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Synthesis of Novel Imidazole-Containing DNA Minor Groove Binding Oligopeptides Related to the Antiviral Antibiotic Netropsin

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Analysis of the structural and stereochemical requirements for the strict DNA base sequence recognition of $(AT)_4$ and $(AT)_5$ respectively for the oligopeptide minor groove binding agents netropsin and distamycin leads to proposals for the rational structure modification for altered base and sequence recognition. The syntheses of new analogues of the natural oligopeptide antiviral antibiotic netropsin bearing one or more imidazole moieties in place of pyrrole are described in order to test this hypothesis. In this regard nitration of ethyl 1-methylimidazole-2-carboxylate was investigated. During the reduction of nitroimidazole derivatives with stannous chloride, a novel chlorination of the imidazole nucleus was encountered.

Introduction

Several alternative approaches may be considered in attempting to design sequence-specific DNA binding ligands for possible application as gene control agents. One approach, which is being developed by several groups, is to use the inherent specificity of β-oligonucleotides.[1-3] The latter have certain disadvantages associated with difficulties of transport across membranes,[1,2] susceptibility to rapid intracellular nuclease degradation,[4,5] and in requiring single stranded DNA as a cellular target.[1] We are currently addressing the problem of nuclease sensitivity of such probes by exploring the properties of unnatural α-oligonucleotides.[6-8] Alternatively groove selective agents offer advantages as probes for molecular recognition in that they target for duplex DNA and their binding does not incur helix distortion as in the use of intercalators.[9,10] We have chosen as our starting point netropsin[11,12] and distamycin,[13] which are members of a modest family of natural oligopeptides including anthelvencin,[14] kikumycin B,[15] amidinomycin,[16] and noformycin,[17] which exhibit antibiotic, antiviral, and antitumor activity. Evidence from a study of their biochemical pharmacology indicates that they act to block the template function of DNA by binding to specific nucleotide sequences in the minor groove of double helical DNA.[12] These sequences are $(AT)_4$ and $(AT)_5$ respectively for netropsin and distamycin.[12] Examination of the structural requirements for the molecular recognition, deduced, in part, from recent X-ray studies on a complex of netropsin with a dodecamer,[18] suggested that the replacement of one or more pyrrole rings by imidazole, or other appropriate heterocycle, should result in a rational alteration of base recognition from AT to GC.[19] The latter prediction follows from the implied formation of new hydrogen bonds between G(2)-NH_2 in the minor groove and

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(19) R. E. Dickerson, Molecular Biology Institute, UCLA has independently suggested the substitution of imidazole for pyrrole in netropsin on the basis of X-ray diffraction analysis of the antibiotic cocrystallized with a dodecamer:[18] Dr. Dickerson and his co-workers are currently attmpting similar cocrystallization and X-ray analysis with out compounds **26** and **36** bound to a duplex oligodeoxyribonucleotide.

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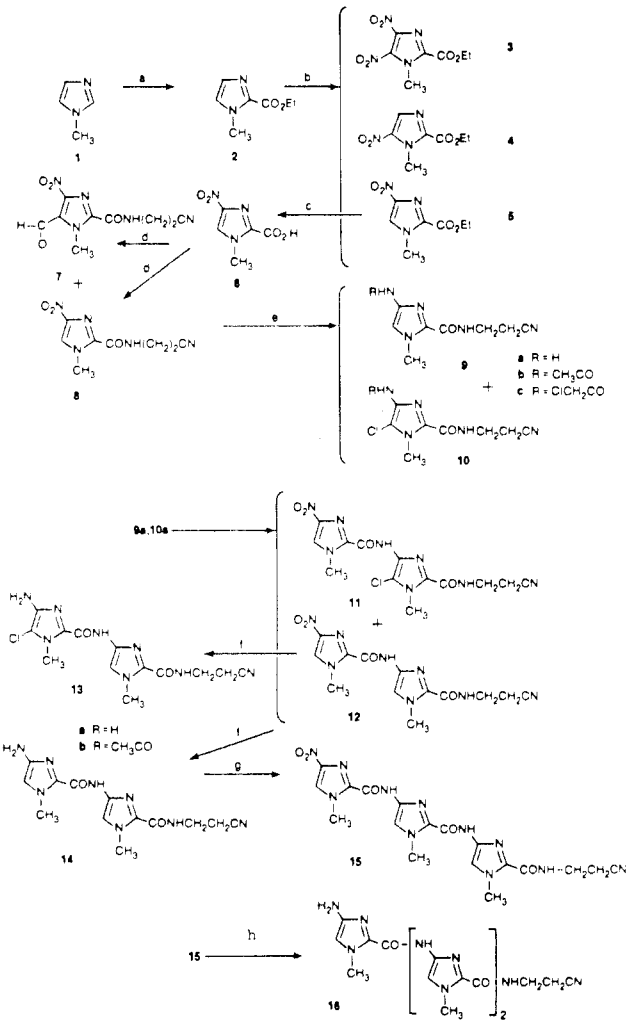
the N3 of the imidazole moiety. We have recently confirmed this prediction[20] and now report details of the syntheses of these novel oligopeptide analogues. This report includes certain synthetic aspects of imidazole nitration, reduction of nitroimidazoles with stannous chloride and the concomitant chlorination of the imidazole ring, as well as efforts to improve the efficiency of introduction of the guanidineacetyl moiety in this class of oligopeptide agents.

Synthetic Strategy. The structure of the antibiotic netropsin was established in 1963 by Julia and Préau-Joseph.[11] Since that time syntheses of both the parent antibiotic[11] and that of certain analogues have been reported. The latter are those bearing additional pyrrole units or in which the pyrrole rings were replaced by benzene, pyridine, trimethylpyrrole, or thiophene,[21] or by changing the mode of pyrrole ring substitution from 2,4 to 2,5.[22] The syntheses of these latter compounds have been based largely on the original method of Julia and Préau-Joseph.[11] The anticipated need to examine a number of new structures in order to establish the molecular recognition characteristics for DNA binding required development of an efficient and flexible general synthesis. Therefore we recently reported new efficient total syntheses of netropsin and distamycin, which are sufficiently adaptable for the present requirements.[23] The strategy adopted involves significant changes in the methods and order of introduction of the amidine and guanidineacetyl moieties from those reported hitherto.[11,21] This resulted in better yields of the final products (Schemes I–III). An additional important advantage was the avoidance of chromatography, which is not suitable for such polar compounds as netropsin, because of contamination of the final products with inorganic salts eluted from the absorbents. The syntheses of the imidazole analogues are essentially based on our method of synthesis of netropsin[23] and distamycin[23] but with modifications necessitated by the presence of imidazole moieties.

Nitration of 1-Methylimidazole-2-carboxylic Acid. Reaction of 1-methylimidazole with ethyl chloroformate in the presence of triethylamine afforded the ester **2**.[24] Nitration of **2** afforded three products, **3**, **4**, and **5**, which were readily separable and of which only the 5-nitro derivative **4** has been described previously[25] (Scheme I). Development of this reaction showed that the highest yield of the desired compound **5** is obtained by using a 1:1 mixture of 100% nitric acid and sulfuric acid at a temperature of 95 °C for 50 min. Longer reaction times tend to give more of the undesired 4,5-dinitro derivative **3**. Reaction of **2** with a mixture of nitric acid and acetic anhydride is very slow whereas a similar reaction using trifluoroacetic anhydride is very rapid and affords largely the dinitro compound **3**. Compound **5** was isolated from the preferred procedure in crystalline form in 46.5% yield. Alkaline hydrolysis of **5** afforded the imidazole-2-carboxylic acid **6** after acidification in 95% yield. Compound **6** proved to be sensitive to decarboxylation upon heating under acidic conditions.

Scheme I[a]



[a] Reaction conditions: (a) $ClCO_2Et$, Et_3N; (b) H_2SO_4 + HNO_3; (c) aqueous NaOH, then HCl; (d) $(COCl)_2$ or pivaloyl chloride and NEt_3, add $NH_2(CH_2)_2CN$; (e) $SnCl_2 \cdot 2H_2O$ + HCl, acetyl chloride, chloroacetyl chloride or acyl chloride of **6**; (f) $SnCl_2 \cdot 2H_2O$ + HCl; (g) $EtN(i\text{-}Pr)_2$, acyl chloride of **6**; (h) Pd/H_2.

The initial attempt to condense the acid **6** with aminopropionitrile employed oxalyl chloride, which, however, afforded, in addition to the main product **8** in 80% yield, a small amount of the 5-formyl derivative **7** (Scheme I). The latter plausibly arises from initial oxalylation at position 5 followed by hydrolysis of the acyl chloride and decarboxylation. For this reason an alternative procedure was adopted employing the mixed anhydride of **6** and pivalic acid to give **8** in 91% yield.

Reduction of Nitroimidazole Derivatives with Stannous Chloride. Since catalytic reduction of nitroheterocycles was inconvenient on a larger scale, an alternative procedure was investigated. Reduction of **8** with stannous chloride afforded a mixture of two compounds **9a** and **10a**, which were not separated but were identified by means of their N-acetyl and N-chloroacetyl derivatives **9b**, **10b**, and **9c**, **10c**, respectively (Scheme I). The compositions of the derivatives **9(b,c)**, **10(b,c)** were established unequivocally by MS exact mass measurement.

In the case of chloroacetyl derivatives **9c** and **10c** the ratio of the 5-chloro derivative to the nonchlorinated one was 56:44. However, this ratio depends on the order of addition of the reactants, being higher if $SnCl_2$ is added slowly to the nitro compound dissolved in aqueous HCl, and lower if the nitro compound is added to the solution of $SnCl_2$ in aqueous HCl. Coupling of the compounds **9a**

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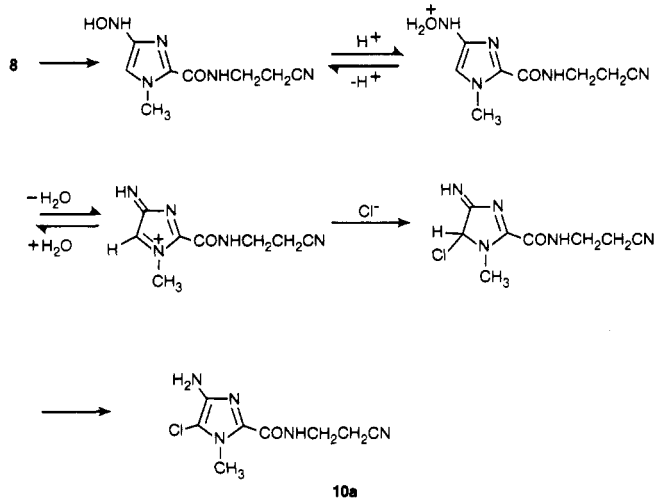
Scheme II. Possible Mechanism for the 5-Chlorination of Imidazole Which Accompanies $SnCl_2$ Reduction of 8



10a

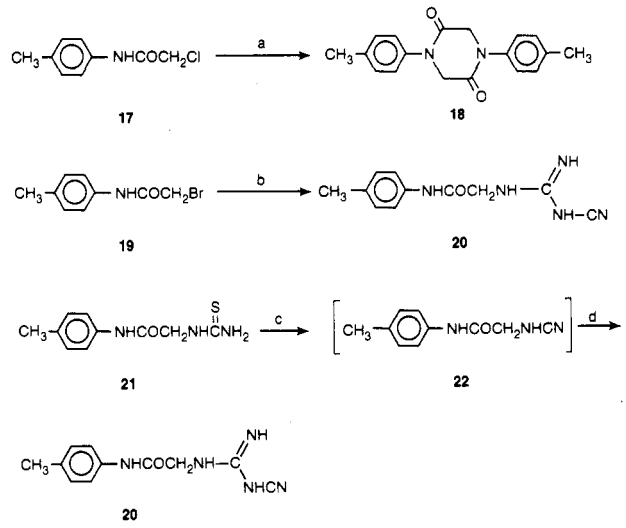
and **10a** (as a mixture) with the acid chloride of **6** gave the corresponding peptides **11** and **12** (Scheme I), which were readily isolable in 50% and 12% yields, respectively, and were identified individually. Stannous chloride reduction of compound **12** also led to two products, **13a** and **14**, which were readily separated by chromatography to give 30% and 57% yields, respectively. No such chlorination of the second imidazole ring of **12** occurs, so this reaction is evidently connected with the process of reduction of the nitro group. A large excess of stannous chloride caused a decrease in the proportion of chlorination products from about 56% to about 2%. However this cannot be due to reduction of the chloro substituent by stannous chloride, similar to that described by Rinkes,[26] because the chloro derivatives **10** and **13b** survive such prolonged treatment. We also found that neither compound **9a** nor **14** reacts with stannous chloride. We suggest that this 5-imidazole chlorination may follow the mechanism suggested in Scheme II, which is analogous to that of the reaction of *N*-phenylhydroxylamine with HF affording *p*-aminofluorobenzene[27] and that of the reduction of nitrobenzene with stannous chloride and hydrochloric acid producing a mixture of 53% of chloroaniline and 47% of aniline.[28] Small amounts of chloro derivatives formed in the presence of a large excess of $SnCl_2$ means that in the case the reduction of the intermediate hydroxylamine group prevails over the rearrangement.

Although the reduction of the nitro compounds with stannous chloride affords some of the side product due to imidazole chlorination, it was faster and more convenient on a larger scale for intermediate stages than catalytic reduction in the presence of palladium. However, in order to avoid chlorination in the step leading to the final three ring compound **15**, the nitro compound **12** was reduced catalytically with hydrogen in the presence of palladium on charcoal to give **14** in an excellent yield of 80% (Scheme I).

Coupling of the dipeptide **14** with the acid chloride of **6** afforded the tripeptide **15** in 68% yield. Similar catalytic reduction of **15** gave the amino tripeptide **16** in 65% yield.

Introduction of the Guanidineacetyl Moiety. With the bis(imidazole) dipeptide **14** and the corresponding tris(imidazole) tripeptide **16** in hand, we now turned to the attachment of the guanidineacetyl end group. The existing

Scheme III[a]



[a] Reaction conditions: (a) $CNNH_2$, NaOEt, dicyclohexano-18-crown-6; (b) CNNHLi; (c) $Hg(OAc)_2$, CNNHNa.

literature method of coupling the amine with guanidineacetic acid hydrochloride in the presence of dicyclohexylcarbodiimide (DCC) was unsatisfactory because of the low yield caused by a side reaction between DCC and aminopyrrole derivatives.[29] Therefore alternative procedures were examined. Attempted couplings of the chloroacetyl group in **9c** with guanidine carbonate, acetate, or free base were unsuccessful. A report on cyanoaminoacetic acid formation from disodium cyanamide and chloroacetic acid[30] suggested an alternative method.[31] Therefore reactions with model compounds **17** and **19** and sodium cyanamide designed to afford **22** were investigated. Reaction of the chloro compound **17** with sodium cyanamide in ethanol afforded the cyclic product **18**. In a similar reaction of the bromo compound **19** in protic solvents the cyanoguanidinyl derivative **20** was isolated (Scheme III), although the desired compound **22** is implicated as an intermediate. Presumably compound **22** is too reactive to survive in the presence of the cyanamide sodium salt. Therefore in order to avoid the basic conditions of the reaction the thiourea derivative **21** was allowed to react with mercuric acetate. Although the desired compound **22** was formed, it again proved to be unstable under the reaction conditions. Its formation was confirmed by the reaction with sodium cyanamide to afford **23**.

The above efforts being unsuccessful, we turned back to the condensation in the presence of DCC by optimizing the conditions. The best results were obtained by using 1 equiv of the nitro compound, e.g., **8**, **12**, or **15**, to be reduced, 1.5 equiv of guanidineacetic acid hydrochloride, and 1.5 equiv of DCC.

Introduction of the Amidine Moiety by Modified Pinner Reaction. Formation of the amidine moiety was effected by a modification of the Pinner reaction.[32] Our observations agree with those of Baksheev[33] that the first step of the reaction of the cyano group, i.e., formation of the imino ester with an alcohol in the presence of hydrogen chloride, is completed in 90 min and that longer reaction

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(31) All our attempts to repeat the original reaction[30] failed. Since the cyanoaminoacetic acid has not been reported in the literature subsequently, it appears unlikely that it can be isolated in monomeric form.
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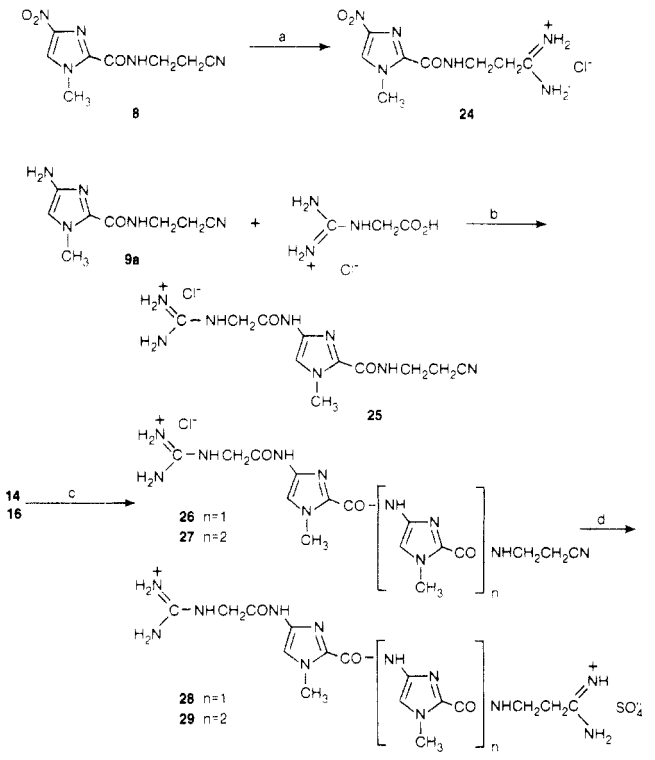
(33) Baksheev, A. N.; Gavrilov, N. I. *Zh. Obshch. Khim.* **1952**, *22*, 2021; *Chem. Abstr.* **1952**, *47*, 864/e.

Scheme IV[a]



[a] Reaction conditions: (a) CH_3OH + HCl, NH_3; (b) DCC; (c) $(H_2N)_2C^+NHCH_2CO_2H$, Cl^- and DCC; (d) EtOH + HCl, NH_3, Na_2SO_4.

times promote side reactions resulting in lower yields. The imino ester reacts readily with ammonia in ethanolic solution within 1 h at ambient temperature. The conditions for both reactions, i.e., Pinner and condensation with guanidineacetic acid, were first optimized with the monoimidazole derivatives **8** and **9a** to give **24** and **25**, respectively.

In the final step of the synthesis, involving compounds **14** and **16**, the latter were first allowed to react with guanidineacetic acid since the products, **26** and **27**, respectively, could be readily purified. Subsequent introduction of the amidine moiety to afford the final target molecules **28** and **29** proceeded almost quantitatively (Scheme IV). The latter products were isolated initially as the hydrochloride salts contaminated with a little ammonium chloride and were very soluble in protic solvents. Exchange of the counterion with sodium sulfate afforded the corresponding sulfate salts which were more readily purified by recrystallization. The composition of these polar compounds was established by FAB-MS.[34] In the case of the chloride salts $(M - Cl)^+$ or $(M - HCl - Cl)^+$ ions were observed for one or two salt functional groups, respectively. For the corresponding sulfates both MH^+ and $(M - HSO_4)^+$ ions were observed.

Synthesis of Oligopeptides Containing Both Pyrrole and Imidazole Moieties. The isomeric imidazole–pyrrole and pyrrole–imidazole dipeptides **32** and **33** were synthesized following similar procedures from **30** and **31** and 1-methyl-4-nitroimidazole-2-carboxylic acid and 1-methyl-4-nitropyrrole-2-carboxylic acid, respectively (Scheme V). After the reduction of the nitro groups, the resulting amino compounds **34** and **35** were condensed with guanidineacetic acid hydrochloride in the presence of DCC to give **36** and **37**. The Pinner reaction conditions on the

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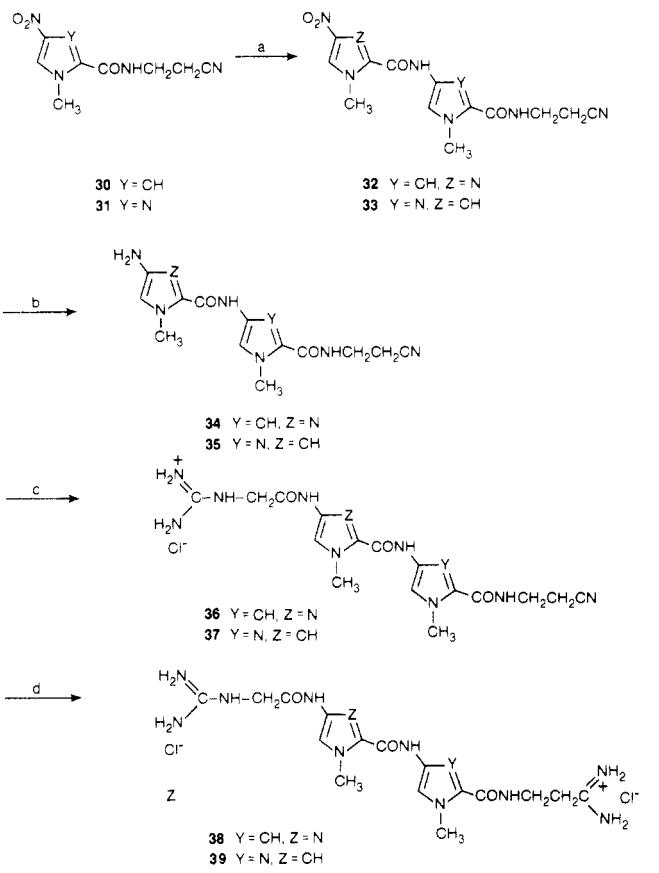
Scheme V[a]



[a] Reaction conditions: (a) 5% $Pd/C/H_2$ acyl chloride of **6** or acyl chloride of 1-methyl-4-nitropyrrole-2-carboxylic acid; (b) 10% $Pd/C/H_2$; (c) $(H_2N)_2C^+NHCH_2CO_2H$, Cl^- and DCC; (d) EtOH + HCl, NH_3.

latter compounds produced the target compounds **38** and **39**.

Initial findings on the altered DNA sequence specificity of these novel oligopeptides have been reported.[20] These studies while confirming the predicted GC-recognizing character of the imidazole moiety in turn led to other predictions concerning the contribution of certain van der Waals contacts in their molecular recognition of DNA. Appropriate models are being synthesized to elucidate these factors and their syntheses and DNA specificity will be reported in due course.

Experimental Section

Melting points were determined on a Fisher-Johns apparatus and are uncorrected. The IR spectra were recorded on a Nicolet 7199 F.T. spectrophotometer, and only the principal sharply defined peaks are reported. The ^1H NMR spectra were recorded on Bruker WH-200 and WH-400 spectrometers. The spectra were recorded on approximately 5–25% (w/v) solutions, depending upon the spectrometers, in appropriate deuteriated solvents with tetramethylsilane as internal standard. Line positions are recorded in ppm from reference. Electron impact and FAB mass spectra were determined on an Associated Electrical Industries (AEI) MS-9 and MS-50 focussing high resolution mass spectrometers. Kieselgel 60 (230–400 mesh) of E. Merck was used for flash chromatography and precoated sheets of Kieselgel 60 F_{254} of E. Merck were used for thin layer chromatography. TLC systems: (i) for covalent peptidic compounds, chloroform–methanol 9:1; (ii) for ionic compounds with one ionic pair, methanol with some acetic acid; (iii) for ionic compounds with two ionic pairs, methanol with some formic acid.

Ethyl 1-Methylimidazole-2-carboxylate (2). A solution of 3.2 mL (40 mmol) of 1-methylimidazole (Aldrich Chem. Co.) in 20 mL of anhydrous acetonitrile and 10 mL of triethylamine was

cooled to −30 °C and a solution of 7 mL (66 mmol) of ethyl chloroformate in 10 mL of acetonitrile added rapidly while maintaining the temperature at <10 °C. The mixture was set aside at room temperature overnight. Triethylamine hydrochloride was collected by filtration, the filtrate was concentrated under reduced pressure, and the residue was dissolved in water and extracted with chloroform. Chromatography on silica gel with ethyl acetate as eluent gave **2**,[24] 3.9 g (63% yield): mp 44 °C (ethyl ether/hexane); ^{1}H NMR ($CDCl_3$) δ 1.32 (t, 3 H), 3.92 (s, 3 H), 4.32 (q, 2 H), 6.95 (s, 1 H), 7.03 (s, 1 H).

When the reaction is run on a larger scale the product is purified by fractionation, bp 97 °C/1 mm. However the larger scale reaction always gives a lower yield of **2** because of less efficient cooling and the longer time required for the addition of the ethyl chloroformate (with a tenfold larger scale the yield of **2** may drop to 30%).

Nitration of Ethyl 1-Methylimidazole-2-carboxylate. A solution of 0.5 g (3.2 mmol) of **2** in 2 mL of concentrated sulfuric acid and 2 mL of 100% nitric acid was warmed for 50 min at 95 °C, and after being cooled the solution was poured onto ice and extracted several times with CCl_4 to give mixture a and then extracted with methylene chloride to give mixture b. Mixture a contains mainly ethyl 1-methyl-4,5-dinitroimidazole-2-carboxylate (**3**) and some ethyl 1-methyl-4-nitroimidazole-2-carboxylate (**5**), while mixture b contains mainly **5**, with some ethyl 1-methyl-5-nitroimidazole-2-carboxylate (**4**) and dinitro derivative **3**. TLC on silica gel with ethyl acetate/carbon tetrachloride (2:7) separates **3**, **4**, and **5** with R_f values of 0.50, 0.45, and 0.25, respectively.

Ethyl 1-Methyl-4,5-dinitroimidazole-2-carboxylate (3). Mixture a was separated on a silica gel column, eluting with ethyl acetate/carbon tetrachloride (2:7). Compound **3** was recrystallized from carbon tetrachloride/hexane as light-sensitive yellow crystals: mp 63 °C; ^{1}H NMR ($CDCl_3$) δ 1.34 (t, 3 H), 4.15 (s, 3 H), 4.37 (q, 2 H); MS, m/z (rel intensity) 244 (21) M^+, 172 (100) $(M - CO_2C_2H_4)^+$; IR ν_{max} (Nujol) 1263, 1312, 1377, 1462, 1541, 1564, 1729 cm^{-1}. Anal. Calcd for $C_7H_8N_4O_6$: C, 34.4; H, 3.3; N, 22.9. Found: C, 34.3; H, 3.4; N, 22.7.

Ethyl 1-Methyl-4-nitroimidazole-2-carboxylate (5). Mixture b was recrystallized from carbon tetrachloride with a little ethanol to give **5**, 300 mg (46.5% yield) as pale cream needles, mp 130–131 °C (EtOH). The mother liquor was concentrated and the residue recrystallized from ethanol to give **4**, 84 mg, mp 80–81 °C (lit.[25] mp 80–81 °C).

Compound **5**: ^{1}H NMR ($CDCl_3$) δ 1.45 (t, 3 H), 4.14 (s, 3 H), 4.46 (q, 2 H), 7.85 (s, 1 H); MS, m/z (rel intensity) 199, (19.3) M^+, 127 (100) $(M - CO_2C_2H_4)^+$; IR ν_{max} (Nujol) 1259, 1310, 1380, 1454, 1497, 1539, 1722, 3138 cm^{-1}. Anal. Calcd for $C_7H_9N_3O_4$: C, 42.2; H, 4.5; N, 21.1. Found: C, 42.1; H, 4.5; N, 21.4.

1-Methyl-4-nitroimidazole-2-carboxylic Acid (6). A solution of 100 mg (0.5 mmol) of **5** and 60 mg of sodium hydroxide in 2 mL of water was heated under reflux for 15 min. The warm solution (~50 °C) was then acidified with aqueous hydrochloric acid. (In hot solutions decarboxylation takes place whereas in cold solution the sodium salt of the acid precipitates together with the free acid.) The free acid **6** was isolated as a white crystalline solid, 81 mg (95% yield). A pure sample was prepared for analysis by recrystallization from water: mp 138 °C dec; IR ν_{max} (Nujol) 1206, 1301, 1340, 1391, 1461, 1503, 1549, 1727, 3150 cm^{-1}. Anal. Calcd for $C_5H_5N_3O_4$: C, 35.1; H, 2.9; N, 24.6. Found: C, 34.8; H, 2.9; N, 24.4.

3-(1-Methyl-4-nitroimidazole-2-carboxamido)propionitrile (8) and 3-(1-Methyl-5-formyl-4-nitroimidazole-2-carboxamido)propionitrile (7). (a) Coupling by means of Oxalyl Chloride. A solution of 2.05 g (12 mmol) of **6** and 10 mL of oxalyl chloride in 20 mL of dry tetrahydrofuran was heated under reflux for 30 min. The solvent and excess of oxalyl chloride were removed under reduced pressure. The residue was dissolved in tetrahydrofuran (30 mL) and cooled to −20 °C, and a solution of 1.7 mL of 3-aminopropionitrile in 10 mL of tetrahydrofuran was added. A white precipitate of the aminopropionitrile hydrochloride was formed. The filtered solution was evaporated to dryness in vacuo and water added. Compound **8** was obtained as a white solid which was collected, 2.34 g, and purified by recrystallization from 150 mL of water to give pure **8**, 2.14 g (80% yield), mp 182–184 °C (see below for characterization).

The aqueous filtrate was concentrated to a small volume and a white precipitate collected. Recrystallization from acetonitrile gave pure **7**, 150 mg: mp 205–207 °C; ^{1}H NMR (CD_3CN) δ 2.73 (t, 2 H, J = 6 Hz), 3.63 (q, 2 H, J = 6 Hz), 4.03 (s, 3 H), 7.95 (br s, 1 H), 8.18 (s, 1 H, CHO); MS (CIMS, m/z, rel intensity), 252 (31, MH^+), 269 (MNH_4^+); IR ν_{max} (Nujol) 1314, 1348, 1376, 1459, 1550, 1664, 1688, 2244, 3140, 3288 cm^{-1}. Anal. Calcd for $C_9H_9N_5O_4$: C, 43.0; H, 3.6; N, 27.9; Found: C, 43.3; H, 3.7; N, 27.6.

(b) Coupling by means of Pivaloyl Chloride. A solution of 114 mg (0.66 mmol) of **6** and 266 μL of ethyldiisopropylamine in 2 mL of anhydrous THF was treated with 92 μL of pivaloyl chloride at room temperature. After 15 min, 53 μL of 3-aminopropionitrile was added to the mixture. After stirring for 30 min, the solvent was removed in vacuo, water was added, and **8** was obtained as a white solid which was collected, washed with water, and dried, 137 mg (91% yield): mp 182–184 °C (water); ^{1}H NMR ($CDCl_3$) δ 2.73 (t, 2 H, J = 7 Hz), 3.70 (q, 2 H, J = 7 Hz), 4.17 (s, 3 H), 7.73 (br s, 1 H), 7.82 (s, 1 H); IR ν_{max} (Nujol) 1309, 1365, 1379, 1441, 1462, 1534, 1668, 2251, 3137, 3284 cm^{-1}. Anal. Calcd for $C_8H_9N_5O_3$: C, 43.1; H, 4.1; N, 31.4. Found: C, 43.1; H, 4.1; N, 31.3%.

Procedures Using a Slight Excess of Stannous Chloride in the Reduction of the Nitro Group. (a) 3-(4-Acetamido-1-methylimidazole-2-carboxamido)propionitrile (9b) and 3-(4-Acetamido-1-methyl-5-chloroimidazole-2-carboxamido)propionitrile (10b). Compound **8** (168 mg, 0.75 mmoles) was added to a cold solution of 900 mg (4 mmol) of $SnCl_2 \cdot 2H_2O$ in 1.5 mL of concentrated hydrochloric acid and the temperature was maintained at 5 °C for 15 min. Then, with efficient cooling, 5.5 mL of 25% aqueous sodium hydroxide was added and the mixture was extracted several times with chloroform. The solvent was removed and the residue was allowed to react with acetic anhydride. Recrystallization of the product from ethanol gave a mixture of **9b** with about 15% of **10b**, 132 mg (about 56% yield): mp 156–160 °C; ^{1}H NMR ($CDCl_3$) δ 2.16 (s, 3 H), 2.69 (t, 2 H, J = 5.5 Hz), 3.65 (q, 2 H, J = 5.5 Hz), 3.99 (s, 3 H), 7.40 (s, 0.85 H), 7.55 (t, 1 H, J = 5.5 Hz), 7.87 (s, 1 H); MS, m/z (rel intensity) 235.1070 (32.8, M^+), calcd 235.1069; 193.0962 [100, $(M - CH_2CO)$], calcd 193.0963; 271.0644 (1.8, $C_{10}H_{12}N_5O_2{}^{37}Cl$), calcd 271.0649; 269.0677 (5.09, $C_{20}H_{12}N_5O_2{}^{35}Cl$), calcd 269.0679; 229.0544 [14.6, $(M - CH_2CO)$ for ^{37}Cl], calcd 229.0543; 227.0573 [44.6, $(M - CH_2CO)$ for ^{35}Cl], calcd 227.0573.

(b) 3-[4-(Chloroacetamido)-1-methylimidazole-2-carboxamido]propionitrile (9c) and 3-[4-(Chloroacetamido)-1-methyl-5-chloroimidazole-2-carboxamido]propionitrile (10c). To a solution of 1200 mg of $SnCl_2 \cdot 2H_2O$ (5.3 mmol) in 2 mL of aqueous HCl was added compound **7** (223 mg, 1 mmol) slowly at 5 °C. After 15 min, with efficient cooling, 7.4 mL of 25% aqueous sodium hydroxide was added and the mixture was extracted several times with chloroform. The solvent was removed, the residue, containing the amino compounds **9** and **10**, was dissolved in 1 mL of anhydrous tetrahydrofuran, 0.2 mL of triethylamine was added, and the mixture was cooled to −20 °C then a solution of 0.1 mL of chloroacetyl chloride in tetrahydrofuran was added. The temperature was allowed to rise to 20 °C and the solvent was removed in vacuo. Water was added and the mixture was extracted with chloroform. The dried chloroform extracts were concentrated to a small volume and passed through a silica gel column using chloroform/methanol (40:1) as eluant. The product thus obtained was recrystallized from ethyl acetate to give 180 mg (62% yield), mp 165–186 °C. The ^{1}H NMR spectrum showed this to be a mixture of 44% of **9c** and 56% of 3-[4-(chloroacetamido)-5-chloro-1-methylimidazole-2-carboxamido]propionitrile (**10c**): ^{1}H NMR (Me_2SO-d_6) δ 2.76 (t, 2 H, CH_2CN), 3.50 (m, 2 H, $NHCH_2CH_2CN$), 3.95 (2 s, 3 H, NCH_3), 4.26 (s, 2 H, CH_2Cl), 7.48 (s, 0.44 H, aryl), 8.34 (t, 0.44 H, amide NH), 8.80 (t, 0.56 H, amide NH); ^{1}H NMR ($CDCl_3$) δ 2.70 (2 t, 2 H, CH_2CN), 3.67 (2 q, 2 H, $NHCH_2CH_2CN$), 4.03 (s, 3 H), 4.18 and 4.23 (2 s, CH_2Cl), 7.43 (s, 0.44 H, aryl), 7.55 (m, 1 H, $NHCH_2$), 8.05 (s, 0.56 H) and 8.58 (s, 0.44 H) both $NHCO$; MS (for **10c**) m/z (rel intensity) 305.0250 [35, M^+ ($^{35}Cl + {}^{37}Cl$)], calcd 305.0260; 303.0290 [52.5, M^+ ($2^{35}Cl$)], calcd 303.0290; 229.0523 [55.9, $(M - ClCHCO)$ for ^{37}Cl], calcd 229.0543; 227.0572 [100, $(M - ClCHCO)$ for ^{35}Cl], calcd 227.0573; MS (for **9c**), m/z (rel intensity) 271.0656 [16.9, M^+ (^{37}Cl)], calcd 271.0649; 269.0683 [49.6, M^+ (^{35}Cl)], calcd 269.0679; 193.0966 [21.5, $(M - ClCHCO)$], calcd 193.0963.

(c) 3-[1-Methyl-(1-methyl-4-nitroimidazole-2-carboxamido)imidazole-2-carboxamido]propionitrile (12) and 3-[1-Methyl-5-chloro-(1-methyl-4-nitroimidazole-2-carboxamido)imidazole-2-carboxamido]propionitrile (11). To a solution of 12 g (53 mmol) of $SnCl_2 \cdot 2H_2O$ in 20 mL of concentrated hydrochloric acid was added 2.24 g (10 mmol) of **8** with cooling to 5 °C. After 15 min, and with very efficient cooling, 73 mL of 25% aqueous sodium hydroxide was added and the mixture was extracted several times with methylene chloride. The solvent was removed in vacuo and the residue was dissolved in 50 mL of anhydrous tetrahydrofuran. A solution of the mixed anhydride of pivalic acid and 1-methyl-4-nitroimidazole-2-carboxylic acid was prepared by mixing 1.71 g (10 mmol) of **6** in 30 mL of dry tetrahydrofuran followed by 4 mL of ethyldiisopropylamine and 1.38 mL of pivaloyl chloride. The solution of the reduced **8** was added to the solution of the acid **6** mixed anhydride and after 1 h of stirring at room temperature the solvent was removed under reduced pressure. Water was added, and the solid product was collected and washed with hot acetonitrile to give pure **12**, 1.7 g (50% yield, mp 268–269 °C (from CH_3NO_2)): ^1H NMR (Me_2SO-d_6) δ 2.71 (t, 2 H, J = 6.5 Hz), 3.39 (q, 2 H, J = 6.5 Hz), 3.95 and 4.01 (2 s, 6 H), 7.37 (s, 1 H), 8.34 (t, 1 H, J = 6.5 Hz), 8.41 (s, 1 H), 10.06 (s, 1 H); IR ν_{max} (Nujol) 1463, 1531, 1545, 1658, 1667, 1680, 2240, 3112, 3160, 3360, 3400 cm^{-1}. Anal. Calcd for $C_{13}H_{14}N_8O_4$: C, 45.1; H, 4.1; N, 32.4. Found: C, 45.4; H, 4.1; N, 32.3.

The filtrate which contained **11** was purified by column chromatography on silica gel using 5% methanol in chloroform as eluant and the resulting solid recrystallized from isopropyl alcohol to give pure **11**, 444 mg (11.5% yield): mp 100 °C; ^1H NMR ($CDCl_3$) δ 2.71 (t, 2 H, J = 6.5 Hz), 3.67 (q, 2 H, J = 6.5 Hz), 4.06 and 4.20 (2 s, 6 H), 7.89 (br s, 2 H), 9.36 (br s, 1 H); MS, m/z (rel intensity) 382.0714 [20.3, M^+ (^{37}Cl)], calcd 382.0718; 380.0740 [59.6, M^+ (^{35}Cl)], calcd 380.0748; 346.1105 [31.9, (M – Cl) H], calcd 346.1137; 345.1057 [100, (M – Cl)$^+$], calcd 345.1059; 340.0578 [9.8, (M – CH_2CN), ^{35}Cl], calcd 340.0561; 311.0289 [7.4, (M – $NHCH_2CH_2CN$), ^{35}Cl], calcd 311.0295; 283.0336 [7.0, (M – $CONHCH_2CH_2CN$), ^{35}Cl], calcd 283.0346; IR ν_{max} (Nujol) 1377, 1461, 1524, 1573, 1670, 1690, 2245, 3140, 3300 cm^{-1}.

(d) 3-[1-Methyl-(4-amino-1-methylimidazole-2-carboxamido)imidazole-2-carboxamido]propionitrile (14) and 3-[1-Methyl-(4-amino-5-chloro-1-methylimidazole-2-carboxamido)imidazole-2-carboxamido]propionitrile (13). Compound **12** (175 mg, 0.6 mmol) was reduced with 600 mg (2.66 mmoles) of $SnCl_2 \cdot 2H_2O$ in 1.0 mL of concentrated hydrochloric acid at 5 °C for 15 min. The mixture was cooled to −20 °C and 4.4 mol of 25% aqueous sodium hydroxide added; then the mixture was extracted with methylene chloride. Evaporation of the solution gave 154 mg as a mixture of **14** and **13** (TLC on silica gel developed with acetonitrile: **14**, R_f 0.2; **13**, R_f 0.6). The mixture was separated on a silica gel column using acetonitrile and then acetonitrile/methanol (95:5) as eluants to afford 3-[1-methyl-(4-amino-5-chloro-1-methylimidazole-2-carboxamido)imidazole-2-carboxamido]propionitrile (**13**), 52 mg (29.7% yield): mp 222 °C dec; ^1H NMR ($CDCl_3$) δ 2.67 (t, 2 H, J = 6.5 Hz), 3.55 (br s) and 3.64 (q, together 4 H, J = 5.4 Hz), 3.96 and 4.01 (2 s, 6 H), 7.41 (s, 1 H), 7.56 (t, 1 H, J = 6.5 Hz), 9.20 (s, 1 H); MS, m/z (rel intensity) 352.0983 (33.2, M^+, ^{37}Cl), calcd 352.0977; 350.1007 (100, M^+, ^{35}Cl), calcd 350.1006; IR ν_{max} (Nujol) 1376, 1463, 1546, 1572, 1630, 1666, 1678, 2244, 3290, 3364, 3420 cm^{-1}. Anal. Calcd for $C_{13}H_{15}ClN_8O_2$: C, 44.5; H, 4.3; N, 31.9; Cl, 10.1. Found: C, 44.0; H, 4.2; N, 31.6; Cl, 10.4.

The N-acetyl derivative of **13** was prepared by refluxing **13** with acetic anhydride for 2 min. It was purified by recrystallization from ethyl acetate: mp 197 °C; ^1H NMR ($CDCl_3$) δ 2.21 (s, 3 H), 2.72 (t, 2 H, J = 6.5 Hz), 3.68 (q, 2 H, J = 6.5 Hz), 4.04 (s, 6 H), 7.38 (m) and 7.45 (s) together 2 H, 7.68 (t, 1 H, J = 6.5 Hz), 9.60 (s, 1 H); MS, m/z (rel. intensity) 392.1118 (55.16, M^+, ^{35}Cl), calcd 392.1112; 394.1094 (19.1, M^+, ^{37}Cl), calcd 394.1083; 350.1012 [46.7, (M – CH_2CO), ^{35}Cl], calcd 350.1006; 352.0985 [16.3, (M – CH_2CO), ^{37}Cl], calcd 352.0977; IR ν_{max} 1375, 1462, 1540, 1567, 1584, 1667, 2244, 3230, 3408 cm^{-1}. The second fraction from the column was 3-[1-methyl-(4-amino-1-methylimdazole-2-carboxamido)-imidazole-2-carboxamido]propionitrile (**14**), 85 mg (57% yield): mp 187–189 °C (recrystallized from CH_3CN); ^1H-NMR (Me_2SO-d_6) δ 2.77 (t, 2 H, J = 6 Hz), 3.48 (q, 2 H, J = 6 Hz), 3.87 and 3.88 (2 s, 6 H), 4.61 (s, 2 H, exchangeable NH_2), 6.46 (s, 1 H), 7.49 (s, 1 H), 8.65 (t, 1 H, J = 6 Hz), 9.29 (s, 1 H); MS, m/z (rel intensity) 316.1397 (M^+), calcd 316.1396; IR ν_{max} (Nujol) 1376, 1462, 1530, 1548, 1571, 1673, 2240, 3260, 3364, 3400 cm^{-1}. Anal. Calcd for $C_{13}H_{16}N_8O_2$ (316.52): C, 49.4; H, 5.1; N, 35.5. Found: C, 49.3; H, 5.2; N, 35.3.

Example of a Procedure Using a Large Excess of Stannous Chloride in the Reduction of the Nitro Group. To a solution of 6 g (26.6 mmol) of $SnCl_2 \cdot 2H_2O$ in 10 mL of concentrated aqueous HCl was added 224 mg (1 mmol) of **8** at 5 °C. After 15 min 36.5 mL of 25% aqueous sodium hydroxide was added with efficient cooling and the mixture was extracted with methylene chloride. The amine thus obtained was allowed to react with the acid mixed anhydride (see above) prepared from 171 mg (1 mmol) of the acid **6**. The product of the reaction was recrystallized from acetonitrile to give pure **12**, 201 mg (58% yield). The acetonitrile filtrate contained no more than 2 mg of the chlorinated compound **11**.

3-[1-Methyl-4-[1-methyl-4-(1-methyl-4-nitroimidazole-2-carboxamido)imidazole-2-carboxamido]imidazole-2-carboxamido]propionitrile (15). Compound **14** was obtained by catalytic reduction of 347 mg (1 mmol) of the nitro compound **12** with hydrogen at atmospheric pressure over 140 mg of 10% palladium on charcoal in a mixture of 3 mL of dimethylformamide and 6 mL of methanol at 40 °C. After absorption of the theoretical quantity of hydrogen (68 mL), the catalyst was collected and the solvents were removed in vacuo. The residual solid was washed with carbon tetrachloride and hexane and dissolved in anhydrous acetonitrile. The solution was cooled to −10 °C and 0.2 mL of ethyldiisopropylamine was added, followed by the solution of the carboxylic acid chloride obtained from 171 mg of the acid **6** in tetrahydrofuran. The mixture was stirred at room temperature for 30 min; then it was concentrated in vacuo, water was added, the mixture was decanted, and the product was washed with hot methanol and filtered off to give pure **15**, 320 mg (68% yield): mp 268 °C; ^1H NMR ($CDCl_3$) δ 2.72 (t, 2 H, J = 6 Hz), 3.70 (q, 2 H, J = 6 Hz), 4.04 (s, 3 H), 4.09 (s, 3 H), 4.21 (s, 3 H), 7.46 (s, 1 H), 7.50 (s, 1 H), 7.66 (t, 1 H, J = 6 Hz), 7.86 (s, 1 H), 9.40 (s, 2 H), 9.64 (s, 1 H): IR ν_{max} (Nujol) 1377, 1460, 1529, 1542, 1563, 1663, 2248, 3110, 3178, 3370, 3480 cm^{-1}. Anal. Calcd for $C_{18}H_{19}N_{11}O_5$: C, 46.0; H, 4.1; N, 32.8. Found: C, 46.0; H, 4.1; N, 32.8.

3-(1-Methyl-4-nitroimidazole-2-carboxamido)propionamidine Hydrochloride (24). A solution of 50 mg (0.22 mmol) of **8** in 1 mL of anhydrous methanol was saturated with dry hydrogen chloride with cooling. The mixture was stirred for 1.5 h at room temperature. The solvent was removed under reduced pressure and the residual solid washed with dry ether, decanted, and dissolved in anhydrous methanol; then dry ammonia was condensed into the mixture. The resulting solution was stirred at room temperature for an hour. The solvents were removed in vacuo and the residue was recrystallized from isopropyl alcohol to give **24**, 40 mg (64% yield): mp 270–272 °C dec; ^1H NMR (Me_2SO-d_6) δ 2.65 (t, 2 H, J = 6 Hz), 3.58 (q, 2 H, J = 6 Hz), 4.04 (s, 3 H), 8.61 (s, 1 H), 8.80 (br s, 2 H, exchangeable NH_2), 8.98 (t) and 9.02 (s) 3 H together (NH, amide, and NH_2 amidine); MS-FAB (glycerol), m/z 241 (M – Cl)$^+$; IR ν_{max} (Nujol) 1314, 1378, 1463, 1541, 1573, 1668, 1685, 3309 cm^{-1}. Anal. Calcd for $C_8H_{13}ClN_6O_3$: C, 34.7; H, 4.7; Cl, 12.8; N, 30.4. Found: C, 34.5; H, 4.9; Cl, 13.0; N, 30.6.

3-(1-Methyl-4-guanidineacetamidoimidazole-2-carboxamido)propionitrile Hydrochloride (25). The nitro compound **8** (224 mg, 1 mmol) was reduced with 1.2 g of $SnCl_2 \cdot 2H_2O$ in 2 mL of concentrated hydrochloric acid at 5 °C for 15 min. The solution was then basified with 7.5 mL of 15% aqueous sodium hydroxide and extracted with methylene chloride. The solvent was removed from the dried extract and the residue dissolved in 1 mL of dimethylformamide. A solution of guanidineacetic acid hydrochloride (230 mg, 1.5 mmol) and dicyclohexylcarbodiimide (DCC) (154 mg, 0.75 mmol) in 2.5 mL of dimethylformamide was added to the solution of the amine. A further quantity of DCC (154 mg) was added in portions during 2 h. After stirring for an additional 1 h, the dimethylformamide was removed over reduced pressure, the residue was triturated with water, and the dicyclohexylurea was collected. The filtrate was evaporated to dryness and the residue was purified by chromatography on acid

washed alumina with methanol as eluant. The fractions containing the product were evaporated to dryness and taken up in 2-propanol. The latter solution was evaporated to dryness and the residue recrystallized from methanol/acetone to give pure **25**, 164 mg (50% yield): mp 155 °C; ^{1}H NMR (Me_2SO-d_6) δ 2.79 (t, 2 H, J = 6 Hz), 3.52 (q, 2 H, J = 6 Hz), 3.94 (s, 3 H), 4.10 (d, 2 H, J = 6 Hz), 7.46 (br s, 5 H, amidine and aromatic), 7.77 (t, 1 H, J = 6 Hz), 8.34 (t, 1 H, J = 6 Hz), 10.56 (s, 1 H); MS, m/z (rel intensity) 275.1111 ($M - NH_4Cl$)$^+$, calcd 275.1130; FAB (glycerol), m/z 293 ($M - Cl$)$^+$; IR ν_{max} (Nujol) 1450, 1464, 1532, 1575, 1613, 1642, 1686, 2260, 3240, 3340, 3438 cm^{-1}.

3-[1-Methyl-4-(4-guanidineacetamido-1-methylimidazole-2-carboxamido)imidazole-2-carboxamido]propionitrile Hydrochloride (26). The nitro compound **12** (2 g) was reduced with 34.6 g of $SnCl_2$·$2H_2O$ in 31.5 mL of concentrated hydrochloric acid to give 1.622 g (5.1 mmol) of the amine **14**. The latter together with 1.576 g (7.65 mmol) of guanidineacetic acid hydrochloride was dissolved in 25 mL of *N,N*-dimethylacetamide (DMA). To this mixture was added a solution of 1.56 g (10.2 mmol) of DCC in 10 mL of DMA gradually during 2 h. After an additional 1 h of stirring, the solvent was removed in vacuo, the water was added, and the mixture was centrifuged to remove dicyclohexylurea. The aqueous solution was evaporated to dryness and the residue recrystallized from methanol. The product was finally purified by preparative TLC on silica gel using methanol containing a few drops of acetic acid as eluant. This afforded **26**, 750 mg (32.6% yield), mp 190–193 °C; ^{1}H NMR (Me_2SO-d_6) δ 2.79 (t, 2 H), 3.48 (q, 2 H), 3.97 (2 s, 6 H), 4.15 (d, 2 H), 7.53 (br s) and 7.79 (t) [together 7 H], 8.66 (t, 1 H), 9.40 [D_2O exchange reveals 2 s at 7.51 and 7.55 (2 H)], 10.80 (s, 1 H); MS-FAB (glycerol), m/z 416 ($M - Cl$)$^+$; IR ν_{max} (Nujol) 1376, 1455, 1532, 1544, 1571, 1611, 1663, 1697, 2240, 3174, 3275, 3318, 3380 cm^{-1}. Anal. Calcd for $C_{16}H_{22}ClN_{11}O_3$: C, 42.5; H, 4.9; Cl, 7.8; N, 34.1. Found: C, 42.1; H, 5.1; Cl, 8.0; N, 33.9.

3-[1-Methyl-4-(4-guanidineacetamido-1-methylimidazole-2-carboxamido)imidazole-2-carboxamido]propionamidine Sulfate (28). We already reported the hydrochloride **28A** and the sulfate **28**.[20] Their physical data are as follows. For the hydrochloride derivative **28A**: mp 190–193 °C; ^{1}H NMR (Me_2SO-d_6) δ 2.70 (t, 2 H), 3.58 (q, 2 H), 3.96 and 3.98 (2 s, 6 H), 4.17 (d, 2 H), 7.51 (br m, 6 H), 7.90 (t, 1 H), 8.54 (t, 1 H), 8.92 and 9.15 (2 br s, 4 H, amidine), 9.34 (s, 1 H), 10.90 (s, 1 H), [D_2O exchange reveals 2 s at 7.52 and 7.56]; MS-FAB, m/z 433 ($M - HCl - Cl$)$^+$. For the sulfate **28** mp 250 °C dec; MS-FAB (glycerol), m/z 531 (MH^+), 433 ($M - HSO_4$)$^+$; IR ν_{max} (Nujol) 1376, 1465, 1530, 1575, 1667, 1688, 3165 cm^{-1}. Anal. Calcd for $C_{16}H_{26}N_{12}O_7S$: C, 36.2; H, 4.4; N, 31.7; S, 6.0. Found: C, 35.9; H, 5.1; N, 31.6; S, 6.2.

3-[1-Methyl-4-[1-methyl-4-(1-methyl-4-aminoimidazole-2-carboxamido)imidazole-2-carboxamido]imidazole-2-carboxamido]propionitrile (16). **15** (1 g, 2.13 mmol) in 15 mL of dimethylacetamide and 25 mL of methanol was hydrogenated in the presence of 400 mg of 10% Pd/C. The solvents were evaporated to dryness under reduced pressure, acetonitrile was added, and a crystalline substance was collected and washed with ethyl acetate and hexane to give 612 mg (65% yield) of pure **16**: mp 193 °C dec; ^{1}H NMR (Me_2SO-d_6) δ 2.77 (t, 2 H), 3.48 (q, 2 H), 3.87, 3.94, and 3.99 (3 s, 9 H), 4.58 (s, 2 H, NH_2), 6.46 (s, 1 H), 7.54 (s, 1 H), 7.58 (s, 1 H), 8.58 (t, 1 H, $NHCH_2$), 9.52 (s, 1 H), and 9.71 (s, 1 H); MS, m/z (rel intensity) 439.1833 (100.00), calcd 439.1829; IR ν_{max} (Nujol) 1376, 1462, 1542, 1575, 1668, 1687, 2250, 3180, 3200, 3320, 3420 cm^{-1}.

3-[1-Methyl-4-[1-methyl-4-(1-methyl-4-guanidineacetamidoimidazole-2-carboxamido)imidazole-2-carboxamido]-imidazole-2-carboxamido]propionitrile Sulfate (27A) and Hydrochloride (27). This is an improved procedure of our previous one.[20] **16** (565 mg, 1.28 mmol) and 396 mg (2.58 mmol) of guanidineacetic acid hydrochloride were dissolved in 7 mL of dimethylacetamide and 396 mg (1.92 mmol) of DCC in 3 mL of dimethylacetamide was added during 2 h; the mixture was stirred for 2 h more. The solvent was evaporated under reduced pressure, the residue was treated with water, and the solid was separated by centrifugation. The aqueous solution was evaporated to dryness, and the residue was dissolved in boiling MeOH, concentrated to allow crystallization. The product was filtered off and redissolved in methanol (hot), and a tetraethylammonium sulfate solution in methanol was added. The precipitate was collected, washed with methanol, 2-propanol, and hexane, and dried at 80 °C under reduced pressure to give 355 mg (53% yield) of **27A**; ^{1}H NMR (Me_2SO-d_6) δ 2.76 (t, 2 H), 3.45 (q, 2 H), 3.94, 3.96, and 3.99 (3 s, 9 H), 4.16 (d, 2 H, CH_2CO), 7.45, 7.46, and 7.52 (3 s, 3 H), 7.75 (br s, 4 H, amidine), 8.15 (t, 1 H), 8.50 (t, 1 H), 9.58 (2 s, 2 H), 10.80 (s, 1 H); MS-FAB, m/z 539 $^1/_2$(2 M – SO_4)$^+$; IR ν_{max} (Nujol) 1310, 1380, 1464, 1541, 1571, 1673, 2250, 3170, 3350 cm^{-1}.

27A (320 mg, 0.27 mmol) was dissolved in hot water and the solution of 62 mg (0.25 mmol) of $BaCl_2$·$2H_2O$ was added. $BaSO_4$ was filtered off and the solution was evaporated to dryness. The residue was dissolved in a large amount of MeOH (hot), filtered from the excess of unreacted sulfate **27A**, and concentrated to a small volume. The solid was collected to give 244 mg of pure **27**: mp 238–241 °C dec; ^{1}H NMR (Me_2SO-d_6) δ 2.78 (5, 2 H, J = 6 Hz), 3.49 (q, 2 H, J = 6 Hz), 3.97, 3.99, and 4.02 (3 s, 9 H), 4.07 (br s, 2 H), 7.28 (br s, 4 H), 7.53, 7.56, and 7.66 (3 br, 4 H), 8.55 (t, 1 H), 9.64 and 9.68 (2 s, 2 H); MS-FAB, m/z 539 ($M - Cl$)$^+$; IR ν_{max} 1376, 1465, 1538, 1659, 1681, 2250, 3112, 3199, 3339 cm^{-1}. Anal. Calcd for $C_{21}H_{27}ClN_{14}O_4$: C, 43.9; H, 4.7; Cl, 6.2; N, 34.1. Found: C, 43.6; H, 4.9; Cl, 6.4; N, 34.2.

3-[1-Methyl-4-[1-methyl-4-(1-methyl-4-guanidineacetamidoimidazole-2-carboxamido)imidazole-2-carboxamido]-imidazole-2-carboxamido]propionamidine Sulfate (29) and Hydrochloride (29A). This is an improved procedure of our previous one.[20] **27** (240 mg, 0.42 mmol) in 15 mL of anhydrous ethanol was saturated with dry HCl with cooling. After 1.5 h at room temperature the solvent was evaporated to dryness; some ethanol was added followed by NH_3 condensed into the reactional vessel. After 1 h the solvent was removed, the residue was dissolved in a large amount of methanol, and impurities were collected. The solution was condensed to a small volume and the pure compound was collected to give 171 mg (65.5% yield) of **29A**: mp 243–245 °C; ^{1}H NMR (Me_2SO-d_6) δ 2.70 (t, 2 H), 3.60 (q, 2 H), 3.98, 4.00, and 4.04 (3 s, 9 H), 4.16 (d, 2 H), 7.58 and 7.69 (2 s situated on a large band of $(NH_2)_2CNH$, 6 H together), 7.83 (t, 1 H), 8.49 (t, 1 H), 8.90 and 9.17 (2 s, 4 H, amidine), 9.72 and 9.76 (2 s, 2 H), 10.83 (s, 1 H); MS-FAB, m/z 556 ($M - HCl - Cl$)$^+$; IR ν_{max} 1376, 1464, 1538, 1566, 1669, 3150, 3340 cm^{-1}.

From the mother liquor some more product precipitated with $(NEt_4)_2SO_4$ to give 53 mg (8% yield) of the sulfate **29**. Anal. Calcd for $C_{21}H_{31}N_{15}O_8S$ (653.64): C, 38.6; H, 4.8; N, 32.1; S, 4.90. Found: C, 38.5; H, 4.9; N, 31.9; S, 4.9; MS-FAB 654 $(MH)^+$, 556 ($M - HSO_4$)$^+$.

3-[1-Methyl-4-(1-methyl-4-nitroimidazole-2-carboxamido)pyrrole-2-carboxamido]propionitrile (32). To condense the acid **6** with the amino derivative of **30**, first the chloride of 4-nitroimidazole-2-carboxylic acid (**6**) was prepared. Thus acid **6** was refluxed in a solution of acetonitrile/chloroform (1:1) with an excess of oxalyl chloride until dissolved. The solvents were evaporated, dry toluene was added, and the solid was collected and dried, which was pure acylchloride of **6**. Separately 222 mg (1 mmol) of **30** was hydrogenated in methanol over 60 mg of 10% Pd/C. The catalyst was filtered off, methanol was evaporated in vacuo, and the evaporation was repeated with some tetrahydrofuran. The residue was dissolved in anhydrous tetrahydrofuran and 180 μL of Hunig's base was added. The mixture was cooled to −20 °C and a solution of 190 mg of the acyl chloride of **6** in dry tetrahydrofuran was added. After 30 min at room temperature the solvent was removed and water added. The precipitate was collected and crystallized from acetonitrile to give a yellow substance, which was pure **32**, 276 mg (80% yield), mp 268–270 °C; ^{1}H NMR (Me_2SO-d_6) δ 2.76 (t, 2 H), 3.43 (q, 2 H), 3.84 (s, 3 H), 4.06 (s, 3 H), 7.12 (s, 1 H), 7.32 (s, 1 H), 8.40 (t, 1 H), 8.63 (s, 1 H), 10.87 (1 H); MS, m/z 345.1193 M^+, calcd 345.1185; IR ν_{max} 1310, 1376, 1464, 1540, 1644, 1658, 2250, 3120, 3310, 3400 cm^{-1}.

3-[1-Methyl-4-(1-methyl-4-aminoimidazole-2-carboxamido)pyrrole-2-carboxamido]propionitrile (34). **32** (900 mg, 26 mmol) was suspended in 100 mL of acetonitrile/methanol (1:1) and hydrogenated over 300 mg of 10% Pd/C at 40 °C. The catalyst was filtered off and the solvent was removed in vacuo. Some acetonitrile was added and the crystalline product was collected to give 743 mg (90.5% yield) of **34**, mp 85–87 °C; ^{1}H NMR (Me_2SO-d_6) δ 2.70 (t, 2 H), 3.38 (q, 2 H), 3.77 and 3.82 (2

s, 6 H), 4.37 (br s, 2 H, NH_2), 6.38 (s, 1 H), 7.01 (d, 1 H), 7.21 (d, 1 H), 8.32 (t, 1 H), 9.98 (s, 1 H); MS 315.1435 (97%) M^+, calcd 315.1443; IR ν_{max} 1274, 1376, 1404, 1527, 1572, 1652, 2250, 3330 cm^{-1}.

3-[1-Methyl-4-(1-methyl-4-guanidineacetamidoimidazole-2-carboxamido)pyrrole-2-carboxamido]propionitrile (36). **34** (525 mg, 1.67 mmol) and 380 mg (2.48 mmol) of guanidineacetic acid hydrochloride were dissolved in 4 mL of DMA and a solution of 515 mg (2.5 mmol) of DCC was added during 2 h. After 2 h of stirring at room temperature, the solvent was removed under reduced pressure. The solid was shaken with several portions of chloroform to rid off the excess of DCC and side product (amine reacted with DCC). The solid was dissolved in a small amount of methanol and dicyclohexylurea was filtered off. The solvent was evaporated and the product crystallized from water to give 410 mg (54.6% yield) of **36**: mp 175–180 °C; ^1H NMR (Me_2SO-d_6) δ 2.76 (t, 2 H), 3.42 (q, 2 H), 3.84 (s, 3 H), 3.98 (s, 3 H), 4.12 (br s, 2 H), 7.07 d, 7.31 d, and 7.49 s over a broad band altogether 7 H, 7.97 (br s, 1 H), 8.48 (t, 1 H), 10.05 (s, 1 H), 10.57 (s, 1 H); MS-FAB, m/z 415 $(M - Cl)^+$; IR ν_{max} 1376, 1462, 1544, 1575, 1640, 1665, 2250, 3160, 3320 cm^{-1}. Anal. Calcd for $C_{17}H_{23}ClN_{10}O_3$: C, 45.3; H, 5.1; Cl, 7.9; N, 31.1. Found: C, 45.0; H, 5.3; Cl, 8.1; N, 31.2.

3-[1-Methyl-4-(1-methyl-4-guanidineacetamidoimidazole-2-carboxamido)pyrrole-2-carboxamido]propionamidine Hydrochloride (38) and Sulfate (38A). These compounds were prepared according to our previous procedure.[20] The sulfate (**38A**) has no distinct melting point (it starts to decompose at 235 °C); IR ν_{max} (Nujol) 1122, 1376, 1461, 1547, 1577, 1672, 3250 cm^{-1}; MS-FAB, m/z 530 $(MH)^+$, 432 $(M - HSO_4)^+$.

The hydrochloride (**38**): mp 184–185 °C; ^1H NMR (Me_2SO-d_6) δ 2.70 (t, 2 H), 3.54 (q, 2 H), 3.84 (s, 3 H), 3.97 (s, 3 H), 4.11 (s, 2 H), 7.07 (d, 1 H), 7.26 (d, 1 H), 7.35 (br s) and 7.46 (s) (together 5 H), 7.76 (br s, 1 H), 8.32 (t, 1 H), 8.78 and 9.07 (2 br s, 4 H), 9.88 (s, 1 H), 10.53 (s, 1 H); IR ν_{max} (Nujol) 1376, 1462, 1559, 1580, 1658, 1679, 3140, 3250, 3320 cm^{-1}; MS-FAB, m/z 432 $(M - HCl - Cl)^+$. Anal. Calcd for $C_{17}H_{27}Cl_2N_{11}O_3$: C, 40.5; H, 5.4; N, 30.5; Cl, 14.1. Found: C, 40.2; H, 5.7; N, 30.1; Cl, 13.7.

3-[1-Methyl-4-(1-methyl-4-nitropyrrole-2-carboxamido)-imidazole-2-carboxamido]propionitrile (33). (a) Acyl chloride of 1-methyl-4-nitropyrrole-2-carboxylic acid. The acid (381 mg, 2.24 mmol) was refluxed in acetonitrile with an excess of thionyl chloride for 15 min. The solvent was removed under vacuum and evaporation was repeated with anhydrous acetonitrile to get rid of thionyl chloride. (b) **8** (500 mg, 2.24 mmol) was reduced in methanol over 100 mg of 5% Pd/C. The catalyst was removed, the solvent was evaporated in vacuo, the evaporation was repeated with dry acetonitrile, and the residue was dissolved in the same solvent. The solution was cooled to 20 °C and 400 μL of Hunig's base was added followed by the solution of acyl chloride (a). The mixture was stirred at room temperature for 0.5 h. A yellow precipitate was collected and washed with water and methanol to give 600 mg of pure **33**. From the mother liquor some 60 mg more of **33** was isolated, together 660 mg (85% yield): mp 257–258 °C; ^1H NMR (Me_2SO-d_6) δ 2.79 (t, 2 H), 3.52 (q, 2 H), 3.97 (2 s, 6 H), 7.56 (s, 1 H), 7.78 and 8.20 (2 d, 2 H), 8.26 (t, 1 H), 10.91 (s, 1 H); MS, m/z 345.1187 for M^+ (44.39%), calcd 345.1185; IR ν_{max} (Nujol) 1212, 1320, 1374, 1465, 1490, 1540, 1550, 1670, 2242, 3120, 3351, 3408 cm^{-1}.

3-[1-Methyl-4-(1-methyl-4-aminopyrrole-2-carboxamido)-imidazole-2-carboxamido]propionitrile (35). **33** (1 g, 2.9 mmol) was hydrogenated in dimethylformamide/methanol (1:1) over 250 mg of 5% Pd/C. The solvents were removed under very low pressure and the lowest temperature possible (the product being very unstable in solutions). The residue was washed with the carbon tetrachloride and was dissolved in acetonitrile. After addition of some ethyl acetate the solid was collected to give 684 mg (75% yield): mp 185 °C; ^1H NMR (Me_2SO-d_6) δ 2.77 (t, 2 H), 3.50 (q, 2 H), 3.72 (s, 3 H), 3.83 (s, 2 H, NH_2), 3.92 (s, 3 H), 6.31 and 6.52 (2 d, 2 H), 7.50 (s, 1 H), 8.28 (t, 1 H), 9.97 (s, 1 H); MS, m/z 315.1448 (M^+) (34.14%), calcd 315.1443; IR ν_{max} (Nujol) 1255, 1302, 1442, 1464, 1521, 1558, 1640, 1670, 2245, 3230, 3285, 3360, 3385 cm^{-1}.

3-[1-Methyl-4-(1-methyl-4-guanidineacetamidopyrrole-2-carboxamido)imidazole-2-carboxamido]propionitrile Hydrochloride (37). **35** (684 mg, 2.17 mmol) and 495 mg (3.25 mmol) of guanidineacetic acid hydrochloride were dissolved in 5 mL of dimethylacetamide under nitrogen. DDC (671 mg, 3.25 mmol) dissolved in 3 mL of dimethylacetamide was added during 2 h and the mixture was stirred at room temperature for 2 h more. The solvent was removed under reduced pressure. A little water was added and the solid was collected. The product was washed out from the solid with methanol. The solvent was removed, the residue was dissolved in hot water, and a solution of Na_2SO_4 was added. On cooling the sulfate crystallized off. It was collected and washed with methanol to get rid of a side product. The remaining solid was dissolved in hot water, an equimolecular amount of $BaCl_2 \cdot 2H_2O$ was added, and $BaSO_4$ was filtered off. The filtrate was condensed to a small volume and the pure product recrystallized to give a 251 mg (25%) yield; mp 168 °C; ^1H NMR (Me_2SO-d_6) δ 2.80 (t, 2 H), 3.50 (q, 2 H), 3.87 (s), 3.97 (s) and 4.05 (br s) together 8 H, 7.03 (d, 1 H), 7.30 (d), 7.43 (br s), and 7.55 (s) together 6 H, 7.72 (t, 1 H), 8.32 (t, 1 H), 10.29 (s) and 10.31 (s) together 2 H; MS-FAB, m/z 415 $(M - Cl)^+$; IR ν_{max} (Nujol) 1376, 1447, 1464, 1535, 1614, 1666, 2245, 3310 cm^{-1}. Anal. Calcd for $C_{17}H_{23}ClN_{10}O_3$: C, 45.3; H, 5.1; Cl, 7.9; N, 31.1. Found: C, 45.1; H, 5.3; Cl, 8.2; N, 31.4.

3-[1-Methyl-4-(1-methyl-4-guanidineacetamidopyrrole-2-carboxamido)imidazole-2-carboxamido]propionamidine Hydrochloride (39) and Sulfate (39A). These compounds were prepared according to our previous procedure.[20] The sulfate **39A**: mp 230–231 °C; IR ν_{max} (Nujol) 1376, 1404, 1453, 1538, 1663, 3318 cm^{-1}; MS-FAB, m/z 530 MH^+, 432 $(M - HSO_4)^+$. The hydrochloride **39**: mp 186 °C; ^1H NMR (Me_2SO-d_6) δ 2.65 (t, 2 H), 3.60 (q, 2 H), 3.86 (s, 3 H), 3.96 (s, 3 H), 4.06 (br s, 2 H), 7.03 (d, 1 H), 7.31 (d, 1 H), 7.46 (br s, 4 H), 7.54 (s, 1 H), 7.77 (br s, 1 H), 8.14 (t, 1 H), 8.96 and 9.12 (2 br s, 4 H), 10.38 (2 s, 2 H); MS-FAB, m/z 432 $(M - HCl - Cl)^+$; IR ν_{max} (Nujol) 1377, 1463, 1539, 1610, 1661, 3258 cm^{-1}. Anal. Calcd for $C_{17}H_{27}N_{11}O_3Cl_2$ (504.38): C, 40.5; H, 5.4; N, 3.05; Cl, 14.1. Found: C, 40.1; H, 5.7; N, 30.2; Cl, 13.8.

***N*-(Chloroacetyl)-*p*-toluidine (17).** A solution of 5.4 g (50 mmol) of *p*-toluidine in 40 mL of anhydrous tetrahydrofuran was treated with 8 mL of triethylamine and cooled to −20 °C. A solution of 4.2 mL of chloroacetyl chloride in 10 mL of tetrahydrofuran was added to the mixture. The solvents were removed in vacuo and the residual solid was washed with water and recrystallized from isopropyl alcohol to give pure **17**, 5.3 g (58% yield): mp 163 °C; ^1H NMR ($CDCl_3$) δ 2.22 (s, 3 H), 4.10 (s, 2 H), 7.05 and 7.30 (2 d, 4 H, J = 6.5 Hz), 8.10 (br s, 1 H). Anal. Calcd for $C_9H_{10}ClNO$: C, 58.9; H, 5.5; N, 7.6; Cl, 19.3. Found: C, 58.9; H, 5.4; N, 7.8; Cl, 19.3.

***N*-Bromo-*p*-toluidine (19).** This compound was obtained as described above, employing bromoacetyl chloride, and afforded **19** in 79% yield: mp 172–173 °C (from isopropyl alcohol); ^1H NMR ($CDCl_3$) δ 2.25 (s, 3 H), 3.59 (s, 2 H), 7.11 and 7.30 (2 d, 4 H, J = 6.5 Hz), 8.03 (br s, 1 H); IR ν_{max} (Nujol) 1461, 1513, 1552, 1613, 1668, 3130, 3199, 3262, 3300 cm^{-1}. Anal. Calcd for $C_9H_{10}BrNO$: C, 47.4; H, 4.4; Br, 35.0; N, 6.1. Found: C, 47.5; H, 4.3; N, 6.0; Br, 35.2.

***N*-(Thioureidoacetyl)-*p*-toluidine (21).** *p*-Toluidine (321 mg, 3 mmol) and 400 mg (3 mmol) of thioureidoacetic acid were dissolved in 1.5 mL of dimethylacetamide. To this mixture was gradually added a solution of 618 mg (3 mmol) of DCC in 1.5 mL of dimethylformamide during 2 h. After an additional 1 h of stirring, water was added and the resulting precipitate collected. The solid was triturated with acetone and filtered. The filtrate was evaporated to dryness and the residual solid was washed with ethyl acetate and recrystallized from acetonitrile to give **21**, 490 mg (73% yield): mp 195–197 °C; ^1H NMR (Me_2SO-d_6) δ 2.26 (s, 3 H, CH_3), 4.25 (d, 2 H, CH_2), 7.10 and 7.46 (2 d, 4 H, aromatics), 7.3 (s, 2 H, exchangeable NH_2), 7.73 (t, 1 H, NHCS), 9.99 (s, 1 H, NHCO); MS, m/z 223.0777 (M^+), calcd 223.0779; IR ν_{max} (Nujol) 1445, 1455, 1614, 1673, 3100, 3238, 3328, 3392 cm^{-1}.

***N*,*N*'-Di-*p*-tolylpiperazine-2,5-dione (18).** (a) A solution of 153 mg (3 mmol) of cyanamide in 5 mL of anhydrous ethanol was treated with a solution of 69 mg (3 mmol) of sodium reacted in 5 mL of dry ethanol. The solvent was removed in vacuo and the residue treated with 366 mg (2 mmol) of **17** in 10 mL of dry toluene, and the mixture was stirred with molecular sieves for 1.5 h. A solution of 100 mg of dicyclohexano-18-crown-6 in 5 mL of toluene was added and the mixture was stirred at 60 °C for

2 h. The solution was evaporated to dryness and extracted with chloroform, and the extract was washed with water and dried (Na_2SO_4). The solvent was removed in vacuo and the residue was recrystallized from ethanol to give white crystals of **18**, 90 mg (30% yield): mp 200 °C; ^1H NMR ($CDCl_3$) δ 2.30 (s, 6 H), 4.38 (s, 4 H), 7.20 (s, 8 H); MS, m/z (rel intensity) 294.1363 (91, M^+), calcd 294.1368; IR ν_{max} 1335, 1429, 1466, 1515, 1658 cm^{-1}.

(b) A suspension of 42 mg (1 mmol) of cyanamide in tetrahydrofuran at -60 °C was treated with 650 μL of a 1.6 M solution of *n*-butyllithium. After 30 min a solution of 228 mg (1 mmol) of **19** in 3 mL of tetrahydrofuran was added and the mixture was stirred at room temperature for 1 h. The solvent was removed in vacuo, water was added, and the resulting solid was collected and purified by recrystallization from isopropyl alcohol to give pure **18**, 50 mg (34% yield).

***N*-(Cyanoguanidineacetyl)-*p*-toluidine (20).** (a) A solution of 76 mg of sodium reacted (3.3 mmol) in 5 mL of absolute ethanol was treated with 252 mg (6 mmol) of cyanamide followed by 684 mg (3 mmol) of **19**, and the mixture was stirred at room temperature. During this process **20** (360 mg) precipitated as a white solid and was collected and purified by recrystallization from isopropyl alcohol or ethylene dichloride and purified by recrystallization from isopropyl alcohol or ethylene dichloride to give pure **20**, 350 mg (50% yield): mp 205 °C dec; ^1H NMR (Me_2SO-d_6) δ 2.25 (s, 3 H, CH_3), 4.11 (s, 2 H, CH_2), 6.10 (s, 2 H, exchangeable NH), 9.72 (s, 1 H, NHCO); MS, m/z (rel intensity) 231.1121 (6.8, M^+), calcd 231.1120; 189.0902 (2.04, M - H_2NCN), calcd 189.0902; 132.0685 (100, M - $COCH_2NHC{=}NHNH$), calcd 132.0687; IR ν_{max} (Nujol) 1398, 1463, 1606, 1660, 1712, 3210 cm^{-1}.

(b) A solution of 44.6 mg (0.2 mmol) of **21** in 2 mL of methanol was treated with a solution of 70 mg (0.22 mmol) of mercuric acetate in 1 mL of methanol. The mixture turned deep red and then black and HgS precipitated. The precipitate was collected and the filtrate evaporated to dryness, water was added, and the solution was extracted with methylene chloride. The solvent was removed in vacuo and the residue dissolved in ethanol and then allowed to react with cyanamide in the presence of sodium ethoxide to give **23**, 30 mg (65% yield). TLC (silica gel, methanol/chloroform, 1:9): R_f **21** = 0.45, for **24** R_f 0.5.

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Registry No. **1**, 616-47-7; **2**, 30148-21-1; **3**, 109012-22-8; **4**, 1564-49-4; **5**, 109012-23-9; **6**, 109012-24-0; **6** (acid chloride), 109012-46-6; **7**, 109012-25-1; **8**, 109012-26-2; **9a**, 109012-27-3; **9b**, 109012-47-7; **9c**, 109012-48-8; **10a**, 109012-28-4; **10b**, 109012-49-9; **10c**, 109012-50-2; **11**, 109012-29-5; **12**, 109012-30-8; **13**, 109012-31-9; **13** (*N*-acetyl derivative), 109012-51-3; **14**, 109012-32-0; **15**, 104394-08-3; **16**, 104394-09-4; **17**, 16634-82-5; **18**, 21303-69-5; **19**, 5343-65-7; **20**, 109012-33-1; **21**, 109012-34-2; **23**, 109012-35-3; **24**, 109012-36-4; **25**, 109012-37-5; **26**, 109012-38-6; **27**, 109012-39-7; **27A**, 109012-53-5; **28**, 101772-43-4; **28A**, 109012-54-6; **29**, 104394-13-0; **29A**, 101772-44-5; **30**, 3185-95-3; **32**, 109012-40-0; **33**, 109012-41-1; **34**, 109012-42-2; **35**, 109012-43-3; **36**, 109012-44-4; **37**, 109012-45-5; **38**, 101809-75-0; **38A**, 104394-05-0; **39**, 104394-12-9; **39A**, 101772-41-2; EtOCOCl, 541-41-3; $H_2NCH_2CH_2CN$, 151-18-8; $ClCH_2COCl$, 79-04-9; $BrCH_2COCl$, 22118-09-8; guanidineacetic acid hydrochloride, 14901-20-3; 3-(1-methyl-4-aminopyrrole-2-carboxamido)propionitrile, 97950-77-1; 1-methyl-4-nitropyrrole-2-carboxylic acid, 13138-78-8; 1-methyl-4-nitropyrrole-2-carboxylic acid chloride, 28494-51-1; *p*-toluidine, 106-49-0; thioureidoacetic acid, 51675-47-9.

Rose Bengal Functionalized Phase-Transfer Catalysts Promoting Photooxidations with Singlet Oxygen. Nucleophilic Displacements on Dioxetanic and Endoperoxidic Intermediates

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Photooxidations with $^1O_2$ promoted by an anionic photosensitizer, such as Rose Bengal, can be readily performed in an aqueous–organic, two-phase system in the presence of catalytic amounts of quaternary onium salts that transfer the anion in the organic phase. This means that one can use low-polar solvents, which are unusual in reactions with this sensitizer. Soluble and silica gel immobilized onium salts were used as the phase-transfer catalysts. If a nucleophile is also present in the reaction mixture, it, too, is transported in the organic phase and reacts via S_N2, if the intermediate is of the dioxetanic or endoperoxidic type. With the heterogenized catalysts it is possible to carry out the reaction in two steps: the photooxidation itself and later the nucleophilic displacement. In addition to being productive with regard to the synthesis, this can clarify the photooxidation reaction pathway. The following organic substrates were photooxidized under PTC conditions: anthracene, 2,3-dimethyl-2-butene, 5,6-dihydro-2,3-diphenyl-*p*-dioxin, 1,3-cyclohexadiene, and indene. In the presence of N_3^- as nucleophile, anthracene gives 10-imino-9(10*H*)-anthracenone while 5,6-dihydro-2,3-diphenyl-*p*-dioxin yields, selectively, two different azido derivatives according to the procedure employed.

Introduction

Singlet oxygen ($^1O_2$) can be generated in various ways, the most important being reaction of $^3O_2$ with the excited state of a photosensitizer: light irradiation of the sensitizer in a solvent in the presence of $^3O_2$ has been reported to produce the oxidation of various substrates.[1]

One of the principal sensitizers is Rose Bengal (3,4,5,6-tetrachloro-2-(2,4,5,7-tetraiodo-6-hydroxy-3-oxo-3*H*-xanthen-9-yl)benzoic acid, disodium salt).[2]

Since Rose Bengal is a disodic salt (Na_2RB) it can only be dissolved in water and strongly polar solvents. It is thus of limited utility in organic synthesis. In addition, on account of its low solubility, Na_2RB forms aggregates in low polar organic solvents with reduced catalytic activity, owing to the formation of excimers.[3] The activity of RB^{2-}

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