Design, Synthesis, and Anticonvulsant Activity of 1-(Pyrid-3-ylsulfonamido)-2-nitroethylenes

Bernard Masereel,*,† Johan Wouters,‡ Lionel Pochet,† and Didier Lambert§

Department of Pharmacy and Laboratory of Molecular Structures, University of Namur, FUNDP, 61, rue de Bruxelles, 5000 Namur, Belgium, and Laboratory of Medicinal Chemistry and Radiopharmacy, Catholic University of Louvain, Avenue Mounier CMFA 73.40, 1200 Brussels, Belgium

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The lipophilic 1-cycloalkylamino-1-(pyrid-3-yl-sulfonamido)-2-nitroethylenes were synthesized as bioisosteres of BM-34, an anticonvulsant sulfonylthiourea. Compound **17** (ip) emerged from the maximal electroshock seizure (MES) test with a 50% effective dose (ED_{50}) of 8.25 mg/kg. Its anticonvulsant profile was similar to that of phenytoin (ED_{50} = 9.51 mg/kg) and of BM-34 (ED_{50} = 1.19 mg/kg): active in the MES test and inactive in seizures induced by subcutaneous injection of pentetrazole, strychnine, bicuculline, picrotoxin, or *N*-methyl-D,L-aspartate. The neurotoxicity of **17** (TD_{50} = 113.8 mg/kg) was lower than that of phenytoin (TD_{50} = 65.5 mg/kg) but higher than that of BM-34 (TD_{50} = 147.2 mg/kg). Crystallographic study revealed that BM-401 (**17**) was a zwitterionic structure. Its sulfonamido nitroethylene side chain adopted a conformation which placed the two cycloalkyl rings face to face to form a single hydrophobic area.

Introduction

Nonclassical bioisosteric molecules are characterized by a different number of atoms, by some similar physicochemical parameters, and by a broadly similar biological activity.[1] This concept has been applied to the thiourea function of metiamide (**1**) and led to the discovery of cimetidine (**2**), a cyanoguanidine with histamine-H_2 antagonism (Figure 1).[2] Classically, the 1,1-diamino-2-nitroethylene is another group considered as a bioisostere of the thiourea function.[1] This bioisosteric replacement has been used to design ranitidine (**3**), an antiulcer nitroethylene derivative (Figure 1).[3] Recently, the acylurea group of phenytoin was modified into an anticonvulsant acylcyanoguanidine.[4] A similar strategy was also dedicated to the sulfonylurea function of torasemide (**4**) and glibenclamide, an ATP-sensitive K^+-channel blocker, which was transformed into a sulfonylcyanoguanidine moiety to give compounds with a similar pharmacological profile.[5−7] In the present work, we prepared lipophilic 1-amino, 1-sulfonamido, 2-nitroethylene compounds (**11**−**23**) regarded as bioisosteres of the sulfonylthiourea BM-34 (**5**) (Figure 1) structurally related to torasemide (**4**), a diuretic sulfonylurea.[8] Interestingly, BM-34 (**5**) was found devoid of diuretic activity in rats but showed, in anticonvulsant models, a higher potency, a lower neurotoxicity and a profile similar to that of phenytoin.[9] Epilepsy is a neurological disorder that involves the depolarization and interaction of ensembles of CNS neurons ultimately to produce the abnormal behavior pattern seen during the epileptic seizure.[10] Valproate, phenytoin, and carbamazepine are still the major drugs used today although phenytoin is much less used in Europe than in the USA. Due to the numerous side effects of these molecules, and the existence of refractory epilepsies, new drugs with different mechanism of actions have recently become available or await their approval: vigabatrin, gabapentin, lamotrigine, oxcarbazepine, tiagabine, and remacemide.[11,12] Because some 20% of all patients remain refractory to the current drugs or suffer unacceptable side effects, there is still a need for new antiepilectic drugs. BM-34 displayed an original template for chemical modifications, and considering its high anticonvulsant potency was a good lead for this study.

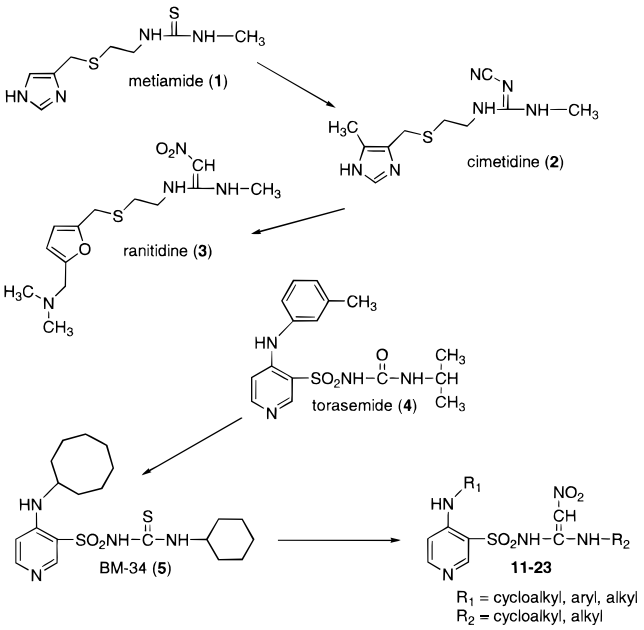

Figure 1. Bioisosterism of the thiourea function.

* Corresponding author: Professor B. Masereel, Department of Pharmacy, University of Namur, FUNDP, 61, rue de Bruxelles, 5000 NAMUR, Belgium. Tel: + 32 (0)81 72 43 38. Fax: + 32 (0)81 72 43 38. E-mail: bernard.masereel@fundp.ac.be.
† Department of Pharmacy, University of Namur.
‡ Laboratory of Molecular Structures, University of Namur.
§ Catholic University of Louvain.

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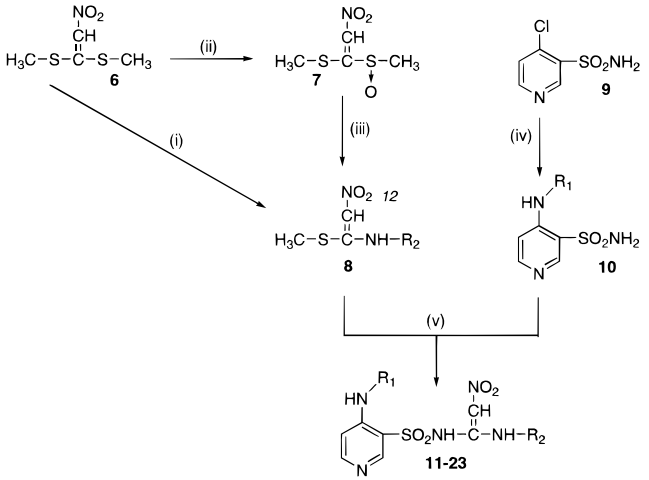
Scheme 1. Synthesis of 1-Amino-1-[(4-cycloalkylaminopyrid-3-yl)sulfonamido]-2-nitroethylenes Related to BM-34[a]



[a] (i) R_2-NH_2, C_2H_5OH; (ii) H_2O_2, CH_3COOH; (iii) R_2-NH_2, CH_3OH; (iv) R_1-NH_2, 2-propanol; (v) 1 equiv of NaOH, DMF, dioxane.

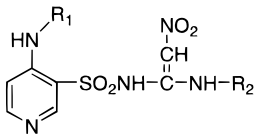
The synthesis, the physicochemical and the anticonvulsant properties of the novel nitroethylene derivatives are discussed.

Results and Discussion

Chemistry. The 1,1-bismethylsulfanyl-2-nitroethylene[13] **6** was refluxed with 1.2 equiv of cycloalkylamine to give the corresponding 1-cycloalkylamino-1-methylsulfanyl-2-nitroethylene **8** (Scheme 1). 1-Methylsulfanyl-1-methylsulfinyl-2-nitroethylene **7** prepared by oxidation of **6**[14] and treatment with isopropylamine at room temperature led to 1-isopropylamino-1-methylsulfanyl-2-nitroethylene. On the other hand, the 4-arylamino- and 4-cycloalkylaminopyrid-3-ylsulfonamides[5,15] **10** were synthesized by the reaction of 4-chloropyrid-3-ylsulfonamide[15] **9** with the corresponding aryl or cycloalkylamine. Finally, the sodium salt of **10** reacted with the required 1-substituted amino-1-methylsulfanyl-2-nitroethylene **8** to obtain the sulfonamidonitroethylenes derivatives **11**−**23** (Tables 1 and 2).

Lipophilicity. The novel nitroethylene derivatives (Table 1) are considered as the bioisosteres of their sulfonylurea and sulfonylthiourea counterparts which are neuroprotective[16] or anticonvulsant[9] drugs. Beside the active carrier systems present in the blood−brain barrier endothelium, the brain penetration is probably related to the overall drug lipophilicity. This later parameter is commonly expressed as the logarithm of the partition coefficient (log *P*) between a lipophilic phase (1-octanol) and an aqueous buffer. To reach the central nervous system, the log *P* value of a drug should be higher than +1.5.[17] The capacity factor (log *k*′) of compounds **21**−**23** and **11** was measured using a RP-HPLC assay (Table 2).[5] Their log *k*′ values were significantly correlated to their partition coefficient (log *P*) determined by using the 1-octanol/phosphate buffer pH 7.40 system (Table 2).[18] The log *P* values of the nitroethylene derivatives **11**−**20** (Table 1) were extrapolated from their log *k*′ values. The log *P* of the molecules bearing two cycloalkyl residues are ranging from +1.67 to +2.86. These values are in the range which can cross the blood−brain barrier. The log *P* increased with the C-atom number included in the R_1 and R_2 cycloalkyl (Figure 2). Whatever the size of the cycloalkyl residues (Table 1), the log *P* values were similar for the compounds having an identical total number of methylenes (compare **12**, **14**; **13**, **15**, **17**; **16**, **18**). The increase of one methylene unit resulted in an enhanced lipophilicity of 0.30 (= Δlog *P* CH_2). The lipophilicity of the *m*-tolyl

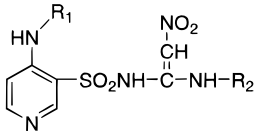
Table 1. Lipophilicity, Anticonvulsant and Diuretic Activity of the Nitroethylene Derivatives



compd	R_1	R_2	log *P*[a]	MES test[b] 0.5 h	MES test[b] 3 h	diuresis[c] (mL/kg)
11	C_6H_{11}	C_6H_{11}	+1.67	0/6	4/7	20
12	C_6H_{11}	C_7H_{13}	+1.97	0/6	5/7	20
13	C_6H_{11}	C_8H_{15}	+2.27	2/6	4/7	21
14	C_7H_{13}	C_6H_{11}	+1.99	1/6	5/6	18
15	C_7H_{13}	C_7H_{13}	+2.30	1/6	5/6	25
16	C_7H_{13}	C_8H_{15}	+2.57	1/6	4/7	23
17	C_8H_{15}	C_6H_{11}	+2.31	1/6	7/7	22
18	C_8H_{15}	C_7H_{13}	+2.59	2/6	5/6	20
19	C_8H_{15}	C_8H_{15}	+2.86	0/6	3/7	22
20	3-$CH_3C_6H_4$	C_6H_{11}	+1.66	1/6	2/6	29
4	torasemide		+0.47	0/6	0/6	88

[a] Values are means of three determinations obtained by the RP-HPLC method. [b] Rate of mice (n = 6−7) protected against electroshock (50 mA; 0.2 s) after ip injection of 30 mg/kg. [c] Urinary volume excreted over 4 h by rats after oral administration of 30 mg/kg. Control is 22 mL/kg.

Table 2. Correlation between the Logarithm of the Partition Coefficient (log *P*) Determined by the Shake-Flask Technique and the Logarithm of the Capacity Factor (log *k*′) Obtained by RP-HPLC



compd	R_1	R_2	log *P*[a]	log *k*′[b]
21	CH_3CH_2	$(CH_3)_2CH$	−0.482	−0.627
22	C_5H_9	$(CH_3)_2CH$	+0.538	−0.243
23	3-$CH_3C_6H_4$	$(CH_3)_2CH$	+1.116	−0.072
11	C_6H_{11}	C_6H_{11}	+1.588	+0.166

[a] Partition coefficient in 1-octanol phosphate buffer pH 7.40. log *P* = (2.662 log *k*′) + 1.206; r = 0.997; n = 4. [b] Capacity factor determined by RP-HPLC. All values are the mean of three determinations.

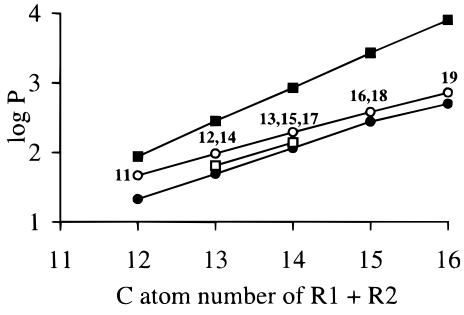


Figure 2. Lipophilicity of the sulfonylureas (●),[15] -thioureas (□),[15] and -cyanoguanidines (■)[5] compared to their nitroethylene counterparts (○).

Table 3. Neurotoxicity (TD_{50}) and Anticonvulsant Profile of BM-401 (**17**), BM-34 (**5**), and Phenytoin

drug	time of peak effect	TD_{50} (mg/kg)	ED_{50} and PI (mg/kg)[a] MES	PTZ	STR	BIC	PIC	NMLDA
BM-401 (**17**)	3 h	113.8 (96.4−121.2)	8.25 (5.1−13.4)[b] PI = 13.8	>100[c]	>100	>100	>100	>100
BM-34 (**5**)	4 h	147.2 (102.4−187.5)	1.19 (0.53−2.68) PI = 123.7	>100	>100	>100	>100	>100
phenytoin	2 h	65.5 (52.5−72.1)	9.51 (2.57−15.4) PI = 6.9	>100	>100	>100	>100	>100

[a] Eight OF-1 male mice per group. [b] 95% confidence interval. [c] No protection up to 100 mg/kg.

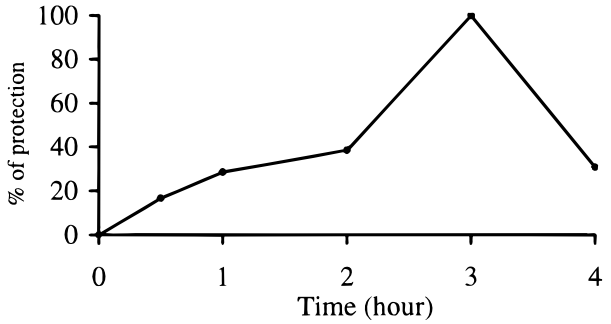


Figure 3. Time-course activity of BM-401 (**17**) (30 mg/kg ip) in the MES test. Eight OF-1 male mice for each time.

moiety was close to that of a cyclohexyl (compare **11** and **20**) (Table 1). At pH 7.40, the nitroethylene compounds were systematically more lipophilic ($\Delta\log P = +0.15$ to $+0.30$) than their urea bioisosteres[9] ($P < 0.01$) as well as the sulfonylthiourea counterparts ($\Delta\log P = +0.16$ to $+0.20$).[9] In the same experimental conditions, the nitroethylenes derivatives were significantly ($P < 0.01$) less lipophilic than the corresponding sulfonylcyanoguanidines ($\Delta\log P = -0.27$ to -1.04).[5] Thus the lipophilicity of these bioisostere functions is ordered as follows: $SO_2NH-C(=N-CN)-NH > SO_2NH-C(=CH-NO_2)-NH > SO_2NH-CS-NH \geq SO_2NH-CO-NH$.

Pharmacology. The anticonvulsant activity of the nitroethylene derivatives was examined at 30 mg/kg (ip) in mice by using the maximal electroshock seizure (MES) test which detect drugs that prevent spread of tonic-clonic seizures.[19] The MES test was applied 0.5 and 3 h after drug injection, and the rate of protected mice noted (Table 1). For each drug, a poor protection against MES was observed 0.5 h after the ip administration. At this time, **13** and **18** exhibited the best protection (33.3%). Evaluated 3 h after the drug injection, the protection rate was higher than 50% for each group treated by the compounds **11**−**18**. Compound **17** provided complete protection against induced seizures in mice at this dose level (Table 1). The weak activity observed at 0.5 h could be related to a lower drug concentration in the central nervous system at that time. The most potent molecule **17**, called BM-401, was the nitroethylene counterpart of BM-34 (**5**), which was the most active compound among the sulfonylureas and sulfonylthioureas previously studied.[9]

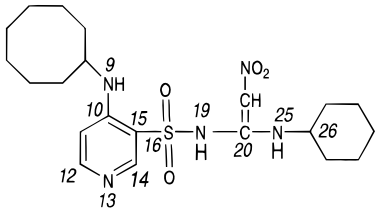
The time-course activity (Figure 3) of **17** (30 mg/kg) gave a peak effect ($T_{max} = 3$ h) shorter than that of BM-34 ($T_{max} = 4$ h) but longer than phenytoin ($T_{max} = 2$ h).[9] At that time, the MES ED_{50} value of **17** (8.25 mg/kg) was similar to that of phenytoin (9.51 mg/kg) but higher than that of BM-34 (1.19 mg/kg) determined in the same test conditions[9] (Table 3).

When examined at doses up to 100 mg/kg (ip), **17**, BM 34, and phenytoin did not protect mice against convulsions induced by subcutaneous injection of pentetrazole (85 mg/kg), strychnine (1.20 mg/kg), picrotoxin (3.15 mg/kg), *N*-methyl-D,L-aspartic acid (NMLDA, 340 mg/kg), or bicuculline (2.70 mg/kg) (Table 3). These results suggested that neurotransmitters such as glycine, GABA, or NMDA were not directly involved in the mechanism of action of **17** but do not preclude downstream modulation of their sites. Thus its pharmacological profile was similar to that of phenytoin and BM-34. As BM-34, **17** was structurally close to torasemide, a diuretic sulfonylurea (Figure 1).[8] Conversly to some lipophilic sulfonylureas and sulfonylthioureas,[9] **17** (30 mg/kg) had no diuretic activity in rats (Table 1).

At the time of peak effect (3 h), the neurotoxicity was evaluated by the rotorod procedure in mice[20] which detects motor coordination deficits. The median toxic dose (TD_{50}) of **17** (113.8 mg/kg) was higher than that of phenytoin (65.5 mg/kg) and lower than that of BM-34 (147.2 mg/kg). The protective index ($PI = TD_{50}/ED_{50}$) of **17** was better than that of phenytoin but lower than that of BM-34 (Table 3).

X-ray Crystallography and Molecular Modeling. In the crystal, **17** adopted a zwitterionic form resulting from the transfer of the sulfonamide proton to the nitrogen atom of the pyridine ring (Table 4). Valence angle $C_{12}-N_{13}-C_{14}$ (119.1°) close to 120° also suggested protonation of the pyridine ring. This zwitterionic structure was in agreement with the intermolecular interaction which revealed an interaction between the protonated nitrogen of the pyridinum ring and an oxygen of the nitro group. An intramolecular H-bond which involved the nitrogen N_9 (Table 4) and one oxygen

Table 4. Crystallographic Data, Optimized and Theoretical Values[a] of the Torsional Angles, and Energy Level of the Four Conformers of **17**



$\phi_1 = C_{10}-C_{15}-S_{16}-N_{19}$ $\phi_3 = S_{16}-N_{19}-C_{20}-N_{25}$

$\phi_2 = C_{15}-S_{16}-N_{19}-C_{20}$ $\phi_4 = N_{19}-C_{20}-N_{25}-C_{26}$

conformer	ϕ_1 (deg)	ϕ_2 (deg)	ϕ_3 (deg)	ϕ_4 (deg)	E (kcal/mol)
crystal	+69.4	+107.5	+172.5	−5.8	
β	+45.5 (+90)	+57.9 (+90)	−176.1 (180)	+3.3 (0)	8.53
α	−117.2 (−90)	+49.3 (+90)	−152.3 (180)	+130.2 (180)	15.56
γ	+62.4 (+90)	+54.1 (+90)	−159.7 (180)	+134.2 (180)	13.10
δ	+49.9 (+90)	+54.4 (+90)	−2.1 (0)	+131.6 (180)	11.94

[a] Theoretical values are in parentheses.

of the sulfonyl group reinforced the molecular packing. Crystallographic studies of several sulfonylureas, sulfonylthioureas, or sulfonylcyanoguanidines related to **17** revealed four different conformations (α, β, γ, δ), defined by typical values of the torsion angles ϕ_1, ϕ_2, ϕ_3, and ϕ_4 (Table 4).[5] In the crystal, the values of the torsion angles of the sulfonamidonitroethylene side chain indicated that **17** adopted a β conformation as observed in torasemide[21] (Table 4). To establish the relative energies of the α, β, γ, and δ conformations, only the torsion angles (ϕ_1 to ϕ_4) defining the different geometries were studies. The other rotable torsion angles, in particular those of the two cycloalkyl rings, were fixed to their initial values measured in the crystal. Conformational analysis showed that the β conformation corresponded to the preferred geometry of **17** as compared to the energy level of the α (ΔE = 7.03 kcal/mol), the γ (ΔE = 4.57 kcal/mol), or the δ conformation (ΔE = 3.41 kcal/mol). In the β conformation, both cycloalkyl rings were close and formed a single hydrophobic area.

In conclusion, the synthesis of the 1-amino, 1-sulfonamido, 2-nitroethylenes as bioisosteres of the anticonvulsant BM-34 led to the discovery a novel anticonvulsant **17**, called BM-401, exhibiting a pharmacological profile similar to that of phenytoin and BM-34. **17** was less neurotoxic and showed anticonvulsant activity similar to that of phenytoin. Additional experiments are warranted to elucidate the mechanism of action of **17**. This nitroethylene derivative presents an interesting alternative to BM-34 for which the thiourea function is well-known to be responsible for agranulocytosis neutropenia and aplastic anemia.[2,22,23]

Experimental Section

Chemistry. Melting points were determined on a Büchi B-540 capillary apparatus. IR spectra were recorded as KBr pellets on a Perkin-Elmer 1750 FT spectrophotometer. The ^{1}H NMR spectra were taken on a Brucker (300 MHz) instrument in DMSO-d_6 with HMDS as an internal standard; chemical shifts were reported in δ values (ppm) relative to internal HMDS. The abbreviation s = singlet, d = doublet, t = triplet, m = multiplet, and b = broad signal were use throughout. Elemental analyses (C, H, N, S) were performed on a Carlo-Erba elemental analyzer and were within ±0.4% of the theoretical values. All reactions were routinely checked by TLC on silica gel Merck 60F254.

1-Methylsulfanyl-1-methylsulfinyl-2-nitroethylene (7). Hydrogen peroxide (30%, 11.3 mL) was added dropwise to a stirred solution of 1,1-bismethylsulfanyl-2-nitroethylene[13] (**6**, 16.5 g, 100 mmol) in acetic acid (450 mL) and heated to 60 °C. After 17 h of reaction, the medium was evaporated under reduced pressure and the crude residue crystallized from methylethyl ketone (50 mL) to give the title compound (7.50 g, 41%): mp 118−120 °C; IR (KBr) 1549 (NO_2) cm^{-1}; ^{1}H NMR (DMSO-d_6, 300 MHz). Anal. ($C_4H_7NO_3S_2$) C, H, N, S.

1-Isopropylamino-1-methylsulfanyl-2-nitroethylene (8). Isopropylamine (3.8 mL, 45 mmol) was added to a solution of 1-methylsulfanyl-1-methylsulfinyl-2-nitroethylene[14] **7** in methanol (120 mL). The mixture was stirred for 30 min at 25 °C. After evaporation of the solvent under reduced pressure, the crude product was crystallized from 2-propanol (10 mL) to give the title compound (2.01 g, 30%): mp 92−94 °C; IR (KBr) 1562 (NO_2) cm^{-1} ; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 1.10 and 1.23 (2s, 6H, C(C*H*$_3$)$_2$), 2.43 (s, 3H, C*H*$_3$S), 3.84 (m, 1H, C*H*(CH$_3$)$_2$), 6.55 (s, 1H, C*H*-NO_2), 10.15 (b, 1H, N*H*). Anal. ($C_6H_{12}N_2O_2S$) C, H, N, S.

1-Cyclohexylamino-1-methylsulfanyl-2-nitroethylene (8). A solution of 1,1-bismethylsulfanyl-2-nitroethylene[13] (**6**, 2.0 g, 12 mmol) in absolute ethanol (25 mL) was refluxed with an excess of cyclohexylamine (2.1 mL, 18 mmol) for 18 h. After cooling, the formed precipitate was collected by filtration, washed with ice-cold ethanol, and dried to give the title compound (2.02 g, 75%): mp 101−103 °C; IR (KBr) 1563 (NO_2) cm^{-1}; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 1.05−1.95 (m, 11H, cyclohexyl), 2.45 (s, 3H, SC*H*$_3$), 6.68 (s, 1H, C*H*NO_2), 10.37 (b, 1H, N*H*). Anal. ($C_9H_{16}N_2O_2S$) C, H, N, S.

1-Cyclohexylamino-1-[(4′-cyclooctylamino)pyrid-3′-ylsulfonamido)]-2-nitroethylene (17). The sodium salt of 4-cyclooctylaminopyrid-3-ylsulfonamide **10**[5,15] (1.08 g, 3.5 mmol)) and 0.90 g (4.2 mmol) of 1-cyclohexylamino-1-methylsulfanyl-2-nitroethylene **8** were dissolved in a mixture of 1,4-dioxane (3 mL) and *N*,*N*-dimethylformamide (2 mL) and refluxed for 6 h. After evaporation of solvents under reduced pressure, the residue was dissolved in water (50 mL) and 2.5N NaOH (2 mL). The solution was extracted three times with diethyl ether (50 mL) and adjusted to pH 3 with dilute HCl. The precipitate was collected by filtration, washed with water, dried, and crystallized from absolute ethanol (50 mL) to afford the title compound (0.76 g, 48%): mp 186−188 °C; IR (KBr) 1577 (NO_2) cm^{-1}; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 0.96−1.97 (m, 26H, cyclohexyl and cyclooctyl), 6.70 (s, 1H, C*H*-NO_2), 7.10 (d, 1H, 5*H*-pyridine), 7.42 (d, 1H, pyridine-N*H*), 8.22 (d, 1H, 6*H*-pyridine), 8.57 (s, 1H, 2*H*-pyridine), 10.25 (d, 1H, N*H*-cyclohexyl). Anal. ($C_{21}H_{33}N_5O_4S$) C, H, N, S.

This procedure was used for the synthesis of the compounds **11**−**23**.

1-Cyclohexylamino-1-[(4′-cyclohexylamino)pyrid-3′-ylsulfonamido)]-2-nitroethylene (11). The title compound was obtained as previously described for the compound **17** by starting from **10** (1.0 g, 4.0 mmol) and 1-cyclohexylamino-1-methylsulfanyl-2-nitroethylene **8** (0.9 g, 4.2 mmol): yield 78% (1.32 g); mp 187−189 °C; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 1.15−1.95 (m, 22H, two cyclohexyl), 6.72 (s, 1H, C*H*-NO_2), 7.20 (d, 1H, 5*H*-pyridine), 7.38 (d, 1H, pyridine-N*H*), 8.27 (d, 1H, 6*H*-pyridine), 8.57 (s, 1H, 2*H*-pyridine), 10.32 (d, 1H, N*H*-cyclohexyl). Anal. ($C_{19}H_{29}N_5O_4S$) C, H, N, S.

1-Cycloheptylamino-1-[(4′-cyclohexylamino)pyrid-3′-ylsulfonamido)]-2-nitroethylene (12). The title compound was obtained as previously described for the compound **17** by starting from **10** (1.0 g, 4.0 mmol) and 1-cycloheptylamino-1-methylsulfanyl-2-nitroethylene **8** (0.96 g, 4.2 mmol): yield 56% (0.98 g); mp 186−188 °C; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 1.18−1.92 (m, 24H, cyclohexyl and cycloheptyl), 6.70 (s, 1H, C*H*-NO_2), 7.19 (d, 1H, 5*H*-pyridine), 7.40 (d, 1H, pyridine-N*H*), 8.26 (d, 1H, 6*H*-pyridine), 8.58 (s, 1H, 2*H*-pyridine), 10.37 (d, 1H, N*H*-cycloheptyl). Anal. ($C_{20}H_{31}N_5O_4S$) C, H, N, S.

1-Cyclooctylamino-1-[(4′-cyclohexylamino)pyrid-3′-ylsulfonamido)]-2-nitroethylene (13). The title compound was obtained as previously described for the compound **17** by starting from **10** (1.0 g, 4.0 mmol) and 1-cyclooctylamino-1-methylsulfanyl-2-nitroethylene **8** (1.1 g, 4.7 mmol): yield 51% (0.90 g); mp 171−174 °C; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 1.12−1.97 (m, 26H, cyclohexyl and cyclooctyl), 6.69 (s, 1H, C*H*-NO_2), 7.19 (d, 1H, 5*H*-pyridine), 7.40 (d, 1H, pyridine-N*H*), 8.27 (d, 1H, 6*H*-pyridine), 8.58 (s, 1H, 2*H*-pyridine), 10.38 (d, 1H, N*H*-cyclooctyl). Anal. ($C_{21}H_{33}N_5O_4S$) C, H, N, S.

1-Cyclohexylamino-1-[(4′-cycloheptylamino)pyrid-3′-ylsulfonamido)]-2-nitroethylene (14). The title compound was obtained as previously described for the compound **17** by starting from **10** (1.0 g, 3.7 mmol) and 1-cyclohexylamino-1-methylsulfanyl-2-nitroethylene **8** (0.96 g, 4.5 mmol): yield 54% (0.88 g); mp 191−193 °C; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 1.02−1.95 (m, 24H, cycloheptyl and cyclohexyl), 6.71 (s, 1H, C*H*-NO_2), 7.15 (d, 1H, 5*H*-pyridine), 7.41 (d, 1H, pyridine-N*H*), 8.24 (d, 1H, 6*H*-pyridine), 8.57 (s, 1H, 2*H*-pyridine), 10.35 (d, 1H, N*H*-cyclohexyl). Anal. ($C_{20}H_{31}N_5O_4S$) C, H, N, S.

1-Cycloheptylamino-1-[(4′-cycloheptylamino)pyrid-3′-ylsulfonamido)]-2-nitroethylene (15). The title compound was obtained as previously described for the compound **17** by starting from **10** (1.0 g, 3.7 mmol) and 1-cycloheptylamino-1-methylsulfanyl-2-nitroethylene **8** (1.0 g, 4.3 mmol): yield 31% (0.55 g); mp 139−141 °C; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 1.10−1.98 (m, 26H, two cycloheptyl), 6.70 (s, 1H, C*H*-NO_2), 7.10 (d,

1H, 5*H*-pyridine), 7.40 (d, 1H, pyridine-N*H*), 8.26 (d, 1H, 6*H*-pyridine), 8.55 (s, 1H, 2*H*-pyridine), 10.39 (d, 1H, N*H*-cycloheptyl). Anal. ($C_{21}H_{33}N_5O_4S$) C, H, N, S.

1-Cyclooctylamino-1-[(4′-cycloheptylamino)pyrid-3′-ylsulfonamido)]-2-nitroethylene (16). The title compound was obtained as previously described for the compound **17** by starting from **10** (1.1 g, 4.1 mmol) and 1-cyclooctylamino-1-methylsulfanyl-2-nitroethylene **8** (1.2 g, 4.9 mmol): yield 63% (1.20 g); mp 177−179 °C; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 1.20−1.97 (m, 28H, cycloheptyl and cyclooctyl), 6.69 (s, 1H, C*H*-NO_2), 7.10 (d, 1H, 5*H*-pyridine), 7.28 (d, 1H, pyridine-N*H*), 8.28 (d, 1H, 6*H*-pyridine), 8.58 (s, 1H, 2*H*-pyridine), 10.39 (d, 1H, N*H*-cyclooctyl). Anal. ($C_{22}H_{35}N_5O_4S$) C, H, N, S.

1-Cycloheptylamino-1-[(4′-cyclooctylamino)pyrid-3′-ylsulfonamido)]-2-nitroethylene (18). The title compound was obtained as previously described for compound **17** by starting from **10** (1.0 g, 3.5 mmol) and 1-cycloheptylamino-1-methylsulfanyl-2-nitroethylene **8** (1.0 g, 4.3 mmol): yield 69% (1.14 g); mp 184−186 °C; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 1.22−1.95 (m, 28H, cyclooctyl and cycloheptyl), 6.70 (s, 1H, C*H*-NO_2), 7.09 (d, 1H, 5*H*-pyridine), 7.42 (d, 1H, pyridine-N*H*), 8.26 (d, 1H, 6*H*-pyridine), 8.57 (s, 1H, 2*H*-pyridine), 10.38 (d, 1H, N*H*-cycloheptyl). Anal. ($C_{22}H_{35}N_5O_4S$) C, H, N, S.

1-Cyclooctylamino-1-[(4′-cyclooctylamino)pyrid-3′-ylsulfonamido)]-2-nitroethylene (19). The title compound was obtained as previously described for the compound **17** by starting from **10** (1.0 g, 3.5 mmol) and 1-cyclooctylamino-1-methylsulfanyl-2-nitroethylene **8** (1.0 g, 4.1 mmol): yield 58% (1.00 g); mp 164−166 °C; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 1.10−1.93 (m, 30H, two cyclooctyl), 6.68 (s, 1H, C*H*-NO_2), 7.10 (d, 1H, 5*H*-pyridine), 7.44 (d, 1H, pyridine-N*H*), 8.27 (d, 1H, 6*H*-pyridine), 8.57 (s, 1H, 2*H*-pyridine), 10.39 (d, 1H, N*H*-cyclooctyl). Anal. ($C_{23}H_{37}N_5O_4S$) C, H, N, S.

1-Cyclohexylamino-1-[4′-(3″-methylphenylamino)pyrid-3′-ylsulfonamido]-2-nitroethylene (20). The title compound was obtained as previously described for the compound **17** by starting from **10** (0.5 g, 1.9 mmol) and 1-cyclohexylamino-1-methylsulfanyl-2-nitroethylene **8** (0.5 g, 2.3 mmol): yield 25% (0.21 g); mp 144−146 °C; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 0.98−1.85 (m, 11H, cyclohexyl), 2.34 (s, 3H, aryl-CH_3), 6.87 (s, 1H, C*H*-NO_2), 7.01−7.45 (m, 5H, phenyl and 5*H*-pyridine), 8.26 (d, 1H, 6*H*-pyridine), 8.59 (s, 1H, 2*H*-pyridine), 9.45 (b, 1H, pyridine-N*H*), 10.33 (d, 1H, N*H*-cyclohexyl). Anal. ($C_{20}H_{25}N_5O_4S$) C, H, N, S.

1-Isopropylamino-1-[(4′-ethylamino)pyrid-3′-ylsulfonamido]-2-nitroethylene (21). The title compound was obtained as previously described for compound **17** by starting from **10** (0.7 g, 3.5 mmol) and 1-isopropylamino-1-methylsulfanyl-2-nitroethylene **8** (0.7 g, 4.0 mmol): yield 37% (0.41 g); mp 163−164 °C; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 1.04 (2s, 6H, C(CH_3)$_2$), 1.23 (1s, 3H, CH_3), 3.40 (m, 2H, CH_2), 4.01 (m, 1H, C*H*<), 6.70 (s, 1H, C*H*-NO_2), 7.08 (d, 1H, 5*H*-pyridine), 7.70 (m, 1H, pyridine-N*H*), 8.21 (d, 1H, 6*H*-pyridine), 8.57 (s, 1H, 2*H*-pyridine), 10.10 (d, 1H, N*H*-isopropyl). Anal. ($C_{12}H_{19}N_5O_4S$) C, H, N, S.

1-Isopropylamino-1-[(4′-cyclopentylamino)pyrid-3′-ylsulfonamido)]-2-nitroethylene (22). The title compound was obtained as previously described for compound **17** by starting from **10** (0.7 g, 2.9 mmol) and 1-isopropylamino-1-methylsulfanyl-2-nitroethylene **8** (0.6 g, 3.4 mmol): yield 25% (0.27 g); mp 172−173 °C; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 1.10 (2s, 6H, C(CH_3)$_2$), 1.23−1.90 (m, 9H, cyclopentyl), 2.05 (m, 1H, C*H*<), 6.75 (s, 1H, C*H*-NO_2), 7.20 (d, 1H, 5*H*-pyridine), 7.50 (d, 1H, pyridine-N*H*), 8.25 (d, 1H, 6*H*-pyridine), 8.58 (s, 1H, 2*H*-pyridine), 10.15 (d, 1H, N*H*-cyclopentyl). Anal. ($C_{15}H_{23}N_5O_4S$) C, H, N, S.

1-Isopropylamino-1-[4′-(3″-methylphenylamino)pyrid-3′-ylsulfonamido]-2-nitroethylene (23). The sodium salt of 4-(3′-methylphenylamino)pyrid-3-ylsulfonamide **10**[5,15] (1.09 g, 3.8 mmol) and 0.75 g (4.3 mmol) of 1-isopropylamino-1-methylsulfanyl-2-nitroethylene **8** were dissolved in a mixture of 1,4-dioxane (3 mL) and *N*,*N*-dimethylformamide (2 mL) and refluxed for 4 h. After evaporation of solvents under reduced pressure the residue was dissolved in water (50 mL) and 2.5 N NaOH (2 mL). The solution was extracted three times with diethyl ether (50 mL) and adjusted to pH 3 with dilute HCl. The precipitate was collected by filtration, washed with water, dried, and crystallized from absolute ethanol (40 mL) to afford the title compound (0.79 g, 53%): mp 168−169 °C; IR (KBr) 1570 (NO_2) cm^{-1}; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 1.05 (2s, 6H, C(CH_3)$_2$), 2.32 (s, 3H, aryl-CH_3), 4.02 (m, 1H, C*H*(CH_3)$_2$), 6.80 (s, 1H, C*H*-NO_2), 6.95−7.50 (m, 5H, phenyl and 5*H*-pyridine), 8.18 (d, 1H, 6*H*-pyridine), 8.63 (s, 1H, 2*H*-pyridine), 9.38 (br s, 1H, pyridine-N*H*), 10.15 (d, 1H, N*H*-isopropyl). Anal. ($C_{17}H_{21}N_5O_4S$) C, H, N, S.

Lipophilicity. The lipophilicity of standard molecules (**21**−**23** and **11**) listed in Table 2 was expressed as the logarithm of the partition coefficient (log *P*) in 1-octanol/phosphate buffer pH 7.40 by using the shake-flask technique.[24] A RP-HPLC system was also loaded to determine their log *k*′ values defined as $(t_r - t_o)/t_o$ where t_r is the sample retention time and t_o the NO_3^- retention time detected at 254 nm.[5] The stationary phase was an octadecyl column (Lichrospher 100RP-18, 15 cm length, 5 μm porosity), and the mobile phase was a mixture of phosphate buffer pH 7.40 with 2-propanol (70:30 v/v). A correlation curve was calculated from log *P* and log *k*′ values of the standards: log $P = (2.662 \log k') + 1.206$; $n = 4$; $r = 0.997$. By using the correlation curve, the log *P* values of other compounds (Table 1) were then obtained by extrapolation of their log *k*′ values evaluated by chromatography.

X-ray Crystallography. Crystals of **17**, $C_{21}H_{33}N_5O_4S$, $M_r = 451.6$, were obtained by slow evaporation of a concentrated solution in EtOH/MeOH and studied by crystallography: monoclinic, $P2_1/c$, $a = 10.606(2)$ Å, $b = 13.231(3)$ Å, $c = 16.776(2)$ Å, $\beta = 102.67(2)°$, $V = 2296.8$ Å^3, $Z = 4$, $\mu = 15.6$ cm^{-1}, $D_x = 1.3060(5)$ g cm^{-3}, λ(Cu Kα) = 1.54178 Å, $F(000) = 968.0$, $T = 290$ K, 4503 unique reflections, R1 = 0.0671 for 1817 $F_o > 4\sigma(F_o)$ and wR2 = 0.1677, GOF = S = 0.835.

Molecular Modeling. Internal geometries were optimized by molecular mechanics (MM) with the Discover program (MSI, San Diego) using the cff91 Force Field. Those parameters reproduce well both ab initio optimized (LCAO-MO, 3-21G*) and experimental (X-ray) geometries. Calculations were performed on the neutral (nonzwitterionic) form of **17**. Theoretical values of the starting geometries for the α, β, γ, and δ conformations were taken from the literature.[5] Those geometries were optimized to a final RMS deviation of 0.01 kcal/mol.

Maximal Electroshock Seizures Test. Each compound was intraperitoneally injected to OF1 male mice (22−25 g; Iffa-Credo, Les Oncins, France) at a dose volume of 10 mL/kg. At the desired time, an electrical stimulus (50 mA; 60 Hz) was delivered for 0.2 s through corneal electrodes. Protection against seizures was defined as the abolition of the hind limb tonic extension.[9,19] The preliminary screening (Table 1) was conducted with groups of six or seven mice at an intraperitoneal dose of 30 mg/kg. The electroshock was applied at 0.5 or 3 h after injection. The time-course activity of **17** was measured after injection of 30 mg/kg. The effective dose protecting 50% of mice (ED_{50}) was determined by treating groups of eight mice with varying doses at the time of peak effect. The ED_{50} and 95% confidence intervals were computed by using the method of Litchfield and Wilcoxon.[25]

Chemically Induced Seizures. Pentetrazole (85 mg/kg), strychnine (1.20 mg/kg), picrotoxin (3.15 mg/kg), and NMLDA (340 mg/kg) were dissolved in 0.9% NaCl (10 mg/kg). Bicuculline (2.70 mg/kg) was dissolved in warmed 0.1 N HCl and 0.9% NaCl (10 mL/kg) and used within 30 min after preparation. The proconvulsive chemicals (pentetrazole, strychnine, bicuculline, picrotoxin, and NMLDA) were administered subcutaneously at the time of peak effect obtained in the MES test and the anticonvulsant activity measured as described.[9,19,26] The ED_{50} and 95% confidence intervals were computed by using the method of Litchfield and Wilcoxon.[25]

Neurotoxicity. Neurotoxicity was defined as the failure of an intraperitoneally treated mouse to maintain its equilibrium on rod rotating (6 rpm).[9,20] The determination of the

median toxic dose (TD_{50}) and confidence intervals were calculated as mentioned previously.[25]

Diuresis. Each drug or vehicle (NaCl 0.9% with methocel 0.1%) was orally administered in male rats (Wistar 193−229 g) in a dose volume of 40 mL/kg. Three rats housed in the same metabolism cage received 30 mg/kg of each drug, and urine was collected over 4 h. The results were expressed in mL/kg of urine excreted over this period.

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Supporting Information Available: X-ray crystallographic data, including positional parameters, bond distances, bond angles, torsion angles, and anisotropic displacement parameter expressions for **17** (13 pages). Ordering information is given on any current masthead page.

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