Pyrrolo[2,1-c][1,4]benzothiazines: Synthesis, Structure–Activity Relationships, Molecular Modeling Studies, and Cardiovascular Activity†

Giuseppe Campiani,‡ Antonio Garofalo,‡ Isabella Fiorini,‡ Maurizio Botta,‡ Vito Nacci,*,‡ Andrea Tafi,‡ Alberto Chiarini,|| Roberta Budriesi,|| Giancarlo Bruni,§ and Maria R. Romeo§

Dipartimento Farmaco Chimico Tecnologico, Banchi di Sotto 55, and Istituto di Farmacologia, Via delle Scotte 6, Universita' di Siena, 53100 Siena, Italy, and Dipartimento di Scienze Farmaceutiche, Via Belmeloro 6, Universita' di Bologna, 40126 Bologna, Italy

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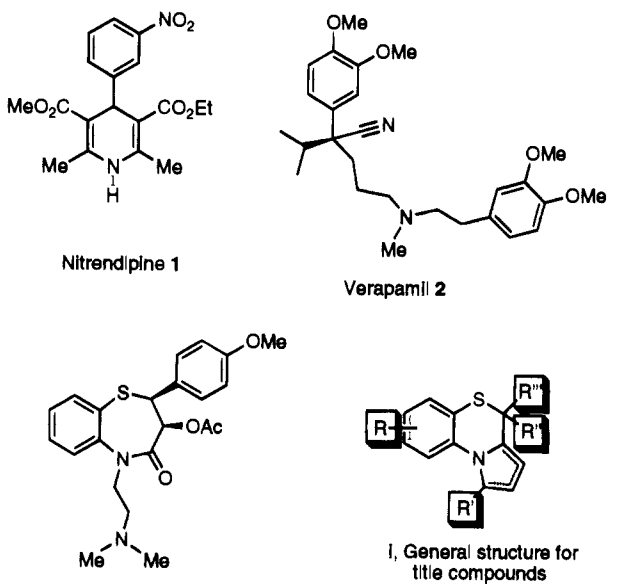
The synthesis and pharmacological evaluation of a series of pyrrolo[1,4]benzothiazine derivatives are described. These compounds, related to diltiazem, have been shown to be representative of a novel series of calcium channel antagonists. The IC_{50}s for inhibition of [^{3}H]nitrendipine binding calculated by radioreceptor assay on rat cortex and rat heart homogenates showed that some of the described compounds possess an affinity equal to or higher than those of the reference calcium antagonists verapamil and *cis*-(+)-diltiazem. Furthermore, the alteration of the benzothiazepinone system of diltiazem to the pyrrolo[1,4]benzothiazine system of the title compounds resulted in a clear-cut selectivity for cardiac over vascular tissue, as shown in functional studies. In fact comparison of calcium antagonist activity on guinea pig aorta strips with the negative inotropic activity, determined by using an isolated guinea pig left atrium, revealed that the compounds examined displayed higher selectivity than the reference standard, within a wide variation of data. A number of structure–activity relationship trends have been identified, and possible explanation is advanced in order to account for the observed differences in selectivity. Prerequisite for in vitro calcium channel-blocking activity is the presence of two pharmacophores, namely, the substitution at C-4 and the substitution on the pyrrole ring. Two of the tested compounds, **8b** and **28a**, were identified as potent calcium antagonists selective for cardiac over vascular tissue.

Introduction

Calcium antagonists are a well-established therapeutic class of agents which now find a wide use in the treatment of angina and hypertension.[1] Calcium entry blockers (CEBs) are structurally classified in two large groups: the dihydropyridines (DHPs), represented by nitrendipine (**1**), and the non-dihydropyridines, represented by verapamil (**2**) and diltiazem (**3**)[2] (Chart **1**). These compounds exert their pharmacological activity by selectively inhibiting the magnitude of Ca^{2+} current through the L-type calcium channels.[3,4] After the introduction of CEBs into clinical practice, many compounds possessing calcium antagonist activity have been reported, most of them being structurally related to the DHPs, the most widely studied class of CEBs. Several DHPs analogues are currently under clinical trials. A further group of active compounds are the phenylalkylamines, represented by verapamil, the binding site of which is allosterically coupled to that of DHPs.[5,6] This class of CEBs is characterized by lower pharmacological selectivity.[7] In fact they possess a variety of actions in addition to their ability to reduce the calcium current through the slow channels. A structurally distinct class of CEBs is the 1,5-benzothiazepines, represented by diltiazem. Similarly to phenylalkylamines, this compound binds to a site that is allosterically coupled to the binding site of DHPs in the calcium channel protein.[5,8] Reducing the calcium current magnitude through the slow channels, CEBs induce inhibition of the cardiac contractile apparatus as well, resulting in a negative inotropic effect.[9] Accordingly, diltiazem is a valuable therapeutic agent for the treatment of angina pectoris and hypertension.[10] Although the structure–activity relationships (SARs) of DHPs and verapamil derivatives have been widely reviewed,[11] very few effective calcium antagonists related to **3** have so far been reported. Therefore, little information is available concerning SARs in this class of CEBs. Recently, Inoue *et al.*[12] described a new series of diltiazem derivatives with a similar pharmacological profile as **3** but with more potent antihypertensive activity. Additionally,

Chart 1



† In honor of Koji Nakanishi's 70th birthday.
* To whom correspondence should be addressed.
‡ Dipartimento Farmaco Chimico Tecnologico, Universita' di Siena.
§ Istituto di Farmacologia, Universita' di Siena.
|| Universita' di Bologna.
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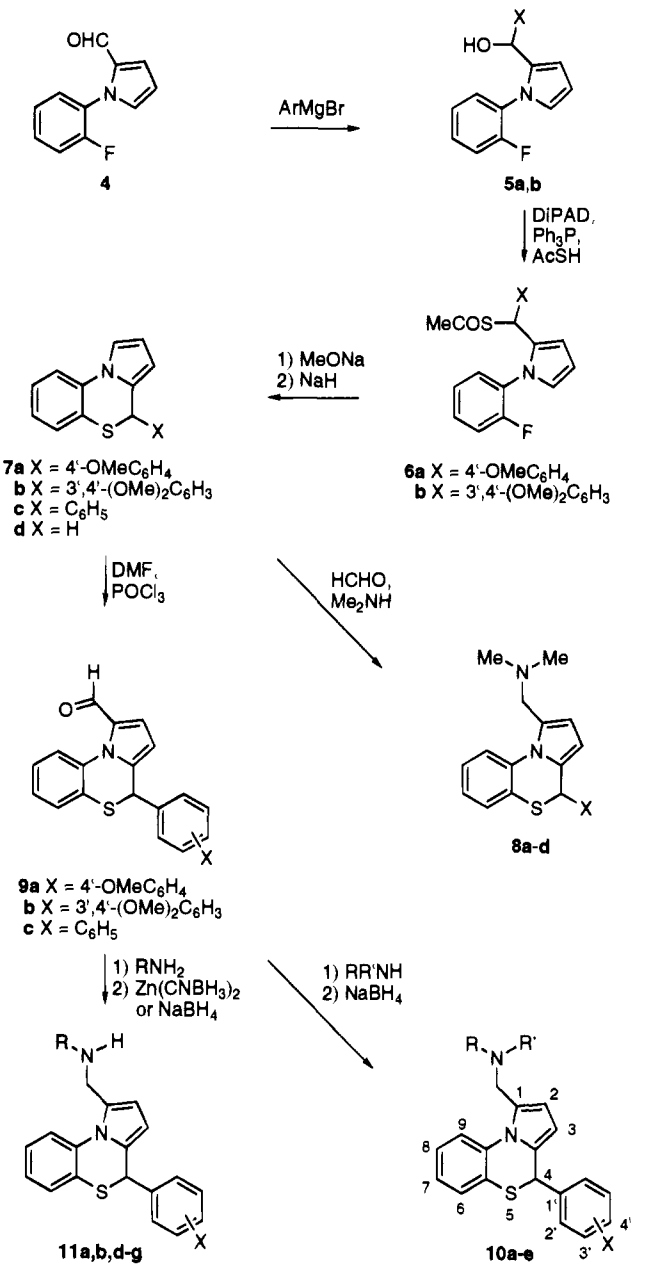
substituted benzazepinone derivatives of **3**, recently described,[13] exhibited longer acting antihypertensive activity than that of 8-Cl-diltiazem reported by Inoue.[14] Anyway, the structural alteration of diltiazem has involved only the fused aromatic ring and the sulfur atom, and in both cases no selectivity between cardiac and vascular tissue was clearly detected. In our previous paper[15] we reported the synthesis of a prototypic compound of a novel series of potential CEBs, related to diltiazem, in which the contraction of the seven-member ring together with the bioisosteric substitution of the amide moiety with a lipophilic pyrrole ring was reported. In the present paper we describe further developments in our research on novel calcium antagonists. We detail herein the synthesis of pyrrolobenzothiazine derivatives **I** (Chart 1) and their SARs for calcium antagonist activity associated with variation of the substituents on the heterocyclic system. Substitutions on the tricyclic skeleton together with alteration of the benzothiazepinone system of **3** to pyrrolobenzothiazine allowed us to identify a novel class of calcium antagonists selective for cardiac over vascular tissue.

Chemistry

The key intermediate 4-substituted pyrrolo[2,1-*c*][1,4]-benzothiazine derivatives **7** have been prepared following an already published procedure.[16] The 1-(*o*-fluorophenyl)pyrrole 2-aldehyde (**4**) was reacted with the appropriate Grignard reagent to provide the corresponding alcohols **5**. Because of their substantial instability, the alcohols were immediately converted into the thiolacetates **6** by a modified Mitsunobu procedure using triphenylphosphine, diisopropyl azodicarboxylate (DiPAD), and thiolacetic acid. Methanolysis followed by in situ cyclization by an intramolecular aromatic nucleophilic displacement of the fluorine atom provided the tricyclic compounds **7** in good overall yields. Mannich condensation on the pyrrole ring of **7** afforded the (dimethylamino)methyl derivatives **8**, while the introduction of a formyl group at C-1, via Vilsmeyer–Haack formylation, gave valuable intermediates **9** for introducing (monoalkylamino) and (dialkylamino)methyl substituents at C-1. In fact, aldehydes **9** underwent facile reaction with a wide variety of primary and secondary amines to afford imines or immonium salts,[17] which were subjected to reduction by means of sodium borohydride or zinc cyanoborohydride[18] to give tertiary and secondary amine derivatives **10 and 11**, respectively (see Scheme 1 and Table 1). Afterward, to assess the role played by the sulfur atom in the heterocyclic ring system, we decided to investigate the benzoxazine analogue **13**. Starting from the alcohol **5a**, intramolecular aromatic nucleophilic displacement of the fluorine provided the benzoxazine **12** which was functionalized with the (dimethylamino)methyl side chain in the usual way (**13**) (see Scheme 2 and Table 1).

In order to better explore the possible binding sites of these compounds, we decided to investigate a synthetic pathway to introduce additional substituents at C-4 and on the fused phenyl ring. These structural modifications were in part dictated by the constraints of synthesis in combination with the results of the modeling studies. Accordingly, we chose to introduce an ester group and an extra phenyl ring at C-4 to evaluate the effect of the hindrance and of extra electrostatic interactions with the binding site. Thus

Scheme 1



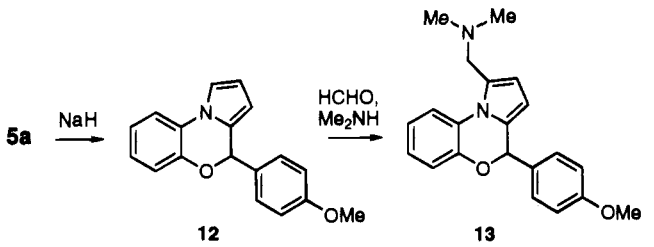
the key thiolactones **18** were synthesized starting from [2'-(methylthio)phenyl]pyrroles **14**[19] and disulfides **19**[20] as outlined in Schemes 3 and 4. To obtain unsubstituted and 8-trifluoromethyl thiolactones **18a**[21] and **18b**, the key intermediate pyrrole-2-carboxylic acids **16a,b** were synthesized via trichloroacetylation of the pyrrole ring[22] of **14a,b** followed by alcoholysis of the trichloroacetyl group (**15a,b**) and hydrolysis of the ester functions (**16a,b**). Chlorination with thionyl chloride of acids **16a,b** followed by intramolecular cyclization under Friedel–Crafts conditions provided the tricyclic compounds **18a,b**, with elimination of methyl chloride. This route gave us the opportunity to investigate a novel intramolecular cyclization to obtain pyrrolobenzothiazinones. On the other hand, this cyclization method proved to be unsuccessful in the case of chloro-substituted phenylpyrroles. Accordingly, we synthesized thiolactones **18c,d** by the action of anhydrous zinc chloride[21] on thiocarbonates **20a,b** which were in turn prepared by reductive alkylation of disulfides **19a,b** with sodium borohydride and ethyl chloroformate (Table 2).

Table 1. Physical Data for Compounds **5**–**13**

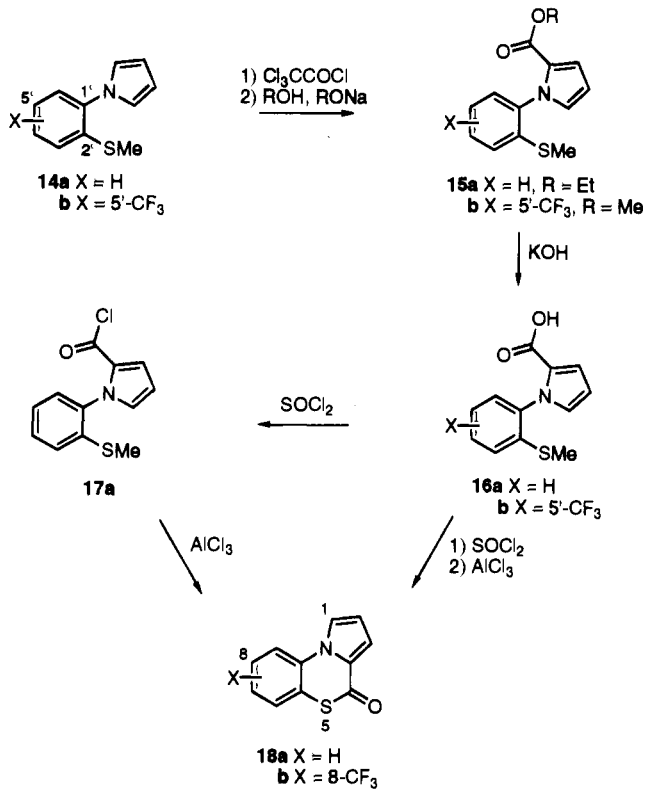
compd	X	R	R′	yield[a] (%)	mp (°C)	recryst solvent	formula	anal.[b]
5a	4′-OMeC$_6$H$_4$			75	*c*			
5b	2′,4′-(OMe)$_2$C$_6$H$_3$			78	*c*			
6a	4′-OMeC$_6$H$_4$			74			$C_{20}H_{18}FNO_2S$	C,H,N
6b	2′,4′-(OMe)$_2$C$_6$H$_3$			68			$C_{21}H_{20}FNO_3S$	C,H,N
7a	4′-OMeC$_6$H$_4$			79	150–151	petroleum ether (60–80 °C)	$C_{18}H_{15}NOS$	C,H,N
7b	2′,4′-(OMe)$_2$C$_6$H$_3$			89	172–173	petroleum ether (60–80 °C)	$C_{25}H_{21}NO_2S$	C,H,N
7c[d]	C$_6$H$_5$							
7d[d]	H							
8a	4′-OMeC$_6$H$_4$			88	174–175	EtOAc	$C_{21}H_{22}N_2OS$	C,H,N
8b	2′,4′-(OMe)$_2$C$_6$H$_3$			87	171–172	EtOAc	$C_{22}H_{24}N_2O_2S$	C,H,N
8c	C$_6$H$_5$			77	141–142	EtOAc	$C_{20}H_{20}N_2S$	C,H,N
8d	H			68	118–119	EtOAc/hexane	$C_{14}H_{16}N_2S$	C,H,N
9a	4′-OMe			84	158–160	MeOH	$C_{19}H_{15}NO_2S$	C,H,N
9b	2′,4′-(OMe)$_2$			91	147–148	MeOH	$C_{20}H_{17}NO_3S$	C,H,N
9c	H			92	168–169	MeOH	$C_{18}H_{13}NOS$	C,H,N
10a	4′-OMe	O(CH$_2$CH$_2$)$_2$		93	180–181	EtOAc/hexane	$C_{23}H_{24}N_2O_2S$	C,H,N
10b	2′,4′-(OMe)$_2$	O(CH$_2$CH$_2$)$_2$		76	166–167	EtOAc/hexane	$C_{24}H_{26}N_2O_3S$	C,H,N
10c	H	3,4-(OMe)$_2$C$_6$H$_3$CH$_2$CH$_2$	Me	80			$C_{29}H_{31}ClN_2O_2S$[e]	C,H,N
10d	4′-OMe	3,4-(OMe)$_2$C$_6$H$_3$CH$_2$CH$_2$	Me	88			$C_{30}H_{33}ClN_2O_3S$[e]	C,H,N
10e	2′,4′-(OMe)$_2$	3,4-(OMe)$_2$C$_6$H$_3$CH$_2$CH$_2$	Me	95			$C_{31}H_{35}ClN_2O_4S$[e]	C,H,N
11a	4′-OMe	*s*-C$_4$H$_9$		94	119–120	MeOH	$C_{23}H_{26}N_2OS$	C,H,N
11b	4′-OMe	4-FC$_6$H$_4$		59	176–177	*i*-PrOH	$C_{25}H_{21}FN_2OS$	C,H,N
11d	4′-OMe	3,4-(OMe)$_2$C$_6$H$_3$CH$_2$CH$_2$		79	132–133	EtOAc	$C_{29}H_{30}N_2O_3S$	C,H,N
11e	2′,4′-(OMe)$_2$	*s*-C$_4$H$_9$		73	128–130	MeOH	$C_{24}H_{28}N_2O_2S$	C,H,N
11f	2′,4′-(OMe)$_2$	4-FC$_6$H$_4$		67	172–173	*i*-PrOH	$C_{26}H_{23}FN_2O_2S$	C,H,N
11g	2′,4′-(OMe)$_2$	3,4-(OMe)$_2$C$_6$H$_3$CH$_2$CH$_2$		73	124–125	EtOAc	$C_{30}H_{32}N_2O_4S$	C,H,N
12				55	98–99	hexane	$C_{18}H_{15}NO_2$	C,H,N
13				91	153–154	petroleum ether (60–80 °C)	$C_{21}H_{22}N_2O_2$	C,H,N

[a] Yields refer to isolated and purified materials. [b] All the compounds were analyzed within ±0.4% of the theoretical values. [c] Not crystallized and used crude. [d] See ref 16. [e] As hydrochloride.

Scheme 2



Scheme 3



The thiolactones were found to be appropriate precursors of 4-acetoxy-4-aryl and 4,4-diaryl derivatives **22** and **23** by using Grignard reaction protocols (see Scheme 5). The behavior of thiolactones with nucleophiles has not been widely investigated, and one of the examples cited[23] is the addition of phenylmagnesium bromide to give unstable adducts which quickly underwent a disproportionation and rearrangement to several final products. This did not occur with the adducts from 4-oxo-4*H*-pyrrolobenzothiazines **18**. In fact, Grignard reaction performed at room temperature or 40 °C with arylmagnesium bromides in a 1:2 molar ratio provided 4-aryl-4-hydroxy derivatives **21a**–**g**: In this case, the carbonyl group of the thiolactones behaved essentially like an isolated ketone, providing little amount of 4,4-diaryl compounds **23** as byproducts (see Scheme 5). On the contrary, the nucleophilic attack of the Grignard reagents carried out at reflux, by adding thiolactones **18** to the nucleophile, in the same molar ratio, provided 4,4-diaryl derivatives **23** and tertiary alcohols **21** in 50–70% and 20–30% yields, respectively. The formation of diaryl compounds may be explained by the process involving the partial intermediate formation of the carbocation (stabilized owing to the high nucleophilicity of sulfur), which combines another molecule of nucleophile. Finally, after examination of several esterification methods, we found that treatment of alcohols **21** with acetyl bromide in the presence of triethylamine at 80 °C afforded the acetyl derivatives **22** in 60–70% yield. Unfortunately, attempts to introduce the (dimethylamino)methyl side chain at C-1 of **22** were unsuccessful. On the contrary, 4,4-diaryl compounds were easily functionalized to give amines **24a**–**d** (Table 3).

Replacement of the lipophilic pyrrole portion with a pyrrolidinone ring was further investigated by the

Scheme 4

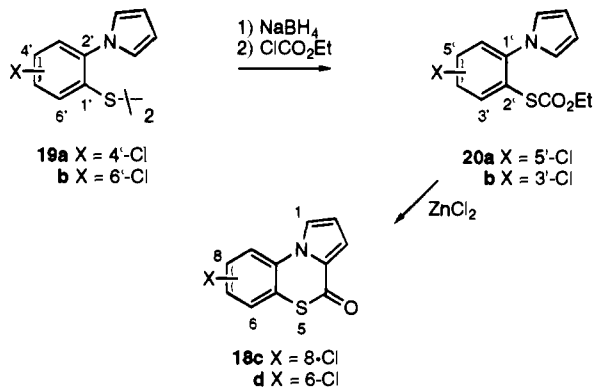


Table 2. Physical Data for Compounds 15–20

compd	X	yield[a] (%)	recryst solvent	mp (°C)	formula	anal.[b]
15a	H	84	hexane	50–53	$C_{14}H_{15}NO_2S$	C,H,N,S
15b	5-CF_3	79			$C_{14}H_{12}F_3NO_2S$	C,H,N
16a	H	94	benzene	165–166	$C_{12}H_{11}NO_2S$	C,H,N,S
16b	5-CF_3	93	EtOAc	171–173	$C_{13}H_{10}F_3NO_2S$	C,H,N
17a	H	67	ligroin	98–100	$C_{12}H_{10}ClNOS$	C,H,N,S,Cl
18a	H	46		156–157[c]		
18b	8-CF_3	53	EtOH	162–163	$C_{12}H_6F_3NOS$	C,H,N
18c	8-Cl	82	EtOAc	221–223	$C_{11}H_6ClNOS$	C,H,N
18d	6-Cl	62	EtOH	188–189	$C_{11}H_6ClNOS$	C,H,N
20a	5-Cl	66			$C_{13}H_{12}ClNO_2S$	C,H,N
20b	3-Cl	70			$C_{13}H_{12}ClNO_2S$	C,H,N

[a] Yields refer to isolated and purified materials. [b] All the compounds were analyzed within ±0.4% of the theoretical values. [c] See ref 21.

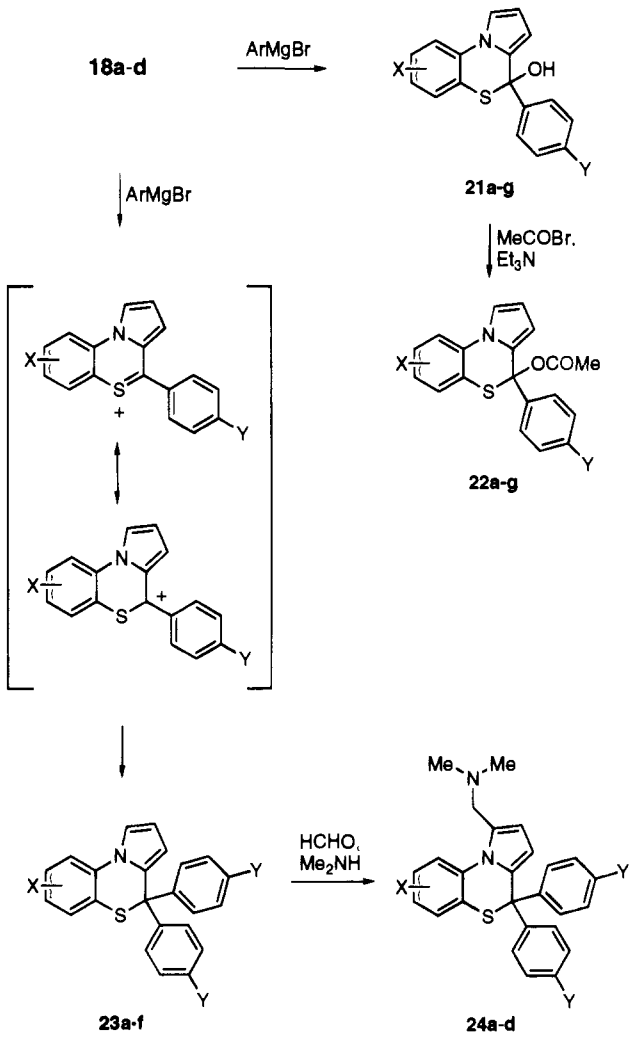
achievement of **28**. Our retrosynthetic analysis of compounds **28** is shown in Chart 2. Disulfide **25**[24] could serve as an appropriate precursor of the hemithioacetal **26** which would be subjected to chlorination followed by Friedel–Crafts reaction. Thus, the main task to be accomplished in this synthesis was the development of a method for the cyclization reaction to **26**. As highlighted in Scheme 6, diastereoselective reductive cyclization of **25** with lithium triethylborohydride[25] afforded the hemithioacetal **26** in 79% yield (85% de, and the separation of the diastereoisomers could be accomplished by fractional crystallization or flash chromatography). Chlorination of **26** with thionyl chloride followed by Friedel–Crafts reaction[26] provided the target compounds **28a,b** in 67% and 84% yields, respectively (Table 3). At this stage the diastereoisomers were separated by fractional crystallization. On the basis of ^{1}H NMR data, the major diastereoisomer showed a *trans* configuration for protons at C-3a and C-4 ($J_{H4\text{-}H3a}$ = 9.9 Hz, see the Experimental Section).

Results and Discussion

In Vitro SAR Study. A series of pyrrolobenzothiazine derivatives was examined for their calcium antagonist activity which was measured in rat cortex and heart homogenates[3,27] by displacing [^{3}H]nitrendipine, while in functional studies their inotropic, chronotropic, and vasorelaxing effects were evaluated, as reported in the Experimental Section. Compounds described herein were obtained in the racemic form. Data for diltiazem and verapamil are included for comparison in Tables 4–8.

1,4-Disubstituted Pyrrolobenzothiazines. In this benzothiazine series the introduction at C-4 of a substituted phenyl ring proved to be critical for activity.

Scheme 5



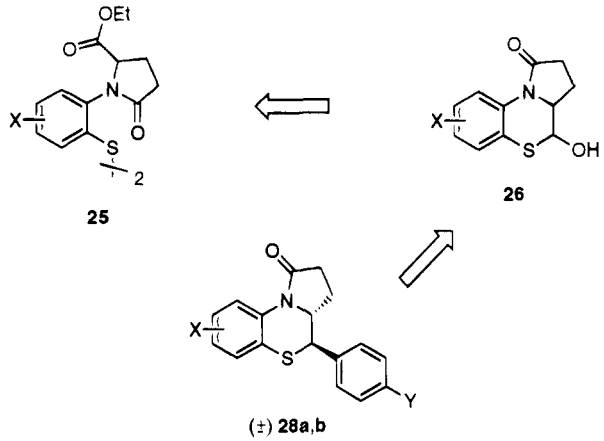
In radioligand-binding studies, the 4-phenyl derivative **8c** as well as the corresponding unsubstituted **8d** showed a large drop in the in vitro activity when compared with the 4′-methoxyphenyl **8a** or the 2′,4′-dimethoxyphenyl analogue **8b**. The alteration of the substituent in the phenyl ring revealed that compounds monomethoxy substituted at C-4′ showed higher affinity than those disubstituted at C-2′ and C-4′ (**8a** > **8b**, **10d** > **10e**, and **11a** > **11e**). Among these compounds, **8a** displayed the highest affinity in radioreceptor assay (Table 4). The further essential pharmacophore is the basic side chain at C-**1** on the pyrrole ring. In fact, pyrrolobenzothiazine **7a**, in which the side chain is lacking, did not show any affinity for the receptor (Table 6). Different amine substituents were introduced, keeping the same distance (two carbon atoms) between the two nitrogen atoms present in these heterocycles. The greatest antagonist activity was found for **8a** which possesses a (dimethylamino)methyl side chain at C-**1**. Modification on the basic character of the amine gave us additional supporting evidence for the critical contribution of the amino group.[28] The interaction with the receptor is linked much more to the basicity of the nitrogen than to the steric perturbation. In fact, while the arylamino derivatives **11b,f** show a large drop in affinity, aralkylamino compounds **10c–e** and **11d,g** maintained the biological activity (Table 4). In the aralkyl series the presence of a tertiary amino group led to more active compounds (**10c–e** vs **11d**). The

Table 3. Physical Data for Compounds 21–28

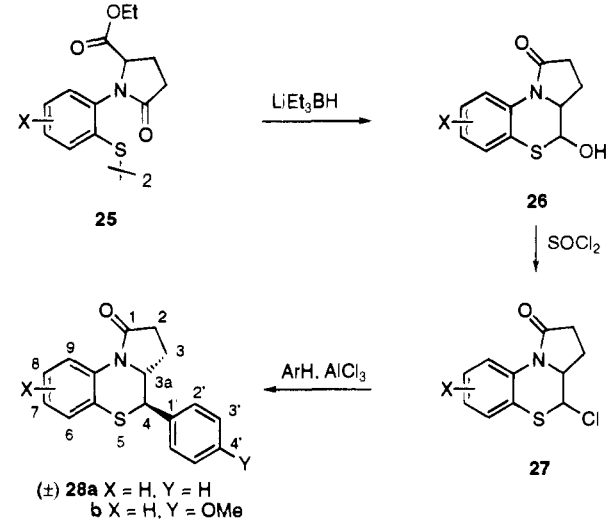
compd	X	Y	yield[a] (%)	recryst solvent	mp (°C)	formula	anal.[b]
21a	H	H	82			$C_{17}H_{13}NOS$	C,H,N
21b	H	4'-OMe	67			$C_{18}H_{15}NO_2S$	C,H,N
21c	8-Cl	H	63			$C_{17}H_{12}ClNOS$	C,H,N
21d	8-Cl	4'-OMe	61			$C_{18}H_{14}ClNO_2S$	C,H,N
21e	8-CF_3	H	64			$C_{18}H_{12}F_3NOS$	C,H,N
21f	8-CF_3	4'-OMe	73			$C_{19}H_{14}F_3NO_2S$	C,H,N
21g	6-Cl	H	59			$C_{17}H_{12}ClNOS$	C,H,N
22a	H	H	61	cyclohexane	69–71	$C_{19}H_{15}NO_2S$	C,H,N
22b	H	4'-OMe	56	hexanes	120–121	$C_{20}H_{17}NO_3S$	C,H,N
22c	8-Cl	H	63		c	$C_{19}H_{14}ClNO_2S$	C,H,N
22d	8-Cl	4'-OMe	71		c	$C_{20}H_{16}ClNO_3S$	C,H,N
22e	8-CF_3	H	71	EtOAc/hexanes	97–98	$C_{20}H_{14}F_3NO_2S$	C,H,N
22f	8-CF_3	4'-OMe	69	EtOAc/hexanes	143–144	$C_{21}H_{16}F_3NO_3S$	C,H,N
22g	6-Cl	H	67	cyclohexane	80–81	$C_{19}H_{14}ClNO_2S$	C,H,N
23a	H	H	77	hexanes	138–139	$C_{23}H_{17}NS$	C,H,N
23b	H	4'-OMe	70	petroleum ether (60–80 °C)	172–173	$C_{25}H_{21}NO_2S$	C,H,N
23c	8-CF_3	H	73	hexanes	157–159	$C_{24}H_{16}F_3NS$	C,H,N
23d	8-CF_3	4'-OMe	68	hexanes	117–118	$C_{26}H_{20}F_3NO_2S$	C,H,N
23e	8-Cl	H	56	petroleum ether (60–80 °C)	124–125	$C_{25}H_{20}ClNO_2S$	C,H,N
23f	6-Cl	H	58	hexanes	187–189	$C_{23}H_{16}ClNS$	C,H,N
24a	H	H	84	EtOAc	176–177	$C_{26}H_{24}N_2S$	C,H,N
24b	H	4'-OMe	68	EtOAc/hexanes	147–149	$C_{28}H_{28}N_2O_2S$	C,H,N
24c	8-CF_3	H	89	EtOAc	118–119	$C_{27}H_{23}F_3N_2S$	C,H,N
24d	6-Cl	H	86	EtOAc	196–197	$C_{26}H_{23}ClN_2S$	C,H,N
26	H		79	EtOAc/cyclohexane	189–190	$C_{11}H_{11}NO_2S$	C,H,N
27	H		77		d		
28a	H	H	84	Et_2O	154–155	$C_{17}H_{15}NOS$	C,H,N
28b	H	4'-OMe	67	i-Pr_2O	149–150	$C_{18}H_{17}NO_2S$	C,H,N

[a] Yields refer to isolated and purified materials. [b] All the compounds were analyzed wtihin ±0.4% of the theoretical values. [c] Amorphous solid. [d] Not crystallized and used crude.

Chart 2



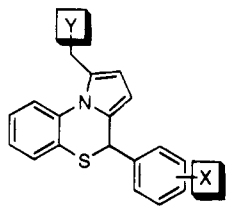
Scheme 6



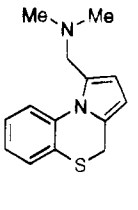
introduction of a morpholinomethyl chain resulted in a loss of activity (**10a,b**). Moreover, the role played by the sulfur atom in the interaction with the receptor protein was evaluated; replacement of the bridged sulfur atom with a bioisosteric oxygen atom resulted in a decrease in affinity (**13** vs **8a**, Table 4). These data support the conclusion that a 4-aryl substitution at C-4, the basic side chain, and the sulfur-bridged atom are critical for the binding interaction at the receptor.

Effect of Double Substitution at C-4. We have also synthesized a series of C-4 disubstituted pyrrolobenzothiazines in which lipophilic, electronic, and steric parameters have been varied. Since C-1-unsubstituted 4-arylpyrrolobenzothiazine **7a** proved to be inactive (Table 6), it was somewhat surprising to find that introduction of an acetoxy group at C-4 provided relatively active compounds (**22**) (see Table 5). In this series the calcium antagonist activity is increased by the presence of a hydrogen bond acceptor (OMe) at C-4' (**22b**). Furthermore, we investigated the effect of substitution in the fused aryl ring. In the 4-acetoxy-4-phenyl analogues, the electron-withdrawing trifluoromethyl group at C-8 led to an active compound (**22e** vs **22a**), while the 6-chloro analogue **22g** did not differ greatly in affinity from the unsubstituted **22a**. The enhanced activity of **22e** containing an electron-withdrawing group at C-8 may indicate a preference at the receptor for a negative dipole at position 8. Unfortunately, the synthesis of the 4'-OMe derivatives of **22e,g**, together with the introduction of (dialkylamino)methyl side chains on **22**, in order to evaluate the contribution of a basic center in the interaction with the receptor protein, was unsuccessful. The introduction of an extra aryl group at C-4 (**23**) revealed that compounds with large lipophilic groups are more potent in vitro than those monosubstituted (**23a** vs **7a**) (see Table 6). These data prompted us to investigate more fully this series of diarylpyrrolobenzothiazines. A substituent on the fused phenyl ring and a basic side chain on the pyrrole

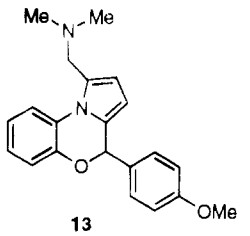
Table 4. Receptor-Binding Affinity: Effect of Substitution at C-1 and on the Phenyl Ring at C-4 and Effect of Replacement of the Bridged Sulfur Atom



8a-c, 10, 11



8d

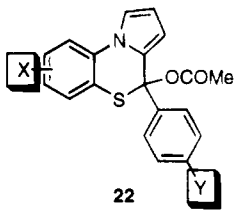


13

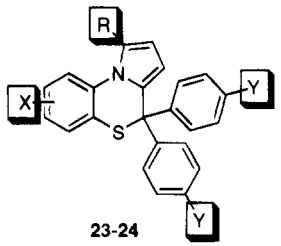
compd	X	Y	IC_{50} ± SEM (nM)[a]	K_i ± SEM (nM)
8a[b]	4'-OMe	$(Me)_2N$	0.42 ± 0.15	0.16 ± 0.06
8b[b]	2',4'-$(OMe)_2$	$(Me)_2N$	150 ± 49.5	56 ± 18.6
8c[b]	H	$(Me)_2N$	315 ± 60.0	118 ± 22.4
8d			783 ± 86.0	290 ± 31.9
10a	4'-OMe	$O(CH_2CH_2)_2N$	>5000	
10b	2',4'-$(OMe)_2$	$O(CH_2CH_2)_2N$	1230 ± 340.0	461 ± 149.9
10c	H	$3,4\text{-}(OMe)_2C_6H_3CH_2CH_2NMe$	198 ± 27.0	74 ± 10.0
10d[b]	4'-OMe	$3,4\text{-}(OMe)_2C_6H_3CH_2CH_2NMe$	53 ± 11.7	19 ± 4.2
10e	2',4'-$(OMe)_2$	$3,4\text{-}(OMe)_2C_6H_3CH_2CH_2NMe$	1600 ± 416.0	740 ± 192.4
11a	4'-OMe	$s\text{-}C_4H_9NH$	12 ± 3.1	4.4 ± 1.1
11b[b]	4'-OMe	$4\text{-}FC_6H_4NH$	>5000	
11d[b]	4'-OMe	$3,4\text{-}(OMe)_2C_6H_3CH_2CH_2NH$	534 ± 192.2	200 ± 72.0
11e[b]	2',4'-$(OMe)_2$	$s\text{-}C_4H_9NH$	15 ± 3.2	5.5 ± 1.1
11f	2',4'-$(OMe)_2$	$4\text{-}FC_6H_4NH$	>5000	
11g	2',4'-$(OMe)_2$	$3,4\text{-}(OMe)_2C_6H_3CH_2CH_2NH$	218 ± 39.9	81 ± 14.8
13			4670 ± 116.7	1750 ± 437.5
verapamil			3.7 ± 1.3	1.4 ± 0.5
diltiazem			46 ± 6.5	21 ± 2.9

[a] The concentration of the tested compounds that inhibited [^{3}H]nitrendipine binding on rat cortex homogenate by 50% (IC_{50}) was determined by log–probit analysis with six concentrations of the displacers, each performed in triplicate. The IC_{50} values obtained were used to calculate apparent inhibition constants (K_i) by the Prusoff method. Values are the mean ± SE of at least three separate experiments performed in triplicate. [b] Reference 15.

Table 5. Receptor-Binding Affinity: Effect of Substitution at C-4 and in the Fused Phenyl Ring



22



23-24

compd	X	Y	R	IC_{50} ± SEM (nM)[a]	K_i ± SEM (nM)
22a	H	H		4700 ± 300.0	1830 ± 116.7
22b	H	4'-OMe		30 ± 1.1	11 ± 0.4
22e	8-CF_3	H		65 ± 2.0	24 ± 0.8
22g	6-Cl	H		1550 ± 400.0	581 ± 150.0
23a	H	H	H	465 ± 100.0	178 ± 38.3
23b	H	4'-OMe	H	>5000	
23c	8-CF_3	H	H	2060 ± 700.0	772 ± 262.3
23d	8-CF_3	4'-OMe	H	>5000	
23e	8-Cl	H	H	4350 ± 159.6	1630 ± 59.8
23f	6-Cl	H	H	320 ± 80.0	120 ± 30.0
24a	H	H	$(Me)_2NCH_2$	>5000	
24b	H	4'-OMe	$(Me)_2NCH_2$	2160 ± 150.0	809 ± 56.1
24c	8-CF_3	H	$(Me)_2NCH_2$	107 ± 30.0	40.1 ± 11.2
24d	6-Cl	H	$(Me)_2NCH_2$	82 ± 20.5	31 ± 7.75
diltiazem				46 ± 6.5	21 ± 2.9
verapamil				3.7 ± 1.3	1.4 ± 0.5

[a] The concentration of the tested compounds that inhibited [^{3}H]nitrendipine binding on rat cortex homogenate by 50% (IC_{50}) was determined by log–probit analysis with six concentrations of the displacers, each performed in triplicate. The IC_{50} values obtained were used to calculate apparent inhibition constants (K_i) by the Prusoff method. Values are the mean ± SE of at least three separate experiments performed in triplicate.

ring were the two structural features taken into account. Introduction in positions 6 and 8 of a chlorine atom or a trifluoromethyl group, respectively, together with the presence of a basic chain provided compounds **24c,d** which are as potent as the reference standards in radioligand-binding studies (Table 5). Pyrrolobenzothiazines lacking substituents at C-6 and C-8 (**24a,b**) showed significant lower affinity than **3**. Anyway, the role played by a methoxy substituent on the phenyl ring is unclear (see Table 5). These C-4-disubstituted compounds were suitable for our understanding the role of the substitution at position 4 in the interaction with the binding site in spite of their lower affinity and activity as CEBs.

Table 6. Comparison of Binding Affinity of Pyrrolobenzothiazines Lacking the C-1 Basic Side Chain

compd	IC_{50} ± SEM (nM)[a]	K_i ± SEM (nM)
7a	>5000	
22a	4700 ± 300.0	1830 ± 116.7
23a	465 ± 100.0	178 ± 38.3
28a[b]	0.78 ± 0.3	0.29 ± 0.11
28b[b]	4.4[c] ± 1.14	2.0[c] ± 0.52
diltiazem	46 ± 6.5	21 ± 2.9
verapamil	3.7 ± 1.3	1.4 ± 0.5

[a] The concentration of the tested compounds that inhibited [^{3}H]nitrendipine binding on rat cortex homogenate by 50% (IC_{50}) was determined by log–probit analysis with six concentrations of the displacers, each performed in triplicate. The IC_{50} values obtained were used to calculate apparent inhibition constants (K_i) by the Prusoff method. Values are the mean ± SE of at least three separate experiments performed in triplicate. [b] Reference 29. [c] Different stock of compound **28b** was previously reported as not active in inhibiting [^{3}H]nitrendipine binding. Although we have no clear explanation for such discrepancy, the present data substitute the previous result.

Effect of Modification of the Pyrrole Ring. Previously[29] we reported preliminary results about novel pyrrolobenzothiazines in which the pyrrole ring was replaced by a pyrrolidinone ring (**28**). Although the basic side chain is lacking and it is replaced by a

Table 7. Comparison of Inhibition of [^{3}H]Nitrendipine Binding in Rat Brain and Heart

compd	K_i ± SEM (nM)	
	cerebral cortex	heart[a]
8a	0.16 ± 0.06	53 ± 13.2
8b	56 ± 18.6	160 ± 40.0
10d	19 ± 4.2	8.3 ± 2.1
11e	5.5 ± 1.1	640 ± 160.0
13	1750 ± 437.5	NA[b]
24d	31 ± 7.75	26 ± 6.5
28b	2.0 ± 0.52	35 ± 8.75
diltiazem	21 ± 2.9	280 ± 70.0
verapamil	1.4 ± 0.5	18 ± 4.5

[a] K_i values for displacement of [^{3}H]nitrendipine binding to rat heart L-type calcium channels. The concentration of the tested compounds that inhibited [^{3}H]nitrendipine binding on rat heart homogenates by 50% (IC_{50}) was determined by log–probit analysis with six concentrations of the displacers, each performed in triplicate. IC_{50}s were converted to K_i values using the Cheng–Prusoff equation. Values are the mean ± SE of at least three separate experiments performed in triplicate. [b] NA = not active.

carbonyl function at C-1, the 4-phenyl derivative showed a subnanomolar affinity for calcium channel receptors (CCRs) in radioreceptor assay on rat cortex homogenate (Table 6). In functional studies **28a** proved to be one of the most active compounds on cardiac tissue. In fact it is 4-fold more potent as a negative inotropic agent than **3** (see the Molecular Modeling section). The apparent superiority of the CO of **28** to the amine of other analogues could suggest an H-bond acceptor role for these functions. Some of the described compounds were also tested in radioligand-binding assays using rat heart homogenate following Ehlert's procedure.[27] Substantially equivalent results were obtained comparing the receptor affinity of the tested compounds on the two tissue homogenates (Table 7), although the tested compounds proved to be less active than in rat cortex homogenate.

Pharmacological Studies. Vasorelaxing activity, which is a measure of calcium antagonism, was assessed on guinea pig aorta strips, while negative inotropy and chronotropy were determined on guinea pig atria. Essentially equivalent results were obtained for the tested compounds in radioligand-binding assays and functional studies, although there is a discrepancy between the negative inotropic effects of **8a,b**. In fact, **8b** appeared to be 5-fold more potent than **8a** in tissue preparation, although **8a** showed higher affinity in radioreceptor assays (see Table 8). Even the negative inotropic potency of **13** was not associated with high affinity for the diltiazem receptor. The reason for these discrepancies between cardiovascular activity and binding affinity is not straightforward, but they may be related to tissue selectivity and the use of different tissues for the two assays. The observed activity suggested that the receptor can accommodate compounds with a wide range of substituents at C-1 without marked adverse effect on activity. The tolerance of the receptor to the presence of bulky substituents at position 1 is reflected by the fact that **10d,e** and **11d–g** are equipotent with diltiazem as negative inotropic agents. Furthermore, these studies revealed a different degree of selectivity of the tested compounds for cardiac over vascular tissue. Significant differences in the relative cardiac depression/smooth muscle relaxant activities between the three major agents verapamil, diltiazem, and nifedipine are recognized. Verapamil and diltiazem

Table 8. Cardiovascular Activity of Tested Compounds

compd	negative inotropic activity on isolated guinea pig left atrium[a] at 10^{-7} M (n = 5–7), percent decrease (M ± SEM)	negative chronotropic activity on isolated guinea pig left right atrium[a] at 5×10^{-6} M (n = 5–7), percent decrease (M ± SEM)	ED_{20} of inotropic negative potency on stimulated guinea pig left atrium		ED_{20} of chronotropic negative potency of tested compounds on guinea pig spontaneously beating right atrium		calcium antagonist activity on K$^+$-depolarized guinea pig aortic strips at 10^{-4} M (n = 4–5), percent inhibition of Ca^{2+} contraction[b] (M ± SEM)	IC_{50} of calcium antagonist potency on K$^+$-depolarized guinea pig aortic strips	
			ED_{20} (μM)	95% conf lim ($\times 10^{-6}$)	ED_{20} (μM)	95% conf lim ($\times 10^{-6}$)		IC_{50} (μM)	95% conf lim ($\times 10^{-6}$)
8a	75 ± 4.1[c]	30 ± 1.7[b]	0.81	0.69–0.94			10 ± 0.7		
8b	68 ± 3.1[c]	20 ± 1.4[b]	0.15	0.13–0.19			5 ± 0.2[c]		
8c	79 ± 3.7	76 ± 5.5	1.5	1.2–1.8	0.51	0.44–0.62	42 ± 3.7		
10d	86 ± 3.9	32 ± 2.8	0.84	0.72–0.95			4 ± 0.3		
10e	62 ± 4.9[c]	18 ± 0.7	0.86	0.75–0.93			10 ± 0.7[c]		
11d	82 ± 4.1	14 ± 1.1	0.45	0.39–0.52			16 ± 0.9[c]		
11e	85 ± 4.0[c]	67 ± 4.8[b]	0.60	0.52–0.70	0.42	0.35–0.51	18 ± 1.4[c]		
11g	83 ± 3.3[c]	10 ± 0.4	0.56	0.51–0.63			9 ± 0.2[c]		
13	69 ± 2.7	22 ± 1.8	0.92	0.88–0.97			24 ± 1.5		
22a	86 ± 2.4[c]	33 ± 2.5	0.38	0.35–0.42			36 ± 3.0		
22b	71 ± 4.3[c]	70 ± 3.0	0.24	0.19–0.3	2.3	1.9–2.8	11 ± 0.5		
24a	56 ± 1.4[c]	10 ± 0.8	0.89	0.82–0.97			3.4 ± 0.1		
24b	78 ± 3.0	48 ± 1.5	0.73	0.64–0.83			12 ± 1.3		
28a	66 ± 2.3[c]	20 ± 1.6	0.23	0.19–0.27			37 ± 2.9	20.0	16–27
28b	60 ± 0.7	39 ± 2.9[c]	1.2	0.9–1.6			32 ± 0.9	16.6	13–21
diltiazem	78 ± 3.4[c]	94 ± 5.6	0.79	0.7–0.85	0.07	0.064–0.075	88 ± 2.3	2.6	2.2–3.1

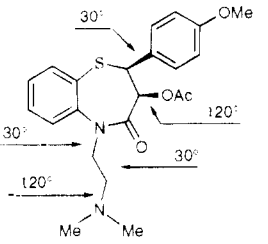
[a] The left atria were driven at 1 Hz. The indicated concentration expressed the maximum effect for most compounds. [b] Pretreatment ranged from 165 to 190 beats/min. The indicated concentration expressed the maximum effect for most compounds. [c] Calculated from log concentration–response curves (Probit analysis by Litchfield and Wilcoxon with $n = 5$–7). When the maximum effect was <50%, the ED_{20}, ED_{50}, and IC_{50} values were not calculated. [d] The 10^{-7} M gave the maximum effect for most compounds. [e] At 10^{-6} M. [f] At 5×10^{-6} M. [g] At 10^{-5} M. [h] At 10^{-4} M.

are approximately equiactive in cardiac and vascular tissue, whereas nifedipine is significantly more active in smooth muscle (vascular and nonvascular).[30] Literature[31] does not evidence examples of clear-cut selectivity for diltiazem-related CEBs.[32] The fused aromatic ring of diltiazem was substituted with different atoms by Inoue,[12] while Floyd[13] described analogues with the sulfur-bridged atom replaced by a methylene group. In both cases the described compounds showed an equal or superior pharmacological profile than **3** (longer acting, higher antihypertensive activity, and cerebral vasodilating). Additionally, in 1991, Fujita[33] described a series of benzothiazines as potent CEBs, the pharmacological profile of which was found to be superior to that of verapamil and diltiazem, although these benzothiazines exhibited lower cardioselectivity. In our case, the alteration of the bicyclic system of diltiazem to tricyclic pyrrolobenzothiazine resulted in potent cardiac depressant activity with selectivity for cardiac over vascular tissue. The mechanism by which the presence of certain structural features alter the tissue selectivity is not fully understood. In fact, the improvement in selectivity may be a consequence of increased potency or lipophilicity (pyrrolobenzothiazines vs **3**) as well as differences in primary Ca^{2+} channel protein structure (α1 subunit) or a consequence of differences in the coupling between the receptor and the effector components. The genetic polymorphism of α1 subunits may explain the significant changes in the sensitivity of the expressed channels against CEBs. Since the primary sequence of the channel subunits has now been established[34,35] and the structure of the binding site of CEBs is not yet elucidated, further advances are required to fully understand the reason of variation of tissue selectivity among CEBs.

Compounds **8a**, **18b**,**c**, **22b**, and **28a** were also used in additional binding studies.[20] The five representative benzothiazines had no affinity for central benzodiazepine receptors, mitochondrial benzodiazepine receptors, and GABA A receptors.

Molecular Modeling. The geometric and conformational properties of the tested compounds were investigated for their possible relevance in structure–affinity relationships. Seventeen pyrrolobenzothiazines (namely, **8a–d**, **11d**,**e**,**g**, **10a**,**b**,**d**, **23c–e**, **22a**,**e**, and **28a**,**b**) were selected from the whole data set with the assumption that the sampling of their conformational space could provide information valid for other closely related analogues. Diltiazem (**3**) as a typical ligand for CCRs was investigated and employed as template for molecular superimpositions. The molecular model of **3** and the studied compounds were constructed in the *S* configuration, although the described compounds were tested in vitro as a racemic mixture. The first step of our computer-assisted molecular modeling (CAMM) investigation was to verify the accuracy of the generalized MM2 force field implemented in MODEL to describe our compounds.[36] Available diltiazem X-ray crystal coordinates[37] were used as reference geometry for CAMM studies. INPUT submode of the program MODEL (version KS 2.99)[38] has been used to build up and optimize the geometry of the diltiazem structure. Statistical conformational analysis (mixed search) has been carried out on this model, using the program

Chart 3. Conformational Analysis of **3**



BKMDL,[38] and each bond of **3** has been rotated as reported in Chart 3.

We chose the mixed search conformational investigation since it is especially suited to provide all the possible conformers in the case of six- and seven-member rings.[39] One hundred seventeen minimum energy conformations have been identified in the range of 3 kcal/mol (ΔE = 3 kcal/mol), and using the COMPARE option of MODEL, we compared these geometries with the X-ray structure of **3**. Superimpositions were performed on all the heavy atoms of the molecules. At least seven of these conformers showed a good value of rms (0.153–0.41 Å). In one case, the conformer named 83, as shown in Figure 1, gave the best match (rms = 0.153 Å) with the diltiazem X-ray structure, with a small energy difference of 1.96 kcal/mol from the minimum energy conformer.

Now MODEL seemed to be correctly parametrized, and because of the lack of X-ray crystal coordinates for the selected compounds, their input geometries were generated and initially minimized by using the program MODEL. First a thorough conformational analysis was carried out for the above derivatives in order to evaluate the putative global minimum energy conformations, as done for **3**. Bonds were rotated by the same increments as those reported for **3**, and a set of conformational minimum energies was obtained for each molecule. In order to superimpose the investigated compounds, we first had to define a set of pharmacophoric elements. The key substructures identified in diltiazem like CCRs, playing an important role in the interaction with the binding site, are (i) the centroid of the condensed benzene ring, (ii) the sulfur atom, (iii) the basic side chain, (iv) the amide moiety, and (v) the ester function. Thus, it was reasonable superimposing our CCR antagonists by following a pharmacophoric scheme proposed earlier for benzothiazepines and benzazepinones, binding at L-type calcium channels. For the sake of simplification, only minimum energy conformers have been superimposed to diltiazem X-ray geometry. Superimpositions between **3** and all the minimum energy conformers of the studied compounds have been performed keeping the pharmacophoric groups superimposed (*O*-methyl ether, sulfur- and nitrogen-bridged atoms, fused aromatic rings, and remote exocyclic nitrogen), using the FIT procedure within SYBYL.[40] This method appeared to be particularly helpful to evaluate a possible 3D arrangement of significant molecular features in comparison with the reference molecule. Taking into account the specific pharmacophoric groups for the matching processes, the fitting experiments between **3** and our most active benzothiazines showed a very good superimposition of our chosen features in the 3D space (rms range: 0.254–0.654 Å). Most studies were performed on our lead compound **8a**.

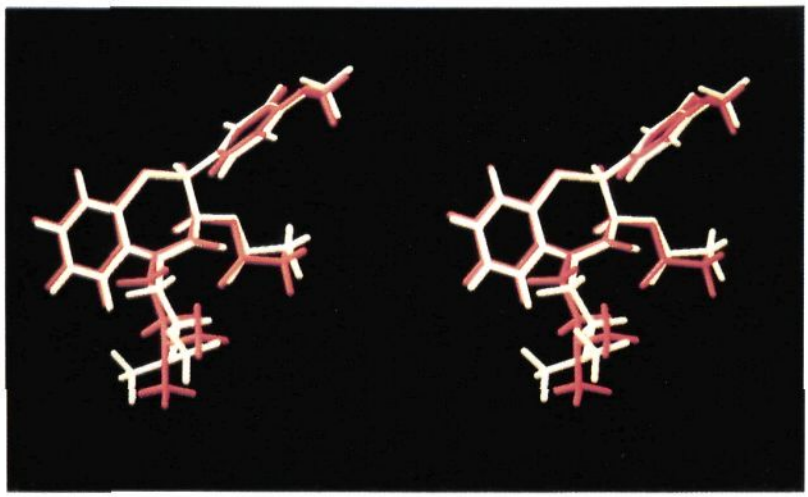


Figure 1. Superimposition of the X-ray-determined structure of diltiazem (orange) and the structure of the conformer named 83 of the same molecule obtained by the conformational analysis (light yellow). ΔE = 1.86 kcal/mol with respect to the minimum energy conformation.

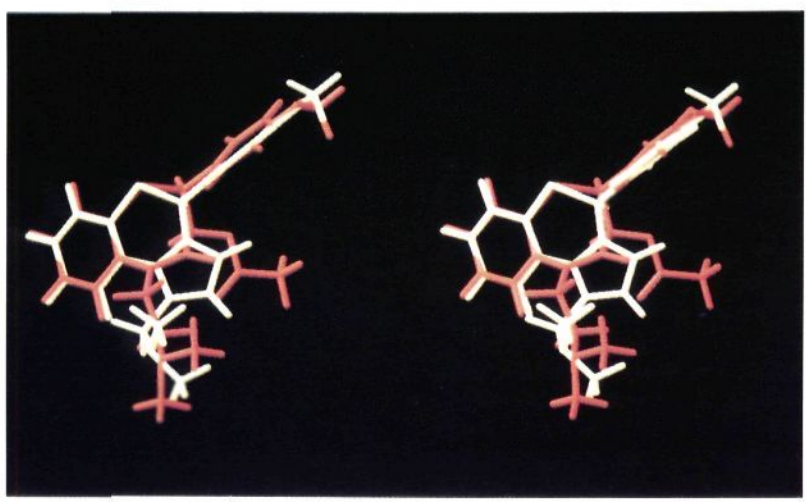


Figure 2. Superimposition of the minimum energy conformer of **8a** (light yellow) and the X-ray-determined structure of diltiazem (orange).

Table 9. Comparison of the Distances between Three Selected Structural Elements

	compound				
distance (Å)	**8a**	**8b**	**10d**	**11e**	diltiazem
N-exo/N-endo	3.139	3.131	3.186	3.291	3.116
N-exo/4′-O	10.240	10.374	10.388	10.692	10.387
N-endo/4′-O	8.011	7.831	7.889	7.993	7.814

Figure 2 shows the good similarity of the pharmacophoric features of **3** and **8a**.

Then, to find a correlation between pharmacological activities and structural features of our compounds, we measured the distances between the exocyclic nitrogen (N-exo), the endocyclic nitrogen (N-endo), and the oxygen atom of the ether function (4′-O). Table 9 reports the values obtained for compounds **8a**,**b**, **10d**, **11e**, and **3**. For sake of simplification, only few representative compounds have been reported in Table 9 since the other analogues showed similar distances between the pharmacophoric groups selected. It appears that the calculated distances between the three pharmacophoric elements are a significant parameter to correlate molecular structure and pharmacological activity. As a further attempt to visualize the high similarity between **3** and the synthesized benzothiazines, we considered their electrostatic potential maps. In order to simplify the computational study, the negative and positive isopotential maps have been considered separately. First we calculated the negative isopotential maps (−12 kcal/mol) of all the selected compounds as minimum energy conformers using the ISOPOTENTIALMAP procedure within SYBYL, the atomic point changes being computed by the program MOPAC.[41] A good match of the most negative areas (*O*-methyl ether, the endocyclic nitrogen, and partially the exocyclic nitrogen) of the benzothiazines and **3** was observed when their key substructures were superimposed, as shown in Figure 3 for **8a** and **3**.

When the positive isopotential map (+12 kcal/mol) of **3** had been constructed and compared to the corresponding map of **8a** (and all the other active compounds), superimposing their key substructures, a good match was also obtained. It is interesting to note that a similar superimposition (negative and positive isopo-

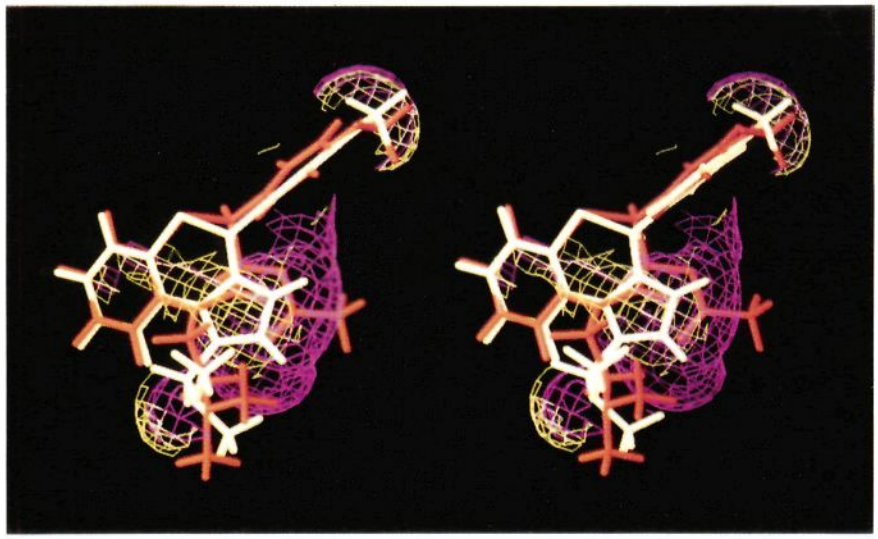


Figure 3. Stereoview of the superimposition of the one-level negative electrostatic isopotential surfaces of compound **8a** and diltiazem. In orange and violet are shown the diltiazem X-ray structure and its −12 kcal/mol isopotential surface, respectively. In light yellow and yellow are shown the minimum energy geometry of **8a** and its −12 kcal/mol isopotential surface, respectively.

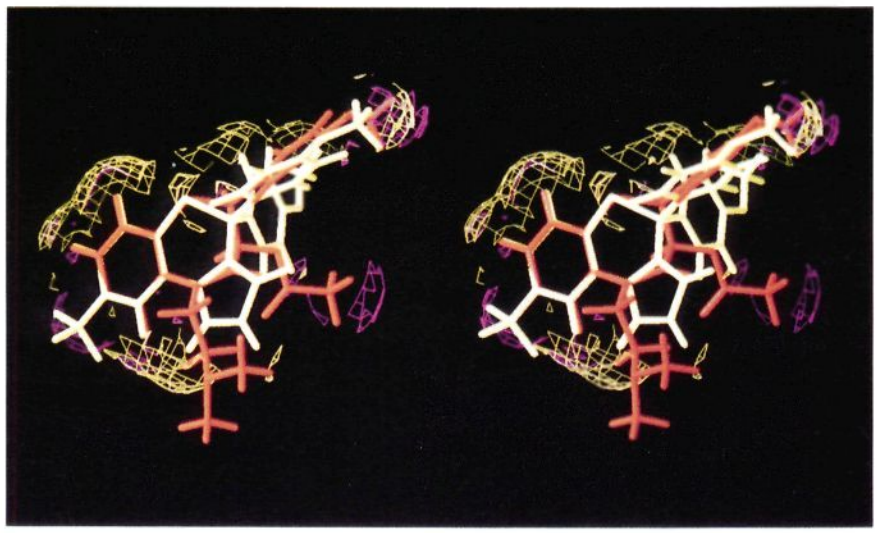


Figure 4. Stereoview of the superimposition of the one-level positive electrostatic isopotential surfaces of compound **23d** and diltiazem. In orange and violet are shown the diltiazem X-ray structure and its +12 kcal/mol isopotential surface, respectively. In light yellow and yellow are shown the minimum energy geometry of **23d** and its +12 kcal/mol isopotential surface, respectively.

tential maps) performed on **23d** gave a poor overlapping, especially evident for positive isopotential maps (see Figure 4).

In order to provide an explanation of the high affinity for CCRs and of the potent negative inotropic activity of compounds **28a**,**b**, we subjected these novel pyrrolobenzothiazines lacking the basic side chain to molecular modeling studies. CAMM studies showed a good match between **3** and **28** (*trans* isomers) when their key substructures were superimposed (rms = 0.60 Å). As a further attempt to rationalize the potent in vitro activity of **28** vs **3**, we considered the negative isopotential maps. Figure 5 shows the good match of the most negative areas (the endocyclic nitrogen, the OMe at C-4′, and the carbonyl group) of **28b** and **3**, although **28a**,**b** do not have the negative area corresponding to that one at C-3 of diltiazem (ester group). The same results have been obtained using **28a** in these CAMM studies.

To verify these last results, in CAMM studies we included the 3-desacetoxy-3-methyldiltiazem recently described by Atwal.[42] In in vitro tests it proved to be 10−20-fold more potent than diltiazem itself. As expected this compound does not show a negative area at C-3, since the ester function of **3** has been replaced by a methyl group. Thus, on the basis of the negative isopotential map studies and structural superimpositions, we conclude that, although the lack of the ester function, the higher in vitro potency of **28** may be due to shape effects. These CAMM studies seem to exclude, in fact, an electrostatic effect for the ester function of **3** in the interaction with the receptor protein. The results of the molecular modeling studies point out that both approaches aim for a very high structural similarity between diltiazem and our benzothiazines. The very high qualitative similarity allowed us to assume that these compounds could interact at the diltiazem-bind-

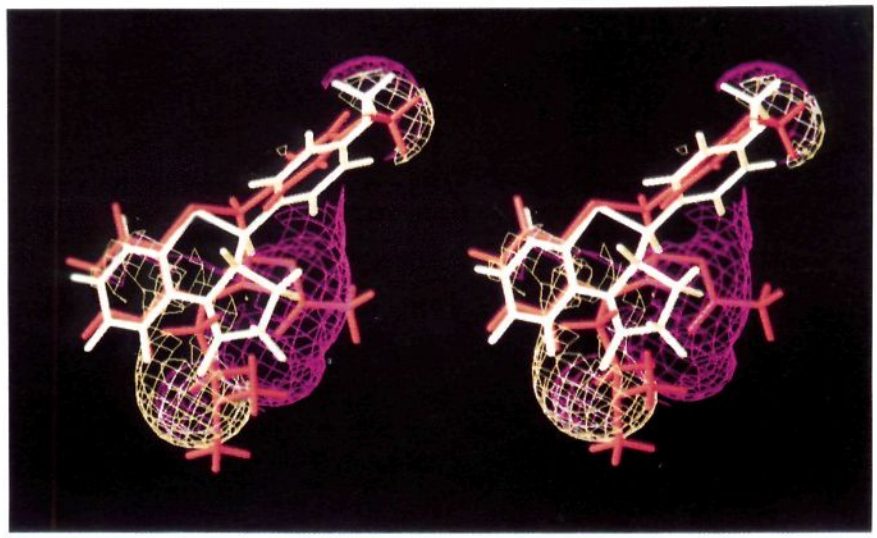


Figure 5. Stereoview of the superimposition of the one-level negative electrostatic isopotential surfaces of compound **28a** and diltiazem. In orange and violet are shown the diltiazem X-ray structure and its −12 kcal/mol isopotential surface, respectively. In light yellow and yellow are shown the minimum energy geometry of **28a** and its −12 kcal/mol isopotential surface, respectively.

ingsite of the protein receptor (L-type calcium channels). Anyway, these results deserve further investigations.

Conclusion

In this paper we have reported the synthesis of a novel class of CEBs related to **3**. The calcium channel activity of pyrrolobenzothiazines is dependent upon two pharmacophores: (1) the substituted phenyl ring at C-4 and (2) the basic side chain at C-1 on the pyrrole ring. The affinity is also influenced by substitution on the fused phenyl ring and by double substitution at C-4. In the **22** and **24** series, the introduction of an electron-withdrawing group in the fused phenyl ring enhances the potency. Replacement of the pyrrole ring with a pyrrolidinone ring lacking the basic side chain led to a novel series of potent cardiodepressant agents (**28**). In functional studies some of the tested compounds showed higher potency and efficacy as negative inotropic agents than diltiazem, without relevant calcium antagonist activity on vascular smooth muscle, revealing a clear-cut selectivity for cardiac over vascular tissue, with a wide variation of data. From these results, we can conclude that this work represents the first example of diltiazem structure alteration that leads to a series of compounds with potent negative inotropic activity and a clear-cut selectivity for cardiac over vascular tissue.

Experimental Section

Melting points were determined using an Electrothermal 8103 apparatus and are uncorrected. IR spectra were taken with Perkin-Elmer 398 and FT 1600 spectrophotometers. ^{1}H NMR spectra were recorded on a Varian XL 200 spectrometer with TMS as internal standard; the values of chemical shifts (δ) are given in parts per million (ppm) and coupling constants (J) in hertz (Hz). All reactions were carried out in an argon atmosphere. Progress of the reaction was monitored by TLC on silica gel plates (Riedel-de-Haen, Art. 37341). Merck silica gel (Kieselgel 60) was used for chromatography (70−230 mesh) and flash chromatography (230−400 mesh) columns. Extracts were dried over $MgSO_4$, and solvents were removed under reduced pressure. Elemental analyses were performed on a Perkin-Elmer 240C elemental analyzer, and the results are within ±0.4% of the theoretical values, unless otherwise noted. Yields refer to the purified products and are not optimized. Physical data for compounds **5**−**13**, **15**−**20**, and **21**−**28** are reported in Tables 1−3, respectively.

(±)-*N*-(2′-Fluorophenyl)-2-(α-hydroxy-*p*-methoxybenzyl)pyrrole (5a). To Mg turnings (0.74 g, 31 mmol) in anhydrous THF (16 mL) was slowly added a solution of *p*-bromoanisole (3.88 mL, 31 mmol) under argon. After refluxing for 2 h, a solution of **4**[16] (2 g, 10.5 mmol) in anhydrous THF (60 mL) was added dropwise, and the mixture was refluxed for 2 h. The cooled reaction mixture was poured into a cold 20% NH_4Cl solution (100 mL), stirred for an additional 30 min, and extracted with chloroform. The combined organic layers were washed with brine, dried, and concentrated. Flash chromatography (CH_2Cl_2) gave 2.32 g (75%) of **5a** as a colorless oil: R_f = 0.22 (CH_2Cl_2); IR (neat) 3480−3380, 1610, 1250, 765 cm^{-1}. This compound was used in the next step without further purification.

(±)-*N*-(2′-Fluorophenyl)-2-(α-hydroxy-2′,4′-dimethoxybenzyl)pyrrole (5b). Starting from **4** (3 g, 15.8 mmol), the title compound was obtained according to the procedure described above (reaction time 4 h) (78%): R_f = 0.26 (CH_2Cl_2). After flash chromatography, **5b** was used crude: IR (neat) 3455−3380, 1605, 773 cm^{-1}.

(±)-2-[α-(Acetylthio)-*p*-methoxybenzyl]-*N*-(2′-fluorophenyl)pyrrole (6a). To a stirred and cooled solution (0 °C) of triphenylphosphine (4.08 g, 15.6 mmol) in anhydrous THF (60 mL) was added DiPAD (3.23 g, 15.6 mmol) under argon. After 30 min a solution of **5a** (2.32 g, 7.8 mmol) and thiolacetic acid (1.12 mL, 15.6 mmol) in anhydrous THF (30 mL) was slowly added. After 1 h at 0 °C the mixture was allowed to stir for 3 h at room temperature. The solvent was removed, and the residue was diluted with ethyl ether. The insoluble material was filtered off, and the filtrate was concentrated to give a residue which was purified by flash chromatography (cyclohexane and benzene) to afford **6a** as a pale yellow oil: R_f = 0.53 (dichloromethane); IR (neat) 1690, 1610, 1250, 715 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 7.62−7.12 (m, 6 H), 6.78 (m, 3 H), 6.25 (m, 2 H), 5.68 (s, 1 H), 3.75 (s, 3 H), 2.17 (s, 3 H).

(±)-2-[α-(Acetylthio)-2′,4′-dimethoxybenzyl]-*N*-(*p*-fluorophenyl)pyrrole (6b). Starting from **5b** (1 g, 3 mmol), the title compound was obtained as an oil, according to the procedure described for **5a** (reaction time 5 h): R_f = 0.48 ($CHCl_3$); IR (neat) 1695, 1615, 1510, 1210, 765 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 7.37−7.08 (m, 5 H), 6.69 (m, 1 H), 6.46−6.18 (m, 4 H), 6.05 (s, 1 H), 3.75 (s, 3 H), 3.63 (s, 3 H), 2.20 (s, 3 H).

(±)-4-(*p*-Methoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (7a). To a stirred and cooled (−20 °C) solution of **6a** (2.06 g, 5.8 mmol) in dry MeOH (59 mL) was added portionwise sodium methoxide (0.35 g, 6.36 mmol) under argon. After 1 h at room temperature, the solution was

neutralized with 10% methanolic HCl and evaporated in vacuo. The residue was taken up in freshly distilled *N,N*-dimethylformamide (DMF) (64 mL), and sodium hydride (0.41 g, 17 mmol) was added. The reaction mixture was heated at 80 °C for 1 h. After neutralization with 1 N HCl and evaporation, the residue was purified by flash chroamtography (EtOAc). Recrystallization gave 1.34 g of **7a** as colorless prisms: R_f = 0.41 ($CHCl_3$); IR (KBr) 1599, 748 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.44–7.00 (m, 7 H), 6.85 (m, 2 H), 6.29 (t, 1 H, J = 3.3 Hz), 5.73 (m, 1 H), 5.29 (s, 1 H), 3.80 (s, 3 H).

(±)-4-(2′,4′-Dimethoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (7b). Starting from **6b** (1.34 g, 3.47 mmol), the title compound was obtained as described above (reaction time 1.5 h): R_f = 0.38 (30% cyclohexane in benzene); IR (KBr) 1602, 720 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.41 (d, 1 H, J = 1.4 Hz), 7.39–7.17 (m, 4 H), 7.05 (m, 2 H), 6.46 (d, 1 H, J = 3.4 Hz), 6.38 (app dd, 1 H, J = 8.4, 3.4 Hz), 6.30 (m, 1 H), 5.84 (m, 1 H), 5.76 (s, 1 H), 3.84 (s, 3 H), 3.77 (s, 3 H).

General Procedure for Preparation of Compounds 8a–d. This procedure is illustrated for the preparation of (±)-1-[(*N,N*-dimethylamino)methyl]-4-(*p*-methoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (**8a**). To a solution of **7a** (0.6 g, 2.04 mmol) in glacial acetic acid (40 mL) was added a mixture of 40% HCHO (1.17 mL) and 40% aqueous dimethylamine (2.1 mL) in glacial acetic acid (5 mL). The solution was stirred at room temperature for 12 h. The solvent was removed under vacuum, and the residue was partitioned between 10% $NaHCO_3$ and EtOAc. The organic solution was washed with brine, dried, and concentrated. The residue was flash chromatographed (EtOAc) and recrystallized to afford 0.65 g of **8a** as a white solid: R_f = 0.31 (EtOAc); IR (KBr) 1380, 1250, 780, 762 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.41 (d, 1 H, J = 8.0 Hz), 7.34–7.24 (m, 4 H), 7.08 (m, 1 H), 6.86 (m, 2 H), 6.14 (d, 1 H, J = 3.3 Hz), 5.61 (d, 1 H, J = 3.1 Hz), 5.17 (s, 1 H), 3.79 (s, 3 H), 3.35 (AB q, 2 H, J = 13.6 Hz), 2.33 (s, 3 H).

(±)-1-[(*N,N*-Dimethylamino)methyl]-4-(2′,4′-dimethoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (8b). Starting from **7b** (0.6 g, 1.85 mmol), the title compound was obtained according to the procedure described for **8a**: R_f = 0.28 (EtOAc); IR (KBr) 1370, 1185, 840, 760 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.46 (d, 1 H, J = 9.4 Hz), 7.38 (dd, 1 H, J = 7.6, 1.6 Hz), 7.33–7.03 (m, 4 H), 6.46 (d, 1 H, J = 2.4 Hz), 6.40 (dd, 1 H, J = 8.5, 2.4 Hz), 6.15 (d, 1 H, J = 3.4 Hz), 5.65 (s, 1 H), 3.81 (s, 3 H), 3.77 (s, 3 H), 3.31 (AB q, 2 H, J = 13.5 Hz), 2.34 (s, 6 H).

(±)-1-[(*N,N*-Dimethylamino)methyl]-4-phenyl-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (8c). Starting from **7c** (0.28 g, 1.06 mmol), the title compound was obtained following an identical procedure as for **8a**: R_f = 0.3 (EtOAc); IR (KBr) 1385, 1140, 765 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.46 (dd, 1 H, J = 8.1, 1.2 Hz), 7.45 (m, 7 H), 7.10 (m, 1 H), 6.15 (d, 1 H, J = 3.5 Hz), 5.61 (d, 1 H, J = 2.9 Hz), 5.21 (s, 1 H), 3.26 (AB q, 2 H, J = 13.6 Hz), 2.33 (s, 6 H).

(±)-1-[(*N,N*-Dimethylamino)methyl]-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (8d). Starting from **7d** (0.2 g, 1.06 mmol), **8d** was obtained according to the procedure described above for **8a** (reaction time 12 h at 30 °C): R_f = 0.42 (EtOAc); IR (KBr) 1525, 1340, 1030 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.60–7.19 (m, 4 H), 6.63 (d, 1 H, J = 3.6 Hz), 6.15 (d, 1 H, J = 3.6 Hz), 4.54 (s, 2 H), 3.90 (s, 2 H), 2.51 (s, 6 H); MS *m/z* 244 (22, M^+), 229, 200 (100), 186, 167, 154, 140.

General Procedure for Preparation of Aldehydes 9a–c. This procedure is illustrated for the preparation of (±)-4-(*p*-methoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine-1-carboxaldehyde (**9a**). Phosphorus oxychloride (86 μL, 0.93 mmol) was slowly added to cold (0 °C) freshly distilled DMF (71 μL, 0.93 mmol). The mixture was allowed to stir at 0 °C for 15 min under argon. Then a solution of **7a** (0.25 g, 0.85 mmol) in DMF (1 mL) was added dropwise. The solution was stirred at 50 °C for 3.5 h under argon and then cooled, the pH was adjusted to 8 with 20% Na_2CO_3, and the mixture was extracted with EtOAc. The organic layers were washed with brine, dried, and concentrated. The residue was flash chromatographed ($CHCl_3$ and EtOAc) and recrystallized to give 250 mg of **9a** as pale yellow prisms: R_f = 0.53 ($CHCl_3$ and EtOAc, 1/1); IR ($CHCl_3$) 1660, 1510, 1240, 760 cm^{-1}; ^1H NMR ($CDCl_3$) δ 9.68 (s, 1 H), 7.56 (dd, 1 H, J = 7.9, 1.3 Hz), 7.45 (dd, 1 H, J = 7.7, 1.7 Hz), 7.40–7.12 (m, 5 H), 6.88 (m, 2 H), 5.89 (d, 1 H, J = 3.4 Hz), 5.11 (s, 1 H), 3.80 (s, 3 H).

(±)-4-(2′,4′-Dimethoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine-1-carboxaldehyde (9b). Starting from 0.5 g (1.54 mmol) of **7b**, the title compound was obtained as colorless prisms (reaction time 3.5 h at 50 °C) according to the procedure described for **9a**: R_f = 0.6 (EtOAc and $CHCl_3$, 1/1); IR (KBr) 1666, 1609, 1377, 824, 751 cm^{-1}; ^1H NMR ($CDCl_3$) δ 9.68 (s, 1 H), 7.53 (dd, 1 H, J = 9.0, 1.0 Hz), 7.43 (dd, 1 H, J = 7.6, 1.5 Hz), 7.32 (dt, 1 H, J = 8.0, 1.6 Hz), 7.21 (m, 2 H), 6.97 (d, 1 H, J = 8.5 Hz), 6.49 (d, 1 H, J = 3.2 Hz), 6.44 (d, 1 H, J = 12.9 Hz), 5.93 (d, 1 H, J = 4.0 Hz), 5.60 (s, 1 H), 3.83 (s, 3 H), 3.78 (s, 3 H).

(±)-4-Phenyl-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine-1-carboxaldehyde (9c). Starting from 0.4 g (1.51 mmol) of **7c**, the title compound was obtained (reaction time 3 h at 50 °C) as colorless plates: R_f = 0.52 ($CHCl_3$); IR (KBr) 1670, 1474, 756 cm^{-1}; ^1H NMR ($CDCl_3$) δ 9.70 (s, 1 H), 7.57 (d, 1 H, J = 6.8 Hz), 7.46 (dd, 1 H, J = 7.6, 1.6 Hz), 7.42–7.15 (m, 8 H), 5.90 (d, 1 H, J = 4.0 Hz), 5.15 (s, 1 H).

(±)-4-(*p*-Methoxyphenyl)-1-(morpholinomethyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (10a). To a solution of **9a** (0.15 g, 0.51 mmol) in glacial acetic acid (9 mL) was added a mixture of morpholine (54 μL, 0.62 mmol), 40% HCHO (86 μL, 1.27 mmol), and glacial acetic acid (1 mL). The reaction mixture was stirred at room temperature for 24 h. The solvent was removed, and the residue was treated with 20% $NaHCO_3$ and extracted with EtOAc. The organic layers were washed with brine, dried, and concentrated. The crude material was purified by flash chromatography (EtOAc and hexanes, 1/1) and recrystallized to give 185 mg of **10a** as a white solid: R_f = 0.58 (EtOAc and hexanes, 1/1); IR ($CHCl_3$) 1599, 761 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.51 (d, 1 H, J = 8.1 Hz), 7.44 (d, 1 H, J = 7.6 Hz), 7.40–7.25 (m, 3 H), 7.13 (t, 1 H, J = 7.5 Hz), 6.88 (m, 2 H), 6.16 (d, 1 H, J = 3.3 Hz), 5.61 (d, 1 H, J = 3.5 Hz), 5.18 (s, 1 H), 3.78 (m, 7 H), 3.51 (0.5 AB q, 1 H, J = 13.5 Hz), 3.31 (0.5 AB q, 1 H, J = 13.6 Hz), 2.60 (m, 4 H).

(±)-4-(2′,4′-Dimethoxyphenyl)-1-(morpholinomethyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (10b). The title compound was obtained starting from **9b** (0.1 g, 0.31 mmol) according to the procedure described for **10a** except that the compound was purified by flash chromatography ($CHCl_3$): R_f = 0.26 ($CHCl_3$); IR (KBr) 1610, 1390, 767 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.53 (d, 1 H, J = 7.9 Hz), 7.50–7.13 (m, 4 H), 6.46 (m, 2 H), 6.18 (d, 1 H, J = 4.2 Hz), 5.63 (m, 2 H), 4.85–4.68 (m, 10 H), 3.44 (0.5 AB q, 1 H, J = 13.2 Hz), 3.38 (0.5 AB q, 1 H, J = 13.3 Hz), 2.61 (m, 4 H).

General Procedure for Preparation of Compounds 10c–e. This procedure is illustrated for the preparation of (±)-1-[[*N*-[2-(3′,4′-dimethoxyphenyl)ethyl]-*N*-methylamino]-methyl]-4-phenyl-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (**10c**). To a solution of aldehyde **9c** (350 mg, 1.19 mmol) dissolved in 12.5 mL of anhydrous MeOH and THF (3/1) was added *N*-[2-(3′,4′-dimethoxyphenyl)ethyl]-*N*-methylamine (0.9 mL, 4.76 mmol) at room temperature, under argon. The solution was then cooled at 0 °C, and a solution of $NaBH_3CN$ (74.8 mg, 1.24 mmol) and anhydrous $ZnCl_2$ (81 mg, 0.57 mmol) in anhydrous MeOH (6 mL) was added. After 30 min at 0 °C, the solution was stirred at room temperature for 3.5 h. The solvent was removed and the residue partitioned between 0.1 N NaOH (12 mL) and EtOAc. The organic solution was dried and concentrated. The crude product was purified by flash chromatography (EtOAc and hexanes, 3/1) to give 0.45 g of **10c** as a colorless oil: R_f = 0.18 ($CHCl_3$); **10c** was characterized as hydrochloride: ^1H NMR (DMSO-d_6) δ 10.78 (br s, 1 H), 7.62 (d, 1 H, J = 7.9 Hz), 7.49 (d, 1 H, J = 7.5 Hz), 7.38 (t, 1 H, J = 7.3 Hz), 7.24 (m, 6 H), 6.86–6.60 (m, 4 H), 5.91 (m, 1 H), 5.59 (s, 1 H), 4.68 (m, 2 H), 3.71 (s, 3 H), 3.69 (s, 3 H), 3.15–2.78 (m, 4 H), 2.57 (s, 3 H); MS ($C_{29}H_{30}N_2O_2S$) *m/z* 470 (5, M^+), 379, 319, 276 (100), 243, 200.

(±)-1-[[*N*-[2-(3′,4′-Dimethoxyphenyl)ethyl]-*N*-methylamino]methyl]-4-(*p*-methoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (10d). Starting from **9a** (80 mg, 0.25 mmol), the title compound was obtained using an identical procedure as for **10c**: R_f = 0.2 ($CHCl_3$); **10d** was characterized

as hydrochloride; ^{1}H NMR (DMSO-d_6) δ 10.48 (br s, 1 H), 7.61 (d, 1 H, J = 9.0 Hz), 7.55 (d, 1 H, J = 7.8 Hz), 7.40 (t, 1 H, J = 8.0 Hz), 7.23 (m, 3 H), 6.98–6.54 (m, 6 H), 5.87 (d, 1 H, J = 3.1 Hz), 5.51 (d, 1 H, J = 3.6 Hz), 4.66 (m, 2 H), 3.72 (s, 3 H), 3.70 (s, 3 H), 3.39 (s, 3 H), 3.20–2.65 (m, 4 H), 2.49 (s, 3 H); MS ($C_{30}H_{32}N_2O_3S$) m/z 500 (7, M^-), 379, 349, 306 (100), 273, 200.

(±)-4-(2′,4′-Dimethoxyphenyl)-1-[[*N*-[2-(3′,4′-dimethoxyphenyl)ethyl]-*N*-methylamino]methyl]-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (10e). Starting from **9b** (0.2 g, 0.57 mmol), the title compound was prepared using an identical procedure as for **10c**: R_f = 0.59 (EtOAc and hexane, 3/1); ^{1}H NMR ($CDCl_3$) δ 8.44 (dd, 1 H, J = 7.2, 1.8 Hz), 7.36 (dd, 1 H, J = 7.1, 1.8 Hz), 7.21–7.00 (m, 3 H), 6.77 (m, 3 H), 6.43 (m, 2 H), 6.17 (d, 1 H, J = 3.4 Hz), 5.65 (m, 2 H), 3.84 (s, 3 H), 3.83 (s, 3 H), 3.81 (s, 3 H), 3.78 (s, 3 H), 3.50 (0.5 AB q, 1 H, J = 13.6 Hz), 3.38 (0.5 AB q, 1 H, J = 13.4 Hz), 2.93–2.75 (m, 4 H), 2.40 (s, 3 H); MS ($C_{31}H_{34}N_2O_4S$) m/z 530 (5, M^-), 379, 336 (100), 306, 200.

(±)-1-[(*N*-*sec*-Butylamino)methyl]-4-(*p*-methoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (11a). A mixture of aldehyde **9a** (126 mg, 0.39 mmol) dissolved in anhydrous MeOH (1.5 mL), *sec*-butylamine (0.11 mL, 0.27 mmol), and 10% methanolic HCl (0.11 mL) was heated at 85 °C for 5 min in a resealable tube under argon. After cooling the solvent was removed and the solid residue was taken up in 1.1 mL of MeOH and THF (1/1). After cooling at 0 °C, sodium borohydride (71 mg, 1.88 mmol) was added and the solution was allowed to stir at 0 °C for 1.5 h. The reaction was quenched with water and the mixture extracted with EtOAc. The organic layers were washed with brine, dried, and concentrated. The residue was flash chromatographed (5% EtOAc in CH_2Cl_2) to give 138 mg of **11a** as colorless prisms: R_f = 0.22 (5% EtOAc in CH_2Cl_2); ^{1}H NMR ($CDCl_3$) δ 8.42 (dd, 1 H, J = 7.8, 2.8 Hz), 7.41 (d, 1 H, J = 8.4 Hz), 7.30 (m, 3 H), 7.10 (m, 1 H), 6.86 (m, 2 H), 6.17 (d, 1 H, J = 3.2 Hz), 5.61 (d, 1 H, J = 3.3 Hz), 5.16 (s, 1 H), 3.92 (0.5 AB q, 1 H, J = 13.1 Hz), 3.79 (s, 3 H), 3.71 (0.5 AB q, 1 H, J = 13.3 Hz), 2.65 (m, 1 H), 1.58–1.40 (m, 3 H), 1.10 (d, 3 H, J = 7.1 Hz), 0.92 (t, 3 H, J = 7.7 Hz).

(±)-1-[[*N*-(4′-Fluorophenyl)amino]methyl]-4-(*p*-methoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (11b). Starting from **9a** (160 mg, 0.5 mmol), and *p*-fluoroaniline, **11b** was prepared following the procedure described for **11a** (reaction time 20 min at 60 °C). The reduction step was performed using sodium borohydride (60 mg, 1.55 mmol) (reaction time 1 h at 0 °C). **11b** was obtained by flash chromatography (10% EtOAc in CH_2Cl_2) as colorless prisms: ^{1}H NMR ($CDCl_3$) δ 7.82 (d, 1 H, J = 8.8 Hz), 7.50–7.12 (m, 5 H), 7.10–6.85 (m, 4 H), 6.66 (m, 2 H), 6.31 (d, 1 H, J = 3.9 Hz), 5.71 (d, 1 H, J = 4.1 Hz), 5.18 (s, 1 H), 4.30 (s, 2 H), 3.81 (m, 4 H); MS m/z 416 (21, M^-), 335, 306 (100), 292, 248, 186.

(±)-1-[[*N*-[2-(3′,4′-Dimethoxyphenyl)ethyl]amino]methyl]-4-(*p*-methoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (11d). Starting from **9a** (100 mg, 0.28 mmol) and 3,4-dimethoxyphenethylamine, the title compound was prepared using the procedure described for **11a** (reaction time 15 min at 80 °C). Sodium borohydride (54 mg, 1.4 mmol) was used as reducing agent (reaction time 3.5 h at room temperature). After flash chromatography (EtOAc and CH_2Cl_2, 1/1) **11d** was obtained as an amorphous solid: R_f = 0.31 (EtOAc and CH_2Cl_2, 1/1); ^{1}H NMR ($CDCl_3$) δ 8.23 (d, 1 H, J = 7.7 Hz), 7.48–7.00 (m, 5 H), 6.93–6.70 (m, 5 H), 6.18 (d, 1 H, J = 3.9 Hz), 5.63 (d, 1 H, J = 3.3 Hz), 5.18 (s, 1 H), 3.92–3.73 (m, 12 H), 3.05–2.75 (m, 4 H); MS m/z 486 (32, M^-), 365, 306 (100), 292, 248.

(±)-1-[(*N*-*sec*-Butylamino)methyl]-4-(2′,4′-dimethoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (11e). Starting from **9b** (100 mg, 0.28 mmol), the title compound was prepared according to the procedure described for **11a**: R_f = 0.18 (10% EtOAc in CH_2Cl_2); ^{1}H NMR ($CDCl_3$) δ 8.42 (d, 1 H, J = 7.9 Hz), 7.38 (d, 1 H, J = 8.4 Hz), 7.29–7.21 (m, 1 H), 7.07 (m, 2 H), 6.45 (d, 1 H, J = 2.1 Hz), 6.38 (dd, 1 H, J = 8.3, 2.2 Hz), 6.17 (d, 1 H, J = 3.5 Hz), 5.66 (d, 1 H, J = 3.6 Hz), 5.64 (s, 1 H), 3.92 (0.5 AB q, 1 H, J = 8.9 Hz), 3.79 (m, 7 H), 2.66 (m, 1 H), 1.57–1.35 (m, 3 H), 1.11 (d, 3 H, J = 6.1 Hz), 0.92 (t, 3 H, J = 7.6 Hz).

(±)-4-(2′,4′-Dimethoxyphenyl)-1-[[*N*-(*p*-fluorophenyl)amino]methyl]-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (11f). Starting from **9b** (100 mg, 0.28 mmol) and *p*-fluoroaniline, **11f** was prepared using the procedure described for **11a** (reaction time 20 min at 60 °C). As reducing agent was used zinc cyanoborohydride (reaction time 3 h at room temperature). **11f** was obtained as a white solid: ^{1}H NMR ($CDCl_3$) δ 7.82 (d, 1 H, J = 7.4 Hz), 7.42–6.82 (m, 6 H), 6.61 (m, 2 H), 6.49 (m, 2 H), 6.35 (d, 1 H, J = 3.7 Hz), 5.75 (d, 1 H, J = 3.8 Hz), 5.68 (s, 1 H), 4.31 (s, 2 H), 3.79 (m, 7 H).

(±)-4-(2′,4′-Dimethoxyphenyl)-1-[[*N*-[2-(3,4-dimethoxyphenyl)ethyl]amino]methyl]-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (11g). Starting from **9b** (100 mg, 0.28 mmol) and 3,4-dimethoxyphenethylamine, **11g** was prepared using the procedure described for **11a**. Sodium borohydride (54 mg, 1.4 mmol) was used for the reduction of the corresponding imine (reaction time 3.5 h at room temperature). After flash chromatography (EtOAc) and recrystallization, **11g** was obtained as colorless prisms: R_f = 0.34 (10% EtOAc in CH_2Cl_2); ^{1}H NMR ($CDCl_3$) δ 8.44 (d, 1 H, J = 7.3 Hz), 7.43 (d, 1 H, J = 7.1 Hz), 7.36–7.00 (m, 3 H), 6.78 (m, 3 H), 6.49 (d, 1 H, J = 1.8 Hz), 6.41 (dd, 1 H, J = 7.3, 1.8 Hz), 6.18 (d, 1 H, J = 3.8 Hz), 5.68 (m, 2 H), 3.85 (m, 15 H), 3.08–2.70 (m, 4 H).

(±)-4-(*p*-Methoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzoxazine (12). To a mixture of sodium hydride (97 mg, 3.9 mmol) in anhydrous benzene (10 mL) was added the alcohol **5a** (1 g, 3.3 mmol) dissolved in 10 mL of anhydrous benzene. After 15 min at room temperature, the reaction mixture was heated at 70 °C for 1 h under argon. DMF (2 mL) was then added, and the red mixture was stirred at 80 °C for 6 h. After cooling the suspension was poured into crushed ice and stirred for 30 min. The aqueous mixture was extracted with ethyl ether, and the organic layers were washed with brine, dried, and concentrated. The residue was purified by flash chromatography (benzene and cyclohexane, 2/1) to give 500 mg of **12** as colorless prisms: R_f = 0.51 (benzene and cyclohexane, 2/1); IR (KBr) 1599, 1520, 1055, 760 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 7.45–7.31 (m, 3 H), 7.20 (d, 1 H, J = 2.7 Hz), 7.15–6.85 (m, 5 H), 6.30 (t, 1 H, J = 3.5 Hz), 6.03 (s, 1 H), 5.72 (d, 1 H, J = 2.6 Hz), 3.81 (s, 3 H).

(±)-1-[(Dimethylamino)methyl]-4-(*p*-methoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzoxazine (13). Starting from **12** (250 mg, 0.86 mmol), the title compound **13** was obtained following the procedure described for **8a** (reaction time 3 h at room temperature): R_f = 0.21 (EtOAc); IR (film) 1600, 1275, 880, 740 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 8.23 (m, 1 H), 7.43 (m, 2 H), 7.06 (m, 3 H), 6.93 (m, 2 H), 6.14 (d, 1 H, J = 3.7 Hz), 5.88 (s, 1 H), 5.58 (d, 1 H, J = 3.1 Hz), 3.83 (s, 3 H), 3.61 (0.5 AB q, 1 H, J = 13.6 Hz), 3.22 (0.5 AB q, 1 H, J = 13.6 Hz), 2.33 (s, 6 H).

Ethyl 1-[2′-(Methylthio)phenyl]pyrrole-2-carboxylate (15a). To a cooled (5 °C) solution of **14a** (0.35 g, 1.8 mmol) and 2,6-lutidine (0.57 mL, 4.9 mmol) in anhydrous dioxane (3 mL) was added trichloroacetyl chloride (0.55 mL, 4.9 mmol), and the reaction mixture was heated at reflux for 3.5 h. After cooling, the mixture was poured into crushed ice and extracted with chloroform. After drying and evaporation, the crude product was dissolved in dry EtOH (2.5 mL), and sodium ethoxide (130 mg, 1.9 mmol) was added. After stirring for 30 min at room temperature, the mixture was adjusted to pH 7 with 0.5 N HCl and extracted with $CHCl_3$. The combined organic layers were washed with brine, dried, and concentrated. The residue was purified by flash chromatography ($CHCl_3$) to afford 395 mg of **15a** as a waxy solid: IR (neat) 1715 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 7.52–7.22 (m, 4 H), 7.13 (m, 1 H), 6.84 (t, 1 H, J = 1.9 Hz), 6.35 (m, 1 H), 4.13 (q, 2 H, J = 7.2 Hz), 2.34 (s, 3 H), 1.17 (t, 3 H, J = 7.1 Hz).

Methyl *N*-[2′-(Methylthio)-5′-(trifluoromethyl)phenyl]pyrrole-2-carboxylate (15b). Starting from **14b** (0.83 g, 1.94 mmol), trichloroacetyl chloride, sodium methoxide, and methanol, **15b** was obtained as a yellowish oil following the procedure described for **15a**: IR (neat) 1734, 1610, 1521, 806, 714 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 7.63 (dd, 1 H, J = 7.4, 1.2 Hz), 7.46

(s, 1 H), 7.32 (d, 1 H, J = 8.3 Hz), 7.10 (m, 1 H), 6.83 (t, 1 H, J = 1.8 Hz), 6.36 (t, 1 H, J = 3.7 Hz), 3.69 (s, 3 H), 2.39 (s, 3 H).

N-[2'-(Methylthio)phenyl]pyrrole-2-carboxylic Acid (16a). To a solution of **15a** (0.5 g, 1.9 mmol) in 10 mL of EtOH and THF (1/1) was added a solution of KOH (0.42 g, 7.6 mmol) in 3 mL of EtOH. The reaction mixture was heated at 60 °C for 20 h under argon. The solution was adjusted to pH 4 with 1 N HCl, the solvent was evaporated, and the mixture was extracted with EtOAc. The organic layers were washed with brine, dried, and concentrated. The residue was crystallized from benzene and gave 420 mg of **16a** as colorless prisms: IR (KBr) 1670 cm^{-1}; ^1H NMR (DMSO-d_6) δ 11.95 (br s, 1 H), 7.48–7.10 (m, 4 H), 7.08–6.81 (m, 2 H), 6.25 (m, 1 H), 2.17 (s, 3 H).

N-[2'-(Methylthio)-5'-(trifluoromethyl)phenyl]pyrrole-2-carboxylic Acid (16b). Similarly to compound **16a**, the acid **16b** was prepared (reaction time 20 h at 60 °C): R_f = 0.29 (EtOAc); IR (film) 1674 cm^{-1}; ^1H NMR (DMSO-d_6) δ 12.15 (br s, 1 H), 7.83 (d, 1 H, J = 8.4 Hz), 7.65–7.53 (m, 2 H), 7.14 (t, 1 H, J = 2.5 Hz), 7.03 (m, 1 H), 6.42 (t, 1 H, J = 2.4 Hz), 2.46 (s, 3 H).

N-[2'-(Methylthio)phenyl]pyrrole-2-carboxylic Acid Chloride (17a). To a solution of acid **16a** (10 g, 43 mmol) in anhydrous benzene (300 mL) was added dropwise a solution of thionyl chloride (15.5 mL, 210 mmol) in anhydrous benzene (100 mL). The reaction mixture was heated at reflux for 2 h under argon. After cooling, the solvent was removed under vacuum, and the residue was crystallized from ligroin to give 7.2 g of **17a** as pale yellow prisms: IR (film) 1745 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.60–7.00 (m, 6 H), 6.43 (m, 1 H), 2.36 (s, 3 H).

4H-Pyrrolo[2,1-c][1,4]benzothiazin-4-one (18a). Anhydrous aluminum chloride (265 mg, 1.98 mmol) was added portionwise to a solution of **17a** (0.5 g, 1.98 mmol) in anhydrous benzene (50 mL), under argon at room temperature. The reaction mixture was stirred at room temperature for 24 h and then the reaction quenched with 4 N HCl. The organic phase was separated, washed with 20% $NaHCO_3$ solution and brine, dried, and concentrated. The crude material was flash chromatographed (30% EtOAc in cyclohexane) to give 180 mg of **18a** as a colorless solid whose spectroscopic data are identical with those reported previously.[21]

8-(Trifluoromethyl)-4H-pyrrolo[2,1-c][1,4]benzothiazin-4-one (18b). To a solution of acid **16b** (0.57 g, 1.9 mmol) in anhydrous benzene (12 mL) was added a solution of 0.89 mL of thionyl chloride in anhydrous benzene (2 mL). The solution was heated at reflux for 4.5 h under argon. After cooling, the solvent was removed, the residue was taken up in anhydrous benzene (3 mL), and anhydrous aluminum chloride (0.265 g, 1.98 mmol) was added. The mixture was refluxed for 6 h under argon. The reaction mixture was worked up as for **18a**. The crude compound was purified by flash chromatography ($CHCl_3$) to give 272 mg of **18b** as colorless prisms: R_f = 0.47 (toluene); IR (film) 1640, 1185, 1115, 745 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.92 (s, 1 H), 7.85 (dd, 1 H, J = 3.0, 1.6 Hz), 7.50 (m, 2 H), 7.32 (dd, 1 H, J = 4.0, 1.5 Hz), 6.71 (dd, 1 H, J = 4.2, 3.0 Hz).

8-Chloro-4H-pyrrolo[2,1-c][1,4]benzothiazin-4-one (18c). A mixture of **20a** (0.852 g, 2.98 mmol), anhydrous zinc chloride (1.19 g, 8.75 mmol), and *o*-dichlorobenzene (4.5 mL) was heated at reflux for 5 h under argon. After cooling the reaction mixture was poured into crushed ice and extracted with $CHCl_3$. The organic layers were washed with brine, dried, and concentrated. The crude material was purified by flash chromatography (toluene) to give 0.58 g of **18c** as white prisms: R_f = 0.51 (toluene); IR (film) 1620, 1370, 780 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.81 (m, 1 H), 7.65 (m, 1 H), 7.31 (m, 3 H), 6.65 (m, 1 H).

6-Chloro-4H-pyrrolo[2,1-c][1,4]benzothiazin-4-one (18d). Starting from 4 g (14 mmol) of ester **20b**, the title compound was obtained following the procedure described for **18c**: R_f = 0.48 (toluene); IR (KBr) 1635, 1590, 1355, 1180, 740 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.75–7.70 (m, 3 H), 7.35–7.28 (m, 2 H), 6.67 (dd, 1 H, J = 3.9, 2.9 Hz).

N-[5'-Chloro-2'-[(ethoxycarbonyl)thio]phenyl]pyrrole (20a). Sodium borohydride (184 mg, 4.8 mmol) was added to a solution of disulfide **19a** (1 g, 2.4 mmol) in anhydrous EtOH (45 mL), the mixture was refluxed for 20 min under argon and then allowed to cool to room temperature, and ethyl chloroformate (5.19 g, 4.8 mmol) in anhydrous EtOH (2.3 mL) was slowly added. After stirring for 1 h at room temperature, the reaction mixture was poured into crushed ice and extracted with $CHCl_3$. The organic layers were washed with brine, dried, and concentrated. The oily residue was purified by flash chromatography (toluene) to give 0.9 g of **20a** as a pale yellow oil: R_f = 0.68 (toluene); IR (neat) 2990, 1725, 1150, 730 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.65–7.15 (m, 3 H), 6.80 (m, 2 H), 6.42 (m, 2 H), 4.35 (q, 2 H, J = 7.1 Hz), 1.40 (t, 3 H, J = 7.0 Hz).

N-[3'-Chloro-2'-[(ethoxycarbonyl)thio]phenyl]pyrrole (20b). Starting from 0.55 g (1.2 mmol) of **19b**, the title compound was prepared over a 1 h reaction time following the procedure described above. **20b** was purified by distillation (bp 143 °C/0.1 mmHg); IR (neat) 2990, 1725, 1580, 1490, 720 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.62 (m, 1 H), 7.40–7.35 (m, 2 H), 6.81 (t, 2 H, J = 2.2 Hz), 6.31 (t, 2 H, J = 2.2 Hz), 4.23 (q, 2 H, J = 7.0 Hz), 1.25 (t, 3 H, J = 7.2 Hz).

General Procedure for Preparation of Alcohols 21. This procedure is illustrated for preparation of (±)-4-hydroxy-4-phenyl-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (**21a**). To a solution of **18a** (1 g, 4.9 mmol) in anhydrous THF (15 mL) was added phenylmagnesium bromide [prepared from Mg turnings (0.48 g, 9.9 mmol) and bromobenzene (1.04 mL, 9.9 mmol)] dissolved in 20 mL of anhydrous ethyl ether within 1 h, and the mixture was heated at 30 °C for 2 h under argon. The cooled mixture was then poured into a cold 20% NH_4Cl solution (100 mL), stirred for an additional 30 min, and extracted with EtOAc. The combined organic layers were washed with brine, dried, and concentrated. Flash chromatography (EtOAc) gave 1.12 g of alcohol **21a** as a colorless oil: R_f = 0.46 (EtOAc); IR (neat) 3450–3300, 1605, 1370, 1020, 760 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.83 (m, 1 H), 7.78–7.00 (m, 9 H), 6.56 (m, 1 H), 6.18 (m, 1 H), 2.63 (br s, 1 H).

(±)-4-Hydroxy-4-(*p*-methoxyphenyl)-4H-pyrrolo[2,1-*c*][1,4]benzothiazine (21b). Starting from **18a** (1.8 g, 8.9 mmol) and *p*-bromoanisole, **21b** was obtained according to the procedure described above (reaction time 2 h at 40 °C) as a colorless oil: R_f = 0.25 (30% $CHCl_3$ in benzene); IR (neat) 3450–3300, 1599, 1040, 745 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.85 (d, 1 H, J = 8.5 Hz), 7.53 (d, 1 H, J = 8.6 Hz), 7.30–7.00 (m, 3 H), 6.95–6.78 (m, 3 H), 6.53 (m, 1 H), 6.19 (m, 1 H), 5.80 (m, 1 H), 3.86 (s, 3 H), 3.72 (br s, 1 H); MS *m/z* 309 (8, M^+), 292 (100), 276, 223, 186, 174, 168.

(±)-8-Chloro-4-hydroxy-4-phenyl-4H-pyrrolo[2,1-*c*][1,4]benzothiazine (21c). Starting from **18c** (0.2 g, 0.8 mmol) and bromobenzene, the title compound was prepared as a colorless oil following the procedure as for **21a** (reaction time 4 h at reflux) and was purified by flash chromatography (5% EtOAc in benzene): IR (neat) 3400–3300, 1589, 1346, 761 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.82 (m, 1 H), 7.60–6.95 (m, 7 H), 6.56 (m, 1 H), 6.28 (m, 1 H), 5.75 (m, 1 H), 3.22 (br s, 1 H).

(±)-8-Chloro-4-hydroxy-4-(*p*-methoxyphenyl)-4H-pyrrolo[2,1-*c*][1,4]benzothiazine (21d). Starting from 0.43 g (0.22 mmol) of thiolactone **18c** and *p*-bromoanisole, the title compound was obtained as a colorless oil (reaction time 30 h at room temperature) following the procedure described for **21a** except that the compound was purified by flash chromatography using benzene as eluent: IR (neat) 3500–3300, 1605, 1495, 1450, 755 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.76 (m, 2 H), 7.70–7.10 (m, 5 H), 6.83 (m, 1 H), 6.28 (m, 1 H), 5.66 (m, 1 H), 4.82 (br s, 1 H), 3.82 (s, 3 H); MS *m/z* 343 (13, M^+), 341, 326, 307, 292 (100), 276, 235, 220, 208.

(±)-4-Hydroxy-4-phenyl-8-(trifluoromethyl)-4H-pyrrolo[2,1-*c*][1,4]benzothiazine (21e). Starting from **18b** (0.54 g, 1.9 mmol) and bromobenzene, the alcohol **21e** was prepared as a pale yellow oil (reaction time 24 h at room temperature) following the procedure described for **21a**: R_f = 0.21 (benzene); IR (neat) 3450–3300, 1605, 1390, 745 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.95–7.65 (m, 3 H), 7.60–7.20 (m, 5 H), 6.70 (m, 1 H), 6.28 (m, 1 H), 5.67 (m, 1 H), 2.92 (br s, 1 H); MS *m/z* 347 (20, M^+), 330, 314, 254, 242 (100), 173.

(±)-4-Hydroxy-4-(*p*-methoxyphenyl)-8-(trifluoromethyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (21f). Starting from 0.2 g (0.74 mmol) of **18b** and *p*-bromoanisole, the title compound was obtained as a colorless oil (reaction time 18 h at room temperature) following the procedure described for **21a**: R_f = 0.18 (benzene); IR (neat) 3450–3300, 1605, 741 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.91 (m, 1 H), 7.63 (m, 1 H), 7.45–6.60 (m, 5 H), 6.51 (m, 1 H), 6.14 (m, 1 H), 5.68 (m, 1 H), 3.75 (s, 3 H), 2.41 (br s, 1 H); MS *m/z* 377 (8, M^+), 344, 301, 242, 223, 149 (100), 135.

(±)-6-Chloro-4-hydroxy-4-phenyl-4*H*-pyrrolo[2,1-*c*][1,4]-benzothiazine (21g). Starting from **18d** (0.7 g, 2.8 mmol), the title compound was obtained as a colorless oil (reaction time 4 h at reflux) following the procedure described for **21a**, except that **21g** was purified by flash chromatography using toluene as eluent: IR (neat) 3450–3300, 1605, 1095, 738 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.95–7.75 (m, 2 H), 7.60–7.25 (m, 6 H), 6.93 (m, 1 H), 6.38 (m, 1 H), 5.63 (m, 1 H), 2.79 (br s, 1 H).

General Procedure for the Preparation of Esters 22. This procedure is illustrated for the preparation of (±)-4-acetoxy-4-phenyl-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (**22a**). To a solution of **21a** (0.27 g, 0.9 mmol) and dry triethylamine (0.4 mL, 2.8 mmol) in anhydrous THF (10 mL) was added acetyl bromide (138 μL, 1.8 mmol) dissolved in anhydrous THF (2 mL). The reaction mixture was heated at reflux for 48 h under argon. The solvent was removed in vacuo, and the residue was partitioned between water and EtOAc. The organic layer was washed with brine, dried, and concentrated. The residue was purified by flash chromatography (15% hexanes in benzene) to give 380 mg of ester **22a** which crystallized as pale yellow prisms: R_f = 0.23 (15% hexanes in benzene); IR (film) 1725 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.85 (m, 2 H), 7.60–7.35 (m, 7 H), 6.93 (m, 1 H), 6.85 (m, 1 H), 6.28 (m, 1 H), 2.24 (s, 3 H).

(±)-4-Acetoxy-4-(*p*-methoxyphenyl)-4*H*-pyrrolo[2,1-*c*]-[1,4]benzothiazine (22b). Starting from **21b** (0.11 g, 0.35 mmol), the title compound was prepared (reaction time 24 h at 80 °C) following the procedure described for **22a** and crystallized as colorless prisms: IR (KBr) 1716, 741 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.87 (m, 2 H), 7.60–7.40 (m, 4 H), 6.92 (m, 3 H), 6.83 (m, 1 H), 6.31 (m, 1 H), 3.88 (s, 3 H), 2.24 (s, 3 H); MS *m/z* 351 (10, M^+), 309, 292, 276 (100), 233, 186, 174.

(±)-4-Acetoxy-8-chloro-4-phenyl-4*H*-pyrrolo[2,1-*c*][1,4]-benzothiazine (22c). Starting from 0.18 g (0.55 mmol) of **21c**, the title compound was obtained (reaction time 24 h at 80 °C) following the procedure as for **22a**. Ester **22b** was obtained as an amorphous solid after trituration with cyclohexane: IR (film) 1720 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.83 (m, 2 H), 7.65–7.35 (m, 6 H), 6.91 (m, 1 H), 6.86 (m, 1 H), 6.32 (m, 1 H), 2.23 (s, 3 H).

(±)-4-Acetoxy-8-chloro-4-(*p*-methoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (22d). Similarly to **22a**, the ester **22d** was prepared starting from 0.18 g (0.5 mmol) of **21d** (reaction time 30 h at 80 °C). An analytical sample was obtained after trituration with cyclohexane: IR (KBr) 1714 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.87 (m, 2 H), 7.46 (m, 2 H), 7.27 (m, 1 H), 6.93 (m, 2 H), 6.89 (m, 1 H), 6.84 (m, 1 H), 6.33 (m, 1 H), 3.88 (s, 3 H), 2.23 (s, 3 H); MS *m/z* 385 (12, M^+), 360, 340, 326, 310 (100), 254, 241, 220, 208.

(±)-4-Acetoxy-4-phenyl-8-(trifluoromethyl)-4*H*-pyrrolo-[2,1-*c*][1,4]benzothiazine (22e). Starting from 0.17 g (0.49 mmol) of alcohol **21e**, the title compound was obtained as colorless prisms (reaction time 48 h at 80 °C) following the procedure described for **22a**. **22e** was purified by flash chromatography (5% EtOAc in benzene): IR (film) 1723 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.84 (m, 2 H), 7.71 (m, 2 H), 7.72–7.38 (m, 4 H), 6.96–6.87 (m, 2 H), 6.37 (m, 1 H), 2.27 (s, 3 H).

(±)-4-Acetoxy-4-(*p*-methoxyphenyl)-8-(trifluoromethyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (22f). Starting from **21f** (50 mg, 0.13 mmol), the title compound was obtained (reaction time 40 h at 80 °C) following the procedure described for **22a**. After flash chromatography (5% EtOAc in benzene), **22f** was obtained as colorless prisms: IR (film) 1725 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.86 (m, 2 H), 7.69 (m, 3 H), 7.05–6.85 (m, 4 H), 6.35 (m, 1 H), 3.88 (s, 3 H), 2.26 (s, 3 H); MS *m/z* 419 (10, M^+), 377, 360, 344 (100), 314, 254, 242, 135.

(±)-4-Acetoxy-6-chloro-4-phenyl-4*H*-pyrrolo[2,1-*c*][1,4]-benzothiazine (22g). Starting from **21g** (107 mg, 0.34 mmol), the title compound was obtained (reaction time 24 h at 80 °C) following the procedure as for **22a**. After purification by flash chromatography ($CHCl_3$), **22g** crystallized as pale yellow prisms: IR (KBr) 1720, 1630, 1410, 1280, 730 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.81 (m, 2 H), 7.63–7.35 (m, 6 H), 6.87 (m, 2 H), 6.31 (m, 1 H), 2.27 (s, 3 H).

General Procedure for Preparation of Pyrrolobenzothiazines 23. This procedure is illustrated for preparation of 4,4-diphenyl-4*H*-pyrrolo[2,1-c][1,4]benzothiazine (**23a**). To Mg turnings (0.23 g, 9.92 mmol) in anhydrous THF (30 mL) was slowly added a solution of bromobenzene (1.04 mL, 9.5 mmol) in anhydrous THF (10 mL) under argon. After refluxing for 1 h, a solution of **18a** (1 g, 4.9 mmol) in anhydrous THF (40 mL) was slowly added under reflux. The reaction mixture was refluxed for 4 h under argon. The cooled mixture was poured into a cold 20% NH_4Cl solution (40 mL), stirred for 30 min, and extracted with EtOAc. The combined organic layers were washed with brine, dried, and concentrated. Flash chromatography (toluene) and recrystallization gave 1.31 g of **23a** as pale yellow prisms (21% of **21a** was recovered): R_f = 0.72 (toluene); IR (KBr) 3090, 2950, 1605, 702 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.40–7.06 (m, 14 H), 6.94 (dt, 1 H, *J* = 7.6, 1.4 Hz), 6.30 (t, 1 H, *J* = 3.2 Hz), 5.57 (m, 1 H); MS *m/z* 339 (46, M^+), 262 (100), 230, 204, 165, 139.

4,4-Bis(*p*-methoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (23b). Starting from 1.8 g (8.9 mmol) of **18a**, the title compound was obtained as white microcrystals (reaction time 2 h at reflux) following the procedure described for **23a**: R_f = 0.63 (toluene); IR (film) 1596, 1360, 745 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.40–7.05 (m, 8 H), 6.95 (m, 1 H), 6.74 (m, 4 H), 6.29 (t, 1 H, *J* = 3.3 Hz), 5.58 (m, 1 H), 3.76 (s, 6 H); MS *m/z* 399 (52, M^+), 384, 310, 292 (100), 264, 249, 311, 173.

4,4-Diphenyl-8-(trifluoromethyl)-4*H*-pyrrolo[2,1-*c*][1,4]-benzothiazine (23c). Starting from 0.5 g (1.87 mmol) of **18b**, the title compound was obtained (reaction time 3 h at 60 °C) (73%) as colorless prisms: IR (film) 3080, 1602, 738 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.35 (s, 1 H), 7.41 (d, 1 H, *J* = 8.2 Hz), 7.24 (m, 12 H), 6.34 (t, 1 H, *J* = 3.7 Hz), 5.59 (m, 1 H); MS *m/z* 407 (28, M^+), 388, 330 (100), 298, 260, 241, 204, 189.

4,4-Bis(*p*-methoxyphenyl)-8-(trifluoromethyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (23d). Starting from 0.5 g (1.87 mmol) of **18b**, the title compound was obtained (reaction time 3 h at reflux) (68%) as colorless prisms: IR (film) 1608, 1360, 1250 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.51 (s, 1 H), 7.44 (d, 1 H, *J* = 8.1 Hz), 7.32–7.08 (m, 6 H), 6.75 (m, 4 H), 6.33 (m, 1 H), 5.61 (m, 1 H), 3.76 (s, 6 H); MS *m/z* 467 (60, M^+), 448, 408, 360 (100), 317, 291, 264, 241, 211.

8-Chloro-4,4-diphenyl-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (23e). Similarly to **23a**, starting from 107 mg (0.45 mmol) of **18c**, the title compound was obtained (ethyl ether as solvent, reaction time 2 h at reflux) as colorless prisms: IR (film) 1599, 1250, 740 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.40–7.10 (m, 13 H), 6.90 (dd, 1 H, *J* = 9.2, 2.1 Hz), 6.31 (t, 1 H, *J* = 3.0 Hz), 5.57 (m, 1 H); MS *m/z* 373 (58, M^+), 296 (100), 261, 204, 165.

6-Chloro-4,4-diphenyl-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (23f). Starting from 1.4 g (5.6 mmol) of **18d**, the title compound was obtained (reaction time 2 h at reflux) following the procedure described for **23a**: R_f = 0.62 (toluene); IR (film) 1605, 740 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.45–7.12 (m, 12 H), 7.08 (m, 2 H), 6.38 (m, 1 H), 5.67 (m, 1 H).

1-[(*N*,*N*-Dimethylamino)methyl]-4,4-diphenyl-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (24a). Starting from **23a** (180 mg, 0.53 mmol), **24a** was obtained as colorless prisms according to the procedure described for **8a**, except that the reaction mixture was heated at 30 °C for 1.5 h: IR ($CHCl_3$) 3335, 2890, 1600 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.26 (m, 1 H), 7.40–6.84 (m, 13 H), 6.12 (d, 1 H, *J* = 3.1 Hz), 5.38 (d, 1 H, *J* = 3.0 Hz), 3.33 (s, 2 H), 2.35 (s, 6 H).

1-[(*N*,*N*-Dimethylamino)methyl]-4,4-bis(*p*-methoxyphenyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (24b). Starting from 150 mg (0.37 mmol) of **23b**, the title compound was obtained using an identical procedure as for **8a** (reaction time 2 h at 30 °C). **24b** recrystallized as colorless prisms: R_f = 0.49 (EtOAc); IR ($CHCl_3$) 1599, 1420, 730 cm^{-1}; ^1H NMR

($CDCl_3$) δ 8.23 (d, 1 H, J = 8.1 Hz), 7.34–7.15 (m, 6 H), 6.92 (t, 1 H, J = 6.4 Hz), 6.71 (m, 4 H), 6.13 (d, 1 H, J = 3.1 Hz), 5.42 (d, 1 H, J = 3.3 Hz), 3.74 (s, 6 H), 3.31 (s, 2 H), 2.32 (s, 6 H).

1-[(*N*,*N*-Dimethylamino)methyl]-4,4-diphenyl-8-(trifluoromethyl)-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (24c). The title compound was prepared according to the procedure described for **8a**, starting from **23c** (50 mg, 0.12 mmol) (reaction time 12 h at room temperature). **24c** was flash chromatographed ($CHCl_3$ and hexanes, 1/1): R_f = 0.56 ($CHCl_3$ and hexanes, 1/1); IR (film) 1600, 1380, 750 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.72 (m, 1 H), 7.53 (m, 1 H), 7.33–7.00 (m, 11 H), 6.23 (d, 1 H, J = 3.2 Hz), 5.39 (d, 1 H, J = 3.1 Hz), 3.31 (s, 2 H), 2.32 (s, 6 H).

6-Chloro-1-[(*N*,*N*-dimethylamino)methyl]-4,4-diphenyl-4*H*-pyrrolo[2,1-*c*][1,4]benzothiazine (24d). The title compound was prepared according to the procedure described for **8a**, starting from **23f** (180 mg, 0.48 mmol) (reaction time 10 h at room temperature). **24d** was flash chromatographed (EtOAc): R_f = 0.22 (EtOAc); IR (film) 1599, 1295, 740 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.25 (m, 1 H), 7.35–7.00 (m, 12 H), 6.13 (d, 1 H, J = 3.1 Hz), 5.43 (d, 1 H, J = 3.5 Hz), 3.27 (s, 2 H), 2.33 (s, 6 H).

(±)-4-Hydroxy-2,3,3a,4-tetrahydro-1*H*-pyrrolo[2,1-*c*][1,4]benzothiazin-1-one (26). A 1 M solution of $LiB(Et)_3H$ in THF (7 mL, 7 mmol) was added dropwise to a solution of disulfide **25** (0.93 g, 1.76 mmol) in anhydrous THF (10 mL) at −40 °C under argon. The reaction mixture was stirred at −40 °C for 1 h and then allowed to warm to room temperature and stirred for an additional 1.5 h. The mixture was poured into ice-cold 2 N HCl (10 mL) and extracted with EtOAc. The collected organic extracts were washed with a little water, dried, and concentrated. The residue was purified by flash chromatography (5% EtOAc in $CHCl_3$) and recrystallized to give 615 mg of hemithioacetal **26** as colorless prisms: R_f = 0.38 (5% EtOAc in $CHCl_3$); IR (KBr) 3460–3400, 1670, 1290, 765 cm^{-1}; ^1H NMR (DMSO-d_6) δ 7.94 (dd, 1 H, J = 8.5, 1.5 Hz), 7.20–7.00 (m, 3 H), 6.79 (d, 1 H, J = 4.8 Hz), 5.36 (dd, 1 H, J = 4.8, 1.5 Hz), 4.07 (m, 1 H), 2.50–1.90 (m, 4 H).

(±)-4-Chloro-2,3,3a,4-tetrahydro-1*H*-pyrrolo[2,1-*c*][1,4]benzothiazin-1-one (27). Thionyl chloride (2.3 mL, 31.5 mmol) dissolved in 10 mL of dry CH_2Cl_2 was added dropwise to a solution of the hemithioacetal **26** (1 g, 4.5 mmol) in 40 mL of dry CH_2Cl_2. The solution was stirred for 10 h at 40 °C under argon. After removal of the solvent by rotary evaporation, the residue was purified by flash chromatography (8% EtOAc in $CHCl_3$) (R_f = 0.43) to give 0.82 g of **27** which was used in the next step without further purification.

(±)-*trans*-4-Phenyl-2,3,3a,4-tetrahydro-1*H*-pyrrolo[2,1-*c*][1,4]benzothiazin-1-one (28a). A mixture of **27** (0.7 g, 2.9 mmol) and anhydrous aluminum chloride (0.46 g, 3.48 mmol) in anhydrous benzene (20 mL) was refluxed for 2 h under argon. After cooling, cold water (15 mL) was added and the organic phase was separated, washed with brine, and dried. After concentration, the crude product was purified by flash chromatography ($CHCl_3$) to give 0.7 g of **28a** which by ^1H NMR analysis consisted of a 85/15 mixture of isomers: the major isomer showed a *trans* configuration for protons at C-3a and C-4. By fractional crystallization *trans*-**28a** was isolated as colorless prisms: IR (KBr) 2935, 1705, 1460, 1200, 755 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.39 (m, 1 H), 7.40 (m, 5 H), 7.20–7.00 (m, 3 H), 4.16 (m, 2 H), 2.52 (m, 2 H), 2.03 (m, 1 H), 1.75 (m, 1 H); ^1H NMR ($CDCl_3$/DMSO-d_6, 5/3) δ 8.38 (d, 1 H, J = 5.6 Hz), 7.54–6.94 (m, 8 H), 4.35 (d, 1 H, J = 9.9 Hz), 4.14 (m, 1 H), 2.52 (m, 2 H), 2.15 (m, 1 H), 1.75 (m, 1 H).

(±)-*trans*-4-(*p*-Methoxyphenyl)-2,3,3a,4-tetrahydro-1*H*-pyrrolo[2,1-*c*][1,4]benzothiazin-1-one (28b). To a cooled (0 °C) mixture of anhydrous aluminum chloride (0.5 g, 3.79 mmol) and anisole (0.6 mL, 5.57 mmol) in anhydrous 1,2-dichloroethane (7 mL) was added a solution of **27** (0.31 g, 1.29 mmol) and nitromethane (70 μL) in anhydrous 1,2-dichloroethane (7 mL). After stirring at 0 °C for 30 min, the solution was heated at 70 °C for 3 h under argon. After cooling, cold water (10 mL) was added and the mixture was extracted with $CHCl_3$. The organic layers were washed with brine and dried. After concentration, the crude product was purified by flash chromatography (10% $CHCl_3$ in benzene) to give 269 mg of **28b** which by ^1H NMR analysis consisted of a 85/15 mixture of isomers; the major isomer showed a *trans* configuration for protons at C-3a and C-4. By fractional crystallization *trans*-**28b** was isolated as colorless prisms: IR (KBr) 2985, 1710, 1620, 1485, 1265, 1125, 750 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.36 (d, 1 H, J = 8.1 Hz), 7.40–6.82 (m, 7 H), 4.12 (m, 2 H), 3.82 (s, 3 H), 2.54 (m, 2 H), 2.04 (m, 1 H), 1.75 (m, 1 H); ^{13}C NMR ($CDCl_3$) δ 21.9, 30.2, 48.7, 55.3, 60.9, 114.5, 122.0, 124.5, 124.8, 124.9, 125.5, 128.2, 129.8, 133.3, 159.8, 173.8; MS m/z 311 (10, M^+), 282, 268, 249, 190 (100), 174, 136.

Molecular Modeling. Computational Procedure. Molecular mechanic calculations were performed on the synthesized molecules and diltiazem with the use of the molecular modeling programs MODEL (version KS 2.99) on a VAX 6610 workstation and SYBYL (version 5.50) on a Silicon Graphics personal IRIS4D/35 workstation. X-ray crystal coordinates were used as input geometry for the diltiazem molecule. All the remaining structures were generated by the DRAW option within MODEL and initially energy minimized by the MM2/M routine of the same program until convergence. Systematic conformational analyses were performed by using the random search mixed method. Only the conformers with a conformational energy window of 3 kcal/mol over the lowest minimum were considered. The final conformationally characterized structures were transferred to the program SYBYL via the TTY/DATA option of the MODEL program. Least-squares matchings were performed in SYBYL by employing the MATCH routine within the COMPARISON menu. Electrostatic isopotential maps were computed by using the POTENTIAL command in SYBYL. One-level surfaces of studied compounds were countered at a potential energy value of −12 and −12 kcal/mol. Atomic point changes used to calculate the isopotential maps were obtained by the MNDO method of MOPAC through a Mulliken electron population analysis. Least-squares fitting of atoms used in the isopotential map superimpositions was carried out by employing the FIT option of SYBYL.

Radioligand-Binding Assays. Tissue homogenate of cerebral cortex and heart containing calcium channel receptors was prepared according to Ehlert *et al.*[27] Cerebral cortices of male Sprague–Dawley rats were homogenized (100 mg/mL in 50 mM Na-Hepes buffer, pH 7.4). Hearts were also removed, perfused through the aorta with ice-cold saline solution, and homogenized (100 mg/mL in 50 mM Na-Hepes buffer, pH 7.4). Subsequently, the cardiac homogenates were filtered through four layers of cheese cloth. Both cortical and cardiac homogenates were washed five times by centrifugation for 10 min at 48000g. The final pellet was resuspended to a concentration of 50 mg of original wet tissue wt/mL of buffer and stored at −70 °C for the assay. Binding determination (RRA): the receptor binding assay was determined as follows. Tissue homogenate (200 μL) was incubated for 90 min in a dark room at 0 °C with 100 μL of [^{3}H]nitrendipine (3×10^{-10} M, 87 Ci/mmol; NEN) and 100 μL of the test compound (dissolved in 5% DMSO) in 50 mM of Na-Hepes buffer, pH 7.4 (total vol 2 mL). The incubations were stopped by adding 4 mL of cold buffer followed by rapid filtration through glass fiber filter disks. The samples were subsequently washed three times with 4.5 mL of the same buffer and placed into scintillation vials; 10 mL of Filter-Count (Packard) liquid scintillation cocktail was then added to each vial, and counting was carried out by a scintillation spectrometer (Packard T.-C. 300C). Nonspecific binding was defined as nondisplaceable binding in the presence of 1×10^{-5} of unlabeled diltiazem and verapamil and specific binding as the difference between total and nonspecific binding. Inhibition of [^{3}H]nitrendipine by diltiazem and verapamil is ca. 70%, according to ref 27. Blank experiments were carried out to determine the effect of the solvent DMSO (5%) on the binding. The concentration of the test compounds that inhibited [^{3}H]nitrendipine binding by 50% (IC_{50}) was determined by log-probit analysis with six concentrations of the displacers, each performed in duplicate. The IC_{50} values obtained were used to calculate apparent inhibition constants (K_i) by the Prusoff method[28] by the following equation: $K_i = IC_{50}/(1 + S/K_d)$, where S represents the

concentration of the ligand used and K_d is its receptor dissociation constant, obtained by Scatchard analysis (K_d value of [^{3}H]nitrendipine was 0.25 nM).

Pharmacology. The pharmacological profile of the compounds was assessed on guinea pig isolated left and right atria to evaluate their negative inotropic and chronotropic effects and K^+-depolarized guinea pig artery strips to test their calcium antagonist activity. All the compounds were first checked at increasing doses to examine the dose-dependent decrease both of the developed tension in the left atria driven at 1 Hz and of the frequency in spontaneously beating right atria and then to measure the inhibitory effect on K^+-evoked contractions in guinea pig helicoidal aortic strips. ED_{50}, ED_{30}, and IC_{50} values were evaluated from log concentration–response curves in the appropriate pharmacological preparation. Guinea pigs (300–400 g male and female) were sacrificed by cervical dislocation. After thoracotomy, the hearts were immediately removed and washed by perfusion through the aorta with oxygenated Tyrode solution of the following composition (mM): 136.9 NaCl, 5.4 KCl, 2.5 $CaCl_2$, 1.0 $MgCl_2$, 0.4 $NaH_2PO_4 \cdot H_2O$, 11.9 $NaHCO_3$, and 5.5 glucose. The physiological salt solution (PSS) was buffered to pH 7.4 by saturation with 95% O_2–5% CO_2 gas, and the temperature was maintained at 35 °C. Isolated guinea pig heart preparations were used, spontaneously beating right atria and left atria driven at 1 Hz. For each preparation, the entire left and right atria were dissected from ventricles, cleaned of excess tissue, and hung vertically in a 15 mL organ bath containing the PSS continuously bubbled with 95% O_2–5% CO_2 gas at 35 °C, pH 7.4. The contractile activity was recorded isometrically by means of a force transducer (FT 0.3, Grass Instruments, Quincy, MA) connected to a pen recorder (KV 380, Battaglia-Rangoni, Bologna, Italy). The left atria were stimulated by rectangular pulses of 0.6–0.8 ms duration and *ca.* 50% threshold voltage through two platinum contact electrodes in the lower holding clamp (Grass S88 stimulator). After the tissue was beating for several minutes, a length–tension curve was determined, and the muscle length was maintained at which elicited 90% of maximum contractile force observed at the optimal length. A stabilization period of 45–60 min was allowed before the atria were challenged by various agents. During this equilibration period, the bathing solution was changed every 15 min and the threshold voltage was ascertained for the left atria. Atrial muscle preparations were used to examine the inotropic and chronotropic activity of the compounds (0.1, 0.5, 1, 5, 10, 50, and 10 μM) first dissolved in DMSO and then diluted with PSS. According to this procedure, the concentration of DMSO in the bath solution never exceeded 0.3%, a concentration which did not produce appreciable inotropic and chronotropic effects. During generation of cumulative dose–response curves, the next higher concentration of the compounds was added only after the preparation reached a steady state. The thoracic aorta was removed and placed in Tyrode solution of the following composition (mM): 118 NaCl, 4.75 KCl, 2.54 $CaCl_2$, 1.20 $MgSO_4$, 1.19 KH_2PO_4, 25 $NaHCO_3$, and 11 glucose, equilibrated with 95% O_2 and 5% CO_2 gas at pH 7.4. Vessel was cleaned of extraneous connective tissue. Two helicoidal strips (10 mm × 1 mm) were cut from each aorta beginning from the end most proximal to the heart. Vascular strips were then tied with surgical thread (6–0) and suspended in a jacketed tissue bath (15 mL) containing aerated PSS at 35 °C. Strips were secured at one end to Plexiglass hooks and connected via the surgical thread to a force displacement (FT 0.3, Grass) transducer for monitoring changes in isometric contraction. Aortic strips were subjected to a resting force of 1 g and washed every 20 min with fresh PSS for 1 h. After the equilibration period, guinea pig aortic strips were contracted by being washed in PSS containing 80 mM KCl (equimolar substitution of K^+ for Na^+). Subsequent to the contraction reaching a plateau (*ca.* 30 min), the compounds (0.1, 0.5, 1, 5, 10, 50, 100, and 500 μM) were added cumulatively to the bath allowing for any relaxation to obtain an equilibrate level of force. Addition of the drug vehicle had no appreciable effect on the K^+-induced level of force (DMSO for all compounds). Data were analyzed by Student's *t*-test. The criterion for significance was a *p* value <0.05. The ED_{50}, ED_{30}, and IC_{50} values were calculated from log concentration–response curves (Probit analysis by Litchfield and Wilcoxon, $n = 6–8$). All data are presented as mean ± SEM.[44]

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