5-Methyl-6-phenyl-1,3,5,6-tetrahydro-3,6-methano-1,5-benzodiazocine-2,4-dione (BA 41899): Representative of a Novel Class of Purely Calcium-Sensitizing Agents

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BA 41899 (5-methyl-6-phenyl-1,3,5,6-tetrahydro-3,6-methano-1,5-benzodiazocine-2,4-dione, **6**) is a structurally novel 1,5-benzodiazocine derivative and represents the prototype of a hitherto unknown class of positive inotropic Ca^{2+}-sensitizing agents. It is completely devoid of phosphodiesterase (PDE) III inhibitory activity or any other known inotropic mechanism. BA 41899 (**6**) exhibits a pharmacological *in vitro* profile comprising Ca^{2+}-sensitizing, positive inotropic, and negative chronotropic effects. CGP 48506 ((+)-**6**), the (+)-enantiomer of BA 41899 (**6**), enantiospecifically carries Ca^{2+} sensitization by up to a full pCa unit and a corresponding positive inotropic effect. Conversely, the negative chronotropic action resides in the corresponding (−)-enantiomer, CGP 48508 ((−)-**6**). All the effects are exerted in the low micromolar range. The positive inotropic action of CGP 48506 ((+)-**6**) is associated with a decelerating effect on contraction and, more prominently, relaxation dynamics in isolated guinea pig atria. In contrast to Ca^{2+}-sensitizing PDE inhibitors, CGP 48506 ((+)-**6**) does not increase maximum Ca^{2+}-activated force in myocardial skinned fibers.

Introduction

Congestive heart failure (CHF) is a common cardiovascular disease. It can be defined as a clinical syndrome caused by an abnormality of the heart and/or peripheral circulation and is manifested in a characteristic pattern of hemodynamic, renal, neural, and hormonal responses. CHF now affects approximately 1−2% of the population of the Western world.[1-3] Approximately 50% of patients die within 5 years of onset of symptoms, and the annual mortality rate for patients with severe heart failure is around 60%.[4] CHF is thus a considerable medical problem and one likely to be aggravated in the foreseeable future, owing to the rising proportion of elderly persons in the population.[5]

All established inotropic agents exert their action via a more or less direct increase in the amount of excitatory free Ca^{2+}. This extra Ca^{2+} may potentially be harmful and lead to unwanted side effects associated with Ca^{2+} overload, like cardiac necrosis, tachycardia, and arrhythmia. In addition, these inotropic agents, with the exception of cardiac glycosides, increase the oxygen requirements of the myocardium, owing both to the greater force production and to the extra energy required for pumping increased systolic Ca^{2+} concentrations back to the diastolic level.[6,7] The clinical use of existing positive inotropic agents, especially in chronic treatment, is therefore limited.

As, with rare exceptions like hypocalcemia, CHF is not due to reduced availability of activator Ca^{2+}, an inotropic regimen not involving an increase in cytoplasmic Ca^{2+} should be much safer. Such considerations led to the concept of pharmacological modulation of Ca^{2+} sensitivity first described by Herzig et al. in 1981.[8] Ca^{2+} sensitization is characterized by a leftward shift of the Ca^{2+} activation curve. Contractility is enhanced at unaltered amplitude of the Ca^{2+} transient by increasing the responsiveness of the contractile myofilaments to a given Ca^{2+} ion concentration. Consequently, maximum force is reached at lower Ca^{2+} concentration, thus reducing both the likelihood of unwanted side effects due to Ca^{2+} overload and the energy necessary for Ca^{2+} cycling within the cell. Several physiological and pathophysiological conditions are known to increase (Frank−Starling mechanism, ischemic arrest, α-adrenergic stimulation, myosin phosphorylation) or decrease (postischemic stunning, hypoxia, troponin I phosphorylation, acidosis, β-adrenergic stimulation, inorganic phosphate, P_i, accumulation) Ca^{2+} sensitivity. Likewise, a variety of pharmacological agents from different structural classes, e.g., sulmazole,[9] isomazole,[10] pimobendan,[11] adibendan,[12] APP 201-533,[13] DPI 201-106,[14] or EMD 53998 and its derivatives,[15-18] with Ca^{2+}-sensitizing properties have been described (Figure 1). However, up to now, all published compounds with considerable Ca^{2+}-sensitizing efficacy and potency were, concomitantly, phosphodiesterase (PDE) III inhibitors. Even the most selective Ca^{2+} sensitizers, Merck−Darmstadt's thiadiazinones (e.g., EMD 57033), were reported to be at best equipotent with respect to Ca^{2+} sensitization and PDE III inhibitory activity. Therefore, their net profiles must include effects due to both increased cAMP and Ca^{2+}.

As a result of our efforts in screening for Ca^{2+}-sensitizing agents devoid of PDE III inhibitory activity using myocardial skinned fibers,[8] a structurally novel 1,5-benzodiazocine derivative, BA 41899 (**6**), was identified. BA 41899 (**6**) represents a hitherto unknown class of Ca^{2+}-sensitizing agents which is completely devoid of PDE III inhibitory activity or any other known inotropic mechanism. Herein we describe the synthesis and biological profile of BA 41899 (**6**) and closely related analogs.

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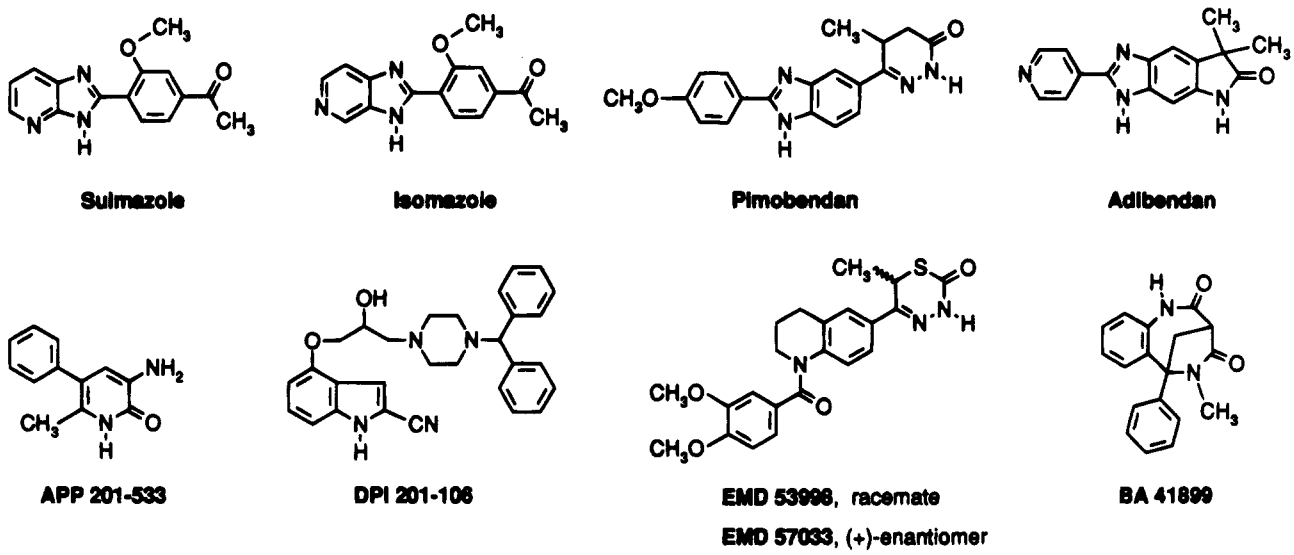


Figure 1. Calcium-sensitizing agents.

Table 1. Physical Data for Compounds 1–12

compd	formula	scheme[a]	mp, °C (solvent[b])	FAB-MS	analysis[c]
1	$C_{17}H_{15}NO_2$	1	190–191[d] (EtOAc)	266 ($M^+ + H$)	C, H, N
2	$C_{17}H_{14}O_3$	1	160–162[e] (Et_2O)	267 ($M^+ + H$)	C, H
(+)-**2**	$C_{17}H_{14}O_3$	3	amorphous[f]	267 ($M^+ + H$)	C, H
(−)-**2**	$C_{17}H_{14}O_3$	3	amorphous[g]	267 ($M^+ + H$)	C, H
3	$C_{17}H_{13}NO_2 \cdot 0.25H_2O$	1	262–263 (EtOAc)	264 ($M^+ + H$)	C, H, N
4	$C_{18}H_{15}NO_2$	1	181–182 (*i*-PrOH)	278 ($M^+ + H$)	C, H, N
(+)-**4**	$C_{18}H_{15}NO_2$	2, 3	208–210 (*i*-PrOH)[h]	278 ($M^+ + H$)	C, H, N
(−)-**4**	$C_{18}H_{15}NO_2$	2, 3	208–210 (*i*-PrOH)[i]	278 ($M^+ + H$)	C, H, N
5	$C_{18}H_{16}N_2O_2$	2	223–224 (EtOAc/hexane)	293 ($M^+ + H$)	C, H, N
6 (BA 41899)	$C_{18}H_{16}N_2O_2$	2	270–271 (Et_2O)	293 ($M^+ + H$)	C, H, N
(+)-**6** (CGP 48506)	$C_{18}H_{16}N_2O_2 \cdot 0.15H_2O$	2	amorphous[j]	293 ($M^+ + H$)	C, H, N
(−)-**6** (CGP 48508)	$C_{18}H_{16}N_2O_2 \cdot 0.2H_2O$	2	138–140 (Et_2O)[k]	293 ($M^+ + H$)	C, H, N
7	$C_{18}H_{16}N_2O_2$	1	236–237 (Et_2O)	293 ($M^+ + H$)	C, H, N
(+)-**7**	$C_{18}H_{16}N_2O_2$	2	221–222 (Et_2O)[l]	293 ($M^+ + H$)	C, H, N
(−)-**7**	$C_{18}H_{16}N_2O_2$	2	225–227 (Et_2O)[m]	293 ($M^+ + H$)	C, H, N
8	$C_{18}H_{15}NO_2$	1	140–142 (EtOAc/hexane)[n]	278 ($M^+ + H$)	C, H, N
9	$C_{18}H_{16}N_2O_2$	1	236–238 (CH_2Cl_2/hexane)	293 ($M^+ + H$)	C, H, N
10	$C_{18}H_{16}N_2O_2$	1	254–257 (CH_2Cl_2/hexane)	293 ($M^+ + H$)	C, H, N
11	$C_{24}H_{27}N_3O_2$	2	125–126 (Et_2O/hexane)	390 ($M^+ + H$)	C, H, N
12	$C_{24}H_{27}N_3O_2$	2	oil	390 ($M^+ + H$)	C, H, N

[a] See the Experimental Section for representative procedures. [b] Recrystallization or trituration solvent. [c] Analysis for C, H, and N within ±0.4%. [d] Lit.[19] mp 187 °C. [e] Lit.[21] mp 161.5–163 °C. [f] $[\alpha]_D$ +29° (c = 1.2, CH_2Cl_2). [g] $[\alpha]_D$ −27° (c = 1.2, CH_2Cl_2). [h] $[\alpha]_D$ +171° (c = 0.72, CH_2Cl_2). [i] $[\alpha]_D$ −170° (c = 0.72, CH_2Cl_2). [j] $[\alpha]_D$ +89° (c = 1.0, CH_2Cl_2). [k] $[\alpha]_D$ −88° (c = 1.0, CH_2Cl_2). [l] $[\alpha]_D$ −141° (c = 1.1, CH_2Cl_2). [m] $[\alpha]_D$ +139° (c = 1.1, CH_2Cl_2). [n] Lit.[24] mp 138–140 °C.

Chemistry

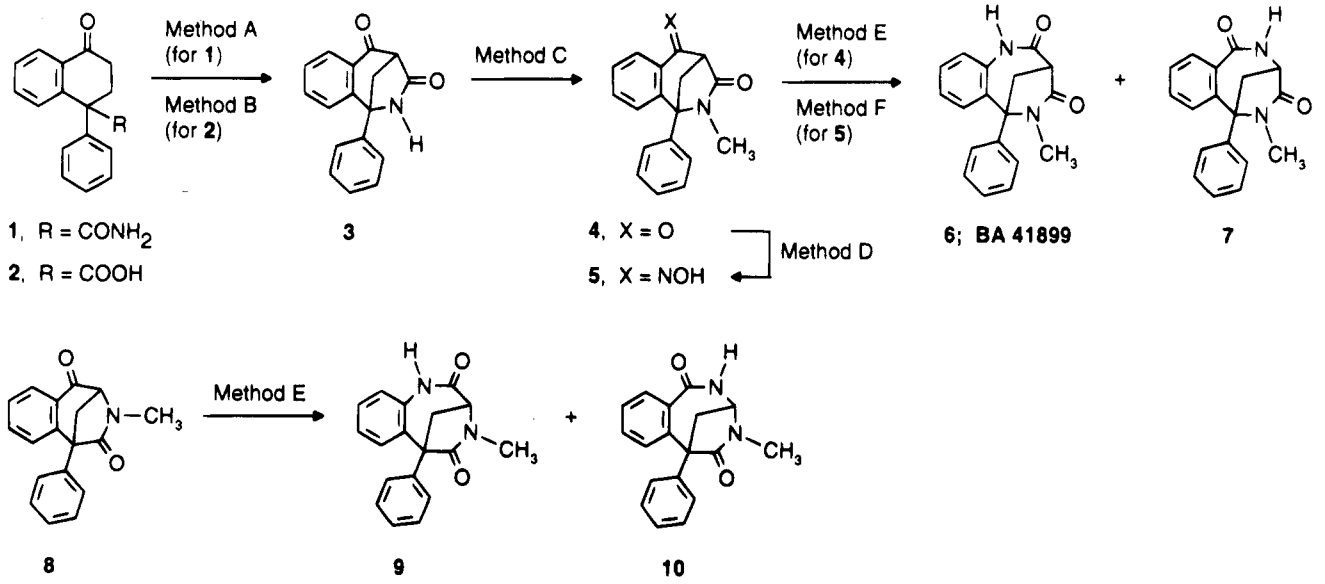
Representative reaction conditions are provided in the Experimental Section describing the preparation and characterization of all the compounds which are comprised in this report. Their structures are shown in Schemes 1–3, and analytical data are listed in Table 1.

1,5- And 2,5-benzodiazocines **6** and **7**, and the corresponding 1,4- and 2,4-isomers **9** and **10**, respectively, are structurally novel and representatives of a hitherto unknown class of compounds. They were prepared as outlined in Scheme 1. Treatment of carboxamide **1**[19] with lead tetraacetate in the presence of triethylamine (method A)[20] or reaction of carboxylic acid **2**[21] with diphenyl phosphorazidate (DPPA)[22] and 1,8-diazabicyclo-[5.4.0]undec-7-ene (DBU) as the base (method B) gave the bicyclic ketone **3** (50–60%) which was converted to the N-methylated amide **4** (90%) using iodomethane and powdered potassium carbonate in DMF (method C). Subsequent Schmidt rearrangement[23] with sodium azide in 98% sulfuric acid (method E) gave the title compound, 1,5-benzodiazocine BA 41899 (**6**) (48%), along with the corresponding 2,5-isomer **7** (37%), which were readily separated by flash chromatography. Analogously, 1,4- and 2,4-diazocines **9** (20%) and **10** (50%), respectively, were prepared starting from ketone **8**.[24]

BA 41899 (**6**) was also available by Beckmann rearrangement.[25] Thus, treatment of ketone **4** with hydroxylamine in pyridine (method D) gave the *E*-configured oxime **5** (83%) which reacted with trifluoromethanesulfonic acid anhydride (Tf_2O) in pyridine to yield the corresponding trifluoromethanesulfonate. Addition of water to the reaction mixture and heating (method F) gave BA 41899 (**6**) as the sole rearrangement product (40%). As expected, isomer **7** was not formed under these reaction conditions. The use of Tf_2O is essential. The reaction completely failed if other activating agents such as *p*-toluenesulfonyl chloride or methanesulfonyl chloride were used, and only unreacted oxime sulfonates were isolated even after prolonged heating.

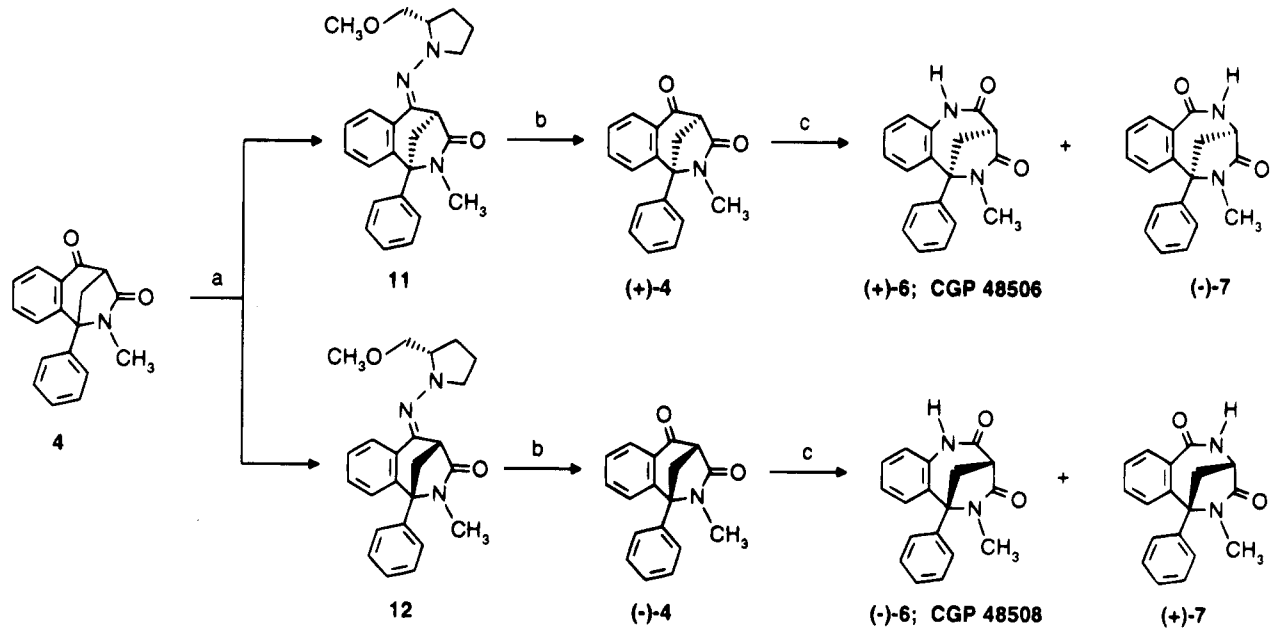
In order to assess the effect of stereochemistry at C(3) and C(6) on the *in vitro* activity of BA 41899 (**6**), the corresponding enantiomers CGP 48506 ((+)-**6**) and CGP

Scheme 1[a]



[a] Reagents and conditions: method A, **1**, $Pb(OAc)_4$, NEt_3, CH_2Cl_2, reflux, 12 h; method B, **2**, DPPA, DBU, toluene, 25 °C, 1 h, then 100 °C, 4 h; method C, CH_3I, K_2CO_3, DMF, 25 °C, 12 h; method D, NH_2OH HCl, pyridine, 80 °C, 6 h; method E, **4**, NaN_3, conc H_2SO_4, 0 → 25 °C, 3 h; method F, 1. **5**, $(CF_3SO_2)_2O$, pyridine, 60 °C, 12 h, 2. H_2O, 60 °C, 12 h.

Scheme 2[a]



[a] Reagents and conditions: (a) 1. (*S*)-(−)-1-amino-2-(methoxymethyl)pyrrolidine (SAMP), toluene, Dean–Stark, reflux, 36 h, 2. separation on silica gel; (b) O_3, CH_2Cl_2, −78 °C, 30 min, then 25 °C; (c) method E.

48508 ((−)-**6**) were prepared as depicted in Scheme 2. The synthesis involved Schmidt rearrangment of key intermediate ketones (+)-**4** and (−)-**4**, respectively, which were available by two different routes. Firstly, condensation of racemic ketone **4** with (*S*)-1-amino-2-(methoxymethyl)pyrrolidine (SAMP)[26] in refluxing toluene followed by flash chromatography provided the diastereomeric hydrazones **11** (40%) and **12** (29%) which were converted to ketones (+)-**4** (78%) and (−)-**4** (82%) by ozonolysis. At this stage, examination of the X-ray structure obtained from crystalline hydrazone **11** (Figure 2) allowed the assignment of absolute configurations of individual compounds. Alternatively, classical resolution of racemic carboxylic acid **2** in the presence of either cinchonidine or cinchonine as the chiral base yielded enantiomerically pure acids (+)-**2** (35%) and (−)-**2** (35%), respectively, in a single crystallization step. Subsequent processing to key intermediates (+)-**4** and (−)-**4** proceeded in a fashion analogous to that presented in Scheme 1.

Biological Evaluation and Discussion

Effects of Benzodiazocines on Porcine Myocardial Skinned Fibers. Porcine right ventricular skinned fibers were activated using Mg-ATP and Ca^{2+} ions. BA 41899 (**6**) and its isomers, compounds **7**, **9**, and **10**, were applied to the skinned fibers in the concentration range between 1 and 100 μM. Of these compounds, only BA 41899 (**6**) fulfilled the activity criteria (i.e., increase in isometric force at 30 μM or below, increase in isometric force at 100 μM by at least 60%, both measured at a free Ca^{2+} concentration of 0.912 μM). Therefore, in the further biological studies described below, we concentrated on BA 41899 (**6**) and its enantiomers, CGP 48506

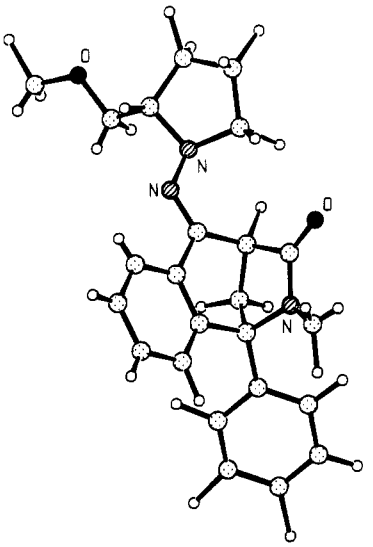


Figure 2. Perspective view of compound **11** from X-ray analysis.

((+)-**6**) and CGP 48508 ((−)-**6**). Figure 3 shows concentration−response curves for all three agents, obtained in skinned fibers partially activated by a constant, EGTA-buffered free Ca^{2+} concentration of 0.912 μM. CGP 48508 ((−)-**6**) was ineffective in this model, whereas its enantiomer, CGP 48506 ((+)-**6**), increased Ca^{2+}-activated force to about 280% of control, with an EC_{50} of close to 10 μM. The racemic mixture, BA 41899 (**6**), was equieffective, with an EC_{50} of ca. 20 μM. These results clearly demonstrate an enantiospecific increase in the Ca^{2+} responsiveness of porcine myocardial skinned fibers by CGP 48506 ((+)-**6**) but not by CGP 48508 ((−)-**6**). By comparison, in the same model, EMD 53998 increases isometric force to about 400% of control at 30 μM.[27]

In a further set of experiments, the effect of the active enantiomer CGP 48506 ((+)-**6**) was investigated at various free Ca^{2+} ion concentrations, ranging from 0.01 to ca. 50 μM. Figure 4 shows that under control conditions, in the absence of CGP 48506 ((+)-**6**), the skinned fibers were maximally activated at ca. 40 μM free Ca^{2+}, the EC_{50} for Ca^{2+} activation being ca. 3 μM. CGP 48506 ((+)-**6**), in concentrations between 1 and 100 μM, led to a concentration dependent left shift of the Ca^{2+} activation curve and a 10-fold reduction of the EC_{50} for Ca^{2+} activation to ca. 0.3 μM free Ca^{2+} at 100 μM CGP 48506 ((+)-**6**). At any given Ca^{2+} concentration between 0.1 and 10 μM, CGP 48506 ((+)-**6**) increased the contractile response of the skinned fiber preparations. These results indicate a strong Ca^{2+}-sensitizing effect of CGP 48506 ((+)-**6**).

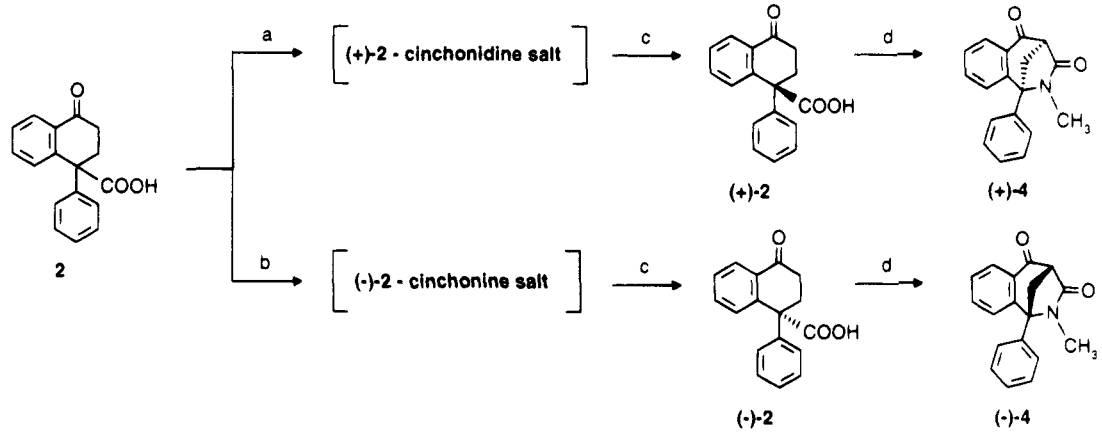
The maximum Ca^{2+}-activated force development was slightly reduced by CGP 48506 ((+)-**6**) in a concentration dependent manner, amounting to a maximum reduction by 17% at 100 μM. This reduction of maximum Ca^{2+}-activated force by CGP 48506 ((+)-**6**) is in remarkable contrast to the effect of the potent Ca^{2+}-sensitizing PDE inhibitor EMD 53998 and its active enantiomer, EMD 57033. These thiadiazinone derivatives induce a similar left shift of the Ca^{2+} activation curve but lead to a significant and concentration dependent increase in maximum Ca^{2+}-activated force in porcine ventricular skinned fibers.[27−30] On the basis of theoretical considerations,[31,32] such an increase in maximum Ca^{2+}-activated force may be indicative of an increase in the amount of force produced by an individual crossbridge. On the other hand, we assume that BA 41899 (**6**) and CGP 48506 ((+)-**6**), which both do not increase maximum Ca^{2+}-activated force, reduce the detachment rate constant, $g_{apparent}$, of the myocardial crossbridges. Such a reduction of the detachment rate should lead to characteristic decelerating effects on the time courses of contraction and relaxation.[33] Therefore, in the series of experiments described below, isometric twitch contractions were elicited in isolated atrial preparations obtained from guinea pig hearts.

Effects of BA 41899 and Its Enantiomers on Isolated Atrial Preparations from Guinea Pig Heart. Left atria from guinea pig hearts were electrically stimulated under isometric conditions at a rate of 2.5 s^{-1}. After equilibration, BA 41899 (**6**) and its enantiomers, CGP 48506 ((+)-**6**) and CGP 48508 ((−)-**6**), were applied in the concentration range between 1 and 100 μM. Figure 5 shows that CGP 48508 ((−)-**6**) was ineffective in this model, whereas CGP 48506 ((+)-**6**) exerted a positive inotropic effect amounting to a maximum increase in isometric tension by 40%, with an EC_{50} of ca. 8 μM. BA 41899 (**6**) was equieffective with an EC_{50} of ca. 15 μM. This clearly demonstrates an enantiospecific positive inotropic effect which corresponds very well with the observations in skinned fibers described above and strongly suggests a mechanistic link between Ca^{2+}-sensitizing effect and positive inotropism in this group of compounds.

In order to exclude other potential mechanisms of positive inotropic activity, the binding of CGP 48506 ((+)-**6**) to β_1-, α_1-, α_2-, and histamine$_1$ receptors and sarcolemmal Ca^{2+} channels was investigated using competition assays employing dehydroalprenolol, prazosine, clonidine, doxepine, desmethoxyverapamil, and nitrendipine, respectively. No measurable affinity of CGP 48506 ((+)-**6**) was found to either of these receptors. Also an activating effect of CGP 48506 ((+)-**6**) on sarcoplasmic reticular Ca^{2+} ATPase and Ca^{2+} uptake was excluded (data not shown). Likewise, investigations of inhibitory activity of CGP 48506 ((+)-**6**) on PDE I−IV showed no effect.[34] Recognizing some structural similarity to benzodiazepines, binding of CGP 48506 ((+)-**6**) to benzodiazepine receptors was investigated at this stage. Using flunitrazepam as ligand, affinity was found negligible. Due to solubility problems at concentrations >100 μM though, an exact IC_{50} value could not be determined. In summary, these results clearly indicate that the Ca^{2+}-sensitizing effect described above is most likely the only mechanism of the positive inotropic action of CGP 48506 ((+)-**6**).

Strikingly, at a concentration of 100 μM, CGP 48506 ((+)-**6**) loses most of its inotropic activity that it had at concentrations up to 30 μM. This 'paradoxical' behavior finds its explanation in a decelerating effect of CGP 48506 ((+)-**6**) (and BA 41899 (**6**)) on contractile dynamics, especially relaxation speed. Figure 6 shows typical isometric twitches in the presence of 100 μM CGP 48506 ((+)-**6**). It is evident that, after an initial phase of positive inotropism (Figure 6b), relaxation velocity is progressively reduced (Figure 6c), so that relaxation becomes incomplete (the next electrical stimulus follows after 400 ms, before full relaxation) and resting tension builds up, leading to an overall loss of developed tension. Allowing longer time for relaxation by reducing the rate of electrical stimulation to 0.25 s^{-1} (stimulus interval

Scheme 3[a]



[a] Reagents and conditions: (a) cinchonidine, NEt_3, CH_3CN, 90 → 25 °C; (b) cinchonine, NEt_3, CH_3CN, 90 → 25 °C; (c) HCl, EtOAc, 25 °C; (d) methods B and C.

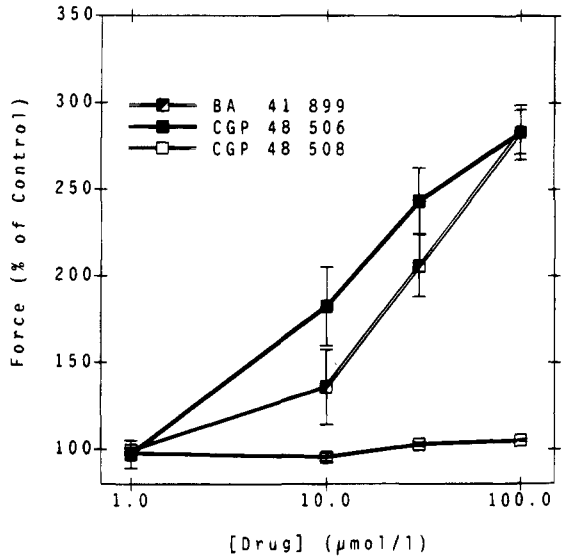


Figure 3. Influence of BA 41899 and its enantiomers on Ca^{2+}-activated isometric force in porcine ventricular skinned fibers at a constant, buffered Ca^{2+} concentration of 0.912 μM. Note that the (+)-enantiomer, CGP 48506, enantiospecifically increases skinned fiber force, whereas the (−)-enantiomer, CGP 48508, is ineffective in this model. Means ± SD, $n = 4$.

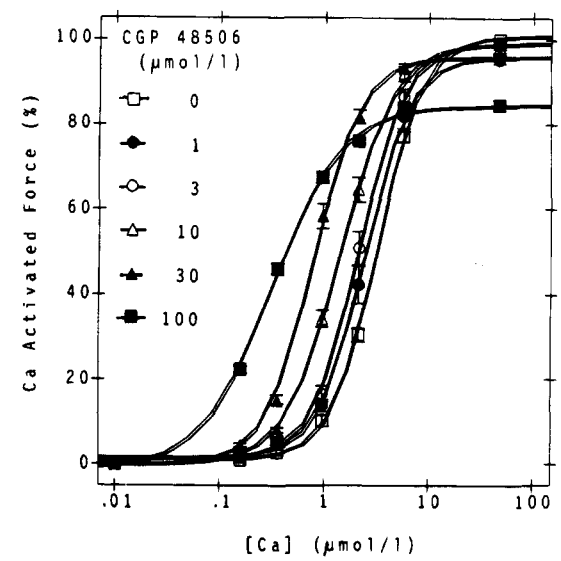


Figure 4. Influence of CGP 48506 on the Ca^{2+} activation curve of force in porcine ventricular skinned fibers. Note that the concentration dependent left shift of the activation curve is associated with a slight reduction of maximum Ca^{2+}-activated force. Means ± SD, $n = 4$.

prolonged to 4000 ms) restores the positive inotropic effect and abolishes resting tension (Figure 6d). Thus, the decelerating effect of high concentrations of CGP 48506 ((+)-**6**) (and BA 41899 (**6**)) on isometric relaxation, at least at a high rate of electrical stimulation corresponding to the physiological heart beat frequency in the guinea pig, may be expected to compromise diastolic relaxation and ventricular filling. It is, though, an open issue whether under *in vivo* conditions, where the heart is certainly better oxygenated by coronary circulation than under *in vitro* conditions by diffusion and is under control of the autonomous nervous system, similarly drastic effects on diastolic relaxation may occur.

In order to investigate effects on heart rate *in vitro*, right atrial preparations from guinea pig hearts were mounted isometrically and spontaneous contractions at their intrinsic sinus node rate were recorded. After equilibration, BA 41899 (**6**) and its enantiomers, CGP 48506 ((+)-**6**) and CGP 48508 ((−)-**6**), were applied in the concentration range between 1 and 100 μM. Figure 7 shows that CGP 48506 ((+)-**6**) was ineffective in this model, whereas its enantiomer, CGP 48508 ((−)-**6**), induced a reduction in spontaneous beating frequency by up to 55%, with an EC_{50} of ca. 15 μM. BA 41899 (**6**) had a similar effect on spontaneous beating frequency but was less potent (concentration−response curve incomplete, therefore no EC_{50} value). This clearly demonstrates an enantiospecific negative chronotropic effect residing in CGP 48508 ((−)-**6**), the enantiomer that is inotropically neutral and does not increase Ca^{2+} sensitivity.

Conclusions

Of the benzodiazocine isomers investigated in this study, BA 41899 (**6**) and its enantiomers, CGP 48506 ((+)-**6**) and CGP 48508 ((−)-**6**), were found to be the only compounds with significant activity in the *in vitro* models employed. In summary, BA 41899 (**6**) exhibits a pharmacological *in vitro* profile comprising a positive inotropic effect based on Ca^{2+} sensitization and a negative chronotropic effect. These two types of action reside in the two respective enantiomers of BA 41899 (**6**), the Ca^{2+}-sensitizing and positive inotropic actions in CGP 48506 ((+)-**6**) and the negative chronotropic action in CGP 48508 ((−)-**6**). At high concentrations, the positive inotropic action of CGP 48506 ((+)-**6**) is associated with prominent decelerating effects on con-

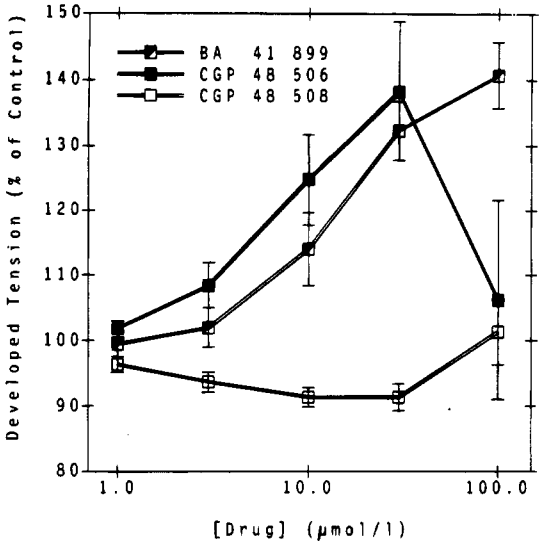


Figure 5. Influence of Ba 41899 and its enantiomers on developed tension in isolated guinea pig left atria, electrically driven at 2.5 s^{-1}. Note that the (+)-enantiomer, CGP 48506, enantiospecifically increases developed tension, whereas the (−)-enantiomer, CGP 48508, is ineffective in this model. For further explanation, see text. Means ± SD, n = 3–4.

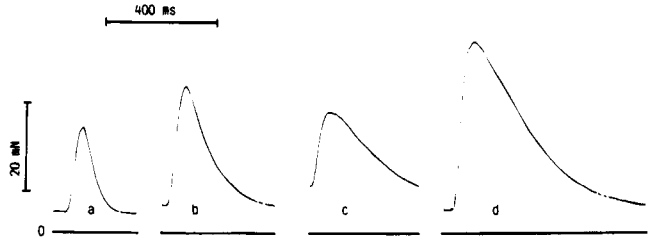


Figure 6. Typical examples of isometric twitch contractions of isolated guinea pig left atria, electrically driven at 2.5 s^{-1} (a–c) and 0.25 s^{-1} (d), respectively, before (a) and after (b–d) application of 100 μM CGP 48506. Note that the positive inotropic effect (b) seen immediately (1 min) after application of 100 μM CGP 48506 is lost after longer exposure (60 min) to the agent (c), due to incomplete relaxation (prolongation of contraction and relaxation phases) and corresponding buildup of resting tension. The positive inotropic effect is restored immediately (1 min) after reduction of the stimulation rate to 0.25 s^{-1} (d).

tractile dynamics, especially relaxation rate. This is in line with the hypothesis that CGP 48506 ((+)-**6**) increases Ca^{2+} sensitivity by a reduction of the dissociation constant, $g_{apparent}$, of myosin crossbridges from actin. Thus, at high rates of stimulation, diastolic relaxation and ventricular filling may be impaired.

To our knowledge, CGP 48506 ((+)-**6**) is the only known pharmacological agent exerting a noteworthy Ca^{2+}-sensitizing effect without concomitant PDE inhibitory activity. Further results obtained in various *in vivo* and in additional *in vitro* models will be published in due course.

Experimental Section

Biology. Materials and Methods. Adenosine triphosphate (ATP), phosphoenol pyruvate, and pyruvate kinase from pig muscle were from Boehringer. $CaCl_2$, $MgCl_2$, KCl, NaCl, NaN_3, $NaHCO_3$, and glycerol were from Merck. Fixanal stock solutions of $CaCl_2$ or $MgCl_2$ were from Riedel-de Haën. DMSO, Triton X-100, and imidazole hydrochloride were from Fluka. Ethylene glycol bis(aminoethyl ether)-*N,N',N,N'*-tetraacetic acid (EGTA) and histidine hydrochloride were from Serva. Dithiothreitol (DTT) was from Sigma. Oxycarbon gas (95% O_2/5% CO_2) was from Carba. All chemicals were of the highest purity degree available.

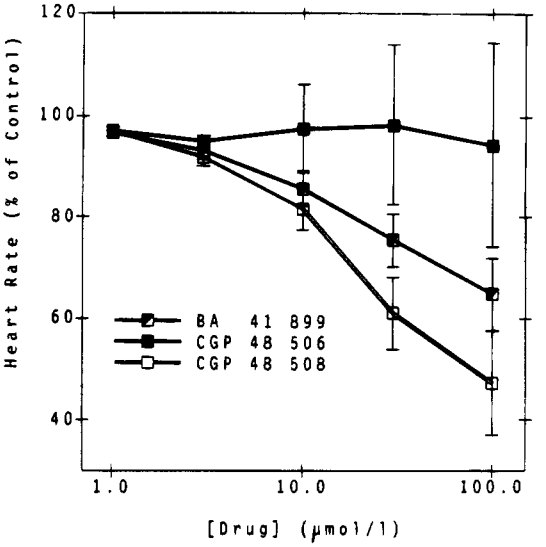


Figure 7. Influence of BA 41899 and its enantiomers on the beating rate of spontaneously beating isolated guinea pig right atria. Note that the (−)-enantiomer, CGP 48508, enantiospecifically reduces the beating rate, whereas the (+)-enantiomer, CGP 48506, is ineffective in this model. Means ± SD, n = 4–5.

Skinned Fiber Experiments. Right ventricular trabecula septomarginalis was taken from fresh pig hearts (slaughterhouse material). Parallel to the length axis of the cardiomyocytes, subendocardial fiber bundles about 10–20 mm in length and less than 200 μm in diameter were dissected and subjected to a detergent/glycerol extraction using Triton extraction solution (see below). By this extraction procedure, sarcolemma and sarcoplasmatic reticulum were destroyed and mitochondria and cellular components were washed out, leaving behind isolated contractile structures, the myofibrils of the fiber bundle. Using a fast setting glue (cellulose nitrate dissolved in acetone), the preparations were attached to a mechanical apparatus designed for the measurement of isometric force. This consisted of an AE 801 force transducer element (Aksjeselskapet Mikroelektronikk, Horten, Norway), a homemade bridge amplifier, and a W + W type 340 two-channel recorder.

In the presence of exogenous Mg-ATP, isometric contractions were elicited by incubation of the preparations in solutions with EGTA-buffered Ca^{2+} concentrations (mixtures of solution T and solution M, see below). Free Ca^{2+} concentrations were calculated using a modified version of a computer program previously described.[35] Stability constants were corrected according to Fabiato.[36] The sensitivity of the force generation of the contractile structures in response to Ca^{2+} was measured by the Ca^{2+} concentration at which one-half of the maximum force was generated (EC_{50}).

CGP 48506 was added to the incubation media from 1000-fold concentrated stock solutions in DMSO. The control incubation media contained 0.1% DMSO.

Triton extraction solution: 50 vol % glycerol, 20 mM histidine hydrochloride, 5 mM EGTA, 10 mM NaN_3, 1 mM DTT, 0.1% (w/v) Triton X-100, pH 7.3, at 4 °C. Fiber bundles were shaken in Triton extraction solution for 30 min on ice and then transferred into and stored at −18 °C and pH 7.0 for up to 1 month in glycerol solution of identical composition with the exception of no added Triton X-100.

T solution (activating solution): 10 mM ATP, 12 mM $MgCl_2$, 5 mM EGTA, 5 mM $CaCl_2$, 20 mM imidazole hydrochloride, 5 mM NaN_3, 5 mM phosphoenol pyruvate, 20 U/mL pyruvate kinase, pH 6.7, at 22 °C.

M solution (relaxing solution): identical in composition with T solution with the exception of no added $CaCl_2$, pH 6.7, at 22 °C.

Experiments in Isolated Guinea Pig Atria. Right, spontaneously beating and left, quiescent atria were dissected from hearts of guinea pigs anesthetized with urethane and incubated in Krebs–Henseleit medium, as previously described.[37] In brief, the spontaneous rate of contraction was

measured in right atria and the force of contraction in left atria electrically stimulated at a rate of 2.5 and 0.25 s^{-1}, respectively, at 32 °C. Potencies of drugs were determined by molar concentrations eliciting half-maximal effects (EC_{50}).

Chemistry. General Methods. Melting points were determined in open capillary tubes and are uncorrected. Standard high-performance liquid chromatography (HPLC) was performed using a Kontron MT 450 apparatus and a C-18 reverse-phase column (5 μm) for routine analysis. Standard conditions were 20% A + 80% B → 100% A within 20 min and then 100% A for 10 min (A = acetonitrile + 0.1% trifluoroacetic acid; B = water + 0.1% trifluoroacetic acid), flow = 1 mL/min, UV detection at 215 nm. Optical purities of individual enantiomers were determined by HPLC using either a Chiralpeak AD or a Chiralcel OJ column. Standard conditions were (1) hexane/2-propanol = 9:1, flow = 1.5 mL/min, UV detection at 230 nm; (2) hexane/2-propanol = 7:3, flow = 1 mL/min. UV detection at 215 nm; (3) hexane/2-propanol/acetic acid = 9:1:0.05, flow = 0.5 mL/min, UV detection at 220 nm. Elemental combustion analyses were within ±0.4% of theoretical values. ^1H NMR were recorded on a Varian Gemini 200, a Varian Gemini 300, or a Brucker WM-360 spectrometer. The chemical shifts are reported in parts per million (δ, ppm) downfield from tetramethylsilane (TMS). Fast-atom-bombardment mass spectra (FAB-MS) were recorded on a VG Manchester apparatus. Optical rotations were measured on a Perkin-Elmer Model 241 polarimeter at ambient temperature. Analytical thin layer chromatography (TLC) was carried out on precoated plates (silica gel 60 F-254; Merck), and spots were visualized with UV light, iodine, or phosphomolybdic acid. Column chromatography was performed with Kieselgel 60 (230–400 mesh) silica gel (Merck). Removal of solvents was performed by rotary evaporation under reduced pressure. All reactions involving air- or moisture-sensitive reagents were performed under a positive pressure of argon. Organic solvents were purified by the methods described by D. D. Perrin, W. L. F. Armarego, and D. R. Perrin (*Purification of Labaratory Chemicals*; Pergamon: Oxford, 1986) or purchased reagent grade from Aldrich or Fluka. Starting materials were purchased from Fluka or Aldrich.

4-Oxo-1-phenyl-1,2,3,4-tetrahydro-1-naphthalenecarboxamide (1). Compound **1** was prepared from diphenylacetonitrile according to the procedure described:[19] mp 190–191 °C (lit.[19] mp 187 °C); HPLC retention time 11.26 min; ^1H NMR (DMSO-d_6) δ 7.97 (d, 1H), 7.65–7.53 (m, 2H), 7.46 (dd, 1H), 7.42–7.26 (m, 3H), 7.16 (d, 2H), 7.02 (d, 2H), 2.85–2.75 (m, 1H), 2.75–2.53 (m, 2H), 2.45–2.30 (m, 1H); FAB-MS *m/z* 266 (M^+ + H). Anal. ($C_{17}H_{15}NO_2$) C, H, N.

4-Oxo-1-phenyl-1,2,3,4-tetrahydro-1-naphthalenecarboxylic Acid (2). Compound **2** was prepared from diphenylacetonitrile according to the procedure described:[21] mp 160–162 °C (lit.[21] mp 161.5–163 °C); HPLC retention time 13.43 min; ^1H NMR ($CDCl_3$) δ 8.16 (d, 1H), 7.60–7.44 (m, 2H), 7.40–7.26 (m, 4H), 7.12 (d, 1H), 3.03–2.92 (m, 1H), 2.90–2.76 (m, 1H), 2.67–2.52 (m, 2H); FAB-MS *m/z* 267 (M^+ + H). Anal. ($C_{17}H_{14}O_3$) C, H.

(+)-(1*S*)-4-Oxo-1-phenyl-1,2,3,4-tetrahydro-1-naphthalenecarboxylic Acid ((+)-2). To a solution of racemic acid **2** (276 g, 1.03 mol) and triethylamine (47.1 g, 0.46 mol) in acetonitrile (1.6 L) was added cinchonidine (167.8 g, 0.57 mol) in small portions at 90 °C, and the clear solution was left at room temperature for 12 h without stirring. The crystalline product was removed by filtration, washed with acetonitrile and diethyl ether, and then dried under vacuum at 40 °C. The cinchonidine salt (202 g) was stirred in 2 N aqueous hydrochloric acid (300 mL) and ethyl acetate (300 mL) at room temperature for 30 min. The organic layer was separated and the aqueous layer reextracted with ethyl acetate (100 mL). The combined ethyl acetate extracts were washed with water (2×), dried over magnesium sulfate, filtered, and concentrated *in vacuo* to yield the title compound (+)-**2** as an amorphous white foam (96 g, 35%): $[\alpha]_D$ +29° (*c* = 1.2, CH_2Cl_2); HPLC retention time 13.43 min; optical purity >99% (HPLC, Chiralcel OJ column, standard condition 3, retention time 43.43 min); ^1H NMR ($CDCl_3$) and FAB-MS identical with that of racemic **2**. Anal. ($C_{17}H_{14}O_3$) C, H.

(−)-(1*R*)-4-Oxo-1-phenyl-1,2,3,4-tetrahydro-1-naphthalenecarboxylic Acid ((−)-2). The 1*R*-enantiomer (−)-**2** was obtained as an amorphous white foam (35%) from the racemic acid **2** and cinchonine as the chiral base by the method described above for the synthesis of compound (+)-**2**: $[\alpha]_D$ −27° (*c* = 1.2, CH_2Cl_2); HPLC retention time 13.43 min; optical purity >99% (HPLC, Chiralcel OJ column, standard condition **3**, retention time 50.26 min); ^1H NMR ($CDCl_3$) and FAB-MS identical with that of racemic **2**. Anal. ($C_{17}H_{14}O_3$) C, H.

1-Phenyl-1,4-dihydro-1,4-methano-2-benzazepine-3,5-(2*H*)-dione (3). General Method A. To a suspension of amide **1** (5 g, 18.8 mmol) in dichloromethane was introduced a solution of lead tetraacetate (85%, contains 15% acetic acid; 11.5 g, 22 mmol) in dichloromethane, and the mixture was stirred at room temperature for 15 min. Triethylamine (9.5 mL, 68.1 mmol) was then added to the solution within 10 min (exothermic reaction), resulting in formation of a white suspension which subsequently dissolved. The clear and almost colorless reaction mixture was stirred at reflux for 12 h and then cooled to room temperature and washed with water (2×). The organic layer was separated, dried over sodium sulfate, filtered, and evaporated *in vacuo*. The brown oil obtained was crystallized from ethyl acetate to yield **3** (2.5 g, 50%) as white crystals: mp 262–263 °C; HPLC retention time 12.83 min; ^1H NMR (DMSO-d_6) δ 9.40 (s, 1H), 7.93 (d, 1H), 7.53–7.38 (m, 7H), 6.53 (d, 1H), 3.56–3.46 (m, 2H), 2.77 (dd, 1H); FAB-MS *m/z* 264 (M^+ + H). Anal. ($C_{17}H_{13}NO_2 \cdot 0.25H_2O$) C, H, N.

General Method B. A suspension of acid **2** (10 g, 37.6 mmol) in toluene (80 mL) was treated with 1,8-diazabicyclo-[5.4.0]undecene (DBU) (6.2 mL, 41.3 mmol) and diphenyl phosphorazidate (DPPA) (11.4 mL, 41.3 mmol), and the resulting solution was stirred at room temperature for 1 h and at 100 °C for 4 h. After evaporation of the solvent *in vacuo*, the residue was dissolved in ethyl acetate and washed with 1 N aqueous hydrochloric acid (1×), water (2×), and brine (1×). The organic layer was separated, dried over sodium sulfate, filtered, and evaporated *in vacuo*. Crystallization from ethyl acetate gave **3** (5.95, g, 60%) as white crystals, identical in all aspects with **3** obtained by General Method A.

2-Methyl-1-phenyl-1,4-dihydro-1,4-methano-2-benzazepine-3,5(2*H*)-dione (4). General Method C. A solution of **3** (5.3 g, 20 mmol) in dimethylformamide (50 mL) was treated with powdered potassium carbonate (5.5 g, 40 mmol) and iodomethane (1.9 mL, 30 mmol). The suspension was stirred at room temperature for 12 h. The reaction mixture was concentrated *in vacuo*; the residue was treated with water and extracted with ethyl acetate. The organic layer was separated, washed with water (3×) and brine (1×), dried over sodium sulfate, filtered, and concentrated *in vacuo*. The residue was recrystallized from 2-propanol to yield compound **4** as white crystals: mp 181–182 °C; HPLC retention time 14.85 min; ^1H NMR (DMSO-d_6) δ 7.97 (d, 1H), 7.63 (dd, 1H), 7.58–7.46 (m, 3H), 7.46–7.33 (m, 3H), 7.18 (d, 1H), 3.60 (d, 1H), 3.26 (d, 1H), 2.54 (dd, 1H), 2.54 (s, 3H); FAB-MS *m/z* 278 (M^+ + H). Anal. ($C_{18}H_{15}NO_2 \cdot 0.25H_2O$) C, H, N.

(+)-(1*S*,4*S*)-2-Methyl-1-phenyl-1,4-dihydro-1,4-methano-2-benzazepine-3,5(2*H*)-dione ((+)-4). From (+)-2: Compound (+)-**4** was obtained as white crystals from acid (+)-**2** using methods B and C: mp 208–210 °C; $[\alpha]_D$ +171° (*c* = 0.72, CH_2Cl_2); HPLC retention time 14.85 min; optical purity >99% (HPLC, Chiralpak AD column, standard condition 1, retention time 13.95 min); ^1H NMR (DMSO-d_6) and FAB-MS identical with that of racemic **4**. Anal. ($C_{18}H_{15}NO_2$) C, H, N.

From 11: Ozone (360 mL) was introduced into a solution of hydrazone **11** (6.3 g, 16.2 mmol) in dichloromethane (150 mL) at −78 °C within 30 min. The blue reaction mixture was warmed to room temperature, and a steady stream of nitrogen was passed through until the solution turned yellow. The reaction mixture was concentrated *in vacuo*, and the residue was flash chromatographed on silica gel using toluene/ethyl acetate (10:1) as eluent. Recrystallization from 2-propanol gave (+)-**4** (3.5 g, 78%), identical in all aspects with (+)-**4** obtained from (+)-**2**.

(−)-(1*R*,4*R*)-2-Methyl-1-phenyl-1,4-dihydro-1,4-methano-2-benzazepine-3,5(2*H*)-dione ((−)-4). From (−)-2: Compound (−)-**4** was obtained as white crystals from acid (−)-**2**

using general methods B and C: mp 208–210 °C; $[\alpha]_D$ −170° (c = 0.72, CH_2Cl_2); HPLC retention time 14.85 min; optical purity >99% (HPLC, Chiralpak AD column, standard condition 1, retention time 16.20 min); ^1H NMR (DMSO-d_6) and FAB-MS identical with that of racemic **4**. Anal. ($C_{18}H_{15}NO_2$) C, H, N.

From 12: Compound (−)-**4**, identical in all aspects with (−)-**4** obtained from (−)-**2**, was obtained from hydrazone **12** using the procedure described for the synthesis of (+)-**4** from **11**.

5-(Hydroxyimino)-2-methyl-1-phenyl-1,4-dihydro-1,4-methano-2-benzazepin-3(2*H*)-one (5). General Method D. Hydroxylamine hydrochloride (1.2 g, 17.3 mmol) was added to a solution of **4** (4 g, 14.4 mmol) in pyridine (40 mL), and the mixture was stirred at 80 °C for 6 h. The solvent was evaporated *in vacuo*, and the residue was treated with cold 1 N aqueous hydrochloric acid and extracted with ethyl acetate. The organic layer was separated, washed with water (2×) and brine (1×), dried over sodium sulfate, filtered, and evaporated *in vacuo*. The residue was recrystallized from ethyl acetate/hexane to yield the *E*-configured oxime **5** (3.6 g, 84%) as white crystals: mp 223–224 °C; HPLC retention time 14.31 min; ^1H NMR (DMSO-d_6) δ 11.75 (s, 1H), 8.0 (d, 1H), 7.53–7.45 (m, 2H), 7.45–7.28 (m, 5H), 7.08 (d, 1H), 4.52 (d, 1H), 2.63 (d, 1H), 2.50 (s, 3H), 2.26 (dd, 1H); FAB-MS m/z 293 (M^+ + H). Anal. ($C_{18}H_{16}N_2O_2$) C, H, N.

5-Methyl-6-phenyl-1,3,5,6-tetrahydro-3,6-methano-1,5-benzodiazocine-2,4-dione (6, BA 41899) and 5-Methyl-6-phenyl-2,3,5,6-tetrahydro-3,6-methano-2,5-benzodiazocine-1,4-dione (7). General Method E. Ketone **4** (42.4 g, 0.15 mol) was added in portions to 98% sulfuric acid (230 mL) at room temperature with stirring. The solution was cooled to 5 °C, and sodium azide (11 g, 0.17 mol) was added in portions over 20 min. After stirring at 5 °C for 30 min and at room temperature for 3 h, the reaction mixture was poured onto crushed ice (600 g). The precipitate was filtered off, dissolved in dichloromethane, and successively washed with water (1×) followed by brine (1×). The organic layer was separated, dried over sodium sulfate, filtered, and concentrated *in vacuo* to approximately 50 mL. The solution was put on top of a silica gel column, and the desired products were separated by flash chromatography eluting with hexane/ethyl acetate (5:2). The less polar isomer was triturated with diethyl ether, filtered, and dried *in vacuo* to yield BA 41899 (**6**) (21.5 g, 48%) as white crystals: mp 270–271 °C; HPLC retention time 12.46 min; ^1H NMR (DMSO-d_6) δ 10.38 (s, 1H), 7.58–7.02 (m, 7H), 6.98–6.86 (m, 2H), 3.57 (d, 1H), 2.89 (d, 1H), 2.56 (s, 3H), 2.18 (dd, 1H); FAB-MS m/z 293 (M^+ + H). Anal. ($C_{18}H_{16}N_2O_2$) C, H, N.

The more polar isomer was triturated with diethyl ether, filtered, and dried *in vacuo* to yield **7** (16.5 g, 37%) as white crystals: mp 236–237 °C; HPLC retention time 11.62 min; ^1H NMR (DMSO-d_6) δ 8.92 (d, 1H), 8.49 (dd, 1H), 7.57–7.25 (m, 6H), 7.12–6.98 (m, 2H), 4.92 (dd, 1H), 2.79 (d, 1H), 2.55 (s, 3H), 2.37 (dd, 1H); FAB-MS m/z 293 (M^+ + H). Anal. ($C_{18}H_{16}N_2O_2$) C, H, N.

General Method F. Trifluoromethanesulfonic acid anhydride (14.8 mL, 88 mmol) was added dropwise to a solution of oxime **5** (8.8 g, 30 mmol) in pyridine (175 mL) at room temperature. The dark reaction mixture was stirred at 60 °C for 12 h, then water (16 mL) was added, and the stirring was continued at 60 °C for 12 h. After concentration *in vacuo*, the residue was treated with 2 N aqueous hydrochloric acid and extracted with ethyl acetate. The organic layer was separated, washed with water (2×) and brine (1×), dried over sodium sulfate, filtered, and evaporated *in vacuo*. Flash chromatography eluting with hexane/ethyl acetate (5:2) followed by trituration with diethyl ether provided BA 41899 (3.5 g, 40%), identical in all aspects with BA 41899 obtained by General Method E.

(+)-(3*S*,6*S*)-5-Methyl-6-phenyl-1,3,5,6-tetrahydro-3,6-methano-1,5-benzodiazocine-2,4-dione ((+)-6, CGP 48506) and (−)-(3*S*,6*S*)-5-Methyl-6-phenyl-2,3,5,6-tetrahydro-3,6-methano-2,5-benzodiazocine-1,4-dione ((−)-7). The two compounds were prepared from (+)-**4** using General Method E. CGP 48506 (compound (+)-**6**): amorphous solid; $[\alpha]_D$ +89° (c = 1.0, CH_2Cl_2); HPLC retention time 12.46 min; optical purity >99% (HPLC, Chiralpak AD column, standard condition 1, retention time 16.20 min); ^1H NMR (DMSO-d_6) and FAB-MS identical with that of BA 41899. Anal. ($C_{18}H_{16}N_2O_2 \cdot 0.15H_2O$) C, H, N.

(−)-**7:** mp 225–227 °C (trituration with diethyl ether); $[\alpha]_D$ −141° (c = 1.1, CH_2Cl_2); HPLC retention time 11.62 min; optical purity >99% (HPLC, Chiralpak AD column, standard condition 2, retention time 61.14 min); ^1H NMR (DMSO-d_6) and FAB-MS identical with that of racemic **7**. Anal. ($C_{18}H_{16}N_2O_2$) C, H, N.

(−)-(3*R*,6*R*)-5-Methyl-6-phenyl-1,3,5,6-tetrahydro-3,6-methano-1,5-benzodiazocine-2,4-dione ((−)-6, CGP 48508) and (+)-(3*R*,6*R*)-5-Methyl-6-phenyl-2,3,5,6-tetrahydro-3,6-methano-2,5-benzodiazocine-1,4-dione ((+)-7). The two compounds were prepared from (+)-**4** using General Method E. CGP 48508 (compound (−)-**6**): mp 138–140 °C (trituration with diethyl ether); $[\alpha]_D$ −88° (c = 1.0, CH_2Cl_2); HPLC retention time 12.46 min; optical purity >99% (HPLC, Chiralpak AD column, standard condition 1, retention time 24.87 min); ^1H NMR (DMSO-d_6) and FAB-MS identical with that of BA 41899. Anal. ($C_{18}H_{16}N_2O_2 \cdot 0.2H_2O$) C, H, N.

(+)-**7:** mp 221–222 °C (trituration with diethyl ether); $[\alpha]_D$ +139° (c = 1.1, CH_2Cl_2); HPLC retention time 11.62 min; optical purity >99% (HPLC, Chiralpak AD column, standard condition 2, retention time 13.97 min); ^1H NMR (DMSO-d_6) and FAB-MS identical with that of racemic **7**. Anal. ($C_{18}H_{16}N_2O_2$) C, H, N.

3-Methyl-1-phenyl-1,4-dihydro-1,4-methano-3-benzazepine-2,5(3*H*)-dione (8). Compound **8** was prepared from amide **1** according to the procedure described:[21] mp 140–142 °C (lit.[21] mp 138–140 °C); HPLC retention time 15.56 min; ^1H NMR (DMSO-d_6) δ 7.99 (d, 1H), 7.84 (br s, 2H), 7.56–7.37 (m, 5H), 6.75 (d, 1H), 4.22 (d, 1H), 3.35 (d, 1H), 2.80 (dd, 1H), 2.77 (s, 3H); FAB-MS m/z 278 (M^+ + H). Anal. ($C_{18}H_{15}NO_2$) C, H, N.

4-Methyl-6-phenyl-1,3,4,6-tetrahydro-3,6-methano-1,4-benzodiazocine-2,5-dione (9) and 4-Methyl-6-phenyl-2,3,4,6-tetrahydro-3,6-methano-2,4-benzodiazocine-1,5-dione (10). The two compounds were prepared from **8** using General Method E. **9** (less polar isomer; 20%): mp 236–238 °C (dichloromethane/hexane); HPLC retention time 12.25 min; ^1H NMR (DMSO-d_6) δ 10.22 (s, 1H), 8.15 (br s, 1H), 7.49–7.30 (m, 3H), 7.25 (d, 1H), 7.20–7.10 (m, 2H), 6.85 (dd, 1H), 6.55 (d, 1H), 4.16 (d, 1H), 3.02 (d, 1H), 2.80 (s, 3H), 2.43 (dd, 1H); FAB-MS m/z 293 (M^+ + H). Anal. ($C_{18}H_{16}N_2O_2$) C, H, N.

10 (more polar isomer; 50%): mp 254–257 °C (dichloromethane/hexane); HPLC retention time 11.72 min; ^1H NMR (DMSO-d_6) δ 8.91 (d, 1H), 8.40 (dd, 1H), 8.09 (br s, 1H), 7.50–7.28 (m, 5H), 7.05 (br s, 1H), 6.79 (dd, 1H), 4.78 (dd, 1H), 2.91 (d, 1H), 2.75 (s, 3H), 2.63 (dd, 1H); FAB-MS m/z 293 (M^+ + H). Anal. ($C_{18}H_{16}N_2O_2$) C, H, N.

(1*S*,4*R*)-5-[[(2*S*)-2-(Methoxymethyl)pyrrolidin-1-yl]imino]-2-methyl-1-phenyl-1,4-dihydro-1,4-methano-2-benzazepin-3(2*H*)-one (11) and (1*R*,4*S*)-5-[[(2*S*)-2-(Methoxymethyl)pyrrolidin-1-yl]imino]-2-methyl-1-phenyl-1,4-dihydro-1,4-methano-2-benzazepin-3(2*H*)-one (12). A mixture of ketone **4** (15 g, 54 mmol) and (*S*)-1-amino-2-(methoxymethyl)pyrrolidine (SAMP) (8 mL, 60 mmol) in toluene (80 mL) was heated under reflux using a Dean–Stark apparatus for 36 h. The solvent was evaporated *in vacuo*, and the residue was purified by flash chromatography eluting with hexane/ethyl acetate (4:1). The less polar diastereomer was crystallized from diethyl ether/hexane yielding **11** (8.4 g, 40%) as yellowish crystals: mp 125–126 °C; HPLC retention time 15.67 min; ^1H NMR (DMSO-d_6) δ 8.05 (d, 1H), 7.48 (dd, 2H), 7.45–7.32 (m, 4H), 7.29 (dd, 1H), 7.04 (d, 1H), 4.25 (d, 1H), 3.79–3.71 (m, 1H), 3.59–3.46 (m, 2H), 3.28 (s, 3H), 3.31–3.25 (m, 1H), 3.14–3.05 (m, 1H), 2.66 (d, 1H), 2.54 (s, 3H), 2.24 (dd, 1H), 2.08–1.97 (m, 1H), 1.95–1.76 (m, 2H), 1.65–1.53 (m, 1H); FAB-MS m/z 390 (M^+ + H). Anal. ($C_{24}H_{27}N_3O_2$) C, H, N.

The more polar isomer **12** (6.1 g, 29%) was isolated as a viscous, yellowish oil: HPLC retention time 13.63 min; ^1H-NMR (DMSO-d_6) δ 8.05 (d, 1H), 7.48 (dd, 2H), 7.45–7.32 (m, 4H), 7.29 (dd, 1H), 7.04 (d, 1H), 4.39 (d, 1H), 3.68–3.57 (m, 2H), 3.45–3.35 (m, 2H), 3.30 (s, 3H), 3.20–3.11 (m, 1H), 2.68

(d, 1H), 2.50 (s, 3H), 2.27 (dd, 1H), 1.96–1.82 (m, 3H), 1.72–1.61 (m, 1H); FAB-MS *m/z* 390 ($M^+ + H$). Anal. ($C_{24}H_{27}N_3O_2$) C, H, N.

Crystal Structure Analysis of Compound 11. A light yellow-colored platelet-shaped crystal of $C_{24}H_{27}N_3O_2$ having approximate dimensions of 0.50 × 0.55 × 0.20 mm was mounted on a glass fiber. All measurements were made on an Enraf-Nonius CAD4 diffractometer with graphite monochromated Cu Kα radiation. The crystal belongs to the orthorhombic space group $P2_12_12_1$ with a = 10.078(1) Å, b = 12.854(1) Å, c = 16.384(2) Å, V = 2122.4(6) Å^3, Z = 4, D_{calc} = 1.219 g/cm^3. The intensities were corrected for Lorentz and polarization effects but not for absorption. A total of 2505 independent intensities were measured of which 2388 were classified as observed with $I > 3\sigma(I)$. The structure was solved by direct methods using the computer program SHELXS-86;[38] 18 of 27 hydrogen atoms were located in a different Fourier map. The positions of the remaining ones were calculated assuming normal geometry. The structure was refined using full matrix least-squares calculations with anisotropic displacement parameters for non-hydrogen atoms and fixed ones for hydrogen atoms. The final R-factor for 343 variables was 0.074. The highest peak in the final difference Fourier map was 0.59 e/Å^3. Positional and thermal parameters, bond lengths, and bond angles have been deposited as supplementary material.

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Supporting Information Available: Positional and thermal parameters, bond lengths, and bond angles of the crystal structure of compound **11** (4 pages). Ordering information is given on any current masthead page.

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