Bioorganic & Medicinal Chemistry Letters, Vol.3, No.2, pp. 299-304, 1993
Printed in Great Britain

0960-894X/93 $6.00 + .00

AMINO ACID BIOISOSTERES: DESIGN OF 2-QUINOLONE DERIVATIVES AS GLYCINE-SITE N-METHYL-D-ASPARTATE RECEPTOR ANTAGONISTS

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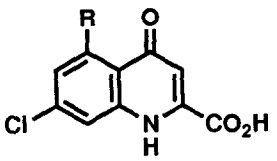
(*Received in USA* 29 *October* 1992)

Abstract: 3-Substituted-2-quinolones (**6-8**) have been identified as glycine-site N-methyl-D-aspartate receptor antagonists. It is proposed that the α-phenyl lactam unit in the potent 4-hydroxy-3-phenyl derivatives (**7d** and **8b**, L-701,315) may act as a glycine bioisostere in receptor recognition.

Antagonists of the N-methyl-D-aspartate (NMDA) subtype of excitatory amino acid receptor have aroused considerable interest as potential agents for the treatment of central nervous system disorders such as cerebral ischaemia, head injury, epilepsy, schizophrenia and Parkinson's disease.[1] Rapid progress has been made in the development of antagonists which act at the glycine modulatory site associated with the NMDA receptor, but existing compounds, including kynurenic acid derivatives (**1a-c**)[2], 2-carboxy tetrahydroquinolines (**2**, L-689,560),[3] quinoxaline-2,3-diones (**3**),[4] and α-amino hydroxamates (**4**, L-687,414),[5] are limited either by poor *in vivo* properties (**1-3**)[6,7] or highly restrictive structure-activity requirements (**4**).[8]



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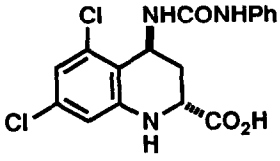
1b R = Cl
1c R = I



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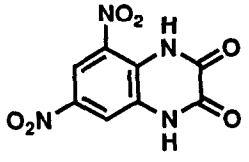

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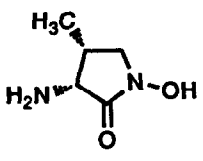

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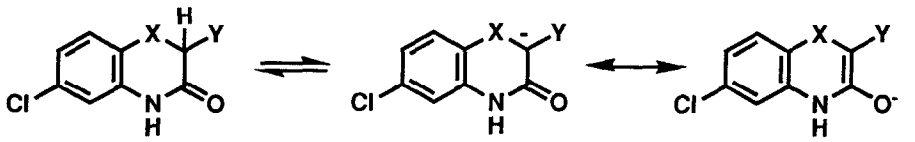
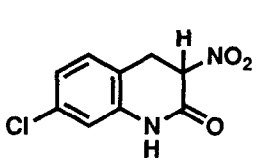
One approach we have explored to identify alternative glycine antagonists has focussed on replacement of the acidic 2-carboxyl in **1** and **2** with suitable bioisosteric groups. In this report we show that placing an acidic moiety within the heterocyclic ring of 2-quinolone derivatives leads to potent glycine antagonists. It is suggested that these compounds, exemplified by the highly active 4-hydroxy-3-phenyl-2-quinolones **7d** and **8b**, contain a novel amino acid bioisostere.

The design strategy employed followed from analysis of structure-activity considerations in compounds **1-3** and their derivatives.[2,3] The key structural features required for activity are: small (usually hydrophobic) 5- and 7-substitutents, the 1-NH group, an optimally positioned carbonyl at the 4- (**1** and **2**) or 3- (**3**) positions, and an in-plane anionic group, represented by the carboxylic acids of **1** and **2** (pKa ~ 4) and the 4-NH of **3** (pKa 6.5). These features led us to propose a glycine antagonist pharmacophore[2,3] in which the non basic 1-NH and acidic moieties were suggested to mimic the receptor binding of glycine. The activities of **2** and **3** indicate that relative location of the acidic group may not be critical in receptor recognition, and we therefore considered structures containing alternative acidic functionality within the fused heterocyclic ring. In compounds of the general structure **5**, the 1,2-lactam unit found in **3** is combined with an ionizable CH group at the adjacent 3-position. We anticipated that selection of suitable X and Y groups in **5** would permit manipulation of 3-CH pKa and the distribution of negative charge in the anion (**5a**), as well as providing possible scope for optimization of hydrogen bonding and hydrophobic interactions with the receptor site.



5 **5a**



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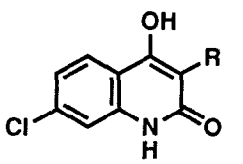
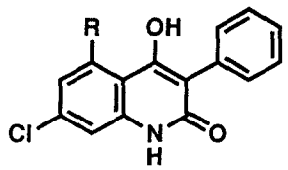


7a R = H
7b R = CO_2CH_3
7c R = COPh
7d R = Ph
7e R = c-hexyl

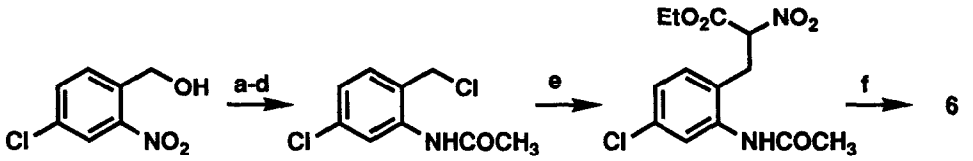


8a R = Cl
8b R = Et

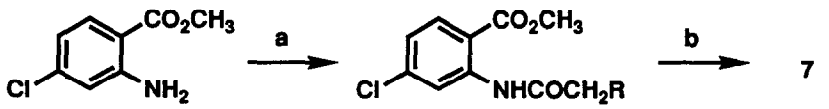
We tested the concept by synthesis of two classes of 2-quinolones **5** where: (a) X is methylene and Y is a strong electron withdrawing group, for example 3-nitro-3,4-dihydro-2-quinolone (**6**) and (b) X is carbonyl and Y a range of substitutents, compounds which exist as the 4-hydroxy-

2-quinolone tautomers (**7a-e**). 3-Substituents (Y) in molecules **7a-e** were chosen to both modify acidity and to attempt to exploit the bulk tolerance site on the receptor revealed by the 4-substituent of **2** and its derivatives.[3]

The 3-nitro derivative **6** was prepared from 2-nitro-4-chlorobenzyl alcohol (Scheme 1). The synthesis of 4-hydroxy-2-quinolones (**7**) is well documented[9] and an improved method was developed, allowing access to the desired compounds in two steps from methyl 2-amino-4-chlorobenzoate and the appropriate substituted acetic acids (Scheme 2). 5-Substituted compounds **8a,b** were prepared in a similar manner.



Scheme 1. a, H_2, Pt S-C; b, CH_3COCl, Et_3N; c, NaOH; d, CCl_4, Ph_3P; e, $NaCH(NO_2)CO_2Et$; f, HCl, MeOH.



Scheme 2. a, RCH_2CO_2H, $(COCl)_2$, catalytic Me_2NCHO; b, 2 equiv. $KN(SiMe_3)_2$.

The measured pKa of the α-nitro carbonyl derivative **6** (5.8) confirms its expected acidity and shows that the nitronate anion is the major species at physiological pH (7.4). The pKa values of the 4-hydroxy-2-quinolones **7a** (5.8), **7b** (4.4) and **7d** (5.4) correlate with the electron-withdrawing ability of the 3-substituent but in each case the anion (derived from deprotonation of the 4-hydroxyl) predominates at pH 7.4. *In vitro* binding affinity at the glycine site and NMDA antagonist activity for the new quinolones were determined by established methods[10,11] and are compared with reference compounds **1-4** in the Table.

The comparable activities of compound **6** and 7-chlorokynurenic acid (**1a**) show that the α-nitro carbonyl anion in **6** is an effective bioisosteric replacement of both the 2-carboxylate[12] and 4-oxo substituents in **1a**. Although the 4-hydroxy-2-quinolone derivatives (**7**) may be considered to be "hybrid" structures of the kynurenates (**1**) and quinoxalinediones (**3**), the parent 3-unsubstituted compound **7a** is not active. The results clearly show that the nature of the individual 3-substitutents in **7a-e**, rather than acidity, is the dominant influence on binding.

Table. Glycine-site affinities and NMDA antagonist activities of quinolone derivatives and reference compounds.

Compound	IC_{50} (μM)[a] [^{3}H]L-689,560	K_b (μM)[b] NMDA
6	0.41	6.7
7a	>300	-
7b	6.5	24
7c	3.2	19
7d	0.17	0.88
7e	~100	-
8a	0.097	1.2
8b (L-701,315)	0.0069	0.091
1a	0.32	7.0
1b	0.064	1.9
1c	0.014	0.41
2 (L-689,560)	0.0040	0.11
3	0.17	1.2
4 (L-687,414)	2.7	15

[a] Displacement of [^{3}H]L-689,560 (**2**) from rat brain membranes (see ref. 10). [b] Inhibition of NMDA-induced depolarizations in rat cortical slices (see ref. 11).

The methyl ester **7b** and phenyl ketone **7c** are equipotent with L-687,414 (**4**) and these 3-substituents markedly improve activity relative to the unsubstituted compound **7a**. The phenyl derivative **7d** unexpectedly provided the best activity within this group of 3-substituted compounds, being equipotent with 5,7-dichlorokynurenic acid (**1b**). The large difference in affinity between the 3-phenyl (**7d**) and 3-cyclohexyl (**7e**) derivatives (equivalent to a change in binding energy of >4 kcal.mol^{-1}) suggests that the 3-phenyl substituent may not recognise the receptor solely through an hydrophobic interaction. One explanation, which is consistent with the glycine antagonist pharmacophore,[2,3] is an electrostatic attraction between the 3-phenyl π system of **7d** and a receptor cationic group. This concept is supported by several observations in the literature suggesting the existence of energetically favourable interactions between phenyl ring π systems and cations, particularly quaternary ammonium groups. Such "π-cation" interactions have been predicted theoretically[13] and have been found experimentally in proteins[14] and in acetylcholine binding to both a synthetic receptor[15] and to acetylcholinesterase,[16] and in the binding of tetraethylammonium to potassium channels.[17]

The interaction of **7d** with the glycine recognition site would be expected to be additionally reinforced by ionization of the 4-hydroxyl, resulting in increased negative charge on the 2-carbonyl. Synthesis of the deoxy derivatives shows that both the 2- and 4-oxygen substitutents are essential for activity and we suggest that the 2-carbonyl acts in concert with the 3-phenyl as a mimic of the carboxylate present in glycine itself and in the antagonists **1** and **2**. The geometry of **7d** (determined by X-ray crystallographic analysis) would allow such an interaction, the angle between the phenyl and quinolone rings being 65°. In addition, it seems highly probable that the 4-hydroxyl anion in **7b-d** mimics the 4-carbonyls of **1** and **2** in receptor binding.[2,3] We therefore propose (Figure) that the α-phenyl lactam unit present in **7d** acts as a glycine bioisostere in receptor recognition.

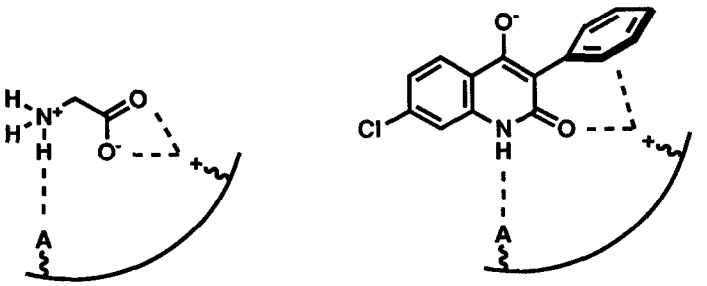


Figure. Hypothetical receptor sites binding glycine zwitterion and 3-phenyl-4-hydroxy-2-quinolone anion. **A** is a hydrogen bond acceptor and + is a receptor cation.

The glycine-site antagonist activity of the 5,7-dichloro derivative **8a** and certain 4'-phenyl substituted derivatives has been reported recently.[18] A dependence of affinity on 4'-substituent electron donation was found[18] and this is consistent with our model, since it may be explained by changes to the π-electron donating ability of the 3-phenyl group.

Extensive studies in this laboratory have shown that both the 3-acyl[19] (**7b** and **7c**) and 3-aryl[20,21] (**7d** and **8b**) series of 4-hydroxy-2-quinolones are particularly amenable to optimization. For example, by analogy with the kynurenic acid derivatives **1a-c**[2] (see Table) it may be predicted that introduction of small hydrophobic 5-substitutents should enhance activity. Although the 5-chloro derivative **8a** does not possess improved affinity relative to **7d**, the 5-ethyl analogue (**8b**, L-701,315, see Table) is equiactive with L-689,560 (**2**), the most potent glycine-site NMDA antagonist found to date.[3b]

Acknowledgements. We thank I. Sanderson for synthetic contributions, R. Herbert for NMR and mass spectra, A. Watt for HPLC and pKa measurements, S. Grimwood and G. Marshall for biological evaluation, and K. Hoogsteen for X-ray crystallography.

REFERENCES AND NOTES

1. Meldrum, B.S. (Ed.). *Excitatory Amino Acid Antagonists*, Blackwell, Oxford, 1991.
2. Leeson; P.D.; Baker, R.; Carling, R.W.; Curtis, NR.; Moore, K.W.; Williams, B.J.; Foster, A.C.; Donald, A.E.; Kemp J.A.; Marshall, G.R. *J. Med. Chem.*, **1991**, *34*, 1243.
3. (a) Carling, R.W.; Leeson, P.D.; Moseley, A.M.; Baker, R.; Foster, A.C.; Grimwood, S.; Kemp, J.A.; Marshall, G.R. *J. Med. Chem.*, **1992**, *35*, 1942. (b) Leeson, P.D.; Carling, R.W.; Moore, K.W.; Moseley, A.M.; Smith, J.D.; Stevenson, G.I.; Chan, T.; Baker, R.; Foster, A.C.; Grimwood, S.; Kemp, J.A.; Marshall, G.R.; Hoogsteen, K. *J. Med. Chem.*, **1992**, *35*, 1954.
4. Randle, J.C.R.; Guet, T.; Bobichon, C.; Moreau, C.; Curutchet, P.; Lambolez, B.; Prado de Carvalho, L.; Cordi, A.; Lepagnol, J.M. *Mol. Pharmacol.*, **1992**, *41*, 337.
5. Leeson, P.D.; Williams, B.J.; Baker, R.; Ladduwahetty, T.; Moore K.W.; Rowley, M. *J. Chem. Soc., Chem. Commun.*, **1990**, 1578.
6. Moore, K.W.; Leeson, P.D.; Carling, R.W.; Tricklebank, M.D.; Singh, L. *Bioorg. Med. Chem. Lett.*, in press.
7. (a) Carling, R.W.; Leeson, P.D.; Moseley, A.M.; Smith, J.D.; Saywell, K.; Tricklebank, M.D.; Kemp, J.A.; Marshall, G.R.; Foster, A.C.; Grimwood, S. *Bioorg. Med. Chem. Lett.*, in press. (b) Rowley, M.; Leeson, P.D.; Grimwood, S.; Foster, A.C., Saywell, K. *Bioorg. Med. Chem. Lett.*, in press.
8. Leeson, P.D.; Williams, B.J.; Rowley, M.; Moore, K.W.; Baker, R.; Kemp, J.A.; Priestley, T.; Foster, A.C.; Donald, A.E. *Bioorg. Med. Chem. Lett.*, in press.
9. Kappe, T.; Abdel, A.S.; Stadlebauer, W. *J. Heterocycl. Chem.*, **1988**, *25*, 857.
10. Grimwood, S.; Moseley, A.M.; Carling, R.W.; Leeson, P.D.; Foster, A.C. *Mol. Pharmacol.*, **1992**, *41*, 923.
11. Foster, A.C.; Kemp, J.A.; Leeson, P.D.; Grimwood, S.; Donald, A.E.; Marshall, G.R.; Priestley, T.; Smith, J.D.; Carling, R.W. *Mol. Pharmacol.*, **1992**, *41*, 914.
12. Nitronates are known to be effective bioisosteres of carboxylates in some enzyme substrates and inhibitors. See Alston, T.A.; Porter, D.J.T.; Bright, H.J. *Acc. Chem. Res.*, **1983**, *16*, 418.
13. Deakyne, C.A.; Meot-Ner (Mautner), M. *J. Amer. Chem. Soc.*, **1985**, *107*, 474.
14. Burley, S.K.; Petsko, G.A.; *FEBS Lett.*, **1986**, *203*, 139.
15. Dougherty, D.A.; Stauffer, D.A. *Science*, **1990**, *250*, 1558.
16. Sussman, J.L.; Harel, M.; Frolow, F.; Oefner, C.; Goldman, A.; Toker, L.; Silman, I. *Science*, **1991**, *253*, 872.
17. Heginbotham, L.; MacKinnon R. *Neuron*, **1992**, *8*, 483.
18. McQuaid, L.A.; Smith, C.R.; Lodge, D.; Pralong, E.; Wikel, J.H.; Calligaro, D.O.; O'Malley, P.J. *J. Med. Chem.* **1992**, *35*, 3423.
19. Baker, R.; Stevenson, G.I.; Leeson, P.D.; Rowley, M. EP 0 459 561 A2 (4 Dec. 1991).
20. Kulagowski, J.J.; Rowley, M.; Leeson, P.D.; Mawer, I.M. EP 0 481 676 A1 (22 Apr. 1992).
21. Carling, W.R.; Leeson, P.D.; Moore, K.W. EP 0 489 458 A (10 June 1992).