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ONE STEP SYNTHESIS OF ORTHOGONALLY PROTECTED DIAMINODICARBOXYLIC ACIDS BY MIXED KOLBE ELECTROLYSIS

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Abstract: Orthogonally protected 2,5-diaminoadipic, 2,6-diaminopimelic, and 2,7-diaminosuberic acid derivatives bearing up to four different protecting groups are prepared in one step by mixed Kolbe electrolysis. The use of 2-[(4-methylphenyl)sulfonyl]ethyl group is crucial for the isolation of the desired orthogonally protected products. © 1997 Elsevier Science Ltd.

In 1988 Laerum *et al.*[1] reported that the dimer **1** [(Pyr-Glu-Asp)$_2$-Cys-(Lys)$_2$] of the hematoregulatory peptide HP5b stimulates myelopoiesis in vitro. Replacement of the metabolic labile cystine of **1** by its dicarbaanlogue *L,L*-2,7-diaminosuberic acid (DAS) resulted in SK&F 107647 [**2**, (Pyr-Glu-Asp)$_2$-DAS-(Lys)$_2$].[2] This peptide displayed higher hematoregulatory activity than compound **1**. SK&F 107647 has been shown to be efficeous in various animal models of bacteria, fungal and viral infections.[2] A structure-activity-relationship study[2] of the novel low molecular weight hematoregulatory peptide **2** resulted in analogue **3**, containing diaminoadipic acid (DAA) instead of 2,7-diaminosuberic acid which was more active than **2**.

Diaminopimelic acid (DAP) is an other important, naturally occurring diaminodicarboxylic acid found in bacteria and higher plants. The biological importance of DAP has stimulated the preparation of orthogonally protected derivatives of *meso*-DAP by a multistep synthesis.[3,4]

Additional examples in which 2,7-*L,L*-diaminosuberic acid was used as substitute for cystine are oxytocin[5] and somatostatin analogues.[6] The resulting dicarba analogues of these biologically active cystine peptides have shown biological activity and enhanced metabolic and chemical stability. This is due to the absence of a reducible disulfide linkage.

In general four different amino acids are connected to the two amino and two carboxylic groups of cystine in the biologically active peptides. Orthogonally protected diaminodicarboxylic acid derivatives are required for the selective synthesis of the corresponding dicarba analogues which are biologically more stable. For a research program we were interested in a short and flexible approach which allows the synthesis of

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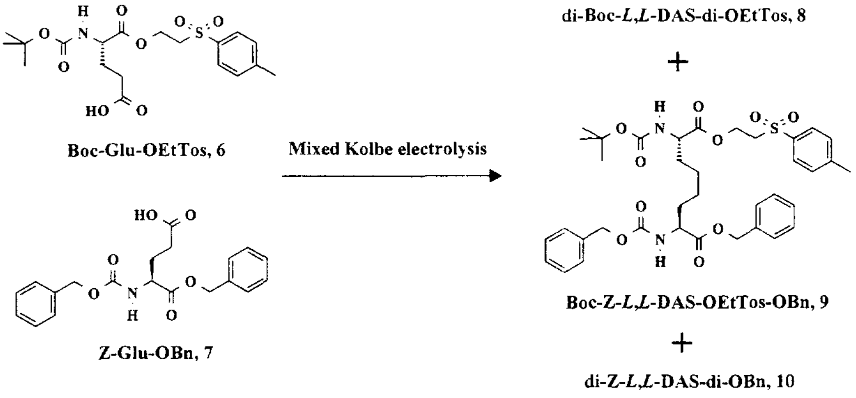
diaminodicarboxylic acid derivatives with i) the desired number of carbons in the skeleton; ii) the desired stereochemistry at the two asymmetric centers; and iii) with the desired protecting groups.

Retro-synthetic analysis revealed that the mixed Kolbe electrolysis fulfills the expected requirements. In the light of the usefulness of the Kolbe synthesis in preparing orthogonally protected diaminodicarboxylic acid derivatives it seems surprising that this reaction is not used more frequently. To the best of our knowledge there is only the paper of Nutt *et al.*[7] who prepared orthogonally protected diaminosuberic acid derivative (Boc-Z-*L*,*L*-DAS-OtBu-OMe, **4**, see Scheme 2) by Kolbe electrolysis. They mentioned that the desired unsymmetrical dimer, **4**, was only difficultly separable from the two symmetrical dimers (di-Z-*L*,*L*-DAS-di-OMe, di-Boc-*L*,*L*-DAS-di-OtBu) because of similar solubility and polarity. When we synthesized Boc-Z-*L*,*L*-DAS-OtBu-OBn (**5**, mp 51-56°C, $[\alpha]_D = -21.5$ (1% in $CHCl_3$), isolated yield by HPLC: 5.5%) by mixed Kolbe electrolysis we also faced the same problem. As the protecting groups determine to a high extent the physical properties of the product the selection of appropriate protecting groups with different polarity in the starting materials should allow to overcome this problem. To prove this hypothesis we carried out the mixed Kolbe electrolysis (see Scheme 1) of Boc-Glu-OEtTos (**6**)[8] and Z-Glu-OBn (**7**). The 2-(4-methylphenylsulfonyl)ethyl (EtTos) group[9] was selected because i) it can be cleaved selectively by nonnucleophilic organic bases (e. g. 1,5-diazabicyclo[4.3.0]non-5-ene, DBU) in the presence of other esters; and ii) it is very polar in comparison to a Boc and Z-group, and Me, tBu, and Bn esters; and iii) the EtTos group was already used successfully in the Kolbe electrolysis in our labs.[8] The commercially not available starting materials Boc-*D*-Glu-OEtTos (mp 96-100°C) , Boc-Asp-OEtTos (mp 108-110°C), and its D-enantiomer were prepared in analogy to **6**.



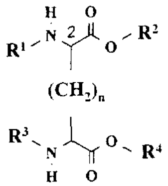
Scheme 1: Mixed Kolbe electrolysis of Boc-Glu-OEtTos and Z-Glu-OBn

The Kolbe electrolysis[10] of **6** and **7** yielded di-Boc-*L*,*L*-DAS-di-OEtTos (**8**)[8], Boc-Z-*L*,*L*-DAS-OEtTos-OBn (**9**), and di-Z-*L*,*L*-DAS-di-OBn (**10**)[8] in a statistical ratio of 1 : 2 : 1 (see Scheme 1). The Rf-values of **8**, **9**, and **10** were 0.2, 0.5, and 0.8 (plate: silica gel 60F254, solvent system: PE / EE = 1 / 1). We were pleased to isolate orthogonally protected product **9** just by flash chromatography in 12.3% yield. Although the yields obtained in the mixed Kolbe reaction (see Table 1) were not as high as might be desired, the yield of this one step reaction is comparable to the overall yield of multistep sequences reported in the literature for the preparation of orthogonally protected diaminopimelic acids (Ref. 4a: 9 steps, overall yield: 9%; Ref. 4d: 6 steps, overall yield: 14%). The mixed Kolbe electrolysis of Boc-*L*-Glu-OEtTos and Boc-*D*-Glu-OBn proved to be very useful for the preparation of di-Boc protected *meso*-diaminosuberic acid **12** free from any contaminants of the other stereoisomers. The protected *meso*-DAS **11** was isolated in 16.3% yield (entry 2, Table 1). Saponification with LiOH yielded **12** (mp 200°C, decomposition) in 83%.

Table 1:Orthogonally Protected Diaminodicarboxylic Acid Derivatives Prepared by Mixed Kolbe Electrolysis

Entry	Starting materials	Product	Yield; mp	$[\alpha]_D$
1	Boc-Glu-OEtTos(**6**) Z-Glu-OBn (**7**)	Boc-Z-*L*,*L*-DAS-OEtTos-OBn (**9**)	12.3%; 90-92°C	+3.45 (4.9% in $CHCl_3$)
2	Boc-*D*-Glu-OBn Boc-*L*-Glu-OEtTos	di-Boc-*D*,*L*-DAS-OBn-OEtTos (**11**)	16.3%; oil	+4.05 (15% in $CHCl_3$)
3	Boc-Asp-OEtTos Z-Asp-OBn	Boc-Z-*L*,*L*-DAA-OEtTos-OBn (**14**)	5.1%; oil	+5.85 (4.4% in $CHCl_3$)
4	Boc-Glu-OEtTos Z-Asp-OBn	Boc-Z-*L*,*L*-DAP-OEtTos-OBn (**15**)	11.2% oil	-2.9 (5.3% in $CHCl_3$)
5	Boc-*L*-Glu-OEtTos Z-*D*-Asp-OBn	Boc-Z-*D*,*L*-DAP-OEtTos-OBn (**16**)	10.1% oil	+2.14 (23.9% in $CHCl_3$)
6	Boc-*D*-Glu-OEtTos Z-*D*-Asp-OBn	Boc-Z-*D*,*D*-DAP-OEtTos-OBn (**17**)	12.9% oil	+2.74 (4.2% in $CHCl_3$)

In contrast the isolation of di-Boc-*meso*-DAS-di-OBn from the mixed Kolbe reaction of Boc-*L*-Glu-OBn and Boc-*D*-Glu-OBn failed due to similarity in Rf-value. On the other hand treatment of **11** with DBU removed selectively the EtTos group yielding compound **13** (di-Boc-*D*,*L*-DAS-OBn-OH, 62%, oil, $[\alpha]_D$ = +14 (6% in $CHCl_3$). The preparation of orthogonally protected diaminoadipic (**14**, entry 3) and diaminopimelic acid derivatives (**15-17**, entries 4-6) by mixed Kolbe electrolysis is summarized in Table 1. Compound **14** represents the first orthogonally protected diaminoadipic acid derivative.[11]



4: n = 4, (2*L*, 7*L*); R^1 = Boc, R^2 = tBu, R^3 = Z, R^4 = Me
5: n = 4, (2*L*, 7*L*); R^1 = Boc, R^2 = tBu, R^3 = Z, R^4 = Bn
8: n = 4, (2*L*, 7*L*); R^1 = Boc, R^2 = EtTos, R^3 = Boc, R^4 = EtTos
9: n = 4, (2*L*, 7*L*); R^1 = Boc, R^2 = EtTos, R^3 = Z, R^4 = Bn
10: n = 4, (2*L*, 7*L*); R^1 = Z, R^2 = Bn, R^3 = Z, R^4 = Bn
11: n = 4, (2*D*, 7*L*); R^1 = Boc, R^2 = Bn, R^3 = Boc, R^4 = EtTos
12: n = 4, (2*D*, 7*L*); R^1 = Boc, R^2 = H, R^3 = Boc, R^4 = H
13: n = 4, (2*D*, 7*L*); R^1 = Boc, R^2 = Bn, R^3 = Boc, R^4 = H
14: n = 2, (2*L*, 5*L*); R^1 = Boc, R^2 =EtTos, R^3 = Z, R^4 = Bn
15: n = 3, (2*L*, 6*L*); R^1 = Boc, R^2 = EtTos, R^3 = Z, R^4 = Bn
16: n = 3, (2*L*, 6*D*); R^1 = Boc, R^2 = EtTos, R^3 = Z, R^4 = Bn
17: n = 3, (2*D*, 6*D*); R^1 = Boc, R^2 = EtTos, R^3 = Z, R^4 = Bn

Scheme 2: Synthesized diaminodicarboxylic acids

In summary we have demonstrated that the mixed Kolbe electrolysis allows the selective generation of molecules with two chiral centers and bearing up to four different protecting groups. The EtTos group simplifies the isolation of the orthogonally protected diaminodicarboxylic acid derivatives and avoids any contamination with unwanted stereoisomers in the mixed Kolbe electrolysis. This one step synthesis provides the medicinal chemist with versatile starting materials for the replacement of cystine in peptides and proteins. As diaminodicarboxylic acids have two amino and two carboxylic functionalities in the molecule, the orthogonally protected derivatives are new and unique building blocks for combinatorial chemistry

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REFERENCES AND NOTES

1 Laerum, O. D.; Sletvold, O.; Bjerknes, R.; Eriksen, J. A.; Johansen, J. H.; Schanche, J. S.; Tverteraas, T.; Paukovitz, W. R. *Exp. Hematol.* **1988**, *16*, 274-280.

2 Bhatnagar, P. K.; Agner, E. K; Alberts, D.; Arbo, B. E.; Callahan, J. F.; Cuthbertson, A. S.; Engelsen, S. J.; Fjerdingstad, H.; Hartmann, M.; Heerding, D.; Hiebl, J.; Huffman, W. F.; Hysben, M.; King, A. G.; Kremminger, P.; Kwon, C.; LoCastro, S.; Løvhaug, D.; Pelus, L. M.; Petteway, S.; Takata, J. S. *J. Med. Chem.* **1996**, *39*, 3814-3819.

3 Cox, R. J.; Sherwin, W. A.; Lam, L. K. P.; Vederas, J. C. *J. Am. Chem. Soc.* **1996**, *118*, 7449-7460 and references cited therein.

4 a) Jurgens, A. R. *Tetrahedron Lett.* **1992**, *33*, 4727-4730. b) Williams, R. M.; Yuan, C. *J. Org. Chem.* **1992**, *57*, 6519-6527. c) Bold, G.; Allmendinger, T.; Herold, P.; Moesch, L.; Schär, H.-P.; Duthaler, R. *Helv. Chim. Acta* **1992**, *75*, 865-882. d) Holcomb, R. C.; Schow, S.; Ayral-Kaloustian, S.; Powell, D. *Tetrahedron Lett.* **1994**, *35*, 7005-7008.

5 Keller, O. *Dissertation*, E.T.H. Zürich, 1974.

6 Nutt, R. F; Veber, D. F.; Saperstein, R. *J. Am. Chem. Soc.* **1980**, *102*, 6539-6545.

7 Nutt, R. F.; Strachan, R. G.; Veber, D. F.; Holly, F. J. *J. Org. Chem.* **1980**, *45*, 3078-3080.

8 Hiebl, J.; Blanka, M.; Gutman, A.; Kollmann, H.; Leitner, C.; Mayrhofer, G.; Rovenszky, F.; Winkler, K. submitted to *Tetrahedron.*

9 HOEtTos is available from Loba Feinchemie AG, A-2401-Fischamend, Austria.

10 Typical experimental procedure: Boc-Glu-OEtTos (34.68g, 80.78mmol) and Z-Glu-OBn (30.0g, 80.78mmol) were dissolved in a 3 / 1 mixture of methanol / pyridine (240ml). The solution was transferred to an electrolysis cell equipped with a cooling jacket and Pt gauze electrodes arranged in cylinder form. A methanolic solution of sodium methoxide (30% w/w, 0.5ml) in methanol was added and the electric current (5 A at 50-80 Volts) passed through the cell keeping the temperature between 18 to 25 °C by cooling with a cryostat. The reaction was sampled periodically (TLC: ethyl acetate (EA) / petroleum ether (PE) = 1 / 1) and once all the starting material was consumed the electrolysis was stopped. The solution was concentrated at reduced pressure, dissolved in EA, washed with diluted HCl-solution, saturated $NaHCO_3$, and with brine until neutral. The organic phase was dried (Na_2SO_4), filtered, and the filtrate was concentrated. The residue was filtered over silica gel using EA / PE = 1 / 4 as eluent. The crude material was purified over RP-18 using acetonitrile / water =2 / 1 to yield 3.54g (12.3 %) of **9**. MS: 611.6 (M^++1-Boc-group). $^1H(CDCl_3$, 400MHz): 1.18(m, 4, 2 CH_2), 1.35(s, 9, $(CH_3)_3C$), 1.49-1.80(m, 4, 2 CH_2) 2.36(s, 3, 2 tolyl-CH_3), 3.33(m, 2, $OC\mathit{H}_2CH_2SO_2C_7H_7$), 3.98(m, 1, CH), 4.31(m, 1, CH), 4.35(t, 2, J = 6.2Hz, $OCH_2C\mathit{H}_2SO_2C_7H_7$), 4.78(m, 1, NH), 5.02(s, 2, benzyl-CH_2), 5.09(AB-system, 2, J = 11Hz, benzyl- CH_2), 5.3(m, 1, NH), 7.2-7.32(m, 12, 10 phenyl-H, 2 tolyl-H), 7.70(d, 2, J = 8.2Hz, 2-tolyl-H).

11 Please note that diaminoadipic acid derivatives are not available by Schöllkopf route according Ref. 4c (footnote 3 on page 868).

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