A Selective Electrocatalytic Cleavage of the Benzyloxycarbonyl Group from Peptides

M. Antonietta Casadei,[a] D. Pletcher*[b]

[a] Instituto di Chimica Farmaceutica e Tossicologica, Università "La Sapienza", P. le Aldo Moro 5, I-00185 Roma, Italy

[b] Department of Chemistry, The University, Southampton SO9 5NH, UK

An electrosynthetic procedure for the cleavage of the benzyloxycarbonyl group from protected amino acids and peptides is described. It is based on the use of a high surface area palladium cathode in methanol/acetic acid and gives an excellent selectivity under very mild conditions.

Benzyloxycarbonyl (the Z group) is a widely used protecting group in peptide synthesis in homogeneous solution.[1,2] Its removal from protected peptides has been commonly carried out by catalytic hydrogenation on a palladium on carbon powder.[3] The reaction, however, requires the use of hydrogen gas, occasionally at elevated pressure, and to simplify the experimental procedure, catalytic transfer hydrogenation[4] has also been used for cleavage of the benzyloxycarbonyl group.[5,6] In such reactions, the protected peptide is refluxed in the presence of palladium on carbon in methanol, ethanol or in alcohol/acetic acid mixtures containing cyclohexene as the hydrogen donor.

The present paper describes a convenient electrocatalytic approach where the cleavage occurs selectively and rapidly on a palladium cathode prepared by electrodeposition onto graphite. Again it avoids the use of hydrogen gas and may also be carried out at room temperature or below. Moreover it should show interesting differences from catalytic hydrogenation with respect to the stability of other protecting groups to the reaction. Since the first studies by Horner and Neumann[7,8] of the cathodic cleavage of protecting groups, many papers have reported the use of electrolysis for such reactions,[9,10] but only at non-catalytic electrodes. The benzyloxycarbonyl group is only cleaved at a very negative reduction potential[11,12] which makes the use of such reactions in synthesis quite impractical.

The appropriate conditions for the cleavage were sought using benzyloxycarbonylmorpholine as a model compound. Early electrolyses with platinised platinum and Raney nickel cathodes led only to the recovery of the starting material but with a palladised graphite electrode, cleavage always occurred. The faradaic efficiency increased when in methanol as solvent, the electrolyte was changed from sodium methoxide to sodium perchlorate to sodium perchlorate and acetic acid. In the final solution, the formation of toluene and morpholine was linear with charge passed and the yields of toluene and morpholine were 90% and 92% respectively after 2 F; the conversion could be made quantitatively by increasing the charge passed to 2.8F. The cathode potential at the chosen current density, 15 mA cm^{-2}, was in the range -0.5 V to -0.9 V *vs* SCE (cf. -2.8 V at a vitreous carbon cathode in dimethylformamide[12]). The cleavage reaction was equally successful in ethanol or using as cathode, palladium on carbon powder sprinkled onto a nickel plate for electrical contract.

Electrolyses were carried out for the benzyloxycarbonyl derivatives of two amines and six peptides: in each case the electrode was palladised carbon, the current density 15 mA cm^{-2} and the catholyte methanol/2.5% acetic acid containing 0.5 mol/L sodium perchlorate. The yields of products were determined by weighing the extracted peptides whose identities were checked by mass spectroscopy, IR and NMR and comparison of their melting points (or simple derivatives) with literature values or

Table. Electrocatalytic Deprotection of *N*-Benzyloxycarbonyl Compounds[a]

Protected Compound	Solvent	Charge/F	Yield[b] Toluene (%)	Yield[c,d] of Peptide (%)	m.p. (°C)[e]	Lit. m.p. (°C)
Z-morpholine	CH_3OH	2.0	90	92[b,f]	–	–
		2.8	99	~100[b,f]		
Z-NHC_6H_{11}	CH_3OH	2.0	82	81[b,f]	–	–
		3.5	100	99[b,f]		
Z-Leu-Ala-OC_2H_5	CH_3OH	2.0	80	–	–	–
		4.0	83	–		
	CH_3OH[g]	2.0	82	–		
		4.0	98	92	238–240 (dec.)	238–240[h]
Z-Phe-Gly-OC_2H_5	CH_3OH[g]	2.0	80	–		
($[\alpha]_D^{20}$: $-16°$)[i]		4.0	97	99[j]	132–134[k]	135–136[13]
Z-Phe-Leu-OC_2H_5	CH_3OH[g]	2.0	78	–		
		4.0	94	95	158–160[k]	158–160[14]
	C_2H_5OH[g]	2.0	72	–		
		4.0	90	–		
Z-Leu_2-OCH_3	CH_3OH[g]	2.0	78	–		
		4.0	99	95	182–184[k]	181–184[15]
Z-Gly_3OH	CH_3OH[g]	2.0	75	–		
		4.0	92	90	260–264 (dec.)	262–265[16]
Z-Leu_4OCH_3	CH_3OH[g]	2.0	74	–		
($[\alpha]_D^{20}$ $-73°$)[i]		4.0	90	96[l]	146–148	146–148[h]

[a] Electrolysis conditions: Pd/C cathode in a divided cell. Catholyte was alcohol/2.5% acetic acid + $NaClO_4$ (0.5 mol/L). Substrate concentration 0.1 mol/L. I = 15 mA cm^{-2}; cathode potentials were in the range -0.6 to -1.0 V SCE.

[b] Determined by GLC.

[c] Yield of isolated product.

[d] All products gave satisfactory IR and NMR data.

[e] Uncorrected.

[f] Product is amine.

[g] To maintain activity of the cathode, the current was reversed for 20 seconds each 10 min.

[h] m. p. of sample prepared by literature method.[6]

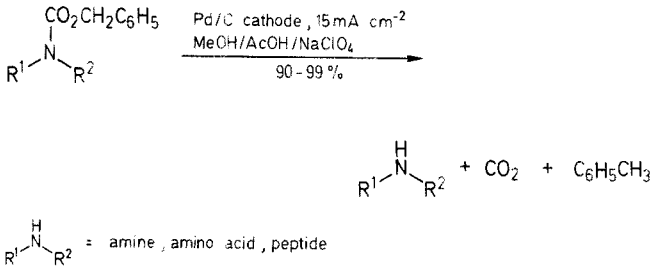
[i] $c = 2$, MeOH.

[j] $[\alpha]_D^{20}$ $-15.5°$.[i]

[k] m. p. of the hydrobromide.

[l] $[\alpha]_D^{20}$ $-73.5°$.[i]

those of standard samples; optical rotations were measured for the starting material and products from two reactions. The yields of both simple amines and peptides are excellent (Table).



The electrolyses of the peptide derivatives differed slightly from those of the model compound in two respects. Firstly, the reactions tended to stop at about 80% conversion and in order to obtain complete cleavage it was necessary to reactivate the catalytic surface of the cathode by reversing the current for 20 seconds every 10 minutes. If this activation is carried out, the cathode may be reused many times. Secondly, late in the electrolyses, when the concentration of substrate has dropped, the current efficiency for the cleavage drops markedly (hydrogen evolution can be seen) and it was necessary to reduce the current density or to pass 3–5 F of charge in order to carry out the reaction to completion: this does not diminish the organic yield. It should also be noted that the product isolation is straightforward from this electrolysis medium.

Of course, it is necessary to examine the stability of other protecting groups in these reaction conditions. Preliminary experiments have demonstrated that the tosyl group is not cleaved by electrolysis in the conditions described above. Similarly, it appears that the benzyl group is not cleaved from benzyl methyl ether or benzyl methylthio ether. On the other hand, the benzyl group is readily cleaved from benzyl acetate and more slowly from benzyldimethylamine and hence the procedure should be used with caution when the peptide includes a benzyl ester or a tertiary benzylamine group.

It is interesting to note the unique properties of palladium as a catalyst for the cleavage of the benzyloxycarbonyl group, whether the reaction is carried out catalytically or electrolytically. The potential for the cleavage and the form of cyclic voltammograms confirm this reaction to occur by an electrocatalytic mechanism. It is probable that the benzyl group adsorbs well or in an appropriate configuration on a palladium surface.

Preparation of the Palladium/Graphite Cathode:[17]

In these experiments, the electrode is a graphite disc, area 2 cm^2, mounted in polytetrafluoroethylene so that only one face of the disc is exposed to the catholyte. The plating bath is $PdCl_2$ (5 g/L) in 1 mol/L aq. HCl. Initially the graphite disc is rubbed on fine emery paper and then electroplated with a current density of 35 mA cm^{-2} for 15 min. Periodically fresh palladium is added to the surface by passing 10 mA cm^{-2} for 5 min.

Deprotection of Benzyloxycarbonyl (Z) Amino Acids and Peptides; Typical Procedure:

Z–Phe–Leu–OC_2H_5 (0.3 g, 0.78 mmol) and nonane (50 mg, internal standard for GC) are dissolved in MeOH (8 mL) containing $NaClO_4$ (0.5 mol/L) and acetic acid (0.4 mol/L). This solution is placed in the catholyte compartment of a divided cell, deoxygenated with a stream of N_2 and then electrolysed at the palladium/graphite cathode with a current density of 15 mA cm^{-2} (at room temperature). The formation of toluene is monitored by GC. After 4 F of charge, the yield of toluene is 94% and the reaction is terminated. The catholyte is evaporated *in vacuo* at 40 °C on a rotary evaporator. The residue is dissolved in water and this solution is made alkaline by the addition of KOH before being extracted with CH_2Cl_2 (3 × 20 mL). The organic phase is then dried (Na_2SO_4) and the solvent evaporated; yield: 180 mg (95%). The product is converted to the hydrobromide; m.p. 158–160 °C (acetone/petroleum ether) (Lit.[14] 158–160 °C).

The authors would like to express sincere thanks to Professor H.L.S. Maia of the Universidade do Minho, Braga, Portugal, for the gift of the benzyloxycarbonyl derivatives used in this work. One of us (M.A.C.) would like to thank NATO for a Science Fellowship (NATO-CNR 215.18/03).

Received: 3 April 1987; revised: 6 July 1987

(1) Greenstein, J.P., Winitz, M. *Chemistry of the Amino Acids*, Vol. 2, John Wiley and Sons, New York 1961.
(2) *The Peptides, Vol. 3: Protection of Functional Groups in Peptide Synthesis*, Gross, E., Meienhofer, J. (eds.), Academic Press, Orlando FL, 1981.
(3) Bergmann, M., Zervas, L. *Ber. Dtsch. Chem. Ges.* **1932**, *65*, 1192.
(4) Brieger, G., Nestrick, T.J. *Chem. Rev.* **1974**, *74*, 567.
(5) Jackson, A.E., Johnstone, R.A.W. *Synthesis* **1976**, 685.
(6) Anantharamaiah, G.M., Sivanandaiah, K.M. *J. Chem. Soc. Perkin Trans 1*, **1977**, 490.
(7) Horner, L., Neumann, H. *Chem. Ber.* **1965**, *98*, 1715.
(8) Horner, L., Neumann, H. *Chem. Ber.* **1965**, *98*, 3462.
(9) Mairanovsky, V.G. *Angew. Chem.* **1976**, *88*, 283; *Angew. Chem. Int. Ed. Engl.* **1976**, *15*, 281.
(10) Montenegro, M.I. *Electrochim. Acta* **1986**, *31*, 607.
(11) Maia, H.L.S., Medeiros, M.J., Montenegro, M.I., Pletcher, D. *Portuguese Electrochim. Acta* **1984**, *2*, 1.
(12) Maia, H.L.S., Medeiros, M.J., Montenegro, M.I., Pletcher, D. *J. Electroanal. Chem.* **1986**, *200*, 363.
(13) Vaughan, J.R. *J. Am. Chem. Soc.* **1952**, *74*, 6137.
(14) Yang, C.-S., Blaha, K., Rudinger, J. *Collect. Czech. Chem. Commun.* **1964**, *29*, 2633.
(15) Mazur, R.H., Schlatter, J.M. *U.S. Patent* 3280099 (1966), G.D. Searle Co; *C.A.* **1968**, *68*, 22258.
(16) Dunn, M.S., Brophy, T.W. *J. Biol. Chem.* **1932**, *99*, 221.
(17) Nonaka, T., Takahashi, M., Fuchigami, R. *Bull. Chem. Soc. Jpn.* **1983**, *56*, 2584.