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(2) We have determined the structures of $Rh_2(bridge)_4^{2+}$ (ref 1b: and Mann, K. R.; Thich, J. A.; Bell, R. A.; Coyle, C. L.; Gray, H. B., to be submitted for publication) and $Rh_2(bridge)_4Cl_2^{2+}$ (ref 1c; and Mann, K. R.; Bell, R. A.; Gray, H. B. *Inorg. Chem.*, in press) in crystals.

(3) In a typical experiment 0.012 mmol of $Rh_2(bridge)_4^{2+}$ produced 0.005 mmol of H_2 upon reaction with 12 M HCl (5 mL) in the dark. Irradiation (>520 nm) of the resultant blue solution yielded an additional 0.006 mmol of H_2. Independent confirmation of the oxidation level of $[Rh_2(bridge)_4Cl^{2+}]_n$ was obtained in redox titrations. Thus, in 6 M HCl 1 equiv of Cr^{2+} reduces $Rh_2(bridge)_4Cl_2^{2+}$ to $(1/n)[Rh_2(bridge)_4Cl^{2+}]_n + Cl^-$, and 1 equiv of Ce^{4+} $(+Cl^-)$ reacts with $(1/n)[Rh_2(bridge)_4Cl^{2+}]_n$ to produce $Rh_2(bridge)_4Cl_2^{2+}$ (Sigal, I. S.; Gray, H. B., to be submitted for publication).

(4) Higher sulfate concentrations result in spectral shifts consistent with sulfate binding; thus, $\lambda_{max} = 590$ nm in 32 N H_2SO_4. We distinguish ligation from ion pairing, which our data suggest to be extensive for $M(H_2SO_4) > 10^{-2}$.

(5) (a) Flash photolysis experiments at Santa Cruz employed a homemade apparatus. Dilute (10^{-6}–10^{-5} M) degassed samples held in 15-cm pathlength cells were excited by a coaxialed xenon flash lamp (output $\lesssim$90 J, $\tau_{1/2} \sim 20$ μs), and monitored with a DC quartz/iodine lamp whose output was filtered to isolate appropriate spectral regions. (b) Laser flash photolysis measurements were made at the University of Southern California on an instrument that has been described previously (Gutierrez, A. R.; Adamson, A. W. *J. Phys. Chem.* **1978**, *82*, 902).

(6) Decay was second order for over 5 half-lives, and the rate constant was independent of total rhodium concentration and flash pulse intensity.

(7) The rate is nearly independent of ionic strength for >1 N H_2SO_4.

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(11) Fe(II) product was determined as the tris(*o*-phenanthroline) complex. The quantitative formation of the Rh(II) product, which has been independently characterized as the product of Ce(IV) thermal oxidation of the H_2SO_4(aq) solutions and has an absorption maximum at 311 nm (ϵ 33 600), was determined by the spectral changes that occur during the photoreaction, and by the clean reduction back to starting material that occurs upon addition of a large excess of Fe^{2+}.

(12) Assuming our kinetic scheme (eq 1, 2, 4), the limiting product quantum yield for (3) should be twice the primary quantum yield ϕ_0 for (1). Therefore, $\phi_0 \simeq 0.01$. Measurements of ϕ_0 from the magnitude of transient signal in the flash photolysis experiments were in reasonable agreement. The transient quantum yield increases at lower ionic strength and is much larger in CH_3CN, suggesting that cage recombination is important.

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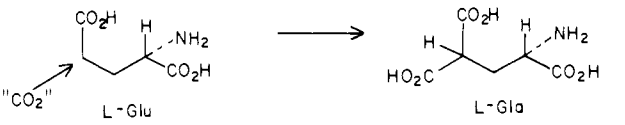
A Simple Synthesis of L-γ-Carboxyglutamate and Derivatives Thereof

Sir:

The curious amino acid, L-γ-carboxyglutamate (Gla), has been encountered in several of the vitamin K dependent blood clotting factors, including prothrombin.[1] Activation of prothrombin to thrombin apparently involves preliminary calcium ion mediated binding of the former to membrane phospholipids,[2] thereby increasing the effective concentrations of the necessary factors. Gla units, which comprise 10 of the first 38 amino acid residues in prothrombin, are thought to be implicated in the calcium binding sites of prothrombin.[3]

Not surprisingly, interest in the chemical synthesis of Gla (and derivatives that are suitably modified for convenient incorporation in peptides) has been high. Early approaches, however, resulted in racemic products.[4] Schwyzer did report the resolution of a synthetic Gla derivative,[5] as well as a multistep preparation of another optically active analogue by a chirally specific Strecker-like reaction.[6]

It was our intention to solve the challenge implicit in the synthesis of L-Gla by providing for the introduction of a carboxyl group on the 4 position of the readily available L-glu-

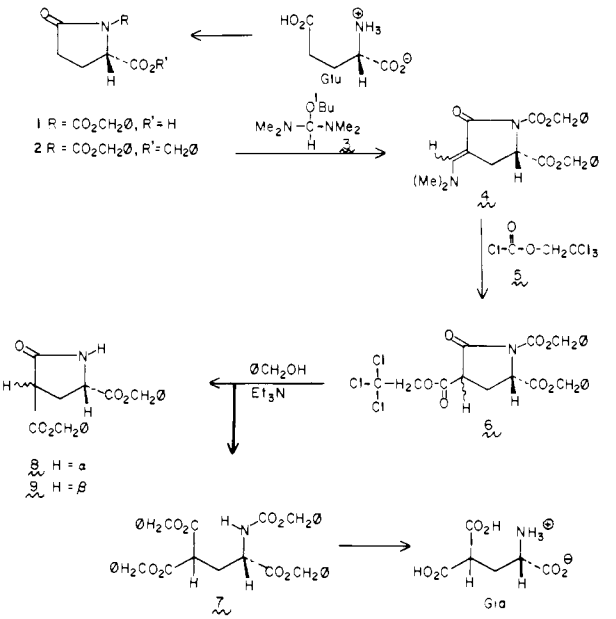


tamic acid (L-Glu), while preserving its *S* chirality. The successful realization of this goal is described below.

Our starting material was the commercially available (Sigma) Glu derivative, *N*-carbobenzyloxy-L-pyroglutamate (**1**), which was converted (triethylamine-benzyl chloride-acetone) into the benzyl ester **2**: mp 107–108 °C; $[\alpha]_D$ −40.7° (*c* 1.1, ethanol); lit.[7] mp 110 °C; $[\alpha]_D$ −39.5°; 93% yield. Compound **3** reacts with Bredereck's reagent, **3**[8,9] (3 equiv of **3**; dimethoxyethane; 70 °C; 3.5 h), to afford a 95% yield of enamine **4**[10a,b] (mp 92–93°; $[\alpha]_D$ −32.8° (*c* 1.4, chloroform), which is apparently a single compound, though of undetermined geometric configuration (see Scheme I). An interesting transformation ensues when **4** is exposed to the action of 2,2,2-trichloroethoxycarbonyl chloride (**5**) (3 equiv of **5** in benzene; reflux; 36 h). Filtration and silica gel chromatography afford a 41% yield of the diastereomeric lactams, **6**.[10a] *It should be emphasized that this transformation was discovered by chance in a closely related series and its scope and limitations have not yet been ascertained.*

Compound **6** was treated with excess benzyl alcohol in the presence of triethylamine (3.80 g of **6**; 150 mL of benzyl alcohol; 0.5 mL of dry triethylamine; 104 °C; 27 h). Evaporation of the excess benzyl alcohol followed by chromatography of the residue on silica gel afforded the tribenzyl ester of N-Cbz-Gla (**7**):[10a] $[\alpha]_D$ +7.8° (*c* 1.6, $CHCl_3$); 62% yield. The infrared and NMR spectra as well as the chromatographic mobility of **7** were identical with those of an authentic sample of the racemic product, prepared according to Weinstein[4f] and

Scheme I



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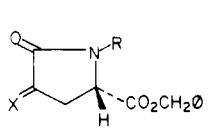
purified by high-pressure liquid chromatography (LC) collection.

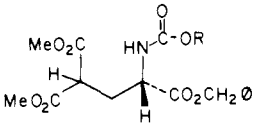
This reaction also afforded a 10% yield of a 3:2 mixture of *trans*- and *cis*-pyro-Gla derivatives **8**[10a] and **9**,[10a] which were purified by LC collection. These compounds do not noticeably react further with benzyl alcohol under the above-described conditions. Thus, the first step in the reaction of **6** with benzyl alcohol is an exchange of the trichloroethyl ester for a benzyl ester. This is then followed by opening of the pyrrolidine ring with minor loss from competitive decarbobenzyloxylation[11] of the N-Cbz function.

Hydrogenolysis of **7** in methanol over 10% palladium/charcoal afforded a 93% yield of L-Gla: mp 154–155 °C dec;[12] $[\alpha]_D$ +33.9° (*c* 1.2, 6 N HCl); lit.[5] $[\alpha]_D$ +34.6°; mp 167–167.5 °C dec. Its NMR spectrum (600 MHz) was identical with that of racemic material prepared from racemic **7** and, in accord, with the published spectrum[5] at 360 MHz.

This methodology lends itself very nicely to the preparation of analogues of L-Gla. Thus, reaction of **6** with methanol in the presence of triethylamine (room temperature, 2.5 h) afforded a 62% yield of **10**:[10a] $[\alpha]_D$ +9.5° (*c* 1.0, $CHCl_3$). A more differentiated analogue was prepared as follows.

Reaction of benzylpyroglutamate (**11**) with **5** in the presence of pyridine–DMAP afforded **12**: mp 61–63 °C; $[\alpha]_D$ −46.2° (*c* 1.03, ethanol). The latter reacts with the Bredereck reagent **3**[8,9] at *room temperature* in dimethoxyethane, providing a 43%[13] yield of **13**:[10a,b] mp 118.5–119 °C; $[\alpha]_D$ −66.7° (*c* 1.0, CH_2Cl_2). Treatment of **13** with **5** in toluene at 95 °C, followed





11 R = H, X = H_2

12 R = C(O) OCH_2CCl_3, X = H_2

13 R = C(O) OCH_2CCl_3, X = C(H)N(Me)$_2$

14 R = C(O) OCH_2CCl_3, X = H,C(O) OCH_2CCl_3

6 → **10** R = $CH_2\emptyset$

14 → **15** R = CH_2CCl_3

by chromatography on silica gel, gave a 41% yield of the trans:cis (ca. 2:1) diastereomers **14**.[10a] Reaction of **14** with methanol, as above, gave **15**:[10a] $[\alpha]_D$ +7.8° (*c* 5.2, CH_2Cl_2); 35%[13] yield.

It is expected that this methodology will find ready application in the synthesis of peptides containing modified L-Gla residues.

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(9) In spite of numerous publications by Bredereck and his associates[8b] on the reactions of orthoformamide derivatives with "active" carbon–hydrogen bonds, the value of this beautiful methodology has perhaps still not been fully comprehended. For some recent applications of Brederecks findings to general synthetic problems, see: (a) secoalkylation: Trost, B. M.; Preckel, M. *J. Am. Chem. Soc.* **1973**, *95*, 7862; (b) formation of α-dicarbonyl compounds: Wasserman, H. H.; Ives, J. L. *J. Org. Chem.* **1978**, *43*, 3238; (c) formation of α-methylene lactones: Danishefsky, S.; Hirama, M.; Gombatz, K.; Harayama, T.; Berman, E.; Schuda, P. F. *J. Am. Chem. Soc.* **1978**, *100*, 6536; (d) formation of α-phenylthiomethylene lactones: Martin, S. F.; Moore, D. R. *Tetrahedron Lett.* **1976**, 4459; (e) formation of α-formyl lactones: Gutzwiller, J.; Pizzolato, G.; Uskokovic, M. *J. Am. Chem. Soc.* **1971**, *93*, 5907; Ban, Y.; Taga, N.; Oishi, T. *Tetrahedron Lett.* **1974**, 187.

(10) The structure of this compound is consistent with (a) its infrared, NMR, and mass spectra and (b) its combustion analysis.

(11) Small amounts of dibenzyl carbonate were isolated by chromatography.

(12) The racemate of Gla is reported[4f] to melt from 90 to 92 °C. In our hands, L-Gla decomposes when heated at 154–155 °C by decarboxylation. This decomposition point does not change materially on recrystallization. The discrepancy between our decomposition temperature and that reported might be a consequence of a small amount of racemate in our material, but more likely it is a consequence of subtle differences in heating.

(13) The lower yields of the *N*-trichloroethoxycarbonyl series arise from the greater lability of this group toward nucleophiles in the sequence.

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Structure of Elasnin, a Novel Elastase Inhibitor

Sir:

A novel human granulocyte elastase inhibitor, elasnin, has been isolated from the culture broth of *Sm. noboritoensis* KM-2753.[1] This inhibitor with low toxicity is of interest in respect to the control of pathological processes such as acute arthritis, various inflammations, pulmonary emphysema, and pancreatitis, which are known to be caused by elastase.[2] In the present report, we describe the structural elucidation of elasnin based on chemical degradations and biosynthetic means using ^{13}C-labeled precursor.

Elasnin (**1**) is a lipophilic colorless and viscous oil possessing the following physical and spectroscopic properties: n^{17}_D 1.4983; $[\alpha]^{18}_D$ −0.9° (*c* 1, EtOH); λ^{EtOH}_{max} 291 nm (ε 7760); ν 3430 (OH), 2960 and 2860 (CH_2, CH_3), 1715 (ketone carbonyl), 1665 (conjugated ester carbonyl), and 1636 cm^{-1} (double bond). The molecular formula $C_{24}H_{40}O_4$ (M^+, *m/e* 392) was established for **1** by high-resolution mass spectral and elemental analyses. The 25.2-MHz ^{13}C NMR spectrum indicated the presence of a ketone carbonyl (δ 207.0), an ester carbonyl (either δ 165.5 or 164.7), and four quarternary olefinic carbons (either δ 165.7 or 164.7, 153.8, 115.0, 104.3). In addition, four methyl carbons (completely overlapped at δ 13.9), several methylene carbons (δ 22.4–40.3), and a methyne (δ 54.7) in the high-field region, as shown by an off-resonance decoupling experiment, were consistent with the presence of four linear C_4 and C_5 alkyl chains in **1**.

Reduction of **1** with $NaBH_4$ afforded dihydroelasnin (**2**) [$C_{24}H_{42}O_4$; M^+, *m/e* 394; λ^{EtOH}_{max} 292 nm (ε 6400)], indicating the presence of a ketone carbonyl at the position isolated from the chromophore in **1**. Ozonolysis of **2** followed by oxidative degradation with 30% H_2O_2 and glacial acetic acid gave an oily acid **3**, which was then treated with CH_2N_2 to give methyl ester **4**: $C_{12}H_{24}O_3$; *m/e* 213 ($M^+ - OH$), 199 ($M^+ - OCH_3$); ν 3520 (OH), 1730 cm^{-1} (ester carbonyl). The ^{13}C NMR and mass spectral analyses[3] of **3** and the corresponding monoacetate **5**, obtained by acetylation of **3**, suggested 2-butyl-3-hydroxyoctanoic acid for the structure of **3**.

Acidic degradation of **1** with 2.5 N HCl in acetic acid gave a colorless oil, **6**: $C_{23}H_{40}O_2$; M^+ *m/e* 348 (348.300; calcd for

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