Synthesis, NMDA Receptor Antagonist Activity, and Anticonvulsant Action of 1-Aminocyclobutanecarboxylic Acid Derivatives

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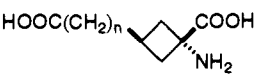
A range of *cis*- and *trans*-3-substituted 1-aminocyclobutane-1-carboxylic acids has been synthesized and evaluated for antagonism at excitatory amino acid receptor sites and for anticonvulsant activity. Potent and selective antagonist activity at *N*-methyl-D-aspartate (NMDA) receptor sites in neonatal rat motoneurones was shown by compounds in which the 3-substituent was, or contained, a 2′-carboxyethyl or 2′-phosphonoethyl moiety. Substances **4b**, **24**, **35**, and **40** were more potent than the standard NMDA receptor antagonist, D-2-amino-5-phosphonopentanoate (D-AP5) as NMDA antagonists in this preparation, and about equipotent with [3-(±)-2-carboxypiperazin-4-yl)-1-propyl]phosphonate (CPP). Anticonvulsant activity, as assessed following intracerebroventricular injection into audiogenic DBA/2 mice, generally paralleled NMDA receptor antagonist activity.

Introduction

Excitatory amino acid receptors represent the predominant excitatory synaptic transmitter receptors in the mammalian central nervous system.[1,2] There are two major families: metabotropic receptors, linked to second messenger systems through G proteins, and ionotropic receptors linked to ion-channels. It is likely that the transmitter activating these receptors is L-glutamate. In each family several major types have been recognized and, for the ionotropic excitatory amino acid receptors, these comprise the *N*-methyl-D-aspartate (NMDA), α-amino-3-hydroxy-5-methylisoxazole-4-propionic acid (AMPA), and kainate receptors, named after their classical selective agonists.[1,3] Such receptors have been recongized as potential targets for therapeutic intervention in a number of neurological conditions.[4–6] In particular, antagonists of the NMDA receptor have well-documented anticonvulsant, antispastic, and neuroprotective properties.[7] The starting point for the present study was the finding that the glutamic acid analogue *trans*-1-aminocyclobutane-1,3-dicarboxylic acid[8,9] (**1a**) is an extremely potent NMDA receptor agonist. Conventionally, NMDA receptor agonists can be converted to antagonists by increasing the length of the connecting chain between the α- and ω-acidic groups so that it becomes from one to three atoms longer than that in glutamic acid itself.[3,10] Usually,[3,10] but not always,[11,12] the α-amino carboxylic acid moiety in these compounds has the (*R*) configuration. The most effective ω-acidic group for NMDA receptor antagonist activity is the phosphonic group, and the prototypes for NMDA receptor antagonist activity are thus (*R*)-2-amino-5-phosphonopentanoate (D-AP5; **5a**) and (*R*)-2-amino-7-phosphonoheptanoate (D-AP7; **5c**).[3,10] Surprisingly, however, neither *trans*- nor *cis*-1-amino-3-(phosphonomethyl)cyclobutane-1-carboxylic acid (**3a**, **4a**) nor the propyl analogues (**3c**, **4c**) showed potent antagonist activity at NMDA receptors in previous studies.[9] We nevertheless felt it worthwhile to explore the structure–activity relations of this series further, initially with the thought of retaining the two carboxyl groups of compound **1a** (to link with agonist binding sites in the receptor) and introducing a further substituent in the 3-position to link with an additional antagonist binding site that has been postulated to exist in the receptor.[3,13,14] We report here the synthesis of such a new series of potent NMDA antagonists, the crucial structural feature of which is not, however, the retention of the agonist 3-carboxy group (which can be omitted) but the introduction of a carboxyethyl or phosphonoethyl group, preferably in the *cis* configuration, and yielding analogues of 2-amino-6-phosphonohexanoic acid (AP6; **5b**) rather than of AP5 (**5a**) or AP7 (**5c**) analogues.



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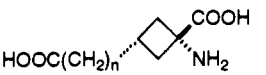
1b: n = 1
1c: n = 2
1d: n = 3



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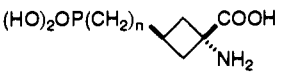

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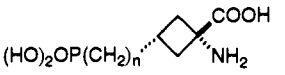
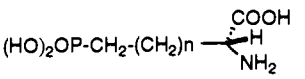
3c: n = 3



4a: n = 1
4b: n = 2
4c: n = 3



5a: n = 2
5b: n = 3
5c: n = 4

Chemistry

The synthesis of the presently described cyclobutane amino acids is based on a general scheme for the preparation of substituted cyclobutanes from 1-(phenylsulfonyl)bicyclo[1.1.0]butanes. Up to four functional-

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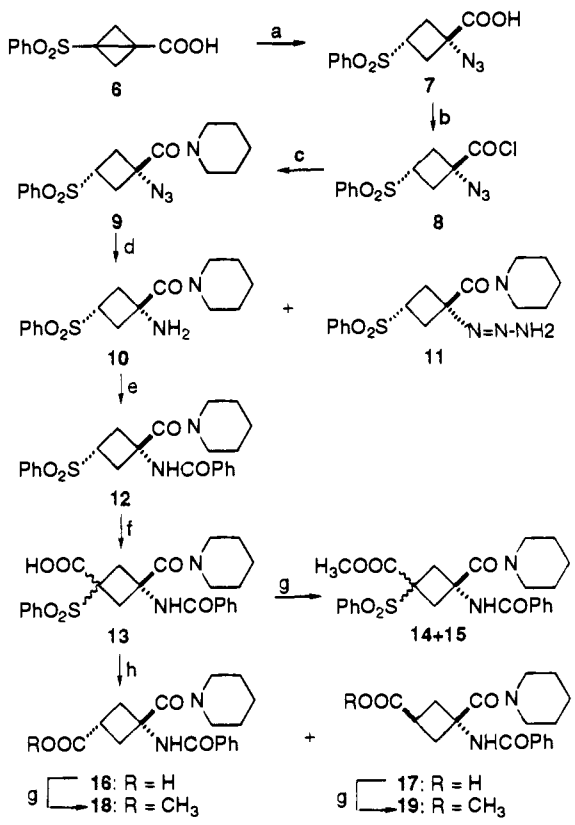
Scheme 1[a]



[a] (a) NaN_3, DMF; (b) $SOCl_2$; (c) piperidine, CH_2Cl_2; (d) H_2, Pd/C; (e) PhCOCl, NEt_3, CH_2Cl_2; (f) BuLi, CO_2; (g) CH_2N_2; (h) 6% Na–Hg.

ized groups can be introduced into the original four-membered ring of the bicyclobutane by a sequence of reactions comprising (1) substitution of the bridghead proton, (2) addition of a nitrogen nucleophile across the central bond, (3) substitution α to the sulfone, mainly by a carboxyl, and (d) eventual substitution, after desulfonylation, α to the latter carboxyl.[15,16]

Scheme 1 outlines the synthesis of intermediates **18** and **19**, used in the preparation of all cyclobutane amino acids described in this work. By carrying out the azidation of acid **6**[16] with sodium azide in DMF–tetramethylguanidine (TMG), product **7** was obtained regioselectively and stereoselectively in 86–92% yield. The stereoselectivity results, probably, from the kinetic protonation of an incipient α-sulfonyl anion by the TMG bound to the carboxyl group. Thus, while the azide ion adds from the endo side of **6**,[17] intramolecular protonation necessarily occurs from the exo side, leading to *cis*-**7**. Azidation of the piperidine amide of **6**[16] with tetramethylguanidinium azide in *N*-methyl-2-pyrrolidone produced both **7** and its geometrical *trans* isomer.

Adduct **7** was converted to the piperidine amide **9** via acid chloride **8**, using standard procedures. Catalytic reduction of the azide group to the amine was more elaborate, since azide **9** produced a stable triazene intermediate which needed extra treatment for conversion to the amine. Following the hydrogenation by TLC, it is observed that while the starting azide disappears, two more polar spots appear, the less polar of which converts slowly to the more polar one. By stopping the hydrogenation after a few hours, solid, stable triazene **11** could be separated from amine **10** by chromatography and be fully characterized.[19] Amine **10**, free of **11**, was obtained in 93–96% yield after prolonged stirring under hydrogen over the palladium catalyst, followed by reflux under air atmosphere until total conversion of the triazene to the amine.

The benzoylated derivative **12**, obtained from **6** in about 70% overall yield, is an important intermediate for the preparation of a large number of 3-substituted 1-aminocyclobutane-1-carboxylic acids via its dilithiated derivative. Carboxylation leads to a mixture of *cis* and *trans* acids **13**. Separation of the isomers is not required before desulfonylation, since the geometry of the separated individual isomers is, anyway, not conserved in the desulfonylated product. Separations were, however, carried out in some cases for characterization purposes. Thus, after esterification of **13**, isomers **14** and **15** were separated and individually characterized.

Key intermediates **18** and **19** were obtained by desulfonylation of total crude **13** followed by esterification and chromatographic separation, ester **19** being slightly more abundant. Alternatively, almost pure acids **16** and **17** could be obtained by fractional crystallization of the total acid product of desulfonylation and further purification by crystallization from ethanol. The total overall yield of **18** and **19** from **6** is 50–55%.

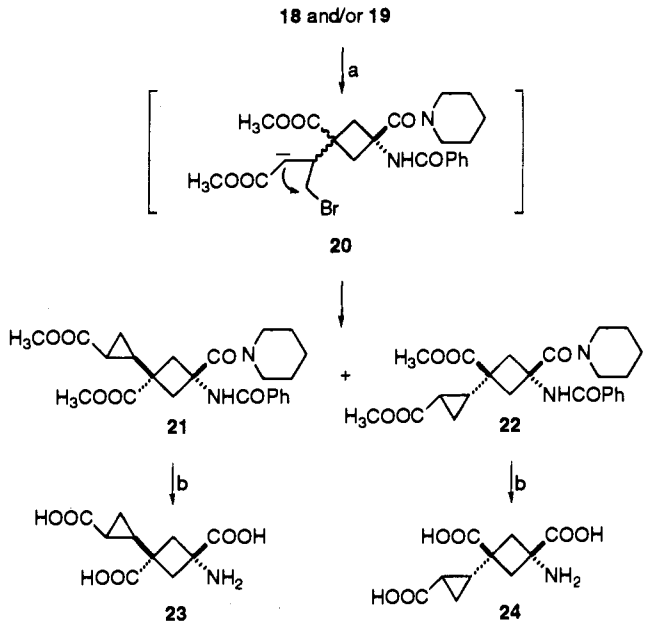
Configurational assignments of all compounds are based on correlations of certain intermediates with compounds the structure of which has been determined by X-ray crystallography[18] and on spectral proton NMR considerations.[16] In particular, the geometry of **18** and **19** was established by acid hydrolysis to the corresponding amino dicarboxylic acids. Thus, hydrolysis of **18** furnished the known *atelia-herbert-smithii* acid **2a**, namely, *cis*-1-aminocyclobutane-1,3-dicarboxylic acid,[18,20] while **19** furnished *trans*-1-aminocyclobutane-1,3-dicarboxylic acid **1a**, the structure of which has been established by X-ray crystallography.[18] Amino acid **1a** was later found to be a potent and selective agonist of the NMDA subtype of glutamate receptors of the central nervous system.[8,9]

Intermediates **18** and **19**, of well-established configuration, were used to prepare the NMDA antagonists which constitute the subject of this publication. Active products were obtained either by substitution α to the methyl carboxylate, using LDA and an electrophile, or by modifying the carboxylate group itself. The following schemes describe the specific transformations used in each case.

Treatment of **18** or **19**, or of a mixture of both with lithium diisopropylamide (LDA) followed by addition of methyl γ-bromocrotonate provided in a total 78% yield a mixture of adducts **21** and **22**, produced probably via the conjugate addition intermediate product **20** (Scheme 2). No expected ethylenic product was detected. The relative geometry on the cyclopropane ring of **21** and **22** was unique, and most probably *trans*, because of an expected steric congestion in a *cis* isomer. A *trans* selectivity was observed also in sterically less demanding cases where a similar conjugate addition–cyclization sequence has been applied.[21,22] Acid hydrolysis of the separated product isomers provided amino acids **23** and **24** in about 70% yield.

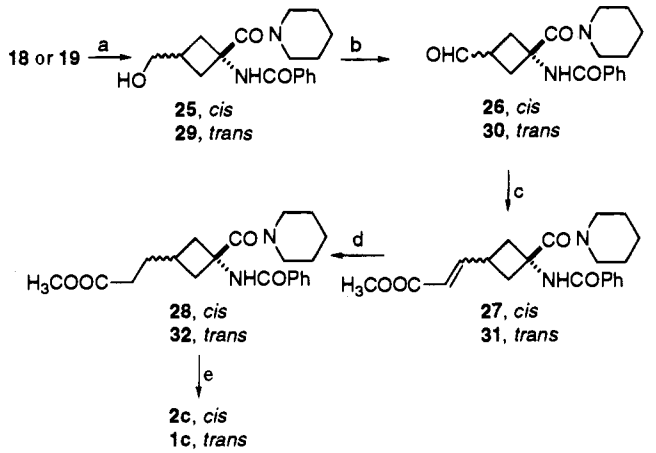
The route followed from **18** to **2c** and from **19** to **1c** is shown in Scheme 3. It involves, in the *cis* series, the reduction of ester **18** with sodium borohydride to alcohol **25** followed by oxidation with activated DMSO to

Scheme 2[a]



[a] (a) LDA; $BrCH_2CH{=}CHCOOCH_3$; (b) 6 M HCl.

Scheme 3[a]



[a] (a) $NaBH_4$; (b) DMSO–TFAA; (c) $Ph_3P{=}CHCOOCH_3$; (d) H_2, cat.; (e) 6 M HCl.

aldehyde **26**, carboxyolefination to **27**, hydrogenation to **28**, and hydrolysis to **2c** (ca. 30% overall yield from **18**). In the *trans* series, amino acid **1c** was similarly obtained from **19**.

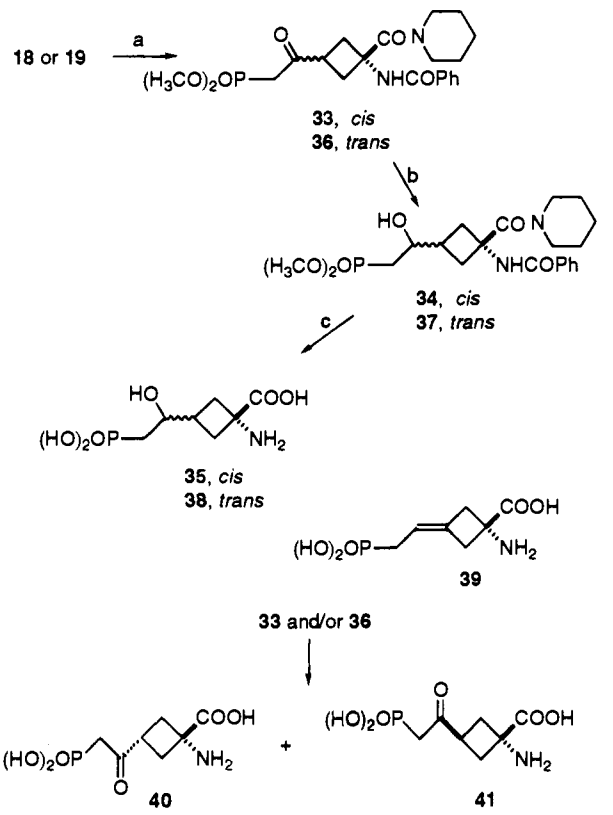
Intermediate **25** is also a direct precursor of the *atelia-herbert-smithii* acid *cis*-1-amino-3-(hydroxymethyl)cyclobutane-1-carboxylic acid,[23] while **29** is a precursor of the corresponding, nonnatural *trans* isomer.[24]

Scheme 4 describes the preparation of amino acids **35**, **38**, **39**, **40**, and **41**. The reaction of ester **18** with dimethyl (lithiomethyl)phosphonate produced ketone **33**. Reduction of **33** with sodium borohydride in ethanol furnished racemic alcohol **34** which was hydrolized to the racemic amino acid **35**. The overall yield from **18** was 50–53%.

Amino acid **38** was similarly obtained from **19**. However, in this case, it was accompanied by the elimination product **39**, which could be separated from **38** and fully characterized.

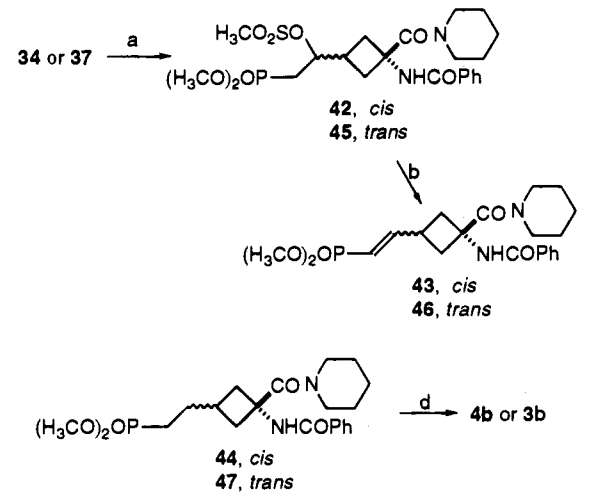
Hydrolysis of either **33** or **36** produced a similar mixture of amino acids **40** and **41**, in a ratio of ca. 3:2, respectively, and in a total 75–80% yield. Equilibration

Scheme 4[a]



[a] (a) $(H_3CO)_2POCH_2Li$; (b) $NaBH_4$; (c) 6 M HCl.

Scheme 5[a]



[a] (a) CH_3SO_2Cl, Et_3N; (b) BuLi; (c) H_2, cat.; (d) 6 M HCl.

of the isomers occured probably by enolization toward the ring. Amino acid **40** was obtained pure by crystallization of this mixture, or of chromatographically enriched mixtures, from water. Its geometry was established by catalytic hydrogenation in water and acetic acid to acid **35**. Pure **41** was obtained in small amounts from the very last chromatography fractions.

Scheme 5 describes the preparation of **4b** from **34** and of **3b** from **37**. In order to eliminate the hydroxyl group of **34** or **37** toward the phosphonate group and not toward the ring, it was first mesylated and then treated with butyllithium, producing **43** and **46**, respectively. Hydrogenation and hydrolysis provided amino acids **4b** and **3b** in similar yields of ca. 50% from the corresponding alcohols.

Table 1. Antagonism of NMDA-Induced Depolarization and Anticonvulsant Activity of Cyclobutane Derivatives

compd	EPMR[c]	anticonvulsant action (ED_{50})[a] (range[b]) μg	nmol
5a	1.0		
DL-CPP	0.20		
23	inact. (1 mM)		
24	0.28	0.089 (0.059–0.134)	0.36 (0.23–0.55)
2c	1.8	16.09 (14.39–18.01)	86.0 (77.0–96.3)
1c	7.9		
35	0.39	0.032 (0.024–0.048)	0.13 (0.10–0.18)
38	1.0	0.124 (0.093–0.165)	0.52 (0.39–0.69)
40	0.33	0.106 (0.071–0.157)	0.53 (0.035–0.78)
41	4.2		
4b	0.32	0.032 (0.024–0.048)	0.14 (0.11–0.22)
3b	1.1		

[a] Antagonist activity following intracerebroventricular administration against sound-induced clonic convulsions in DBA/2 mice; 7–11 mice per drug. [b] 95% confidence limits. [c] Equipotent molar ratio of cyclobutane derivatives relative to D-AP5 = 1.0 on neonatal rat motoneurones.

Pharmacology

Neonatal Rat Spinal Cord *in Vitro*. Previous studies have indicated that neither 3-(phosphonomethyl)- nor 3-(phosphonopropyl)-1-aminocyclobutane-1-carboxylic acid had pronounced NMDA receptor antagonist activity.[9] In the present study we confirmed this lack of potent activity for a mixture of *cis and trans* isomers of 3-(phosphonopropyl)-1-aminocyclobutane-1-carboxylic acid (**3c**, **4c**). However, marked antagonist activity was observed with a series of 1-aminocyclobutane-1-carboxylic acids bearing a carboxyethyl or phosphonoethyl substituent in the 3-position (Table 1). The most potent substances (**24**, **35**, **40**, and **4b**) were approximately equipotent with (±)-CPP and about 3–4 times more potent than D-AP5 (**5a**). All of these compounds had the same configuration and were more active than their genometric isomers (**23**, **38**, **41**, and **3b**). The same isomeric preference (with the 1-amino and substituted 3-ethyl group *cis* to one another) was shown in another pair of moderately active isomers (**1c**, **2c**). Direct comparison of compounds with either carboxyethyl or phosphonoethyl substituents (**1c**, **3b**; **2c**, **4b**) indicated the latter compounds to be the more potent. However, among the most potent compounds was **24** which contained both a *trans*-3-carboxy and a *cis*-3-(2-carboxycyclopropyl) substituent relative to the 1-amino group. Interestingly, the corresponding *cis*-3-carboxy-*trans*-3-(2-carboxycyclopropyl)-substituted compound **23** was completely inactive.

Anticonvulsant Activity. Six of the novel NMDA antagonists were tested for protection against audiogenic seizures in DBA/2 mice. Intracerebroventricular administration of these compounds led to dose-dependent suppression of sound-induced seizures. The ED_{50} values for five of these substances (**24**, **35**, **38**, **40**, **4b**) all fell within 4-fold range of 0.032–0.124 μg (0.13–0.52 nmol); the other compound (**2c**) was 100–500-fold less potent (Table 1). The choice of pretreatment time (60 min) was based on preliminary results using compound **24**, where 30 or 60 min pretreatment periods (icv) resulted in closely similar anticonvulsant ED_{50} values.

Little or no acute overt behavioral effects of the drugs were observed at the pharmacologically effective doses used. Slight ataxia was observed at the highest doses used of **24** (0.2 μg); the latter case was also associated with a significant drug-induced decrease in rectal temperature. At doses 5–10-fold higher than those required for anticonvulsant protection (1 μg of **24** and 50–100 μg of **2c**) the mice exhibited mild to moderate ataxia.

With one exception (compound **2c**), these studies on the suppression of audiogenic seizures in DBA mice showed a close parallelism between the NMDA receptor antagonist activity of the compounds and their anticonvulsant activity (Table 1). Although **2c** was the least active compound in both the *in vitro* and *in vivo* studies, the relative potency of this compound as an anticonvulsant was much lower than would have been predicted from its spinal cord NMDA receptor antagonist activity.

In these studies the compounds were injected intracerebroventricularly because of the small quantities of the substances available. It is not known how well the substances would pass the blood–brain barrier if injected systemically, but it would be expected that their ability to penetrate into the brain would resemble that of CGP 37849.[25] In this case, esterification of the α-carboxyl group may improve the ability of the compounds to pass the blood–brain barrier, though probably lowering its potency at the target sites, as shown in the case of CGP 37849.[25] Further studies are required to elucidate this question.

Structure–Activity Discussion

The high activity of compound **24** (Table 1) as an NMDA receptor antagonist in neonatal rat motoneurones indicated the apparent success of our original strategy of retaining the *trans*-3-carboxyl group of the agonist *trans*-1-aminocyclobutane-1,3-dicarboxylic acid (**1a**). However, our subsequent exploration of related substances (Y. Gaoni, P. C.-K. Pook, and J. C. Watkins, unpublished observations) seemed to indicate that the crucial feature of this activity was not the presence of the agonist-essential *trans*-3-carboxyl moiety; for instance, the analogue of **2c** containing an additional *trans*-3-carboxy group had the same activity as **2c**. Indeed, the consistent structural feature of all the most active substances was the presence of a 2′-carboxyethyl or a 2′-phosphonoethyl group in the 3-position of the cyclobutane ring. The other structural features to emerge from the results reported here were (a) a phosphonoethyl group was more effective than a carboxyethyl group, (b) the *cis* orientation of the acidic 3-ethyl substituent was preferred to the *trans* orientation, and (c) a 1′-keto, 1-hydroxy or 1′, 2′-methylene group were well tolerated in the 3-ethyl side chain.

The most unusual feature of these structure–activity relations is the finding that high activity was shown by "AP6-length" compounds. Allan and colleagues[9] had previously reported only low activity of *cis/trans* mixtures of AP5- and AP7-length cyclobutane analogues, which, in other groups of compounds[3,10,13] usually have higher activity than AP6-length compounds. We have confirmed this low activity of AP5-length and AP7-length cyclobutane analogues in our current study (unpublished observations of Y. Gaoni, P. C.-K. Pook, and J. C. Watkins). An explanation of these results were sought in computer-assisted molecular modeling studies.

Molecular Modeling Studies on AP5–AP7-Length Cyclobutane Analogues. Two template molecules

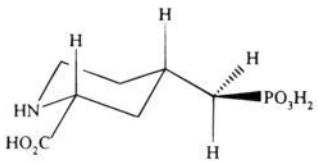


Figure 1. (−)-Gauche conformation of the CGS19755 template.

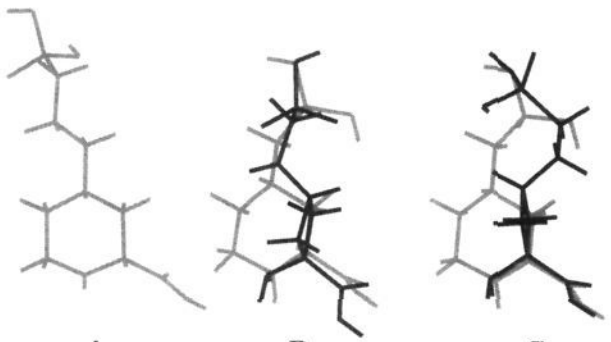


Figure 2. (A) Conformation of CGSAP7 used in the modeling study. (B) Superimposition of **4b** and CGS19755. (C) Superimposition of **3b** and CGS19755.

Table 2. Results of Superimposition of Cyclobutane Analogues on Both AP5- and AP7-Length Templates

	RMS deviation in aligned positions	
compd	CGS19755 template	CGSAP7 template
4a	0.860	No Fit
3a	0.515	0.490
4b	0.155	0.131
3b	0.147	0.182
4c	0.403	0.359
3c	No Fit	No Fit

were selected to represent both AP5- and AP7-length analogues. In agreement with other molecular modeling studies of the NMDA receptor antagonist site,[11,26,27] the (−)-gauche conformation of CGS19755 (Figure 1) was used as the AP5-length template. The analogue of CGS19755 with the AP7-length (CGSAP7; Figure 2A)[26,27] was chosen as the AP7-length template.

In this study all molecules were assembled using INSIGHT II (Version 2.2.0),[33] and then low-energy conformations were obtained by performing energy minimization calculations using DISCOVER (steepest descents followed by the quasi-Newton−Raphson algorithm). The molecules were modelled in the un-ionized state. The N atom of the amino group, an oxygen atom of the carboxyl group, and an oxygen atom of the phosphonic acid group of the cyclobutane analogues (selected to represent relatively fixed sites of interaction with complementary sites in the receptor) were superimposed on the corresponding atoms in either the CGS19755 or the CGSAP7 template. The RMS deviation in aligned positions of these atoms was then recorded (see Table 2). The CGSAP7 template could be superimposed on to the CGS19755 template with a high degree of precision using the strategy outlined above (RMS deviation in aligned positions is 0.111).

The model suggests that both *cis* and *trans* forms of the AP6-length cyclobutane analogues (**4b** and **3b**, respectively) might be expected to fit either AP5-*preferring or* AP7-*preferring subtypes of the NMDA* receptor better than would the corresponding AP5 or AP7 cyclobutane analogues. In the case of an AP7-preferring subtype, the model would also predict the higher activity of the *cis* relative to the *trans* form of the AP6 cyclobutane analogue, as observed (Table 1). In the case of an AP5-preferring receptor subtype, however, the *trans* form of the AP6-length analogue would have been predicted to be of similar potency to the *cis* form. If an AP5-preferring subtype of NMDA receptor is a major contributor to the excitatory amino acid receptor population in neonatal rat motoneurones, the observed lower activity of the *trans* cyclobutane AP6 analogue may possibly be explained by steric hindrance to accommodation of the ethyl chain in the *trans* form (Figure 2B,C). The current model differs from that of Ortwine and colleagues[27] in that, although a very similar conformation of the CGS19755 template was utilized, a more distal oxygen atom of the phosphonate group (relative to the carboxylate group) was required as the primary receptor interaction point in order to obtain good superimposition of the CGSAP7 template and the *cis* cyclobutane AP6 analogue (**4b**) on to the CGS19755 template. Limitations of this preliminary study may be more readily rationalized later when selective agonists and antagonists for specific subtypes of NMDA receptors become available. In this respect, it will be informative to test the compounds of the present investigation on various assemblies of cloned NMDA receptor subunits.[28]

Experimental Section

Chemistry. General Procedures. Melting points were taken on a Fisher-Johns apparatus and were not corrected. Proton NMR spectra were measured with a Bruker AMX400 spectrometer in $CDCl_3$ at 400 MHz, unless otherwise indicated. Shifts are given in δ units downfield from internal TMS, and J values are given in hertz. For spectra taken in D_2O, shifts are given as above, based on locating the HDO signal at d 4.80 ppm. TLC was done on Merck Kieselgel precoated aluminum plates. Silica gel for column chromatograhy was Merck Kieselgel 60 (70−230 mesh). The solvent of crystallization is indicated in paretheses after the indicated melting points. Elemental analyses were performed at the Microanalytical Laboratory of the Hebrew University, Jerusalem. Analytical values were within ±0.4% of the calculated theoretical values.

***cis*-1-Azido-3-(phenylsulfonyl)cyclobutane-1-carboxylic Acid (7).** The bicyclic acid **6** (2.38 g, 10 mmol) was warmed in DMF (30 mL) with TMG (2 mL) and NaN_3 (0.74 g, 15 mmol) at 90 °C during 6 h. The cooled solution was diluted with water, extracted twice with ether to remove nonacidic material, and then acidified with 3 M HCl, NaCl was added to saturation, and the mixture was extracted three times with ether. (*Caution*! *Conduct these operations in a well ventilated hood—hydrazoic acid may be liberated*). The ether solution was washed three times with water and dried over $MgSO_4$. Evaporation of the solvent, trituration of the solid product with hexane, and filtration furnished from 2.4 to 2.6 g (86−92%) of pure **7**. Azide **7**: mp 102−103 °C (benzene); NMR (250 MHz) 2.76−2.94 (symmetrical m, 4 H), 3.86 (pent, 1 H), 7.6−7.9 (5 H, aromatic protons), 9.80 (br, acidic H). Anal. ($C_{11}H_{11}N_3O_4S$) H, N; C: calcd, 46.98; found, 46.52.

***cis*-1-Azido-3-(phenylsulfonyl)cyclobutane-1-*N*,*N*-pentamethylenecarboxamide (9).** Acid **7** was warmed with excess $SOCl_2$ (ca. 1 mL/mmol of **7**) at 75 °C for 1 h. Excess $SOCl_2$ was then evaporated at reduced pressure, and the residue was treated twice by addition of hexane and reevaporation to dryness. The acid chloride **8** was dissolved in CH_2Cl_2 (10 mL/mmol of **7**) and cooled in ice. Piperidine (0.2 mL/mmol of **7**; 2 equiv) was added to the solution, and the reaction was allowed to proceed for 1 h at room temperature. The reaction mixture was washed with dilute acid and water, dried, and evaporated to provide **9** in near quantitative yield. Azide

9: mp 94–95 °C (CH_2Cl_2–hexane); NMR (250 MHz) 1.60 (br, 6 H), 2.78–2.99 (symmetrical m, 4 H), 3.35 (br, 2 H, two of CH_2 NCH_2 protons), 3.48–3.61 (m, 3 H, two of CH_2 NCH_2 protons plus tertiary ring proton), 7.5–7.9 (5 H). Anal. ($C_{16}H_{20}N_4O_3S$) C, H, N.

***cis*-1-Amino-3-(phenylsulfonyl)cyclobutane-1-*N,N*-pentamethylenecarboxamide (10).** Hydrogenation of **9** was carried out in ethyl acetate under atmospheric pressure with 10% Pd/C as the catalyst. The catalyst (10% by weight of **9**) was first stirred in ethyl acetate under hydrogen for 0.25 h. A solution of **9** in the same solvent (10 mL/mmol of **9**) was then added, and stirring under hydrogen was resumed and maintained for 20 h. If the reaction mixture still showed the triazene spot, it was refluxed until total conversion of 11 to **10**. Filtration of the catalyst and evaporation of the solvent furnished solid **10** in 93–96% yield: mp 109–110 °C (CH_2Cl_2–hexane); NMR (250 MHz) 1.55 (br, 6 H), 1.89 (s, NH2), 2.44–2.52 and 3.03–3.11 (two m, 4 H), 3.44–3.53 (m, 5 H, CH_2 NCH_2 plus tertiary ring proton), 7.5–7.9 (5 H). Anal. ($C_{16}H_{22}N_2O_3S$) C, H.

***cis*-1-(Benzoylamino)-3-(phenylsulfonyl)cyclobutane-1-*N,N*-pentamethylenecarboxamide (12).** Benzoylation of **10** was carried out in CH_2Cl_2 with Et_3N and PhCOCl (4 mL, 0.15 mL and 0.116 mL, respectively, per mmol, 322 mg, of **10**). After addition of the benzoyl chloride at 0 °C, the reaction flask was warmed to room temperature for 1 h, during which time the product precipitated. Most of the solvent was then evaporated at reduced pressure, and the residue was taken in water, filtered, washed with water and with ethanol–water, and then air-dried. The highly insoluble product **12** thus obtained (80–86% yield relative to acid **6**) is suitable for all further uses. An analytical sample was obtained by crystallization from ethanol: mp 277–278 °C; NMR 1.43 and 1.55 (two br s, 6H), 2.79 and 3.31 (two m, 4 H), 3.39 and 3.56 (two br s, 4 H), 3.73 (pent, 1 H), 7.38 (s, NH), 7.4–7.9 (10 H). Anal. ($C_{23}H_{26}N_2O_4S$) C, H, N.

***cis*- and *trans*-3-(Benzoylamino)-3-(pentamethylenecarbamoyl)-1-(phenylsulfonyl)cyclobutane-1-carboxylic Acid (13) and *cis*- and *trans*-Methyl 3-(Benzoylamino)-3-(pentamethylenecarbamoyl)-1-(phenylsulfonyl)cyclobutane-1-carboxylate (14 and 15).** A solution–suspension of **12** (5 g, 11.7 mmol) in THF (140 mL) was stirred at −40 to −50 °C with 2.2 equiv of BuLi during 1 h. Pieces of solid CO_2 were then added to the solution, and the reaction flask was allowed warm out of the cooling bath to room temperature. Water was then added, and most of the THF was evaporated under reduced pressure. The residue was dissolved in additional water (total volume ca. 100 mL) and extracted twice with CH_2Cl_2 for any nonacidic material, including unreacted **12** which may be recovered later. The water layer is then acidified and extracted several times with CH_2Cl_2 for the product. Crude **13**, a mixture of *cis* and *trans* isomers, is usually obtained as a solid foam in 86–94% yield (4.8–5.2 g) and is ued as such for desulfonylation. For characterization purposes, a sample of crude **13** was esterified in THF with an ethereal solution of diazomethane and the esters were chromatographically separated (silica gel, 50 times the weight of product EtOAC–CH_2Cl_2–hexane, 1:1:1). The first eluted product was the *cis*-S,N compound **14**: mp 256–257 °C dec (MeOH); NMR (80 MHz) 1.6 (br s, 6 H), 3.24 and 3.66 (ABq, 4 H), 3.55 (br s, 4), 3.66 (s, Me) 7.4–8.0 (10 H). Anal. ($C_{25}H_{28}N_2O_6S$) C, H, N.

The second eluted product was the trans-S,N ester **15**, mp 261–262 °C (dec.; MeOH); NMR (80 MHz) 1.53 (br s, 6 H), 3.09 and 3.72 (ABq, 4 H), 3.5 (br s, 4 H), 3.63 (s, Me), 7.4–8.0 (10 H). Anal. ($C_{25}H_{28}N_2O_6S$) C, H, N.

***cis*- and *trans*-1-(Benzoylamino)-1-(pentamethylenecarbamoyl)cyclobutane-3-carboxylic Acid (16 and 17) and *cis*- and *trans*-Methyl 1-(Benzoylamino)-1-(pentamethylenecarbamoyl)cyclobutane-3-carboxylate (18 and 19).** Crude **13** (ca. 5 g), obtained by carboxylation of 5 g of **12**, was stirred in a mixture of 30 mL of THF and 90 mL of MeOH with 12 g of 6% sodium amalgam during 3 h at room temperature. The reaction mixture was filtered on Celite, the solution was slightly acidified with 3 M HCl, and most of the solvent was evaporated at reduced pressure. Saturated aqueous NaCl (20 mL) was added to the residue, which was then acidified further with HCl and extracted three times with CH_2Cl_2 (total volume: ca. 75 mL). On washing the CH_2Cl_2 solution with water to remove excess HCl, the product begins to precipitate. After completion of the precipitation (ca. 1 h), the mixture is filtered to recover the solid product. This is washed on the filter with water and then air-dried to give 1.2–1.5 g of almost pure **17** (as indicated by esterification of a small sample). The filtrate is then further washed with water, dried, and evaporated to give a residue which can be induced to precipitate 0.3–0.4 g of almost pure **16**, or else may be esterified and chromatographed for esters **18** and **19**. Alternatively, the total acid product can be esterified with ethereal diazomethane in CH_2Cl_2–THF by stirring the reaction mixture until all the solid is dissolved, while ensuring the presence of excess diazomethane. The total yield in acids and esters is 74–79% relative to **12**.

Pure acid **16** was obtained by crystallization of the above product from ethanol, mp 207–208 °C. Anal. ($C_{18}H_{22}N_2O_4$) C, H.

Pure acid **17** was similarly obtained by crystallization of the above described product from ethanol, mp 209–210 °C. Anal. ($C_{18}H_{22}N_2O_4$) H; C: calcd. 62.05; found, 62.47.

For the separation of **18** and **19**, the total product from carboxylation and desulfonylation of 6 g of **12** was esterified and chromatographed (200 g of silica gel; CH_2Cl_2–EtOAc–ether, 8:5:6). This provided 1.65 g of **18**, 0.4 g of a mixture of **18** and **19**, and 1.7 g of **19** (total: 3.75 g, 75% from **12**).

Product **18**: mp 185–186 °C (CH_2Cl_2–hexane); NMR 1.55 (br, 6 H), 2.49–2.56 and 3.22–3.30 (two unsymmetrical m, 4 H), 3.06 (m, 1 H), 3.50 (br, 4 H), 3.71 (s, Me), 7.08 (s, NH), 7.4–7.8 (5 H). Anal. ($C_{19}H_{24}N_2O_4$) C, H, N.

Product **19**: mp 225–226 °C (CH_2Cl_2–hexane); NMR 1.57 (br, 6 H), 2.83–2.91 and 3.00–3.08 (two symmetrical m, 4 H), 3.71 (pent, 1 H), 3.53 (br s, 4 H), 3.69 (s, Me), 6.61 (s, NH), 7.4–7.8 (5 H). Anal. ($C_{19}H_{24}N_2O_4$) C, H, N.

***cis*- and *trans*-1-Aminocyclobutane-1,3-dicarboxylic Acid (2a and 1a).** Product **18** (1.6 g, 4.65 mmol) was warmed in 6 M HCl (15 mL) at 130 °C for 24 h. Water (15 mL) was added to the cooled solution which was then extracted twice with ether to remove benzoic acid. The water solution was evaporated to dryness, and the residue was taken in water and reevaporated. The same operation was repeated twice more with water. The final hydrochloride residue was dissolved in ethanol (10 mL) and added with propylene oxide (2–3 mL). Precipitation of **2a** started after a while and was over after ca. 3 h. The crystalline solid was collected by filtration and recrystallized from water, yielding pure *cis* isomer **2a** (0.7 g, 85%), identical with an authentic sample.[18] NMR (D_2O) 2.54–2.59 and 2.75–2.81 (symm. m, 4 H), 3.36 (pent, 1 H).

The trans isomer **1a** was similarly obtained from **19**, and in similar yield. It was identical in all respects with an authentic sample.[18] NMR (D_2O) 2.57–2.63 and 2.88–2.93 (m, 4 H), 3.41 (m, 1 H).

Methyl *r*-3-(Benzoylamino)-*t*-1-[*trans*-2-(methoxycarbonyl)cyclopropyl]-3-(pentamethylenecarbamoyl)cyclobutane-1-carboxylate (21) and Methyl *r*-3-(Benzoylamino)-*c*-1-[*trans*-2-(methoxycarbonyl)cyclopropyl]-3-(pentamethylenecarbamoyl)cyclobutane-1-carboxylate (22). To a solution of a mixture of **18** and **19** (1.71 g, 4.97 mmol) in THF (80 mL) was added at −78 °C a solution of LDA, prepared at 0 °C from BuLi (8.4 mL of 1.47 M solution, 12.5 mmol; 2.5 equiv) and diisopropylamine (2 mL, 13 mmol) in THF (15 mL). After stirring at −78 °C for 0.5 h, excess methyl γ-bromocrotonate (1.0 mL, ca. 9.0 mmol) was added, and stirring was continued in an ice bath for 1 h. Saturated aqueous ammonium chloride solution (5 mL) was added, and most of the THF was evaporated under reduced pressure. Water was added to the residue, slightly acidified, and extracted with EtOAc. After washing and drying, the solution was added with ethereal diazomethane to account for any hydrolized acidic product. Evaporation of the solvent and chromatography (200 g of silica gel; CH_2Cl_2–ether–EtOAc, 4:2:1) separated **21** (1.14 g) from **22** (0.43 g), with intermediate mixed fractions (0.15 g; total: 1.72 g, 78%).

Compound **21**: mp 212–213 °C (EtOH); NMR 1.01–1.06, 1.18–1.23, 1.63–1.68, and 1.79–1.84 (four m, 4 H, cyclopropane protons), 1.50–1.56 (br, 6 H), 2.58–2.80 (m, 4 H, cyclobutane protons), 3.39 and 3.52 (br, 4 H), 3.68 and 3.73 (two s, two Me), 7.17 (s, NH), 7.4–7.8 (5 H). Anal. ($C_{24}H_{30}N_2O_6$) C, H.

Compound **22**: mp 230–231 °C (EtOH); NMR 1.07–1.12, 1.20–1.25, 1.76–1.80, and 1.85–1.90 (four m, 4 H, cyclopropane protons), 1.50 and 1.59 (br, 6 H), 2.56–2.60 and 3.25–3.30 (two m, 4 H, cyclobutane protons), 3.47 (br s, 4 H), 3.63 and 3.70 (two s, two Me), 6.80 (s, NH), 7.4–8.0 (5 H). Anal. ($C_{24}H_{30}N_2O_6$) C, H.

***r*-1-Amino-*c*-3-(*trans*-2-carboxycyclopropyl)cyclobutane-1,3-dicarboxylic Acid (24).** Product **22** (86 mg) was treated with 6 M HCl (10 mL) as described above for the conversion of **18** to **2a** to yield acid **24** (32 mg, 68%): mp darkening from above 200 °C, no melting (H_2O); NMR (D_2O) 1.09–1.14, 1.21–1.26, 1.66–1.70, and 1.81–1.86 (four m, 4 H, cyclopropane protons), 2.30–2.38 and 2.90–3.00 (dd and symm. m, respectively; two AB systems of the cyclobutane 4 H, the lower field protons showing long range coupling with two of the cyclopropane protons). Anal. ($C_{10}N_{13}NO_6 \cdot H_2O$) C, H, N.

***r*-1-Amino-*t*-3-(*trans*-2-carboxycyclopropyl)cyclobutane-1,3-dicarboxylic Acid (23)** was similarly prepared, in a similar yield, from **21**: mp decomposition at ca. 295 °C (H_2O); NMR (D_2O) 1.17–1.22, 1.25–1.30, 1.72–1.76, and 1.94–2.00 (four m, 4 H, cyclopropane protons), 2.5–2.7 (unsymm. m, 4 H, cyclobutane protons). Anal. ($C_{10}H_{13}NO_6$) C, H.

***cis*- and *trans*-1-(Benzoylamino)-3-(hydroxymethyl)-cyclobutane-1-*N*,*N*-pentamethylenecarboxamide (25 and 29).** To a solution of ester **18** (3.44 g, 10 mmol) in dioxane–water (100 mL each) were added 3.8 g (100 mmol) of $NaBH_4$, and the mixture was stirred for 20 h at room temperature. To the ice-cooled flask was then added carefully 3 M HCl until the solution was slightly acidic. The volume of the solution was reduced by about two-thirds at reduced pressure, whereby the product started to precipitate or was induced to precipitate. It was collected by filtration, washed with water, and air-dried to provide 2.28 g of crude product. Further concentration of the filtrate provided a further crop of solid which was similarly treated. The total solid (2.75 g) was purified by chromatography (60 g of silica gel; CH_2Cl_2–10% MeOH) to furnish 2.2 g of pure *cis*-**25** (70% yield): mp 228–229 °C (EtOAc–MeOH); NMR 1.49 and 1.57 (br, 4 and 2 H), 2.44 (m, 3 H), 2.64 (br, O*H*), 3.00 (m, 2 H), 3.50 (br, 4 H), 3.68 (br s, 2 H), 7.22 (s, N*H*), 7.4–7.8 (5 H). Anal. ($C_{18}H_{24}N_2O_3$) C, H.

Alcohol **29** was similarly prepared from ester **19**, in a similar yield: mp 212–213 °C (EtOAc–MeOH); NMR 1.46 and 1.54 (br, 4 and 2 H), 2.5–2.7 (m, 5 H), 2.75 (O*H*), 3.47 (br s, 4 H), 3.56 (d, J = 5.7, CH_2OH), 7.4–7.8 (N*H* and aromatic protons, 6 H). Anal. ($C_{18}H_{24}N_2O_3$) C, H.

***cis*- and *trans*-1-(Benzoylamino)-3-formylcyclobutane-1-*N*,*N*-pentamethylenecarboxamide (26 and 30).** To a solution of 0.25 mL of DMSO in CH_2Cl_2 (1.5 mL) cooled to −75 °C was added dropwise via syringe a solution of 0.2 mL of trifluoroacetic anhydride (TFAA) in 1 mL of CH_2Cl_2. Stirring was continued for 10 min, and then a solution of **25** (200 mg) in CH_2Cl_2 (ca. 20 mL) was added to the reaction flask (the highly insoluble **25** was dissolved with warming in 40 mL of CH_2Cl_2, and the solution was then concentrated to ca. 20 mL). Stirring was continued for 3 h at that temperature and then for 0.5 h out of the cold bath. Triethylamine (0.5 mL) was then added, and stirring was continued for 5 min. Workup involved washing with water, with dilute acid, and with a saturated sodium chloride solution. The water layer was saturated with NaCl and extracted three times with CH_2Cl_2. The combined organic extracts were dried on $MgSO_4$, filtered, and evaporated to furnish 181 mg of crude aldehyde. Chromatography (20 g of silica gel; CH_2Cl_2–5% MeOH) provided pure **26** (125 mg, 63%): mp 176–177 °C (EtOAc); NMR 1.51, 1.59, and 1.68 (three br s, 6 H), 2.67 (m, 2), 3.15 (m, 3), 3.51 (br s, 4), 6.70 (s, NH), 7.4–7.8 (5 H), 9.85 (s, 1). Anal. ($C_{18}H_{22}N_2O_3$) C, H.

Aldehyde **30** was similarly prepared from **29**, in a similar yield: mp 195–196 °C (EtOAc); NMR 1.60 (br, 6 H), 2.85 and 3.13 (d AB q, $J_{A,B}$ = 13.4, $J_{A,X}$ = 7.3, $J_{B,X}$ = 9.6, 4 H), 3.36 (m, 1 H), 3.52 (br s, 4 H), 6.53 (br s, N*H*), 7.4–7.8 (5 H). Anal. ($C_{18}H_{22}N_2O_3$) C, H.

***cis*- and *trans*-1-(Benzoylamino)-3-[2-(methoxycarbonyl)ethenyl]cyclobutane-1-*N*,*N*-pentamethylenecarboxamide (27 and 31).** Aldehyde **26** (110 mg) was warmed in benzene (20 mL) with methyl (triphenylphosphoranylidene)-acetate (160 mg) at 80 °C for 20 h. The solvent was evaporated to dryness, and the residue was triturated twice with ether which dissolved the phosphorus-containing compounds and left behind crude solid **27**. Recrystallization from EtOAc–hexane provided pure **27** (100 mg, 77%): mp 218–219 °C; NMR 1.52 and 1.59 (br, 2 and 4 H) 2.49 and 3.17 (two m, 4 H), 2.99 (m, 1 H), 3.53 (br, 4 H), 3.72 (s, Me), 5.80 (d, J = 15.6, 1 H) and 7.07 (dd, J = 15.6 and 7.2, 1 h), 6.37 (br s, NH), 7.4–7.8 (5 H). Anal. ($C_{21}H_{26}N_2O_4$) C, H.

Ester **31** was similarly prepared from **30** in a similar yield: mp 232–233 °C (EtOAc); NMR 1.53 and 1.61 (br, 4 and 2 H), 2.70 and 2.83 (two symm. m, 4 H), 3.31 (sextet, 1 H), 3.52 (br s, 4 H), 3.73 (s, Me), 5.79 (d, J = 15.6, 1 H) and 7.00 (dd, J = 15.6 and 7.2, 1 H), 7.4–7.8 (5 H). Anal. ($C_{21}H_{26}N_2O_4$) C, H.

***cis*- and *trans*-1-(Benzoylamino)-3-[2-(methoxycarbonyl)ethyl]cyclobutane-1-*N*,*N*-pentamethylenecarboxamide (28 and 32).** Compound **27** (90 mg) in EtOH (20 mL) was introduced into prereduced PtO_2 catalyst (20 mg in 5 mL EtOH), and the mixture was stirred under hydrogen for 2 h. Filtration of the catalyst and evaporation of the solvent furnished solid **28** in quantitative yield (91 mg): mp 169–170 °C (EtOAc–hexane); NMR 1.51 and 1.58 (br, 4 and 2 H), 1.84 and 2.26 (q and t, 4 H, side-chain methylenes), 2.14 and 3.06 (two m, 3 and 2 H), 3.51 (br, 4 H), 3.66 (s, Me), 6.35 (s, NH), 7.4–7.8 (5 H). Anal. ($C_{21}H_{28}N_2O_4$) C, H.

Hydrogenation of **31** similarly furnished **32**: mp 197–198 °C (CH_2Cl_2–hexane); NMR 1.51 and 1.60 (br, 4 and 2 H), 1.76 and 2.25 (q and t of side-chain methylene protons), 2.45 and 2.60 (two m, 3 and 2 ring protons), 3.51 (br, 4 H), 3.65 (s, Me), 6.54 (s, N*H*), 7.4–7.8 (5H). Anal. ($C_{21}H_{28}N_2O_4$) C, H.

***cis*-1-Amino-3-(2-carboxyethyl)cyclobutane-1-carboxylic Acid (2c).** Hydrolysis of **28** (75 mg) was carried out as described above for **18** providing crystalline **2c** (32 mg, 85%), pure by TLC (cellulose plates; CH_3CN–H_2O–AcOH–pyridine, 90:20:5:1.5) and by NMR: mp ca. 260 °C dec; NMR (D_2O) 1.76 and 2.30 (q and t, 4 H, side chain methylenes), 1.97–2.03 and 2.59–2.65 (two symmetrical m, 4 H), 2.47 (sept, 1 H). Anal. ($2C_8H_{13}NO_4 \cdot H_2O$) C, H, N.

***trans*-1-Amino-3-(2-carboxyethyl)cyclobutane-1-carboxylic Acid (1c)** was similarly prepared by hydrolysis of **32**: mp darkening from about 240 °C, no melting up to 300 °C; NMR 1.54 and 1.76 (two m, 4 H, side-chain methylenes), 2.43 (d, J = 8.2, 4 H), 2.64 (pent, 1 H). Anal. ($2C_8H_{13}NO_4 \cdot H_2O$) C, H, N.

***cis*- and *trans*-1-(Benzoylamino)-3-[2-(dimethoxyphosphoryl)acetyl]cyclobutane-1-*N*,*N*-pentamethylenecarboxamide (33 and 36).** A solution of dimethyl (lithiomethyl)-phosphonate was prepared at −78 °C from dimethyl methylphosphonate (3.5 mL, 31 mmol) in THF (160 mL) by stirring for 1 h with BuLi (12.4 mL of a 2.5 M solution in hexane, 31 mmol). Solid ester **18** (3.2 g, 9.4 mmol) was introduced all at once into the reaction flask, and stirring was continued for 40 min at −78 °C and for 1 h in an ice bath. Addition of water, evaporation of the THF, acidification of the water layer, and extraction with CH_2Cl_2 provided a crude product that was purified by chromatography (65 g of silica gel; CH_2Cl_2–5% MeOH) to furnish pure *cis*-**33** (3.76 g, 92%): mp 173–174 °C (CH_2Cl_2–hexane); NMR 1.4–1.6 (br, 6 H), 2.63 and 3.22 (two m, 4 H), 3.13 (d, J = 22.6, POCH_2CO), 3.30 (m, 1 H), 3.4–3.6 (br, 4 H), 3.73 and 3.76 (two s, two Me), 7.37 (s, N*H*), 7.4–7.8 (5 H). Anal. ($C_{21}H_{29}N_2PO_6$) C, H.

Ketone **36** was similarly prepared from **19** in a similar yield: mp 196–197 °C ($CHCl_3$–hexane); NMR 1.52 and 1.59 (br, 4 and 2 H), 2.77 and 3.05 (two m, 4 H), 3.09 (d, J = 22.5, 2 H), 3.49 (br, 4 H), 3.55 (pent, 1 H), 3.74 and 3.77 (two s, two Me), 7.08 (s, N*H*), 7.4–4.8 (5 H). Anal. ($C_{21}H_{29}N_2PO_6$) C, H.

***cis*- and *trans*-1-(Benzoylamino)-3-[2-(dimethoxyphosphoryl)-1-hydroxyethyl]cyclobutane-1-*N*,*N*-pentamethylenecarboxamide (34 and 37).** Ketone **33** (1090 mg, 2.5

mmol) was dissolved with warming in ethanol (20 mL). The solution was cooled back to room temperature, and solid $NaBH_4$ (70 mg, 1.84 mmol) was added. After 0.25 h at room temperature, AcOH was added dropwise until gas evolution stopped. The solvent was evaporated to dryness under reduced pressure, and the residue was taken in CH_2Cl_2, filtered on Celite, and chromatographed (30 g of silica gel; CH_2Cl_2–7% MeOH) to provide pure *cis*-**34** (1020 mg, 93%): mp 169–170 °C (EtOAc); NMR 1.50 and 1.56 (br, 4 and 2 H), 1.74–1.86 (m, 2 H, POC*H*$_2$), 2.25–2.42 and 2.99–3.09 (two m, 3 and 2 H), 3.50 (br, 4 H), 3.74 and 3.77 (two d, J = 5, two Me), 4.09 (m, 1 H C*H*OH), 4.20 (s, O*H*), 7.32 (s, N*H*), 7.4–7.8 (5 H). Anal. ($C_{21}H_{31}N_2PO_6$) C, H.

Alcohol **37** was similarly prepared from **36** in a similar yield: mp 191–192 °C (EtOAc–MeOH); NMR 1.49 and 1.57 (br, 4 and 2 H), 1.75–1.95 (m, 2 H, POC*H*$_2$), 2.45–2.79 (m, 5 H), 3.48 (br, 4 H), 3.72 and 3.75 (two d, J = 7.7, two Me), 3.93 (m, 1 H, C*H*OH), 7.4–7.8 (5 H). Anal. ($C_{21}H_{31}N_2PO_6$) C, H.

***cis*-1-Amino-3-(2-phosphono-1-hydroxyethyl)cyclobutane-1-carboxylic Acid (35).** Hydrolysis of **34** (160 mg; 6 M HCl, 130 °C, 24 h) and workup with propylene oxide gave after filtration an amorphous, sticky solid. After being induced to crystallize in some water, ethanol was added to complete precipitation. Recrystallization from water–ethanol provided pure **35** (66 mg, 61%): mp 236–237 °C (decomposition); NMR (D_2O) 1.69–1.85 (m, 2 H), 2.20–2.34 and 2.64–2.72 (two m, 2 and 3 H), 3.95 (br, 1 H, C*H*OH). Anal. ($C_7H_{14}NPO_6$) C, H, N.

***trans*-1-Amino-3-(2-phosphono-1-hydroxyethyl)cyclobutane-1-carboxylic Acid (38) and 1-Amino-3-(2-phosphonoethylidene)cyclobutane-1-carboxylic Acid (39).** Similar hydrolysis of **37** provided **38**, accompanied by **39**. Partial separation of **38** from **39** was achieved by crystallization from water, whereby **39** precipitated free of **38**. Acid **38** was further purified by chromatography, as described above for **35**, and by crystallization from water–ethanol: mp, decomposition above 200 °C; NMR (D_2O) 1.68–1.86 (m, 2 H), 2.35–2.4 and 2.6–2.8 (two m, 2 and 3 H), 4.03 (br, 1 H, C*H*OH). Anal. ($C_7H_{14}NPO_6$) C, H, N.

Acid **39** was obtained pure by a second crystallization from water: mp, rapid decomposition above 250 °C; NMR (D_2O) 1.95–2.06 and 2.17–2.29 (two symm. m, 2 H, PC*H*$_2$), 2.33, 2.61, 2.71, and 3.01 (four m, 4 H), 5.05 (m, 1H, CH_2CH=C). Anal. ($C_7H_{12}NPO_5$) C, H, N.

***cis*- and *trans*-1-Amino-3-(2-phosphonoacetyl)cyclobutane-1-carboxylic Acid (40 and 41).** Hydrolysis of either **33** or **36** (6 M HCl, 130 °C, 24 h; work up with propylene oxide) gave a similar mixture of **40** and **41** in a ratio of ca. 3:2, respectively, and in a total 75–80% yield. By crystallization of the mixture from water, a varying amount pure isomer **39** could be obtained, as evidenced by 400 MHz ^{1}H NMR (TLC did not distinguish between the isomers). Further separation of the isomers was achieved by chromatography of the mixture on Dowex 1 ion-exchange resin (acetate form; elution with 1 M AcOH) and fractional crystallization from water of the solid obtained from the first half of product-containing fractions. This provided again isomer **40** free of **41**. Mixtures rich in **41**, which are obtained from the second half of product-containing fractions, can be re-equilibrated to the initial 3:2 composition of the mixture by warming in 3 M HCl at 130 °C for 20 h. Pure isomer **41** could be obtained from the very last chromatography fractions by crystallization from water.

Acid **40**: mp 230–231 °C (decomposition; H_2O); NMR (D_2O), measured ca. 1 h after dissolution) 2.60–2.66 and 2.81–2.87 (two symm. m, 4 H), 3.05 and 3.07 (two d of unequal intensity, $J_{H,P}$ = 21.5, unequal distribution of the partly deuterium-exchanged P-C*H*$_2$-CO protons; the two protons are totally exchanged with deuterium after 24 h), 3.76 (pent, 1 H). Anal. ($C_7H_{12}NPO_6$) C, H, N.

Acid **41**: mp, decomposition above 175 °C (H_2O); NMR (D_2O) 2.60–2.66 asnd 3.00–3.06 (two m, 4 H; two d due to PC*H*$_2$-CO, and partly superimposed on the latter m, disappear slowly by exchange with deuterium), 3.80 (m, 1 H). Anal. ($C_7H_{12}NPO_6$) C, H, N.

The ratio of the two isomers in their mixtures can be determined by the integration ratio of the 2.81–2.87 multiplet of **40** to the 2.60–2.66 multiplet common to **40** and **41**.

Acid **40** (180 mg) was hydrogenated in water (30 mL) and acetic acid (10 mL) over prereduced PtO_2 hydrogenation catalyst (33 mg). The mixture was stirred under hydrogen for 20 h and then filtered and evaporated to provide acid **35** (175 mg), pure by NMR.

***cis*- and *trans*-1-(Benzoylamino)-3-[2-(dimethoxyphosphoryl)-1-[(methylsulfonyl)oxy]ethyl]cyclobutane-1-*N*,*N*-pentamethylenecarboxamide (42 and 45).** Alcohol **34** (400 mg) was mesylated in CH_2Cl_2 by treatment with CH_3SO_2Cl (0.33 mL) in the presence of Et_3N (0.6 mL) for 20 h. The crude product was chromatographed (20 g of silica gel; CH_2Cl_2–5% MeOH) to yield pure **42** (343 mg, 70%): mp 197–198 °C (CH_2Cl_2–MeOH); NMR 1.98–2.05 and 2.15–2.22 (two ddd, 2 H, ABq of POC*H*$_2$, further split by C*H*OMs and by P), 2.50–2.56 and 3.01–3.06 (two m, 4 H), 2.91 (m, 1 H), 3.13 (s, SO_2CH_3), 3.74 and 3.77 (two d, 2 Me), 5.05 (m, 1 H, C*H* OMs), 7.36 (s, NH), 7.4–7.9 (5 H). Anal. ($C_{22}H_{33}N_2PO_8S$) C, H.

Mesylate **45** was similarly prepared from alcohol **37**: mp 160–161 °C (CH_2Cl_2–hexane); NMR 1.5 and 1.7 (br, 4 and 2 H), 2.1–2.3 (m, 2 H, PC*H*$_2$), 2.56–2.69, 2.76–2.85, and 3.00–3.06 (three m, 2, 2, and 1 H), 3.15 (s, OSO_2CH_3), 3.50 (br, 4 H), 3.74 and 3.77 (two d, two Me), 4.93 (m, 1 H, C*H*OMs), 6.85 (br, N*H*), 7.4–7.9 (5 H). Anal. ($C_{22}H_{33}N_2PO_8S$) C, H.

***cis*-1-(Benzoylamino)-3-[2-(dimethoxyphosphoryl)ethenyl]cyclobutane-1-*N*,*N*-pentamethylenecarboxamide (43) and *cis*-1-(Benzoylamino)-3-[2-(dimethoxyphosphoryl)-ethyl]cyclobutane-1-*N*,*N*-pentamethylenecarboxamide (44).** Mesylate **42** (320 mg) was treated in THF (20 mL), at −78 °C, with BuLi (2.2 equiv) for 3 h. After quenching with aqueous NH_4Cl and extractive workup with EtOAc, the crude product was chromatographed (15 g of silica gel; CH_2Cl_2–5% MeOH) to yield **43** (230 mg, 93%), which was used in the following hydrogenation step. A sample was recrystallized from ethyl acetate to give pure **43**: mp 183–184 °C; NMR 2.42–2.48 and 3.06–3.11 (two m, 4 H), 3.32 (m, 1 H), 3.66 and 3.69 (two Me), 5.62 and 6.76 (t and m, 2 H; olefinic protons), 7.4–7.9 (5 H), 8.37 (N*H*). Anal. ($C_{21}H_{29}N_2PO_5$) C, H.

Product **43** (200 mg) was hydrogenated in ethanol (10 mL) over prereduced PtO_2 (40 mg) for 3 h. Filtration of the catalyst and evaporation of the solvent furnished oily **44** (160 mg) which showed no unsaturation (NMR) and which was used in the following hydrolysis step (see below).

***trans*-1-(Benzoylamino)-3-[2-(dimethoxyphosphoryl)-ethenyl]cyclobutane-1-*N*,*N*-pentamethylenecarboxamide (46) and *trans*-1-(benzoylamino)-3-[2-(dimethoxyphosphoryl)ethyl]cyclobutane-1-*N*,*N*-pentamethylenecarboxamide (47)** were similarly prepared from **45**, in similar yields. Phosphonate **46**: mp 165–166 °C; NMR 1.5 and 1.6 (br, 6 H), 2.71–2.74 and 2.79–2.82 (two symm. m, 4 H), 3.32 (m, 1 H), 3.52 (br s, 4 H), 3.70 and 3.73 (two s, two Me), 5.61 and 6.85 (dd and ddd, J_{AB} = 17.1, $^2J_{PH}$ = 20.2, 2 H), 6.66 (s, N*H*), 7.4–7.8 (5 H). Anal. ($C_{21}H_{29}N_2PO_5$) C, H.

Hydrogenation of **46**, as described for **43**, furnished again an oily product which was used as such for hydrolysis to **3b**.

***cis*-1-Amino-3-(2-phosphonoethyl)cyclobutane-1-carboxylic Acid (4b).** Hydrolysis of **44** (140 mg; 6 M HCl, 130° C, 24 h) and workup with propylene oxide gave a sticky solid which was dissolved in water and purified by chromatography (20 cm column of Dowex 1, acetate form; elution with 1 M AcOH) to provide **4b** (70 mg, 75%): mp decomposition from ca. 250 °C (H_2O); NMR (D_2O) 1.49–1.58 and 1.66–1.74 (two m, 4 H, side chain protons), 2.04–2.10 and 2.69–2.74 (two m, 4 H), 2.55 (pent, 1 H). Anal. ($C_7H_{14}NPO_5$) C, H, N.

***trans*-1-Amino-3-(2-phosphonoethyl)cyclobutane-1-carboxylic acid (3b)** was similarly obtained from **47**: mp, no melting below 300 °C, decomposition (H_2O); NMR (D_2O) 1.46–1.55 and 1.69–1.77 (two m, 4 H, side chain protons), 2.39 (d, J = 8.5, 4 H), 2.61 (m, 1 H). Anal. ($2C_7H_{14}NPO_5 \cdot H_2O$) C, H, N.

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