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BIOLOGICAL ACTIVITY OF 3-AMINOPROPYL (METHYL) PHOSPHINIC ACID, A POTENT AND SELECTIVE $GABA_B$ AGONIST WITH CNS ACTIVITY

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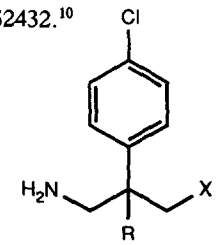
Abstract : The $GABA_B$ receptor affinity of a number of GABA analogues, where the carboxylic acid group has been replaced with a range of acidic moieties, were determined. The phosphinic acid analogue (1) was identified as a potent selective $GABA_B$ ligand which is CNS penetrating and 10-fold more active than baclofen (2).

The amino acid GABA (3) is the major inhibitory neurotransmitter in the mammalian central nervous system.[1] Receptors for GABA are subdivided into $GABA_A$ and $GABA_B$. The $GABA_A$ receptors (the classical GABA receptor) has been exploited therapeutically in the benzodiazepine anxiolytics which modulate it.[2]

Evidence for the $GABA_B$ receptor was first described by Bowery and colleagues[3] in the early eighties, but there is still a dearth of appropriate chemical tools. The only selective potent $GABA_B$ agonists are baclofen (2)[4] and the recently introduced phosphonous acid, CGP 27492 (4).[5] Until recently there were no potent $GABA_B$ antagonists described, only a number of weak compounds which include phaclofen (5),[6] saclofen (6),[7] 2-hydroxysaclofen (7)[8] and a series of phosphinic acids represented by CGP 35348 (8).[9] Futher studies around compounds such as 8 have led Fröstl et al to describe a series of new potent $GABA_B$ antagonists exemplified by CGP 52432.[10]



	R	X
(2)	H	CO_2H
(5)	H	PO_3H_2
(6)	H	SO_3H
(7)	OH	SO_3H

Our interest in this field, prompted us to undertake a SAR study concerning bioisosteric replacements for the carboxylic acid moiety in GABA, as $GABA_B$ ligands. This led to the discovery of 3-aminopropyl(methyl)phosphinic acid, SK&F 97541 (1)[11] as the most potent, selective $GABA_B$ agonist to date. Here we describes the biological activity of this compound, and a comparison with the $GABA_B$ binding affinity of other GABA analogues where the carboxylic acid has been replaced (Table I).

The phosphinic acids **1**, **9**, **10**, the phosphonous acid **4**, and the (diethoxymethyl) phosphinic acid **8** were prepared by the methods of Howson,[11] Dingwall[5] and Bayliss[9] respectively.

Using an in vitro binding assay[12] (displacement of [^{3}H]-GABA from $GABA_B$ sites in rat brain membranes) the affinity of the compounds were assessed; the results are shown in Table I. This led to the following conclusions concerning structure and affinity. The compounds GABA (**3**) and baclofen (**2**), which possess the planar carboxylic acid functionality where the charge is spread over just two heteroatoms, were shown to have affinities in the 0.1 - 0.01 μM range for the $GABA_B$ receptor. All compounds which have an acidic moiety where the charge is spread over more than two heteroatoms have significantly weaker affinity. These include derivatives with distorted tetrahedral acidic groups i.e. **11-14**, and the planar monocharged tetrazole GABA analogue.[13] If the acidic moiety is a singly charged distorted tetrahedral group, with the charge spread over just two oxygen atoms, such as in **1** and **4**, high affinity agonists for the $GABA_B$ receptor are obtained.[14] Increasing the size of the alkyl substituent on the phosphorous atom affords weakly active antagonists (**8**, **9**, **10**) at $GABA_B$ receptors.[9,14b]

The phosphinic acid **1** was subsequently shown to be highly selective for the $GABA_B$ receptor vs the $GABA_A$, binding to the $GABA_B$ receptor with IC_{50} = 0.001 μM and to the $GABA_A$ receptor with IC_{50} > 100 μM, a selectivity ratio of 10^5.[11] Its agonist activity[15] on a number of in vitro peripheral $GABA_B$ receptor assays (guinea-pig ileum, rat vas deferens and anococcygeus) was invariable an order of magnitude greater than baclofen, but similar to the phosphonous acid **4**.

While in two in vitro CNS $GABA_B$ preparations (rat substantia nigra and striatum slices) differences between **1** and **4** were observed.[14b] Here, **1** was ten times more potent than **4** and **4** was equipotent or less potent than baclofen, **4** also showed partial agonist activity.

Further studies have shown the major difference between the two acids **1** and **4** is their apparent ability to penetrate the CNS. Hypothermia in the mouse is known to be a feature of the central action of baclofen.[16] In studies[17] comparing the hypothermic effects of **1** and **4** with baclofen, it was found that **1** (0.1 - 1 mg/Kg i.p.) and baclofen (1 - 10 mg/Kg i.p.) caused a marked and dose related decrease in body temperature. While **4**, studied at doses up to 5 mg/Kg i.p. was without effect on mouse body temperature. All of the agonist effects of the phosphinic acid **1** described above could be antagonised by the hexyl analogue **10**.[14b,17]

In summary, the affinity of a number of GABA analogues where the acidic group has been replaced have been studied. This led to the identification of the phosphinic acid **1**, a potent selective $GABA_B$ ligand which is CNS penetrating and some ten-fold more active than baclofen.

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TABLE 1. Carboxylic Acid Bioisosteres of GABA and their $GABA_B$ Binding Affinity.

No	n	$H_2N(CH_2)_nX$ X	$GABA_B$ receptor binding, IC_{50} (μM)
1	3	$\begin{matrix} O \\ \| \\ POH \\ \| \\ CH_3 \end{matrix}$	0.001
2 ((-)Baclofen)			0.03
3	3	CO_2H	0.06
4	3	PO_2H_2	0.003
8	3	$\begin{matrix} O \\ \| \\ POH \\ \| \\ CH(OEt)_2 \end{matrix}$	2.5
9	3	$\begin{matrix} O \\ \| \\ POH \\ \| \\ CH_2CH_3 \end{matrix}$	3
10	3	$\begin{matrix} O \\ \| \\ POH \\ \| \\ \text{n-hexyl} \end{matrix}$	1
11	3	PO_3H_2	2
12	2	OPO_3H_2	> 100
13	3	SO_3H	2
14	2	OSO_3H	> 100

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