114772-45-1; **103**, 114772-60-0; **104**, 124750-36-3; **105**, 37455-55-3; **106**, 68282-41-7; **107**, 68282-59-7; **108**, 124750-54-5; **109**, 124750-55-6; **110**, 133379-10-9; **111**, 114772-46-2; **112**, 114772-62-2; **113**, 114772-63-3; **114**, 114772-83-7; **115**, 114772-84-8; **116**, 114772-85-9; **117**, 124779-26-6; **118**, 114772-77-9; **119**, 124750-43-2; **120**, 114772-51-9; **121**, 114772-70-2; **122**, 114772-75-7; **123**, 114772-76-8; **124**, 120568-15-2; **125**, 120568-16-3; **126**, 124750-65-8; **127**, 133910-04-0; **128**, 16191-28-9; **129**, 124750-63-6; **130**, 124750-64-7; **131**, 133910-05-1; **132**, 133910-06-2; **133**, 133910-07-3; **134**, 124750-42-1; **135**, 114772-81-5; **136**, 114772-82-6; methyl 2-iodobenzoate, 610-97-9; 4-iodotoluene, 624-31-7; 1-bromo-2-nitrobenzene, 577-19-5; 2-methoxybenzoic acid, 579-75-9; 2-amino-2-methyl-1-propanol, 124-68-5; 4-bromotoluene, 106-38-7; 3-bromobenzonitrile, 6952-59-6; 2-butyl-4(5)-chloro-5(4)-(hydroxymethyl)imidazole, 79047-41-9; 4-[[3-[*N*-(*tert*-butoxycarbonyl)-*N*-isopropylamino]-2-hydroxypropyl]oxy]indole-2-carboxylic acid, 103221-84-7; 2-butyl-4(5)-(hydroxymethyl)-imidazole, 68283-19-2; butyraldehyde, 123-72-8; acetol, 116-09-6; (phenylsulfonyl)acetonitrile, 7605-28-9; ethyl 3-(4-methylphenyl)-3-oxopropanoate, 7116-41-8; allyl bromide, 106-95-6.

GABA-Uptake Inhibitors: Construction of a General Pharmacophore Model and Successful Prediction of a New Representative

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A model for the pharmacophore of GABA-uptake inhibitors was established using published structure–activity data and molecular modeling. The model accounted for the activities of different classes of GABA-uptake inhibitors. Analogues of guvacine substituted at position 6 were synthesized in order to confirm the model. 6-(3,3-Diphenylpropyl)guvacine (**30f**), which fit well with the pharmacophore, had an in vitro IC_{50} of 0.1 μM. This value is as good as those of the best GABA-uptake inhibitors known today.

γ-Aminobutyric acid (GABA, 1) is a major neurotransmitter in mammals.[1-9] Dysfunctioning of GABA-ergic synapses has been invoked for diseases such as Parkinson's disease,[10-13] Huntington's chorea,[14] epilepsy,[15,16] and some forms of schizophrenia.[17-19] One of the possible ways to palliate GABA deficiency lies in the inhibition of uptake mechanisms of this neurotransmitter.[20-24] Taking in account the results of various studies devoted to this GABA-ergic regulation approach[25-27] allowed us to establish a model pharmacophore by means of computer modeling. The validity of the model pharmacophore was confirmed through its ability to rationalize the activity of some newly described uptake inhibitors and to guide the design of a novel, potent GABA uptake inhibitor.

Identification and Construction of the Pharmacophore

Studies on GABA-uptake inhibitors have been developed in several directions. Analogues of GABA itself have been prepared in order to modulate the chain length or to introduce substituents or unsaturation. Although these modifications yielded active compounds, only 2-fluoro-GABA (**2**)[28] (Figure 1) showed uptake-inhibitory potency comparable to that of GABA itself. Most of these acyclic GABA analogues are not selective for GABA-uptake inhibition but also possess agonist properties.

Conformationally restricted cyclic GABA analogues have also been studied, e.g., where the methylenes have been incorporated into homocyclic systems such as cyclopropyl,[29] cyclobutyl,[29] cyclopentyl,[30] cyclopentenyl,[30] cyclohexyl,[29,31] cyclohexenyl,[29] and cyclohexadienyl[29] rings. The best activities were found for cyclopentyl and cyclopentenyl rings, and high stereoselectivity was observed for the (+)-4(*S*)- versus the (−)-4(*R*)-aminocyclopentene-1-carboxylic acids[30] **3** and **4**, respectively (Figure 1). Some heterocyclic amino acids were also described as GABA-uptake inhibitors, especially in the pyrrolidine[29] and pi-

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[‡] Sanofi Recherche.

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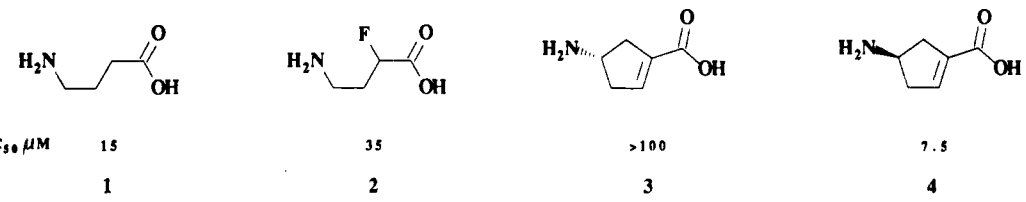

Figure 1. GABA uptake inhibitory potency of GABA (1), 2-fluoro-GABA (2), and (*S*)- and (*R*)-4-aminocyclopentene-1-carboxylic acids (3 and 4).[28,30]

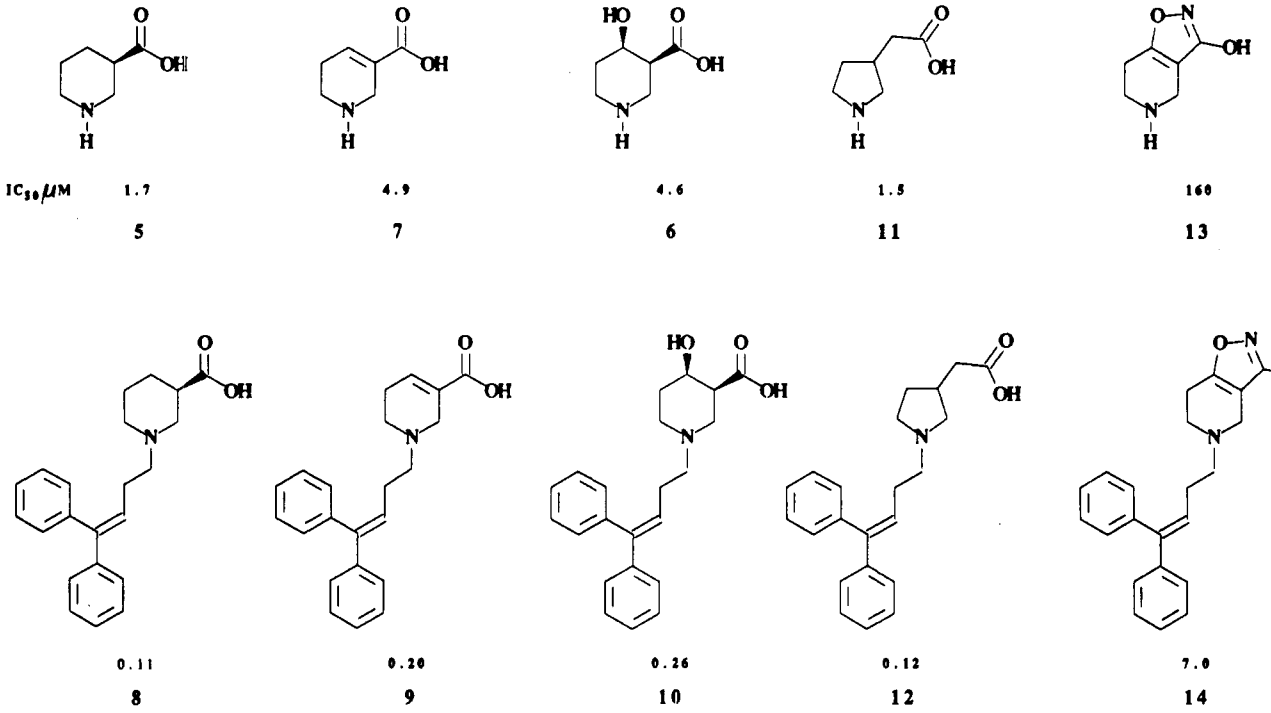

Figure 2. GABA uptake inhibitory potency of (*R*)-nipecotic acid (5), guvacine (7), *cis*-4-hydroxynipecotic acid (6), 3-pyrrolidineacetic acid (11), THPO (13), and their *N*-4,4-diphenyl-3-butenyl derivatives.[33]

peridine.[27,29,32-34] series. The most active compound in the piperidine series, (−)-(*R*)-nipecotic acid (5) (Figure 2), has an IC_{50} value of 1.7 μM on GABA-uptake inhibition.[33,34] For this substance again some enantiomeric selectivity was

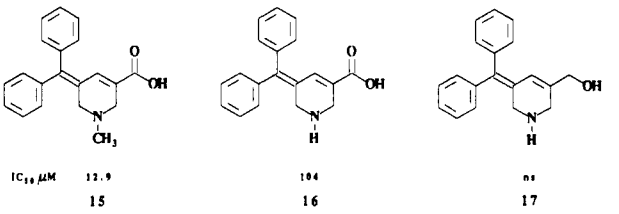

Figure 3. GABA uptake inhibitory potency of 5-benzhydrylideneguvacine derivatives.[44]

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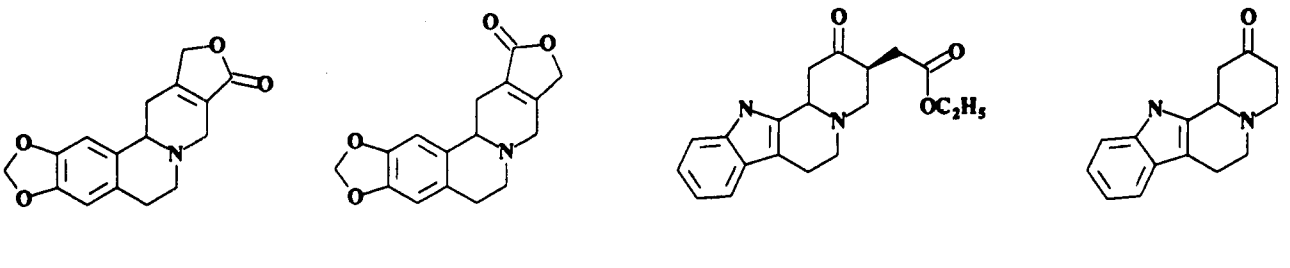

Figure 4. GABA uptake inhibitory potency of polycyclic compounds.[45]

observed insofar that (+)-(*S*)-nipecotic acid is approximately 6 times less potent than the (−)-*R* enantiomer.[34] X-ray analyses, as well as NMR studies, demonstrated that the carboxylic function of (−)-(*R*)-nipecotic acid stayed in an equatorial position.[35] In such an orientation, the acidic group is coplanar with the mean plane of the molecule.

On the basis of its potency and selectivity as a GABA-uptake inhibitor, nipecotic acid became the focuse of additional research. Thus, substitution[29,36] or unsaturation[31] of nipecotic acid resulted in (±)-*cis*-4-hydroxynipecotic acid[29] (**6**) (Figure 2) and the 3,4-unsaturated analogue, guvacine[29] (**7**) (Figure 2), which showed potencies comparable to that of nipecotic acid.

The carboxylic acid function of GABA was also replaced by bioisosteric functions such as isoxazoles[37] or tetrazoles,[38] leading to less active compounds. In 1984, a group at Smith Kline and French Laboratories reported the potent GABA uptake inhibitory properties of nipecotic acid, guvacine, *cis*-4-hydroxynipecotic acid, and homo-β-proline analogues, N-substituted with a diphenylbutenyl group.[34,39] These compounds, **8** (SKF 89976-A), **9** (SKF 100330-A), **10** (SKF 100591), and **12** (SKF 100561) (Figure 2), are approximately 20 times more potent than the unsubstituted amino acids. The authors explained the observed activity increase by the existence of additional binding sites to which the aromatic rings could fit. Lengthening or shortening the alkenyl chain, which links the piperidine ring to the aromatic groups, as well as saturation of the double bond in the side chain of **8** leads to an important loss of activity.[33] Further studies demonstrated that the replacement of one or both of the benzene rings by furan, pyrrole, or thiophene rings leads to slightly more active molecules.[40,41] Also, the replacement of one of the sp^2 carbon atoms of the side chain by an oxygen atom, bearing two p doublets, led to an equipotent product.[42]

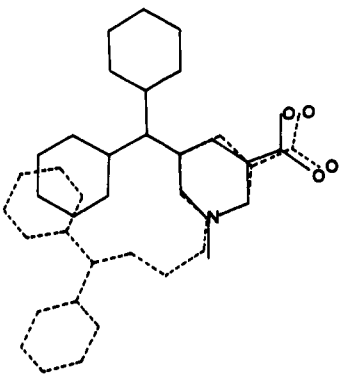

Figure 5. Superimposition of SKF 89976-A (**8**) and *N*-methyl-5-benzhydrylideneguvacine (**15**).

Krogsgaard-Larsen et al. grafted the diphenylbutenyl chain to other GABA-uptake inhibitors such as tetra-

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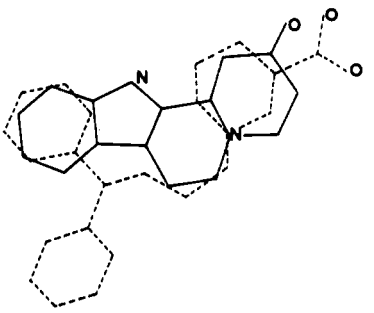

Figure 6. Superimposition of SKF 89976-A (8) in the same conformation as in Figure 5 with piperidone 21.

hydroisoxazolopyridinol (THPO) 13 to give 14. These authors again noted an activity increase comparable to that observed for 8–10 and 12[37] (Figure 2). All the above results support the hypothesis of the existence of additional binding sites for the aromatic rings. However, the side-chain flexibility of these molecules allows a large number of energetically equivalent conformations. With the help of the SYBYL program[43] we searched the optimal spatial localization for the additional binding sites.

Two papers devoted to rigid GABA-uptake inhibitors were helpful for this study. The paper by Müller-Uri et al.[44] describes guvacine derivatives substituted by a benzhydrylidene group in the 5-position, 15–17 (compound 15 is also N-methylated) (Figure 3). Compound 15 shows activities similar to those of the unsubstituted guvacine.

The second paper, by Kardos et al.,[45] observed that the benzo[*a*]perhydroquinolizines 18 and 19 and the indolo-[3,2-*d*]perhydroquinolizines 20 and 21 have inhibitory activities similar to that of nipecotic acid (Figure 4). If we consider these published results as signifying GABA-uptake activity, they suggest that the presence of aromatic rings in the vicinity of the 6-position of the piperidine ring of nipecotic acid analogues is well tolerated by the GABA-uptake system.

On the contrary, substitution at the 2-position of the nipecotic acid skeleton seems to be detrimental. Thus, during earlier studies on GABA-uptake inhibitors, we observed that substituting nipecotic acid with a methyl or a phenyl group at the 2-position yielded practically inactive compounds.[36]

Applied to 8 and 9 these observations suggest placement of the aromatic rings of the diphenylbutenyl side chain in the vicinity of position 6 rather than position 2 of the piperidinic ring.

Molecular modeling shows that there are unstrained conformations of 8 and 9 in which the phenyl rings have a partial overlap with the benzhydrylidene group of the guvacine derivative 15 when the amine functions and the carboxylic acid groups are overlaid (Figure 5). The lower activities observed for the benzhydrylideneguvacine derivative 15 in comparison with derivatives 8 and 9 can be attributed at least to two factors: (i) Compound 15 could only partially occupy the binding sites reached by the side chain of 8–10 and 12. (ii) The larger volume at the top of position 5 of compound 15 could be inappropriate with regard to the additional binding site prerequisites. The same unstrained conformation of the SKF compounds

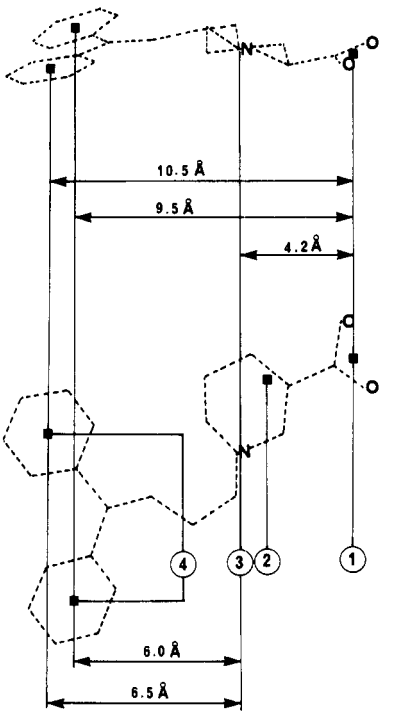

Figure 7. Parameters of the proposed pharmacophore for GABA-uptake inhibitors (---) SKF 89976-A (8) in its active conformation (see text for explanations).

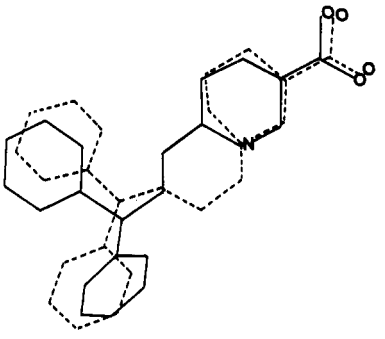

Figure 8. Fitting of 6-(3,3-diphenylpropyl)guvacine with the proposed pharmacophore.

8–10 and 12 also allowed a good overlap of one of their aromatic groups with the end of the aromatic region of the perhydroquinolizines 18–21 (Figure 6).

These observations, integrated with the recent findings of Wise[46] and Krogsgaard-Larsen,[37] allowed us to define important parameters associated with a possible GABA uptake inhibition pharmacophore (Figure 7):

1. The acidic group in position 3 of the piperidino ring should be situated in the pharmacophore mean plane.
2. A double bond in position 3,4 is often beneficial for the activity. Such an unsaturation also forces the carboxylic function into an orientation toward the general pharmacophore plane.
3. The amine function should be located at a distance of 4.2 Å from the center of the acidic group.
4. The two aromatic rings should occupy a region of space essentially coplanar with the piperidine average plane. The centers of these aromatic rings are then located 9.5 Å from the acidic center and 6.0 Å from the amino group for the first ring, and 10.5 Å from the acidic center and 6.5 Å from the amine for the second ring.

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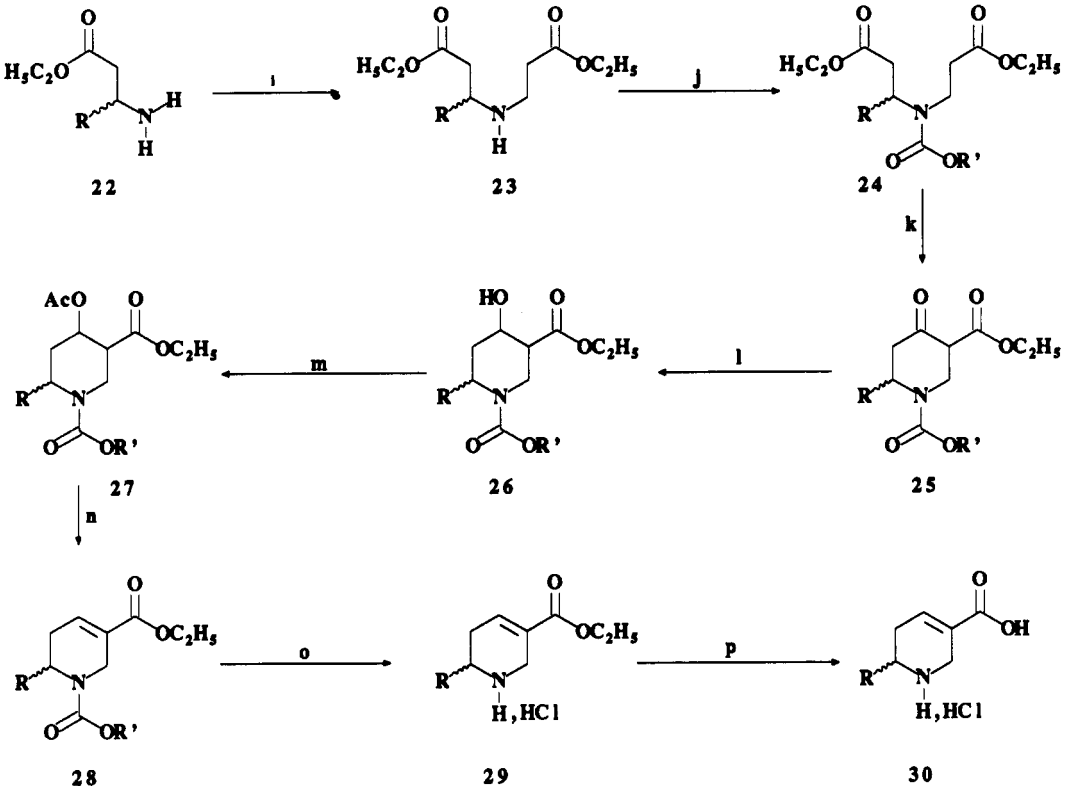
Scheme I[a,b]



[a] (i) CH_2=$CHCO_2Et$, EtOH; (j) $ClCO_2Et$, K_2CO_3, H_2O or $(BOC)_2O$, K_2CO_3, dioxane–H_2O; (k) Na, EtOH, C_6H_6; (l) H_2–Raney Ni EtOH; (m) $(CH_3CO)_2O$; (n) DBN, THF; (o) HCl, EtOH; (p) HCl, H_2O. [b] R = (a) methyl, (b) ethyl, (c) propyl, (d) phenyl, (e) phenethyl, (f) 3,3-diphenylpropyl; R′ = Et or tBu.

Predictive Value of the Pharmacophore Model

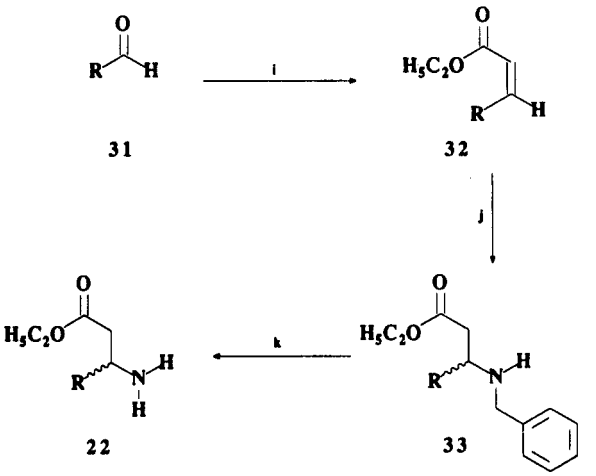
To confirm the real predictive value of our model, we prepared a series of guvacine derivatives in which the π binding sites of the pharmacophore were filled by substituting on position 6 of the guvacine ring (Table I). By adjustment of the parameters of our model pharmacophore and for synthetic reasons, simple hydrocarbon chains with different lengths and chains substituted with one or two phenyl groups were chosen. Molecular modeling suggested that a 3,3-diphenylpropyl side chain would represent a good fit with the pharmacophore (Figure 8), whereas a monophenylethyl side chain should give less potent derivatives comparable to the benzhydrylideneguvacine 15 or the quinolizine derivatives 18–21.

Synthesis

The syntheses of the 6-substituted guvacine derivatives can be divided into two main parts: (1) the construction of the piperidino ring using appropriate β-substituted β-amino esters and (2) the preparation of the β-substituted β-amino esters. These latter compounds were prepared by a simple and general synthetic method starting from aldehydes. The starting aldehydes are generally commercially availble except for β,β-diphenylbutanal, whose preparation will also be described here.

The piperidino ring construction starting from β-substituted β-amino esters was done by analogy with the work of Bishop[47] and Krogsgaard-Larsen.[48] The addition of amino ester 22 to ethyl acrylate yielded diester 23. The amine function was then protected as a carbamate 24 by the action of ethyl chloroformate under the conditions reported by Krogsgaard-Larsen[48] or as a BOC derivative by reaction with di-*tert*-butyl pyrocarbonate, as described by Grzonka.[49] N-Protected diester 24 was cyclized to 3-carbethoxy-4-piperidone 25 by a selective Dieckmann reaction. The catalytic hydrogenation of piperidone 25 gave hydroxypiperidine 26. After acetylation of the hy-

Scheme II[a,b]



[a] (i) $(C_6H_5)_3P^+CH_2CO_2EtBr^-$, C_6H_6; (j) $C_6H_5CH_2NH_2$, EtOH; (k) H_2–Pd/C, EtOH. [b] R = (a) methyl, (b) ethyl, (c) propyl, (d) phenyl, (e) phenethyl, (f) 3,3-diphenylpropyl.

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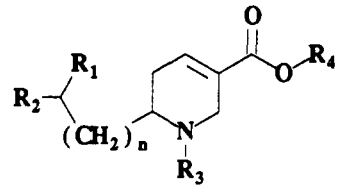
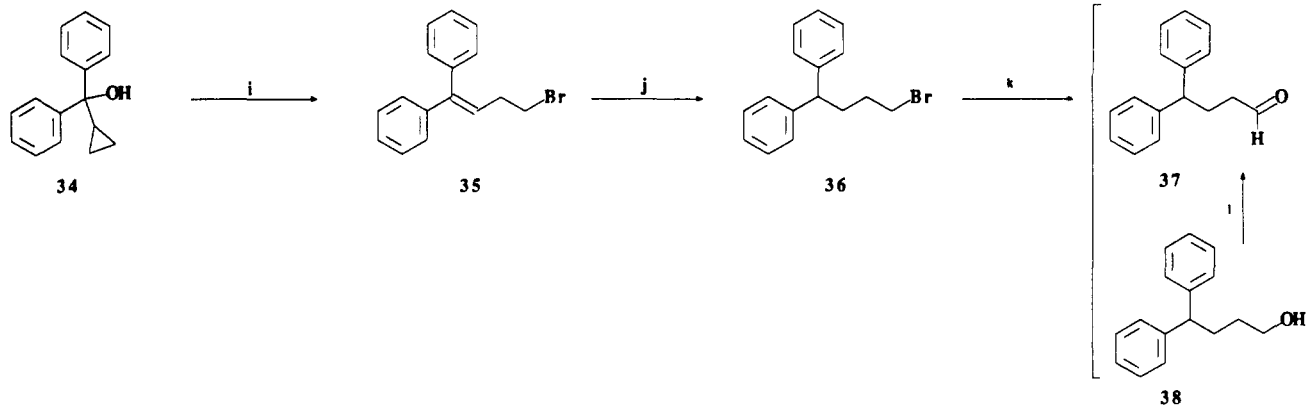
Table I. 6-Substituted Guvacine: Biochemical Data



R_1	R_2	R_3	R_4	n	no.	anal.		IC_{50}, μM
H	H	H	H	0		(37)		40
H	H	H	H	1	**30b**	$C_8H_{13}NO_2$·HCl	C H N	>100
H	H	H	C_2H_5	1	**29b**	$C_{10}H_{17}NO_2$·HCl	C H N	>100
C_6H_5	H	H	H	1	**30e**	$C_{14}H_{17}NO_2$·HCl	C H N	100
H	H	H	H	2	**30c**	$C_9H_{15}NO_2$·HCl	C H N	>100
H	H	H	C_2H_5	2	**29c**	$C_{11}H_{19}NO_2$·HCl	C H N	>100
C_6H_5	C_6H_5	H	H	2	**30f**	$C_{21}H_{23}NO_2$·HCl	C H N	0.1
C_6H_5	C_6H_5	H	C_2H_5	2	**29f**	$C_{23}H_{27}NO_2$·HCl·$^1/_2H_2O$	C H N	37
C_6H_5	H	H	H	3	**30g**	$C_{16}H_{21}NO_2$·HCl	C H N	>100
C_6H_5	H	CH_3	H	3	**39**	$C_{17}H_{23}NO_2$·HCl	C H N	>100

Scheme III[a]



[a] (i) HBr, H_2O; (j) H_2, 5% Pd/charcoal, EtOH; (k) DMSO, NaI, $NaHCO_3$; (l) DMSO, $(CO)_2Cl_2$, CH_2Cl_2.

droxy group, treatment of acetate **27** with 1,5-diazabicyclo[4.3.0]non-5-ene (DBN) created the conjugated double bond. Total deprotection using hydrochloric acid yielded the expected 6-substituted guvacine **30** (Scheme I). The synthesis described above (Scheme I) uses β-substituted β-amino esters as starting material. The published methods for the preparation of these amino esters are often very long and/or tedious as well as suffering from a lack of generality.[50-55] We have found a simple and general method for the preparation of β-substituted β-amino esters in three steps starting from aldehydes[48,56] (Scheme II).

The reaction of aldehydes **31** with (carbethoxymethyl)-triphenylphosphonium bromide according to Delmas and Lebigot[57] gave β-substituted unsaturated esters **32**. The addition of benzylamine to conjugated ester **32** according to Morsch[57,58] yielded *N*-benzylamino esters **33**, which were submitted to hydrogenolysis to produce the expected β-amino esters **22**.

4,4-Diphenylbutanal was prepared by starting from cyclopropyldiphenylmethanol (**34**). The action of hydrobromic acid upon this alcohol gave unsaturated bromide **35**. Catalytic hydrogenation in the presence of palladium on charcoal gave 4,4-diphenyl-1-bromobutane (**36**), which was oxidized to aldehyde **37** via a Kornblum modified reaction.[59] The expected aldehyde was accompanied by 22% of the corresponding alcohol **38**. This side product could also be oxidized into the expected aldehyde via a Swern reaction[60] (Scheme III).

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Table II

compd	pK_{a_1}[a]	pK_{a_2}[a]	pK_a,[a,b] ester	I/U ratio	log P_{oct}[c]	log P_{cyc}[c]	ΔP
30f	3.79	9.25	7.64	7000	0.71	-2.00	2.71
SKF 89976-A	3.57[d]	9.23[d]	7.36	6000	0.99[e]	-0.43	1.43

[a] Determined by potentiometric titration. [b] pK_a of the corresponding ester. [c] Measured by a conventional shake flask technique at room temperature. The concentrations of the compound in the aqueous phase (0.15 M phosphate buffer, pH 7.4) were determined spectrophotometrically. [d] pK_{a_1} = 3.32 and pK_{a_2} = 9.36.[36] [e] Log P_{oct} = 1.14.[36] All the values are the means of at least two determinations.

Biochemistry

The in vitro studies on GABA uptake were conducted by using rat brain synaptosome preparations.[61] The IC_{50} values were established by using concentration ranges up to 10^{-4} M in tested compound. The results of the in vitro tests are given in Table I.

Discussion

The results expressed in Table I demonstrate the predictive value of the proposed pharmacophore. In the guvacine series we had previously observed that position 6 tolerates a methyl substituent. However, replacing the methyl group with an ethyl or a propyl chain (**30b** and **30c**) led to a loss of activity. Presumably the ethyl or propyl groups cannot occupy the π binding area to provide additional binding, and the net result is a loss of potency. However, the phenethyl derivative **30e** showed some activity, presumably reflecting the ability of the phenyl to partially fill the π area. A further, important increase in potency was seen with the diphenylpropyl derivative **30f** wherein the phenyl rings are quite ideally positioned to fill the π site. A further lengthening of the alkyl chain to give the phenylbutyl derivative **30g** led to a loss of activity; this can be interpreted by assuming that the longer side chain interacts negatively with an area of bulk intolerance on the GABA-uptake carrier. Yunger et al. observed the same variations of activity related to alkyl chain length in their series of N-substituted nipecotic acids.[39]

Compound **30f** was inactive in classical in vivo screening models for anticonvulsants, particularly in antagonizing bicuculline-induced convulsions. It is generally accepted that, in the absence of a specific carrier, the rate and extent of entry of a compound into the brain are primarily related to its proton dissociation constant (K_a),[62] partition coefficient P,[63,64] and molecular size.[65] The inactivity in vivo of compound **30f** in comparison to its N-substituted analogue SKF 89976-A cannot be attributed to a difference in molecular size; therefore, we measured the dissociation constants and the partition coefficients of both derivatives. As seen in Table II, very similar pK_a values are found for **30f** and SKF 89976-A (8). The I/U ratios between the zwitterionic (I) and the unionized (U) species, calculated according to Wegscheider,[66] do not furnish a satisfactory explanation for the in vivo inactivity of **30f**, even if they are slightly in favor of compound 8.

The partition coefficients for **30f** and 8 were then measured in two solvent systems: 1-octanol/phosphate buffer pH 7.4 (P_{oct}) and cyclohexane/phosphate buffer pH 7.4 (P_{cyc}). The P_{oct} coefficient accounts for protein binding in the bloodstream[67] whereas P_{cyc} accounts for the permeability through lipid membranes or for the partitioning process into nonpolar brain areas.[68] Thus, the difference between log P_{oct} and log P_{cyc}, defined as ΔP,[69] can be correlated with the logarithm of the brain/blood concentration ratio[69] and accounts for the overall hydrogen-binding ability of the drug.[69] High ΔP values limit permeability across the blood–brain barrier. As shown in Table II, the P_{oct} value of **30f** is slightly lower than that of 8. The P_{cyc} of **30f** shows, however, a value 40 times lower than that of 8, which gives a greater ΔP for **30f** than for 8. This ratio corresponds to an insufficient brain/blood concentration ratio, thus accounting for the lack of in vivo activity.

Besides these physicochemical considerations, one cannot exclude the possibility of metabolic inactivation as another contribution to in vivo inactivity.

Experimental Section

Melting points were measured on a calibrated Kofler hot-stage apparatus and are uncorrected. IR spectra were meaured with a Beckman Acculab-4 spectrophotometer, and ^1H NMR spectra were recorded on Bruker WP-60 or AC 200 spectrometers using the δ scale with reference to Me_4Si. All the new compounds gave satisfactory C,H,N analyses.

Ethyl *N*-(2-Carbethoxyethyl)-3-aminoalkanoates or -aralkanoates 23. General Procedure. A solution of 0.12 mol of the appropriate β-substituted β-amino ester **22** and 0.13 mol of ethyl acrylate in 200 mL of absolute ethanol was stirred overnight at room temperature. The solvent was evaporated under reduced pressure. Water (100 mL) was added to the residue and the solution acidified (pH 4) by addition of diluted hydrochloric acid. The aqueous phase was washed with ethyl acetate (2 × 200 mL), neutralized with a saturated solution of sodium hydrogen carbonate, and extracted with ethyl acetate (2 × 200 mL). The organic layer was washed with water, dried over magnesium sulfate, and evaporated to dryness. The product was pure enough to be used for the next step.

Ethyl *N*-(2-carbethoxyethyl)-3-aminohydrocinnamate (23d): starting from ethyl 3-aminohydrocinnamate (**22d**); yield, 90%; IR ($CHCl_3$) 1720 (C=O); ^1H NMR (60 MHz, $CDCl_3$), 1.22

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(t, J = 6.8, 3 H, OCH_2CH_3), 1.24 (t, J = 6.8, 3 H, OCH_2CH_3), 2.0–3.0 [m, 7 H, containing at 2.14 (br s, exchangeable with D_2O, 1 H, N*H*), 6 H, O=CCH_2CH_2, O=CCH_2], 3.9–4.5 [m, 5 H, containing at 4.12 (g, J = 6.8, 2 H, OCH_2CH_3), at 4.14 (q, J = 6.8, 2 H, OCH_2CH_3), 1 H, O=CCH_2CH], 7.1–7.6 (m, 5 H, C_6H_5).

Ethyl *N*-(2-carbethoxyethyl)-3-amino-5-phenylvalerate (23e): starting from ethyl 3-amino-5-phenylvalerate (**22e**); yield, 88%; IR ($CHCl_3$) 1720 (C=O); ^1H NMR (60 MHz, $CDCl_3$) 1.24 (t, J = 6.8, 6 H, 2 OCH_2CH_3), 1.57 (br s, exchangeable with D_2O, 1 H, N*H*), 1.6–2.0 (m, 2 H, $C_6H_5CH_2CH_2$), 2.3–3.1 (m, 9 H, O=CCH_2CHNHCH_2CH_2C=O, $CH_2C_6H_5$), 4.12 (q, J = 6.8, 4 H, 2 OCH_2CH_3), 7.21 (s, 5 H, C_6H_5).

Ethyl *N*-(2-carbethoxyethyl)-3-amino-6,6-diphenylcaproate (23f): starting from ethyl 3-amino-6,6-phenylcaproate (**22f**); yield, 95%; IR ($CHCl_3$) 1725 (C=O); ^1H NMR (200 MHz, $CDCl_3$) 1.21 (t, J = 6.8, 3 H, OCH_2CH_3), 1.23 (t, J = 6.8, 3 H, OCH_2CH_3), 1.3–1.5 (m, 2 H, CH_2CH_2CHNH), 1.59 (br s, exchangeable with D_2O, 1 H, N*H*), 2.0–2.2 (m, 2 H, $CHCH_2CH_2CHNH$), 2.37 (d, J = 6.8, 2 H, O=CCH_2CH), 2.43 (t, J = 6.8, 2 H, O=CCH_2CH_2), 2.78 (t, J = 6.8, 2 H, O=CCH_2CH_2NH), 3.00 (qu, J = 6.8, 1 H, CH_2CHCH_2), 3.85 (t, J = 6.8, 1 H, $C_6H_5CH(C_6H_5)CH_2$), 4.08 (q, J = 6.8, 2 H, OCH_2CH_3), 4.10 (q, J = 6.8, 2 H, OCH_2CH_3), 7.0–7.4 (m, 10 H, 2 C_6H_5).

Ethyl *N*-(*tert*-Butyloxycarbonyl)-*N*-(2-carbethoxyethyl)-3-aminoalkanoates or -aralkanoates 24. General Procedure. A mixture containing 0.1 mol of a secondary amine **23**, 54 mL of dioxane, 108 mL of water, and 54 mL of 5% potassium carbonate solution was cooled in an ice bath. Di-*tert*-butyl pyrocarbonate (0.13 mL) was added slowly with stirring. The stirring was maintained for 15 min at 0 °C and then continued at room temperature for 3 h more. After concentration under reduced pressure, the residue was extracted with ether (2 × 200 mL). The ethereal phase was washed with 1 N hydrochloric acid (50 mL) and water and dried over magnesium sulfate. After evaporation of the solvent, the crude product was pure enough to be used in the next step. The product was crystallized in a mixture of hexane–ethyl acetate. The yield was >95%.

Ethyl *N*-(*tert*-butyloxycarbonyl)-*N*-(2-carbethoxyethyl)-3-aminovalerate (24b): IR ($CHCl_3$) 1725 (C=O ester), 1680 (C=O carbamate); ^1H NMR (200 MHz, $CDCl_3$) 0.87 (t, J = 6.8, 3 H, $CHCH_2CH_3$), 1.24 (t, J = 6.8, 3 H, OCH_2CH_3), 1.25 (t, J = 6.8, 3 H, OCH_2CH_3), 1.4–1.8 [m, 11 H, containing at 1.45 and 1.51 (2 s, 9 H, $(CH_3)_3C$), 2 H, $CHCH_2CH_3$], 2.3–2.7 (m, 4 H, O=CCH_2, O=CCH_2), 3.3–3.7 (m, 3 H, CH_2CH_2N, CH_2CHCH_2), 4.12 (q, J = 6.8, 2 H, OCH_2CH_3), 4.13 (q, J = 6.8, 2 H, OCH_2CH_3).

Ethyl *N*-(*tert*-butyloxycarbonyl)-*N*-(2-carbethoxyethyl)-3-aminocaproate (24c): IR ($CHCl_3$) 1725 (C=O ester), 1680 (C=O carbamate); ^1H NMR (60 MHz, $CDCl_3$) 0.7–1.7 [m, 22 H, containing at 1.26 (t, J = 6.8, 6 H, 2 OCH_2CH_3), at 1.46 (s, 9 H, $(CH_3)_3C$), 7 H, $CHCH_2CH_2CH_3$], 2.3–2.8 (m, 4 H, O=CCH_2CH_2, O=CCH_2CH), 3.2–3.7 (m, 3 H, O=CCH_2CH_2N, CH_2CHCH_2), 4.13 (q, J = 6.8, 4 H, 2 OCH_2CH_3).

Ethyl *N*-(*tert*-butyloxycarbonyl)-*N*-(2-carbethoxyethyl)-3-amino-5-phenylvalerate (24e): IR ($CHCl_3$) 1730 (C=O ester), 1685 (C=O carbamate); ^1H NMR (60 MHz, $CDCl_3$) 1.24 (t, J = 6.8, 6 H, 2 OCH_2CH_3), 1.46 (s, 9 H, $(CH_3)_3C$), 1.6–2.2 (m, 2 H, $CHCH_2CH_2C_6H_5$), 2.2–2.7 (m, 6 H, $CH_2C_6H_5$, O=CCH_2CH_2, O=CCH_2CH), 3.2–3.7 (m, 2 H, NCH_2CH_2), 3.7–4.4 [m, 5 H, containing at 4.13 (q, J = 6.8, 4 H, 2 OCH_2CH_3), 1 H, CH_2CHCH_2], 7.22 (s, 5 H, C_6H_5).

Ethyl *N*-(*tert*-butyloxycarbonyl)-*N*-(2-carbethoxyethyl)-3-amino-6,6-diphenylcaproate (24f): IR ($CHCl_3$) 1725 (C=O ester), 1680 (C=O carbamate); ^1H NMR (200 MHz, $CDCl_3$) 1.20 (t, J = 6.8, 3 H, OCH_2CH_3), 1.23 (t, J = 6.8, 3 H, OCH_2CH_3), 1.42 and 1.52 (2 s, 9 H, $(CH_3)_3C$), 2.00 (q, J = 6.8, 2 H, $CHCH_2CH_2CHN$), 2.3–2.6 (m, 4 H, O=CCH_2CH_2, O=CCH_2CH), 3.2–3.6 [m, 4 H, containing at 3.47 (q, J = 6.8, 2 H, $CHCH_2CH_2CHN$) 2 H, O=CCH_2CH_2N], 3.8–4.5 [m, 6 H, containing at 4.09 (q, J = 6.8, 4 H, 2 OCH_2CH_3), 1 H, $C_6H_5CHCH_2$, and 1 H, CH_2CHCH_2], 7.0–7.4 (m, 10 H, 2 C_6H_5).

Ethyl *N*-Carbethoxy-*N*-(2-carbethoxyethyl)-3-aminoalkanoates 24. General Procedure. A 50% solution of secondary amine **23** in water was cooled at 0 °C. A 5% potassium carbonate solution (1.1 equiv) was cooled and added with vigorous stirring. Ethyl chloroformate (1.1 equiv) was added, and stirring was maintained for 30 min at 0 °C and 1 h at room temperature. After the same workup as for ethyl *N*-(*tert*-butyloxycarbonyl)-*N*-(2-carbethoxyethyl)-3-aminoalkanoate, the ethyl carbamate was obtained with a yield greater than 90%.

Ethyl *N*-carbethoxy-*N*-(2-carbethoxyethyl)-3-aminobutyrate (24a): IR ($CHCl_3$) 1725 (C=O ester), 1680 (C=O carbamate); ^1H NMR (60 MHz, $CDCl_3$) 0.8–1.4 [m, 12 H, containing at 1.24 (t, J = 6.8, 3 H, OCH_2CH_3), at 1.26 (t, J = 6.8, 6 H, 2 OCH_2CH_3), 1.26 (d, J = 6.8, 3 H, $CHCH_3$)], 2.5–2.8 (m, 4 H, O=CCH_2CH, O=CCH_2CH_2), 3.4–3.9 (m, 2 H, O=CCH_2CH_2N), 4.0–4.3 [m, 7 H, containing at 4.14 (q, J = 6.8, 2 H, OCH_2CH_3), at 4.16 (q, J = 6.8, 4 H, 2 OCH_2CH_3), 1 H, CH_3CHCH_2].

Ethyl *N*-carbethoxy-*N*-(2-carbethoxyethyl)-3-aminohydrocinnamate (24d): IR ($CHCl_3$) 1725 (C=O ester), 1680 (C=O carbamate); ^1H NMR (200 MHz, $CDCl_3$) 1.1–1.4 [m, 9 H, containing at 1.21 and 1.22 (2 t, J = 7.5, 6 H, 2 OCH_2CH_3), at 1.27 (t, J = 6.8, 3 H, OCH_2CH_3)], 2.0–2.6 (m, 2 H, O=CCH_2CH_2), 2.8–3.1 (d, J = 6.8, 2 H, O=CCH_2CH), 3.2–3.5 (m, 2 H, CH_2N), 3.9–4.3 [m, 7 H, containing at 4.06 (q, J = 7.5, 4 H, 2 OCH_2CH_3), 4.18 (q, J = 6.8, 2 H, OCH_2CH_3), 1 H, CH_2CHCH_2], 7.32 (br s, 5 H, C_6H_5).

***N*-Carbalkoxy-2-alkyl- or -2-aralkyl-5-carbethoxy-4-piperidones 25. General Procedure.** A mixture of 0.095 mol of diester **24**, 0.096 g-atom of sodium, and 0.2 mL of absolute ethanol in 200 mL of dry benzene was stirred at room temperature for 24–48 h. The benzene phase was washed with 100 mL of 1 N hydrochloric acid and dried over potassium carbonate. The solvents were evaporated under reduced pressure, and the crude product was purified by silica gel column chromatography with a mixture of hexane–ethyl acetate, 9/1, as eluent.

***N*-Carbethoxy-2-methyl-5-carbethoxy-4-piperidone (25a)**: yield, 90%; IR ($CHCl_3$) 1720 (C=O ester), 1690 (C=O carbamate), 1620 (C=O); ^1H NMR (60 MHz, $CDCl_3$) 1.14 (d, J = 6.8, 3 H, $CHCH_3$), 1.25 (t, J = 6.8, 3 H, OCH_2CH_3), 1.26 (t, J = 6.8, 3 H, OCH_2CH_3), 1.5–3.8 (m, 4 H, O=CCH_2CH, $CHCH_2N$), 3.9–5.0 [m, 5 H, containing at 4.12 (q, J = 6.8, 2 H, OCH_2CH_3), at 4.20 (q, J = 6.8, 2 H, OCH_2CH_3), 1 H, CH_2CHCH_3], 12.00 and 12.27 (2 s, exchangeable with D_2O, 1 H, O*H*).

***N*-(*tert*-Butyloxycarbonyl)-2-ethyl-5-carbethoxy-4-piperidone (25b)**: yield, 91%; IR ($CHCl_3$) 1725 (C=O ester), 1700 (C=O ketone), 1680 (C=O carbamate), 1650 (C=C); ^1H NMR (200 MHz, $CDCl_3$) 0.8–1.0 (m, 3 H, $CHCH_2CH_3$), 1.2–1.9 [m, 14 H, containing at 1.27 (t, J = 6.8, 3 H, OCH_2CH_3), at 1.45 (s, 9 H, $(CH_3)_3C$), 2 H, $CHCH_2CH_2$], 2.3–2.8 (m, 2 H, O=CCH_2), 3.2–3.8 (m, 2 H, CH_2N), 4.0–5.2 [m, 3 H, containing at 4.20 (q, J = 6.8, 2 H, OCH_2CH_3), 1 H, CH_2CHCH_2], 12.04 and 12.23 (2 s, exchangeable with D_2O, 1 H, O*H*).

***N*-(*tert*-Butyloxycarbonyl)-2-propyl-5-carbethoxy-4-piperidone (25c)**: yield, 85%; IR ($CHCl_3$) 1620–1700 (C=O ester, ketone, carbamate; C=C enol); ^1H NMR (60 MHz, $CDCl_3$) 0.8–1.8 [m, 19 H, containing at 1.30 (t, 6.8, 3 H, OCH_2CH_3), at 1.47 (s, 9 H, $(CH_3)_3C$), 7 H, $CHCH_2CH_2CH_3$], 2.0–3.8 (m, 4 H, CH_2N, O=CCH_2), 4.0–5.0 [m, 3 H, containing at 4.25 (q, J = 6.8, 2 H, OCH_2CH_3), 1 H, CH_2CHCH_2], 11.90 and 12.10 (2 s, exchangeable with D_2O, 1 H, O*H*).

***N*-(*tert*-Butyloxycarbonyl)-2-phenethyl-5-carbethoxy-4-piperidone (25e)**: yield, 80%; IR ($CHCl_3$) 1620–1730 (C=O, ester, ketone, carbamate; C=C, enol); ^1H NMR (200 MHz, $CDCl_3$) 1.1–1.6 [m, 12 H, containing at 1.30 (t, J = 6.8, 3 H, OCH_2CH_3), at 1.44 (s, 9 H, $(CH_3)_3C$)], 1.6–1.8 (m, 2 H, $CHCH_2CH_2$), 2.2–2.8 (m, 4 H, $CHCH_2CH_2$, O=CCH_2CH), 3.3–3.8 (m, 2 H, CH_2N), 4.0–5.2 [m, 3 H, containing at 4.28 (q, J = 6.8, 2 H, OCH_2CH_3), 1 H, CH_2CHCH_2], 7.0–7.3 (m, 5 H, C_6H_5), 12.06 and 12.36 (2 s, exchangeable with D_2O, 1 H, O*H*).

***N*-(*tert*-Butyloxycarbonyl)-2-(3,3-diphenylpropyl)-5-carbethoxy-4-piperidone (25f)**: yield, 87%; IR ($CHCl_3$) 1620–1730 (C=O, ester, ketone, carbamate; C=C, enol); ^1H NMR (200 MHz, $CDCl_3$) 1.0–1.6 [m, 14 H, containing at 1.25 and 1.26 (2 t, J = 6.8, 3 H, OCH_2CH_3), at 1.42 and 1.47 (2 s, 9 H, $(CH_3)_3C$), 2 H, $CHCH_2CH_2CHN$], 1.9–3.1 (m, 4 H, O=C$CH_2CHCH_2CH_2CH$), 3.3–5.0 [m, 5 H, containing at 4.15 and 4.16 (2 q, J = 6.8, 2 H, OCH_2CH_3), 3 H, CH_2N, $C_6H_5CHCH_2$], 7.1–7.4 (m, 10 H, 2 C_6H_5) 11.98 and 12.26 (2 s, exchangeable with D_2O, 1 H, O*H*).

***N*-Carbalkoxy-2-alkyl- or -2-aralkyl-5-carbethoxy-4-piperidinols 26. General Procedure.** A 20% ethanolic solution

of a piperidone **25** was hydrogenated at 80 °C under 60 atm in the presence of 20% Raney nickel. The cooled mixture was filtered through a Celite pad. Complete reaction was indicated by a negative enol test using ferric chloride. The solvent was evaporated under reduced pressure. The infrared spectrum did not show ketone or enol absorption but only the ester carbonyl band at 1720 cm^{-1} and the carbamate carbonyl band at 1685 cm^{-1}. The ^1H NMR spectra, measured at 200 MHz, were very complex due to the presence of diastereoisomers and could not be interpreted.

4-Acetoxy-*N*-carbalkoxy-2-alkyl- or -2-aralkyl-5-carbethoxy-5-piperidines 27. General Procedure. A 10% solution of the appropriate piperidinol **25** in acetic anhydride was refluxed overnight. After evaporation under reduced pressure, water was added to the residue. The aqueous solution was alkalinized by addition of saturated sodium hydrogen carbonate and extracted with ethyl acetate. The organic layer was washed with water and dried over magnesium sulfate. After evaporation of the solvents under reduced pressure, the crude product was purified by silica gel column chromatography with a mixture of hexane–ethyl acetate, 2/1, as eluent. The IR spectra showed the esters and carbamate bonds at 1735 and 1685 cm^{-1}. The ^1H NMR spectra, measured at 200 MHz, were too complex to be interpreted.

***N*-Carbalkoxy-2-alkyl- or -2-aralkyl-5-carbethoxy-1,2,3,6-tetrahydropyridines 28. General Procedure.** A 10% solution of an acetate **27** in freshly distilled THF was stirred at room temperature for 2–3 h in the presence of 1 equiv of 1,5-diazabicyclo[4.3.0]non-5-ene (DBN). The solvent was evaporated and water added to the residue. The aqueous solution was acidified with 1 N hydrochloric acid and extracted with ethyl ether. The organic layer was washed with water and dried over magnesium sulfate. The crude product obtained after evaporation of the solvent was purified by silica gel column chromatography with a mixture of hexane–ethyl acetate, 1/1, as eluent.

***N*-Carbethoxy-2-methyl-5-carbethoxy-1,2,3,6-tetrahydropyridine (28a)**: yield, 94%; IR ($CHCl_3$) 1700 (C=O), 1690 (C=C); ^1H NMR (60 MHz, $CDCl_3$) 0.9–1.5 (m, 9 H, $CHCH_3$, 2 OCH_2CH_3), 2.0–3.0 (m, 2 H, $C=CHCH_2$), 3.4–5.0 [m, 7 H, containing at 4.03 (q, J = 6.8, 2 H, OCH_2CH_3), at 4.07 (q, J = 6.8, 2 H, OCH_2CH_3)], 6.8–7.2 (m, 1 H, $CH_2CH=C$).

***N*-(*tert*-Butyloxycarbonyl)-2-ethyl-5-carbethoxy-1,2,3,6-tetrahydropyridine (28b)**: yield, 80%; IR ($CHCl_3$) 1710 (C=O ester), 1680 (C=O carbamate), 1650 (C=C); ^1H NMR (60 MHz, $CDCl_3$) 0.6–1.6 [m, 17 H, containing at 0.90 (t, J = 6.8, 3 H, $CHCH_2CH_3$), at 1.30 (t, J = 6.8, 3 H, OCH_2CH_3), at 1.45 (s, 9 H, $(CH_3)_3C$), 2 H, $CHCH_2CH_3$], 2.0–2.6 (m, 2 H, $C=CHCH_2$), 3.2–3.8 (m, 2 H, CH_2N), 4.0–4.8 [m, 3 H, containing at 4.20 (q, J = 6.8, 2 H, OCH_2CH_3), 1 H, CH_2CHCH_2], 6.8–7.2 (m, 1 H, $CH_2CH=C$).

***N*-(*tert*-Butyloxycarbonyl)-2-propyl-5-carbethoxy-1,2,3,6-tetrahydropyridine (28c)**: yield, 80%; IR ($CHCl_3$) 1705 (C=O ester), 1675 (C=O carbamate), 1640 (C=C); ^1H NMR (60 MHz, $CDCl_3$) 0.7–1.7 [m, 19 H, containing at 1.29 (t, J = 6.8, 3 H, OCH_2CH_3), at 1.46 (s, 9 H, $(CH_3)_3C$), 7 H, $CHCH_2CH_2CH_3$], 2.0–2.6 (m, 2 H, $C=CHCH_2$), 3.2–3.8 (m, 2 H, CH_2N), 4.0–4.8 [m, 3 H, containing at 4.21 (q, J = 6.8, 2 H, OCH_2CH_3), 1 H, CH_2CHCH_2], 6.8–7.2 (m, 1 H, $CH_2CH=C$).

***N*-(*tert*-Butyloxycarbonyl)-2-phenethyl-5-carbethoxy-1,2,3,6-tetrahydropyridine (28e)**: yield, 75%; IR ($CHCl_3$) 1705 (C=O ester), 1680 (C=O carbamate), 1650 (C=C); ^1H NMR (60 MHz, $CDCl_3$) 0.7–3.0 [m, 18 H, containing at 1.29 (t, J = 6.8, 3 H, OCH_2CH_3), at 1.47 (s, 9 H, $(CH_3)_3C$), at 1.59 (t, J = 6.8, 2 H, $CH_2CH_2C_6H_5$), 4 H, $=CHCH_2CHCH_2$], 3.2–3.8 (m, 2 H, CH_2N), 4.0–4.8 [m, 3 H, containing at 4.20 (q, J = 6.8, 2 H, OCH_2CH_3), 1 H, CH_2CCH_2], 7.0–7.3 (m, 1 H, $C=CHCH_2$).

***N*-(*tert*-Butyloxycarbonyl)-2-(3,3-diphenylpropyl)-5-carbethoxy-1,2,3,6-tetrahydropyridine (28f)**: yield, 90%; IR ($CHCl_3$) 1705–1680 (C=O, C=C); ^1H NMR (200 MHz, $CDCl_3$) 0.7–1.8 [m, 16 H, containing at 1.26 (t, J = 6.8, 3 H, OCH_2CH_3), at 1.46 (s, 9 H, $(CH_3)_3C$), 4 H, $CHCH_2CH_2CHN$] 1.8–2.6 (m, 2 H, $C=CHCH_2$), 3.3–4.5 (m, 2 H, CH_2N), 3.87 (t, J = 6.8, 1 H, $(C_6H_5)_2CHCH_2$), 4.2–4.7 [m, 3 H, containing at 4.20 (q, J = 6.8, 2 H, OCH_2CH_3), 1 H, CH_2CHCH_2], 6.8–7.0 (m, 1 H, $C=CHCH_2$), 7.1–7.4 (m, 10 H, 2 C_6H_5).

2-Alkyl- or -2-aralkyl-5-carbethoxy-1,2,3,6-tetrahydropyridine Hydrochlorides 29. General Procedure. A protected amine **28** as a 20% solution in absolute ethanol was treated with an equal volume of a saturated solution of hydrogen chloride in ethanol. The solution was heated under reflux for 2 h. Evaporation of the solvent under reduced pressure and crystallization with a mixture of 2-propanol and ethyl ether gave the expected hydrochlorides.

2-Ethyl-5-carbethoxy-1,2,3,6-tetrahydropyridine hydrochloride (29b): yield, 60%; mp 189 °C; IR (KBr) 1725 (C=O), 1660 (C=C); ^1H NMR (200 MHz, D_2O) 1.03 (t, J = 6.8 3 H, $CHCH_2CH_3$), 1.30 and 1.31 (2, t, J = 6.8, 3 H, OCH_2CH_3), 1.6–1.9 (m, 2 H, $CHCH_2CH_3$), 2.40 (ddq, J = 20.0, J = 9.5, J = 3.0, 1 H, H_{3a}), 2.75 (dt, J = 20.0, J = 3.0, 1 H, H_{3e}), 3.3–3.5 (m, 1 H, H_2), 3.9–4.0 (m, 2 H, containing the AB corresponding to H_{6a} and H_{6e}), 4.27 and 4.28 (2 q, J = 6.8, 2 H, OCH_2CH_3), 7.0–7.3 (m, 1 H, H_4).

2-Propyl-5-carbethoxy-1,2,3,6-tetrahydropyridine hydrochloride (29c): yield, 64%; mp 132.8 °C; IR (KBr) 1725 (C=O), 1660 (C=C); ^1H NMR (200 MHz, D_2O) 0.95 (t, J = 6.8, 3 H, $CH_2CH_2CH_3$), 1.30 (t, J = 6.8, 3 H, OCH_2CH_3), 1.45 (q, J = 6.8, 2 H, $CHCH_2CH_2CH_3$), 1.70 (sext, J = 6.8, 2 H, $CH_2CH_2CH_3$), 2.40 (ddq, J = 20.0, J = 9.5, J = 3.0, 1 H, H_{3a}, 2.75 (dt, J = 20.0, J = 3.0, 1 H, H_{3e}), 3.3–3.5 (m, 1 H, H_2), 3.8–4.1 (m, 2 H, H_{6a}, H_{6e}), 4.27 (q, J = 6.8, OCH_2CH_3), 7.0–7.3 (m, 1 H, H_4).

2-(3,3-Diphenylpropyl)-5-carbethoxy-1,2,3,6-tetrahydropyridine hydrochloride (29f): yield, 58%; mp 114 °C; IR (KBr) 1725 (C=O), 1660 (C=C); ^1H NMR (200 MHz, D_2O) 1.29 (t, J = 6.8, 3 H, OCH_2CH_3), 1.5–1.9 (m, 2 H, $CHCH_2CH_2CHN$), 2.0–2.6 (m, 3 H, H_{3a}, $CHCH_2CH_2CHN$), 2.75 (dt, J = 20.0, J = 3.0, H_{3e}), 3.3–3.6 (m, 1 H, H_2), 3.6–4.2 [m, 3 H, containing at 4.07 (t, J = 6.8, 1 H, $C_6H_5CHCH_2$), 2 H, H_{6a}, H_{6e}], 4.24 (q, J = 6.8, 2 H, OCH_2CH_3), 7.0–7.3 (m, 1 H, H_4), 7.3–7.5 (m, 10 H, 2 C_6H_5).

6-Alkyl- or -6-aralkyl-1,2,5,6-tetrahydropyridine-3-carboxylic Acid Hydrochlorides 30. General Procedure. A 3 N hydrochloric acid solution containing ester **28** or **29** was refluxed for 3 h. After evaporation of the solvent under reduced pressure, the crude product was recrystallized in a mixture of 2-propanol and ethyl ether.

6-Ethyl-1,2,5,6-tetrahydropyridine-3-carboxylic acid hydrochloride (6-ethylguvacine hydrochloride) (30b): yield, 46%; mp 218 °C dec; IR (KBr) 1725 (C=O), 1660 (C=C); ^1H NMR (200 MHz, D_2O) 1.01 (t, J = 6.8, 3 H, $CHCH_2CH_3$), 1.6–1.9 (m, 2 H, $CHCH_2CH_3$), 2.40 (ddq, J = 20.0, J = 9.5, J = 3.0, 1 H, H_{5a}), 2.72 (dt, J = 20, J = 3.0, 1 H, H_{5e}), 3.2–3.4 (m, 1 H, H_6), 3.7–4.1 (m, 2 H, H_{2a} and H_{2e}), 7.0–7.3 (m, 1 H, H_4).

6-Propyl-1,2,5,6-tetrahydropyridine-3-carboxylic acid hydrochloride (6-propylguvacine hydrochloride) (30c): yield, 50%; mp 220–222 °C dec; IR (KBr) 1720 (C=O), 1665 (C=C); ^1H NMR (200 MHz, D_2O) 0.93 (t, J = 6.8, 3 H, CH_2CH_3), 1.45 (q, J = 6.8, 2 H, $CHCH_2CH_2CH_3$), 1.70 (sext, J = 6.8, 2 H, $CH_2CH_2CH_3$), 2.36 (ddq, J = 20.0, J = 9.5, J = 3.0, 1 H, H_{5a}), 2.70 (dt, J = 20.0, J = 3.0, 1 H, H_{5e}), 3.3–3.6 (m, 1 H, H_6), 3.6–4.2 (m, 2 H, H_{2a} and H_{2e}), 7.0–7.4 (m, 1 H, H_4).

6-Phenethyl-1,2,5,6-tetrahydropyridine-3-carboxylic acid hydrochloride (6-phenethylguvacine hydrochloride) (30e): yield, 63%; mp 220 °C; IR (KBr) 1700 (C=O), 1650 (C=C); ^1H NMR (200 MHz, D_2O) 1.9–2.3 (m, 2 H, CH_2CH_2CH), 2.3–3.0 [m, 4 H, containing at 2.45 (ddq, J = 20.0, J = 9.5, J = 3.0, 1 H, H_{5a}), H_{5e}, CH_2CH_2CH], 3.2–3.5 (m, 1.4 H, 0.8 H_2 and 0.6 H_{6a}), 3.6–4.1 (m, 1.2 H, H_{2a} and H_{2e}), 4.30 (dd, J = 7.5, J = 1.5, 0.4 H_{6e}), 7.0–7.2 (m, 1 H, H_4), 7.2–7.5 (m, 5 H, C_6H_5).

6-(3,3-Diphenylpropyl)-1,2,5,6-tetrahydropyridine-3-carboxylic acid hydrochloride [6-(3,3-diphenylpropyl]guvacine hydrochloride) (30f): yield, 70%; mp 263 °C; IR (KBr) 1710 (C=O), 1655 (C=C); ^1H NMR (200 MHz, D_2O), 1.5–2.0 (m, 2 H, $CHCH_2CH_2CHN$), 2.1–2.7 (m, 4 H, $CHCH_2CH_2CHN$, H_{5a}, H_{5e}), 3.2–3.5 (m, 1.4 H, 0.8 H_2 and 0.6 H_{6a}), 3.6–4.2 [m, 2.2 H, containing at 4.00 and 4.01 (2 t, J = 6.8, 1 H, $C_6H_5CHCH_2$), 1.2 H, H_{2a} and H_{2e}], 4.3–4.4 (dd, J = 7.5, J = 1.5, 0.4 H_{6e}), 6.8–7.1 (m, 1 H, H_4), 7.2–7.4 (m, 10 H, 2 C_6H_5).

α,β-Unsaturated Esters 32. General Procedure. A flask containing 1 L of benzene, 0.9 mol of aldehyde **31**, 0.9 mol of triphenyl(carbethoxymethyl)phosphonium bromide, and 0.92 mol of crushed potassium hydroxide was heated at 40 °C for 2 h with stirring. The mixture was filtered, and the filtrate was evaporated under reduced pressure. The residue was taken up in 500 mL of hexane, slightly heated with stirring, and filtered again. The filtrate was cleaned by filtration through silica gel rinsed with a mixture of hexane–ethyl acetate, 1/3. The evaporation of the

solvent yielded the expected compounds.

Ethyl 2-pentenoate (32b):[70] yield, 60%; IR ($CHCl_3$) 1705 (C=O), 1655 (C=C); ^1H NMR (60 MHz, $CDCl_3$) 1.05 (t, J = 6.8, 3 H, CCH_2CH_3), 1.25 (t, J = 6.8, 3 H, OCH_2CH_3), 2.15 (q, J = 6.8, 2 H, CCH_2CH_3), 4.18 (q, J = 6.8, 2 H, OCH_2CH_3), 5.80 (dt, J = 15.0, J = 1.5, 1 H, O=CCH=CH trans), 7.02 (dt, J = 15.0, J = 6.8, 1 H, O=CCH=CHCH$_2$ trans).

Ethyl 2-hexenoate (32c):[71] yield, 65%; IR ($CHCl_3$) 1705 (C=O), 1655 (C=C); ^1H NMR (60 MHz, $CDCl_3$) 0.7–2.5 [m, 7 H, containing at 1.28 (t, J = 6.8, 3 H, OCH_2CH_3), 4 H, $CH_3CH_2CH_2CH$], 4.24 (q, J = 6.8, 2 H, OCH_2CH_3), 5.75 (dt, J = 15.0, J = 1.5, 1 H, O=CCH=CH), 7.00 (dt, J = 15.0, J = 6.8, 1 H, O=CCH=CHCH$_2$).

Ethyl 5-phenyl-2-pentenoate (32e): yield, 80%; IR ($CHCl_3$) 1715 (C=O), 1660 (C=C); ^1H NMR (60 MHz, $CDCl_3$) 1.28 (t, J = 6.8, 3 H, OCH_2CH_3), 2.3–3.0 (m, 4 H, CH_2CH_2), 4.18 (q, J = 6.8, 2 H, OCH_2CH_3), 5.83 (dt, J = 15.0, J = 1.5, 1 H, O=CCH=CH), 7.07 (dt, J = 15.0, J = 1.5, 1 H, O=CCH=CHCH$_2$), 7.24 (s, 5 H, C_6H_5).

Ethyl 6,6-diphenyl-2-hexenoate (32f): yield, 85%; IR ($CHCl_3$) 1710 (C=O), 1660 (C=C); ^1H NMR (200 MHz, $CDCl_3$) 1.26 (t, J = 6.8, 3 H, OCH_2CH_3), 2.0–2.2 (m, 4 H, $CHCH_2CH_2CH$=), 3.92 (t, J = 6.8, 1 H, $CHCH_2$), 4.15 (q, J = 6.8, 2 H, OCH_2CH_3), 5.80 (dt, J = 15.0, J = 1.5, 1 H, O=CCH=CH), 7.03 (dt, J = 15.0, J = 6.8, 1 H, O=CCH=CHCH$_2$), 7.0–7.4 (m, 10 H, 2 C_6H_5).

Ethyl 3-(Benzylamino)alkanoates or -aralkanoates 33. General Procedure. A solution of 0.4 mol of the α,β-unsaturated ester **32** and 0.5 mol of freshly distilled benzylamine in 150 mL of absolute ethanol was heated at reflux for 48 h. After evaporation of the solvent under reduced pressure, the product was purified by silica gel column chromatography with a mixture of hexane–ethyl acetate, 1/1, as eluent.

Ethyl 3-(benzylamino)butyrate (33a): yield, 80%; IR ($CHCl_3$) 1720 (C=O); ^1H NMR (60 MHz, $CDCl_3$) 1.15 (d, J = 6.8, 3 H, $CHCH_3$), 1.24 (t, J = 6.8, 3 H, OCH_2CH_3), 1.60 (br s, exchangeable with D_2O, 1 H, NH), 2.40 (dd, J = 6.8, J = 1.5, 2 H, O=CCH$_2$CH), 2.8–3.4 (m, 1 H, CH_2CHCH_3), 3.80 (s, 2 H, $NHCH_2C_6H_5$), 4.12 (q, J = 6.8, 2 H, OCH_2CH_3), 7.30 (s, 5 H, C_6H_5).

Ethyl 3-(benzylamino)valerate (33b): yield, 40%; IR ($CHCl_3$) 1725 (C=O); ^1H NMR (60 MHz, $CDCl_3$) 0.6–1.8 [m, 9 H, containing at 0.91 (t, J = 6.8, 3 H, $CHCH_2CH_3$), at 1.24 (t, J = 6.8, 3 H, OCH_2CH_3), at 1.49 (q, J = 6.8, 2 H, $CHCH_2CH_3$), at 1.57 (br s, exchangeable with D_2O, 1 H, NH)], 2.43 (d, J = 6.0, 2 H, O=CCH$_2$C$_6$H$_5$), 2.97 (qu, J = 6.0, 1 H, CH_2CHCH_2), 3.77 (s, 2 H, $CH_2C_6H_5$), 4.18 (q, J = 6.8, 2 H, OCH_2CH_3), 7.29 (s, 5 H, C_6H_5).

Ethyl 3-(benzylamino)caproate (33c): yield, 55%; IR ($CHCl_3$) 1725 (C=O); ^1H NMR (60 MHz, $CDCl_3$) 0.6–1.6 [m, 11 H, containing at 1.24 (t, J = 6.8, 3 H, OCH_2CH_3), at 1.51 (br s, exchangeable with D_2O, 1 H, NH) 7 H, $CHCH_2CH_2CH_2CH_3$], 2.43 (d, J = 6.0, 2 H, $CHCH_2C$=O), 2.8–3.2 (m, 1 H, CH_2CHCH_2), 3.77 (s, 2 H, $CH_2C_6H_5$), 4.13 (q, J = 6.8, 2 H, OCH_2CH_3), 7.29 (s, 5 H, C_6H_5).

Ethyl 3-(benzylamino)-5-phenylvalerate (33e): yield, 50%; IR ($CHCl_3$) 1720 (C=O); ^1H NMR (60 MHz, $CDCl_3$) 1.24 (t, J = 6.8, 3 H, OCH_2CH_3), 1.5–2.0 (m, 3 H, CH_2CH_2CHNH), 2.3–3.3 (m, 5 H, $CH_2CH_2CHCH_2C$=O), 3.77 (s, 2 H, $NCH_2C_6H_5$), 4.12 (q, J = 6.8, 2 H, OCH_2CH_3), 7.21 (s, 5 H, C_6H_5), 7.30 (s, 5 H, C_6H_5).

Ethyl 3-(benzylamino)-6,6-diphenylcaproate (33f): yield, 62%; IR ($CHCl_3$) 1725 (C=O); ^1H NMR (200 MHz, $CDCl_3$) 1.20 (t, J = 6.8, 3 H, OCH_2CH_3), 1.45 (q, J = 6.8, 2 H, $CHCH_2CH_2CHNH$), 1.58 (br s, exchangeable with D_2O, 1 H, NH), 2.10 (q, J = 6.8, 2 H, $CHCH_2CH_2CHNH$), 2.42 (d, J = 6.8, 2 H, O=CCH$_2$CH), 3.04 (qu, J = 6.8, 1 H, CH_2CHCH_2), 3.68 (s, 2 H, $CH_2C_6H_5$), 3.83 (t, J = 6.8, 1 H, $C_6H_5CHCH_2$), 4.08 (q, J = 6.8, 2 H, OCH_2CH_3), 7.0–7.4 (m, 10 H, 2 C_6H_5).

Ethyl 3-Aminoalkanoates or -aralkanoates 22. General Procedure. An ethanolic solution of the benzylamino derivative **33** was hydrogenated under a pressure of 4 atm for 3 h in the presence of 5% Pd/C. After filtration of the catalyst through a Celite pad, the solvents were evaporated under reduced pressure. The resulting residue was chromatographed through a silica gel column with ethyl acetate as eluent. The yield of the free secondary amine was generally around 95%.

Ethyl 3-aminobutyrate (22a):[72] IR ($CHCl_3$) 1725 (C=O); ^1H NMR (60 MHz, $CDCl_3$) 1.12 (d, J = 6.8, 3 H, $CHCH_3$) 1.27 (t, J = 6.8, 3 H, OCH_2CH_3), 1.73 (br s, exchangeable with D_2O, 2 H, NH_2), 2.2–2.5 (m, 2 H, O=CCH$_2$CH), 3.1–3.8 (m, 1 H, CH_2CHCH_2), 4.16 (q, J = 6.8, 2 H, OCH_2CH_3).

Ethyl 3-aminocaproate (22c):[73] IR ($CHCl_3$) 1725 (C=O); ^1H NMR (60 MHz, $CDCl_3$) 0.7–1.7 [m, 12 H, containing at 1.26 (t, J = 6.8, 3 H, OCH_2CH_3), at 1.47 (br s, exchangeable with D_2O, 2 H, NH_2), 7 H, $CHCH_2CH_2CH_2CH_3$], 2.2–2.5 (m, 2 H, O=CCH$_2$CH), 3.0–3.8 (m, 1 H, $CHCHCH_2$), 4.14 (q, J = 6.8, 2 H, OCH_2CH_3).

Ethyl 3-amino-6,6-diphenylcaproate (22f): IR ($CHCl_3$) 1725 (C=O); ^1H NMR (200 MHz, $CDCl_3$) 1.22 (t, J = 6.8, 3 H, OCH_2CH_3), 1.3–1.5 (m, 2 H, $CHCH_2CH_2CHNH_2$), 1.65 (br s, exchangeable with D_2O, 2 H, NH_2), 1.8–2.8 (m, 4 H, O=CCH$_2$CHCH$_2$CH$_2$CH), 3.0–3.4 (m, 1 H, CH_2CHCH_2), 3.87 (t, J = 6.8, 1 H, C_6H_5CH, CH_2), 4.12 (q, J = 6.8, 2 H, OCH_2CH_3), 7.0–7.4 (m, 10 H, 2 C_6H_5).

4,4-Diphenylbutanal (37). Method A. A mixture containing 90 g (0.31 mol) of bromide **36**, 600 mL of DMSO, 67.5 g (0.46 mol) of sodium iodide, and 77.8 g (0.93 mol) of sodium hydrogen carbonate was heated at 110 °C with stirring for 2 h. After cooling, water was added (200 mL) and the product was extracted with ethyl ether. The ethereal phase was dried over magnesium sulfate, and the solvents were vaporated under reduced pressure. Aldehyde **37** was purified by silica gel column chromatography with a mixture of hexane–ethyl acetate, 3/1, as eluent. The yield of aldehyde was 60%: IR ($CHCl_3$) 1720 (C=O); ^1H NMR (200 MHz, $CDCl_3$) 2.3–2.5 (m, 4 H, CH_2CH_2), 3.92 (t, J = 6.8, 1 H, $C_6H_5CHCH_2$), 7.0–7.4 (m, 10 H, C_6H_5), 9.73 (s, 1 H, O=CH).

Method B. A flask equipped with a mechanical stirrer and an argon intake, containing 325 mL of dichloromethane and 13 mL (0.15 mol) of oxalyl chloride, was cooled at −60 °C. DMSO (23 mL, 0.30 mol) in 68 mL of dichloromethane was slowly added. After 3 min, a solution of 30 g (0.14 mol) of alcohol **38** in 150 mL of dichloromethane was added during 5 min. After 15 min of stirring, triethylamine (95.5 mL, 0.68 mol) was added. The stirring was maintained for 5 min at −60 °C, and then the reaction mixture was allowed to warm to room temperature. Water was added (750 mL), and the organic layer was separated. The aqueous phase was extracted again with dichloromethane. The organic phases were combined, washed with water and brine, and dried over magnesium sulfate. The solvents were evaporated under reduced pressure, and the crude aldehyde was purified by silica gel column chromatography with ethyl acetate as eluent. The yield of pure aldehyde was 95%.

4,4-Diphenylbutanol (38). This compound was obtained as a side product (22%) in the preparation of 4,4-diphenylbutanal (**37**) using method A described above: ^1H NMR (200 MHz, $CDCl_3$) 1.35 (br s, exchangeable with D_2O, 1 H, OH), 1.53 (qu, J = 6.8, 2 H, CH_2CH_2OH), 2.14 (q, J = 6.8, 2 H, $CHCH_2CH_2$), 3.65 (t, J = 6.8, 2 H, CH_2OH), 3.93 (t, J = 6.8, 2 H, $C_6H_5CHCH_2$), 7.0–7.4 (m, 10 H, 2 C_6H_5).

[^{3}H]GABA Synaptosomal Uptake. Experiments were slightly modified from Ramsey et al.[61] Rats were killed by decapitation, and corpora striata were rapidly dissected. Tissues were pooled and homogenized in 20 volumes of 0.32 M sucrose on a Potter Elvehjem tissue grinder. Homogenates were centrifuged at 1000g at 4 °C for 15 min. The supernatant was centrifuged at 20000g at 4 °C, and the resulting pellet was resuspended in cold 0.32 M sucrose.

Incubation was carried out at 37 °C for 2 min in glass tubes containing 50 μL of synaptosomes (1 mg of protein), 750 μL of pH 7.4 Krebs-Ringer phosphate buffer supplemented with NaCl (0.15 M), and 100 μL of [^{3}H]GABA (25–40 Ci/mmol, New England Nuclear, Boston, MA), in a final concentration of 1.1 μmol, and 100 μL of compound to be tested. Blanks were treated identically

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except that NaCl was not added in the incubation medium. Uptake was determined by dilution with 5 mL of incubation medium without NaCl. Samples were centrifuged at 20000g at 4 °C for 15 min, and radioactivity was evaluated in pellets after dilution in 1 mL of Proposol (New England Nuclear).

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Registry No. 1, 56-12-2; 2, 5130-17-6; 3, 102629-73-2; 4, 102629-74-3; 5, 498-95-3; 6, 68464-00-6; 7, 498-96-4; 8, 85375-85-5; 9, 85375-88-8; 10, 134525-28-3; 11, 89203-64-5; 12, 89203-55-4; 13, 53602-00-9; 14, 132033-96-6; 15, 98943-12-5; 16, 98976-55-7; 17, 134420-65-8; 18, 71622-29-2; 19, 71622-34-9; 20, 134455-45-1; 21, 1217-82-9; 22a, 5303-65-1; 22c, 59663-70-6; 22d, 10039-64-2; 22e, 134420-70-5; 22f, 134420-71-6; 23b, 134420-67-0; 23e, 134420-72-7; 23f, 134420-73-8; 24a, 95629-08-6; 24b, 134420-74-9; 24c, 134420-75-0; 24d, 134420-76-1; 24e, 134420-77-2; 24f, 134420-78-3; 25a, 116140-33-1; 25b, 134420-79-4; 25c, 134420-80-7; 25e, 134420-81-8; 25f, 134420-82-9; 28a, 116140-36-4; 28b, 134420-83-0; 28c, 134420-84-1; 28e, 134420-85-2; 28f, 134420-86-3; 29b, 134420-68-1; 29c, 134420-87-4; 29f, 134420-88-5; 30b, 134420-69-2; 30c, 134420-89-6; 30e, 134420-90-9; 30f, 134420-91-0; 32b, 24410-84-2; 32c, 1552-67-6; 32e, 6048-08-4; 32f, 134420-92-1; 33a, 6335-80-4; 33b, 134420-93-2; 33c, 134455-46-2; 33e, 134420-94-3; 33f, 134420-95-4; cyclopropyldiphenylmethanol, 5785-66-0; 4,4-diphenylbutanol, 56740-71-7; ethyl acrylate, 140-88-5; triphenyl(carbethoxymethyl)phosphonium bromide, 1530-45-6; benzylamine, 100-46-9.

3-Thienyl- and 3-Furylaminobutyric Acids. Synthesis and Binding $GABA_B$ Receptor Studies[1]

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Baclofen (β-(*p*-chlorophenyl)-GABA) is a selective agonist for the bicuculline-insensitive $GABA_B$ receptor. The search for new compounds that bind to the $GABA_B$ receptor is very important to clarify structural requirements. We report herein the synthesis and the binding studies of variously substituted 3-thienyl- and 3-furylaminobutyric acids. 4-Amino-3-(5-methyl-2-thienyl)butyric acid (**5d**) and 4-amino-3-(5-chloro-2-thienyl)butyric acid (**5h**) are potent and specific ligands for $GABA_B$ receptor. The IC_{50} values for the displacement of (*R*)-(−)-[^{3}H]baclofen are 1.34 and 0.61 μM for **5d** and **5h**, respectively, as compared to 0.33 μM for baclofen.

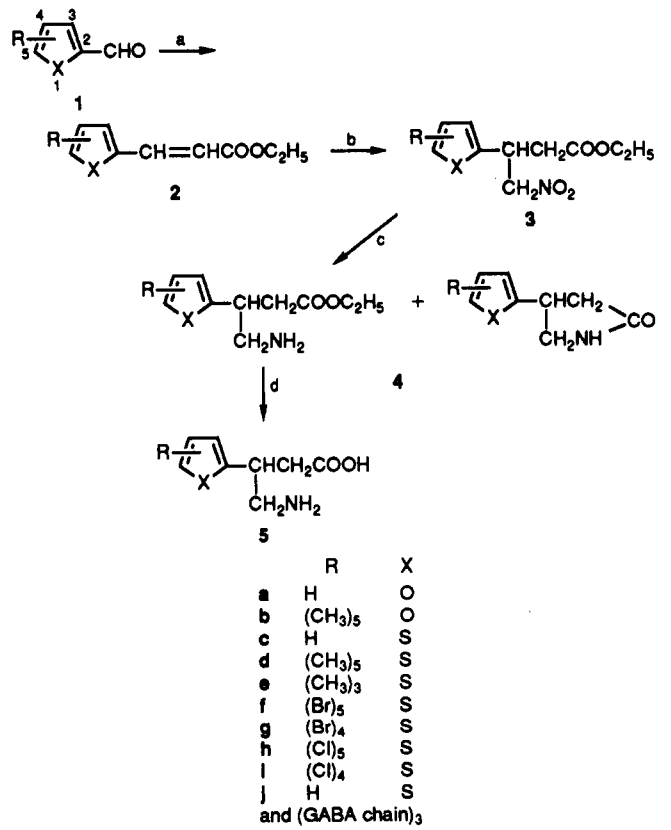
The neutral amino acid γ-aminobutyric acid (GABA) is an inhibitory neurotransmitter concerned with the control of neuronal activity in the mammalian central nervous system and with the regulation of many physiological mechanisms.[2] Within the central and peripheral nervous system, GABA has been shown to act through at least two distinctly different receptor sites,[3] termed $GABA_A$ and $GABA_B$ receptors, with different binding properties.[4,5] Accumulating evidence suggests that $GABA_B$ receptors are predominantly located presynaptically.[6] However, in a recent report, postsynaptically located $GABA_B$ receptor have been described.[7] $GABA_B$ receptors have also been detected and characterized in a variety of tissue preparations of peripheral origin.[8]

Until now, β-(*p*-chlorophenyl)-GABA (baclofen) was the selective agonist for the $GABA_B$ receptor. Analogues of baclofen, saturated or unsaturated, have been synthesized and tested for $GABA_B$ receptor affinity. These compounds showed no selective action at $GABA_B$ receptor sites in vitro.[9] The only new agonist available is (3-aminopropyl)phosphinic acid, which is a potent displacer of baclofen in binding studies.[10] However, this compound can act as a partial agonist under certain conditions.[11]

In recent papers, the phosphonic analogue of baclofen (phaclofen) and two sulfonic analogues (saclofen and 2-hydroxysaclofen) have been shown to be antagonists at $GABA_B$ receptors.[12,13] We recently described the synthesis of 3-(benzo[*b*]furan-2-yl)-GABA, new selective ligands of $GABA_B$ sites,[14] which are specific $GABA_B$ receptor antagonists.[15,16]

In the course of our work and in attempts to elucidate the structural requirements for access to the $GABA_B$ receptor,[14] we report the synthesis and the binding studies of new 3-heteroaryl-GABA analogues. These racemic compounds, especially **5d** and **5h**, are potent and specific

Scheme I[a]



	R	X
a	H	O
b	$(CH_3)_5$	O
c	H	S
d	$(CH_3)_5$	S
e	$(CH_3)_3$	S
f	$(Br)_5$	S
g	$(Br)_4$	S
h	$(Cl)_5$	S
i	$(Cl)_4$	S
j	H	S

and (GABA chain)$_3$

[a] Reagents: (a) $(C_6H_5)PCHCOOC_2H_5/C_6H_6$/reflux; (b) CH_3NO_2/Triton B/85 °C; (c) Raney Ni/H_2; (d) NaOH/EtOH/reflux.

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