Bioorganic & Medicinal Chemistry Letters, Vol.3, No.1, pp. 39-42, 1993
Printed in Great Britain

0960-894X/93 $5.00 + .00

SYNTHESIS AND ANTICONVULSANT ACTIVITY OF THE (+)- AND (-)-ENANTIOMERS OF 1,2,3,4-TETRAHYDRO-5-(2-PHOSPHONOETHYL)-3-ISOQUINOLINECARBOXYLIC ACID, A COMPETITIVE NMDA ANTAGONIST

Christopher F. Bigge,* Jiang-Ping Wu, Thomas C. Malone,
Charles P. Taylor and Mark G. Vartanian

Parke-Davis Pharmaceutical Research Division
Warner-Lambert Company
2800 Plymouth Road
Ann Arbor, Michigan 48105

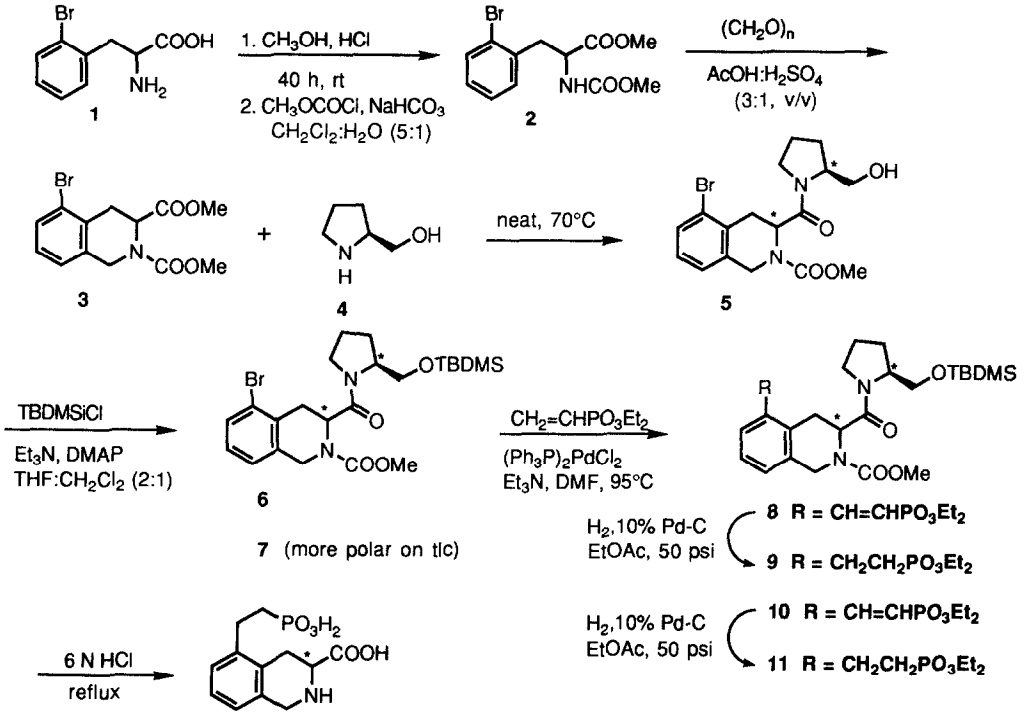
Abstract. The (+)- and (-)-enantiomers of 1,2,3,4-tetrahydro-5-(2-phosphonoethyl)-3-isoquinolinecarboxylic acid (**12** and **13**) were prepared. The anticonvulsant activity of **12** and **13** and their parent (**14**) was compared to the reference agent, CPP, in the maximal electroshock assay in mouse. **12** was similarly potent to CPP, three-fold more potent than the racemate and twenty-fold more potent than **13** in this assay.

Excitatory amino acid (glutamate) receptors are responsible for mediating many synaptic excitation processes in the mammalian brain. The development of selective antagonists has contributed to the rapid advancement of the pharmacology of the N-methyl-D-aspartate (NMDA) receptor subtype, whose ion channel (permeable to Na^+ and Ca^{++}) is voltage dependent, and regulated by the coagonists glutamate and glycine, as well as a number of other modulatory agents.[1] Recent studies have demonstrated that NMDA receptors may play a role in the initiation and/or propagation of epileptiform discharge, and the number of NMDA and glycine receptors has been shown to increase in the kindling model of complex partial epilepsy, which may reveal a role of the NMDA receptor in the etiology of epilepsy.[2] Several types of NMDA receptor antagonists have anticonvulsant properties, including competitive antagonists[3], noncompetitive antagonists[4] and glycine site antagonists.[5] Such compounds have demonstrated this activity in a diversity of models, and with appropriate potency, selectivity and bioavailability may be useful for the treatment of epilepsy, as well as ischemic neurodegeneration.[6] However, in order to have utility in the clinic, NMDA receptor antagonists must be able to cross the blood-brain barrier and must suppress seizures at doses that do not produce unwanted effects on brain function.

Recently, a series of isoquinoline derivatives were synthesized as NMDA receptor antagonists to help define the competitive antagonist binding site on the NMDA receptor complex.[7] In this study, we have synthesized both the (+)- and (-)-enantiomers of 1,2,3,4-tetrahydro-5-(2-phosphonoethyl)-3-isoquinolinecarboxylic acid (**14**), the most potent derivative found in this series, and have examined them in a maximal electroshock model (MES) in mouse. o-Bromophenylalanine **1** was protected as the carbamate methyl ester (**2**). Subsequent amidoalkylation with paraformaldehyde in a mixture of acetic and sulfuric acids (3:1, v/v) gave the requisite dimethyl 5-bromo-3,4-dihydro-2,3(*1H*)-isoquinolinedicarboxylate **3**. Condensation of **3** with (S)-2-hydroxymethylpyrolidine **4**, gave a mixture of the diastereomeric amides **5**. Unfortunately, these amides gave a single spot on tlc, and were not separable by column chromatography. However, after protection of the

hydroxyl group as the *tert*-butyldimethylsilyl ether, separation of the protected diastereomeric amides (**6** and **7**) could be achieved by elution on silica gel with 2:1 hexane/ethyl acetate. The pure diastereomers were carried separately through the rest of the reaction sequence. The arylbromide moeity (**6** or **7**) was palladated using bis(triphenylphosphine)palladium(II) chloride and condensed with diethyl vinylphosphonate under Heck-type conditions. After reduction of the double bond, the protecting groups were removed by hydrolysis to give the desired (+)- and (-)-enantiomers (**12** and **13**).

Scheme. Synthesis of (+)- and (-)-enantiomers of 1,2,3,4-tetrahydro-5-(2-phosphonoethyl)-3-isoquinolinecarboxylic acid.



Maximal electroshock (mouse) has been widely used to determine potential anticonvulsant utility in generalized tonic-clonic seizures in humans.[8] Potent anticonvulsants in this model would seem to predict efficacy against this seizure type in epileptic patients. In addition, animals were examined for ataxia, a measure of undesirable central side effects using an inverted screen ataxia model.[9] Of the isoquinoline derivatives described in our recent publication,[7] the most potent antagonist at the competitive NMDA receptor site as measured in a [^{3}H]-CPP binding assay of rat brain neuronal membranes[10] was the title compound (**14**, IC_{50} = 0.27 μM; compare

with the reference agents AP7[11] and CPP[12], IC_{50}'s = 0.77μM and 0.079 μM, respectively). **14** was calculated to have a log P 1.4 log units higher than the reference agent, CPP (measured log P = -3.4),[12c] because of the fused aromatic functionality, and thus **14** could be expected to have enhanced CNS penetration and biological activity relative to its NMDA site binding affinity. A comparison between **14** and CPP in two models in cell culture showed that the compounds have similar intrinsic activity in two functional models of NMDA antagonist activity: glutamate induced calcium influx[13] and glutamate induced LDH release[14] in cultured neuronal cells (IC_{50}'s = 2.4 μM and 9 μM for CPP; IC_{50}'s = 5.1 μM and 8.8 μM for **14**), but no significant difference was found in their in vivo anticonvulsant activity relative to their binding affinity.

Table. Comparison of Anticonvulsant Effect in Maximal Electroshock in Mouse and CNS Side Effects of Racemate and Enantiomers of 1,2,3,4-tetrahydro-5-(2-phosphonoethyl)-3-isoquinolinecarboxylic acid.

Drug	MES ED_{50} (mg/kg)	Ataxia ED_{50} (mg/kg)	TI
CPP	2.35	5.21	2.21
	[1.52 to 3.62]*	[3.82 to 7.11]*	
14	9.20	15.84	1.72
	[4.41 to 19.22]*	[11.66 to 21.51]*	
12 (+)	3.56	6.29	1.76
	[2.63 to 4.83]*	[4.04 to 9.78]*	
13 (-)	[a]	[b]	----

* - Probit analysis [95% confidence interval]
all animals dosed intravenously; N = 10 mice/dose; MES tested at 1 h and Ataxia tested at 30 min.
a - 8 of 10 mice protected at 100 mg/kg
b - 3 of 10 mice were ataxic at 100 mg/kg

Male CF-1 mice, 22-27 g, from Charles River Laboratories were used in all of the maximal electroshock experiments. Mice were given various doses of **14**, or the (+)- and (-)- enantiomers, **12** and **13**, dissolved in 0.9% NaCl intravenously by injection into the retrobulbar venous sinus. All doses were given in a volume of 10 mL/kg. Mice were given various doses of the compounds intravenously and tested one hour later. Methods used were similar to those published elsewhere (Swineyard).[15] From dose-effect experiments, the doses producing effects in 50% of mice (ED_{50}) were estimated by probit analysis.[16] For the inverted screen ataxia measurements, mice were placed individually on four inch square wire mesh screens. The screens were slowly inverted and animals were observed for one minute. Control animals reliably clung to the screen or climbed to the top. Any animal which fell from the screen was rated ataxic. The therapeutic index (TI) reported in the Table is the ratio of the ED_{50} for ataxia and the ED_{50} to prevent seizures. Results with these competitive NMDA antagonists indicate that there was only a small separation of the anticonvulsant activity and the untoward CNS effects (ataxia). For this class of compound, both actions are predominantly produced by the (+)-enantiomer, which is on the order of twenty-fold more potent than the (-)-enantiomer. Remarkably, the measure of therapeutic index gives identical values for the (+)-enantiomer and the racemate. Similar results have been observed previously with DL-(*E*)-2-4-methyl-5-phosphono-3-pentenoic acid (CGP 37849). In an oral dosing paradigm in maximal electroshock, the *R*-(+)-enantiomer (ED_{50} = 7 mg/kg) was three times more effective than the racemate (ED_{50} = 21 mg/kg), and the *S*-

(-)-enantiomer was ineffective at a 10 fold higher dose (ED_{50} > 60 mg/kg).[3a] Although we have not determined the absolute stereochemistry of **12** and **13**, our molecular modeling studies and comparison with literature compounds of this class suggest that the (+)-enantiomer is of the *R*-configuration.

Our results confirm those previously published with other competitive NMDA antagonists, and indicate that anticonvulsant action is expected at doses that overlap with those producing behavioral side effects such as ataxia. These findings are consistent with those of Loescher[3d], and suggest that competitive NMDA antagonists may have limited utility for clinical treatment of epilepsy because of unwanted effects on central nervous system function.

References

1. For recent reviews of NMDA receptor modulation and function see: a. Wong, E. H. F.; Kemp, J. A. *Annu. Rev. Pharmacol. Toxicol.* **1991**, 31, 401; b. Johnson, G.; Bigge, C. F. Annu. Rep. Med. Chem.; Bristol, J. A., Ed.; Academic Press: **1991** Vol. 26, 11-22.
2. a. Dingledine, R.; McBain, C. J.; McNamara, J. O. *Trends Pharmacol. Sci.* **1990**, 11, 334; b. Yeh, G. C.; Bonhaus, D. W.; Nadler, J. V., McNamara, J. O. *Proc. Natl. Acad. Sci.* **1989**, 86, 8157; c. Nevins, M. E.; Arnold, S. M. *Brain Res.* **1989**, 503, 1; d. Meldrum, B. *Epilepsy Res.* **1991**, 10, 55;
3. a. Fagg, G. E.; Olpe, H.-R.; Pozza, M. F.; Baud, J.; Steinmann, M.; Schmutz, M.; Portet, C.; Baumann, P.; Thedinga, K.; Bittiger, H.; Allgeier, H.; Heckendorn, R.; Angst, C.; Brundish, D.; Dingwall, J. G. *Br. J. Pharmacol.* **1990**, 99, 791; b. Hutchison, A. J.; Williams, M.; Angst, C.; de Jesus, R.; Blanchard, L.; Jackson, R. H.; Wilusz, E. J.; Murphy, D. E.; Bernard, P. S.; Schneider, J.; Campbell, T.; Guida, W.; Sills, M. A. *J. Med. Chem.* **1989**, 32, 2171; c. Ferkany, J. W.; Kyle, D. J.; Willets, J.; Rzeszotarski, W. J.; Guzewska, M. E.; Ellenberger, S. R.; Jones, S. M.; Sacaan, A. I.; Snell, L. D., et al. *J. Pharmacol. Exp. Ther.* **1989**, 250, 100; d. Loescher, W.; Hoenack, D. *J. Pharmacol. Exp. Ther.* **1991**, 256, 432; e. Davies, J.; Evans, R. H.; Herrling, A. W. Jones, A. W.; Olverman, H. J.; Pook, P.; Watkins, J. C. *Brain Res.* **1986**, 382, 169; f. Patel, S.; Chapman, A. G.; Graham, J. L.; Meldrum, B. S.; Frey, P. *Epilepsy Res.* **1990**, 7, 3.
4. a. Leander, J. D.; Rathbun, R. C.; Zimmerman, D. M. Brain Res. **1988**, 454, 368; b. Wong, B. Y.; Coulter, D. A.; Choi, D. W.; Prince, D. A. *Neurosci. Lett.* **1988**, 85, 261; c. Wong, E. H. F.; Kemp, J. A.; Priestley, T. Knight, A. R.; Woodruff, G. N.; Iversen, L. L. *Proc. Natl. Acad. Sci.* (USA) **1986**, 83, 7104.
5. a. Singh, L.; Donald, A. E.; Foster, A. C.; Hutson, P. H.; Iversen, L. L.; Iversen, S. D.; Kemp, J. A.; Leeson, P. D.; Marshall, G. R.; Oles, R. J.; et al. *Proc. Natl. Acad. Sci.* (USA) **1990**, 87, 347; b. Vartanian, M. G.; Taylor, C. P. *Neurosci. Lett.* **1991**, 133, 109.
6. Croucher, M. J.; Collings, J. F.; Meldrum, B. S. *Science* **1982**, 216, 899.
7. Ortwine, D. F.; Malone, T. C.; Bigge, C. F.; Drummond, J. T.; Humblet, C.; Johnson, G.; Pinter, G. W. *J. Med. Chem.* **1992**, 35, 1345.
8. a. Bernasconi, R.; Jones, R. S. G.; Bittiger, H.; Olpe, H.-R.; Martine, P.; Klein, M.; Loo, P.; Braunwalder, A.; Schmutz, M. *J. Neural Transm.* **1986**, 67, 175; b. Löscher, W.; Schmidt, D. *Epilepsy Res.* **1988**, 2, 145.
9. Coughenour, L. L.; McLean, J. R.; Parker, R. *Pharmacol. Biochem. Behav.* **1977**, 6, 351.
10. Murphy, D. E.; Schneider, J.; Boehm, C.; Lehmann, J.; Williams, M. *J. Pharm. Exp. Ther.* **1987**, 240, 778.
11. AP7 (2-amino-7-phosphonoheptanoic acid); Perkins, M. N.; Stone, T. W.; Collins, J. F.; Curry, K. *Neurosci. Lett.* **1981**, 23, 333.
12. CPP (4-(3-phosphonopropyl)-2-piperazine carboxylic acid); a.Harris, E. W.; Ganong, A. H.; Monaghan, D. T.; Watkins, J. C.; Cotman, C. W. *Nature* **1986**, 382, 174; b. Bigge, C. F.; Hays, S. J.; Novak, P. M.; Drummond, J. T.; Johnson, G.; Bobovski, T. P. *Tetrahedron Lett.* **1989**, 30, 5193; c. Hays, S. J.; Bigge, C. F.; Novak, P. M.; Drummond, J. T.; Bobovski, T. P.; Rice, M. J.; Johnson, G.; Brahce, L. J.; Coughenour, L. L. *J. Med. Chem.* **1990**, 33, 2916.
13. Marcoux, F. W.; Goodrich, J. E.; Probert, A. W., Jr.; Dominick, M. A. *Sigma and Phencyclidine-like Compounds as Molecular Probes in Biology*; Domino, E. F.; Kamenka, J.-M., Eds.; NPP Books: Ann Arbor, MI, **1988**, 735-746.
14. Koh, J. Y.; Choi, D. W. *J. Neurosci. Methods* **1987**, 20, 83.
15. Piredda, S. G.; Woodhead, J. H.; Swinyard, E. A. *J. Pharm. Exp. Ther.* **1985**, 232, 741.
16. Litchfield, J. T.; Wilcoxon, F. *J. Pharmacol* **1949**, 96, 99.