methylvinyl (BMV) group for protection of α-amino nitrogen during peptide synthesis. The chief advantage of this group lies in its ready removal under mildly acidic conditions wherein labile side chain protecting groups (e.g., trityl, S-benzhydryl, and O-t-butyl) remain intact.[2a, b] We recently reported[3] that the BMV group and a new benzhydryl resin were useful in solid phase petide synthesis, allowing cleavage of the peptide from the solid support under very mild conditions. Prior to our report[3] BMV amino acids were always synthesized as potassium salts, which we found to be hygroscopic and insoluble in preferred solvents such as methylene chloride, chloroform, etc. The potassium salts also tend to be unstable and in our experience difficult to prepare in high yield and purity. The purpose of this paper is to report the ready synthesis of BMV amino acids as dicyclohexylammonium salts (Table 1) which we find to be non-hygroscopic, stable, soluble in a variety of solvents and can be prepared in high yield and purity.

The spectra of the BMV derivatives in ethanol showed λ_{max} 345 mμ (ε = 18–22,000) and λ_{max} 242 (ε = 9000). Examination of proton resonance spectra taken in deuterated chloroform revealed the presence of a singlet vinyl hydrogen at 5·64 ppm, the vinyl Me at 2·05 ppm and the N—H doublet (J = 7 c/s) at 11·6 ppm. These data and the NMR studies of Dudek[4] on α-β unsaturated β-ketoamines strongly suggest that BMV amino acid dicyclohexylammonium salts exist as Structure I in chloroform.



I

EXPERIMENTAL

All m.ps were determined with a Thomas Hoover Uni-Melt apparatus. Optical rotations were measured on a Rudolph Model 220 polarimeter. TLC was performed with silica gel G plates using the solvent system

TABLE 1. N-(2-BENZOYL-1-METHYLVINYL)AMINO ACID DICYCLOHEXYLAMMONIUM SALTS

Amino acid	Composition Theory C	H	N	S	Found C	H	N	S	M.P.	$[\alpha]_D^{25}$	Yield	Crystallization solvent
L-Alanine	72·42	9·23	6·75	–	72·19	9·30	6·73	—	163–164	+159·1 (C, 1 EtOH)	60	CH_2Cl_2–ether
L-Arginine	60·36	6·96	17·59	—	60·22	7·02	17·35	—	265–266[d]	Insoluble	85	
L-Asparagine	68·24	8·59	9·18	—	68·00	8·37	8·97	—	166–167	+38·03 (C, 2 EtOH)	73	CH_2Cl_2–ether
β-Benzyl-L-Aspartate	72·24	8·08	5·10	—	72·33	8·35	5·38	—	111–115	+16·68 (C, 2 EtOH)	84	„
N-Xanthyl-L-asparagine[a]	73·44	7·43	6·59	—	73·32	7·38	6·72	—	181–183	−79·79 (C, 2 EtOH)	83	DMF–ether
L-Nitro-arginine[a]	61·74	8·14	15·42	—	60·90	8·58	15·55	—	103[d]	+12·02 (C, 2 DMF)	80	CH_2Cl_2–ether
S-Benzhydryl-L-cysteine[c]	74·47	7·89	4·57	5·23	74·67	8·07	4·84	5·45	135–137	−138·7 (C, 1 EtOH)	59	„
S-Trityl-L-cysteine[c]	76·70	7·60	4·06	4·65	76·67	7·49	4·09	4·70	186–188[d]	−151·0 (C, 1 EtOH)	66	„
L-Glutamine	68·47	9·15	8·87	–	68·25	8·93	8·71	—	163–165	+48·05 (C, 2 EtOH)	69	CH_2Cl_2–ether
N-Xanthyl-L-glutamine[d]	73·70	7·58	6·45	—	73·82	7·70	6·54	—	198–200[d]	+8·17 (C, 1 EtOH)	58	DMF–ether
Glycine	71·96	9·05	6·99	—	71·76	9·17	7·27	–	171–173	—	88	CH_2Cl_2–ether
L-Histidine	64·20	5·74	14·05	—	64·18	5·47	13·74	—	225–226[d]	Insoluble	53	DMF–ether
Nim-Benzyl-L-histidine	73·65	8·12	9·82	–	73·40	8·20	10·13	—	165–167	−146·65 (C, 2 MeOH)	55	CH_2Cl_2–ether
L-Isoleucine	73·64	9·71	6·13	—	73·39	9·81	6·18	—	149–151	+145·1 (C, 1 EtOH)	55	„
L-Leucine	73·64	9·71	6·13	—	73·78	9·58	6·03	–	117–119	+118 (C, 2 MeOH)	60	CH_2Cl_2–Pentane
ε-Carbobenzoxy-L-lysine	71·25	8·63	6·92	—	71·21	8·50	7·13	—	135–137	+48·89 (C, 2 EtOH)	57	CH_2Cl_2–ether
L-Methionine	68·31	8·91	5·90	6·75	68·47	8·91	5·68	6·65	168–170	+20·64 (C, 2 EtOH)	87	$CHCl_3$–ether
L-Phenylalanine	76·09	8·62	5·71	—	76·03	8·55	5·61	—	152–154	−276·8 (C, 1 EtOH)	70	CH_2Cl_2–ether
L-Proline	73·78	8·94	6·83	—	73·51	8·91	6·79	–	186–187[d]	+94·10 (C, 1 EtOH)	76	„
L-Serine	70·06	8·47	6·54	–	69·80	8·36	6·26	—	192–193	+98·88 (C, 2 MeOH)	79	Ethanol
O-t-butyl-L-serine[e]	71·56	9·52	5·75	—	71·60	9·29	5·57	—	165–166	−1·139 (C, 1 EtOH)	89	b
L-Threonine	70·23	9·07	6·30	—	70·00	8·93	6·15	–	203–204	+114·18 (C, 2 MeOH)	82	$CHCl_3$–ether
O-t-Butyl-L-threonine[e]	71·96	9·66	5·60	—	71·96	9·50	5·60	—	174–177	−10·7 (C, 2 EtOH)	58	Ether–petether
L-Tryptophane	74·82	8·18	7·93	–	74·71	8·18	7·68	—	195–197[d]	−344·4 (C, 2 MeOH)	85	EtOH–petether
O-t-Butyl-L-tyrosine[e]	73·56	9·35	5·19	—	73·45	9·15	5·09	—	156–159	−339 (C, 2 EtOH)	52	CH_2Cl_2–ether
L-Valine	73·59	9·15	6·35	—	73·39	9·41	6·10	—	161–162	+189·1 (C, 1 EtOH)	90	CH_2Cl_2–ether

[a] BMV nitroarginine DCHA could not be prepared by the general procedure (Experimental)

[b] BMV-O-t-butyl serine DCHA could not be crystallized but could be obtained as a solid by light petroleum trituration

[c] Prepared by the procedure of R. G. Hiskey and J. B. Adams, *J. Org. Chem.* **30**, 1340 (1965)

[d] Prepared by the procedure of S. Akabori *et al.*, *Bull. Chem. Soc. Japan* **34**, 739 (1961)

[e] Prepared by the procedure of E. Wünsch and J. Jentsch, *Chem. Ber.* **97**, 2490 (1964)

isopropanol–acetic acid–water–pyridine (30:6:24:20). All amino acids were of commercial quality except as noted in the Footnotes for the Table.

General procedure. To 95·6 mmoles of amino acid suspended in 10 ml water was added a soln of 18·88 ml (96 mmoles) dicyclohexylamine in 80 ml MeOH followed by the addition of a hot soln of 15·6 g (96 mmoles) benzoylacetone in 80 ml EtOH. The mixture was refluxed with stirring for 3 hr and filtered while hot to remove any unreacted amino acid. The filtrate was concentrated *in vacuo* and the residue twice evaporated to dryness *in vacuo* following the addition of two 100-ml portions of propanol-2. Crystallization was effected from the appropriate solvent system.

N-(2-*Benzoyl*-1-*methylvinyl*)-*nitro-L-arginine dicyclohexylammonium salt.* A mixture of nitro-L-arginine (2·62 g; 11·95 mmoles), dicyclohexylamine (2·36 ml, 12 mmoles)[5] and benzoyl acetone (1·95 g, 12 mmoles) in DMF (100 ml) was stirred at 50° for 120 hr. The insoluble unreacted nitroarginine was removed by filtration and the filtrate was evaporated *in vacuo* to a yellow oil. Trituration with ether and collection yielded 4·39 g (83%) of product, m.p. 101–107, $[\alpha]_D^{25}$ + 12·02 (*c*, 2 DMF). (Found: C, 61·49; H, 8·21; N, 15·15; O, 15·15. Calc for $C_{28}H_{44}N_6O_5$: C, 61·74; H, 8·14; N, 15·42; O, 14·68%).

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