Bioorganic & Medicinal Chemistry Letters 20 (2010) 461–464

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry Letters

journal homepage: www.elsevier.com/locate/bmcl

Synthesis and in vivo evaluation of bicyclic gababutins

David C. Blakemore *, Justin S. Bryans, Pauline Carnell, Christopher L. Carr, Nicola E. A. Chessum, Mark J. Field, Natasha Kinsella, Simon A. Osborne, Andrew N. Warren, Sophie C. Williams

Sandwich Laboratories, Pfizer Global Research and Development, Ramsgate Road, Sandwich, Kent CT13 9NJ, UK

ARTICLE INFO

Article history:
Received 15 November 2009
Revised 22 November 2009
Accepted 24 November 2009
Available online 27 November 2009

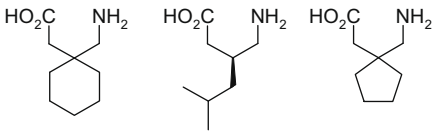
Keywords:
Gabapentin
Gababutin
Pregabalin
Pain

ABSTRACT

Synthesis of a number of bicyclic five-membered ring derivatives of gabapentin led to the identification of two compounds, (−)-(**11A**) and (**20A**) which both had an excellent level of potency against $\alpha_2\delta$ and were profiled in an in vivo model of neuropathic pain.

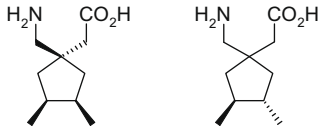
Gabapentin (Neurontin®) (**1**)[1] and pregabalin (Lyrica®) (**2**)[2,3] have been shown preclinically to have utility against neuropathic pain and anxiety; their efficacy against neuropathic pain has also been demonstrated clinically in humans.[4] Pregabalin has been approved for the management of neuropathic pain associated with diabetic peripheral neuropathy and post-herpetic neuralgia and achieved world-wide sales of over one billion dollars in 2007. Gabapentin and pregabalin are thought to mediate their pharmacological actions through binding to the $\alpha_2\delta$ subunit of a voltage gated calcium channel.[5,6]

We, and others, have disclosed SAR investigations around a range of $\alpha_2\delta$ ligands.[7] Recently, our attention has focused on the five-membered ring analogue (**3**) which we have called gababutin.[8,9]



(1) Gabapentin (2) Pregabalin (3) Gababutin

Gababutin has a binding affinity of 420 nM at the $\alpha_2\delta$ binding site compared to 140 nM for gabapentin. This observation suggested to us that the gababutin five-membered ring is not optimally filling the available space in the binding pocket. We have recently reported our investigations into appending alkyl substituents onto the 3-position which demonstrated that methyl, ethyl and *n*-propyl groups enhanced binding affinity, when the stereocentre at the 3-position had the (*R*)-configuration. 3,4-Disubstitution with methyl groups also led to enhanced binding affinity but is again highly dependent on the stereochemistry of the 3-position. Compounds (**4**) and (**5**) have greater binding affinities than gabapentin but are indistinguishable from it in the CCI (chronic constriction injury) weight-bearing model of neuropathic pain.



(4) IC_{50} = 107 nM **(5)** IC_{50} = 22nM

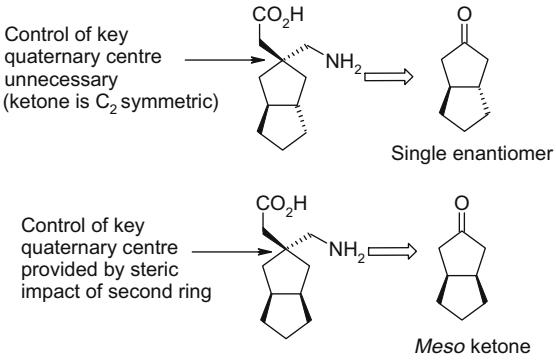
We now report the effect on binding and efficacy of building conformational constraint into these molecules. We have synthesised a number of bicyclic analogues of (**4**) where the ring junction is *cis* and analogues of (**5**) where a *trans* ring junction is present.

With bicyclic gababutin analogues of (**4**) and (**5**), if the ring junction is *trans*, then the stereochemistry of the quaternary centre carrying the aminomethyl group is irrelevant as this centre is *pseudo*-asymmetric; the key problem is synthesis of the C_2 symmetric chiral ketone precursor. If the ring junction is *cis*, then the starting ketone is achiral, or *meso* (as is the final amino acid), but introduction of the stereogenic quaternary centre carrying the aminomethyl group must be controlled (by making use of the differing

* Corresponding author. Tel.: +44 1304 644573.
E-mail address: david.blakemore@pfizer.com (D.C. Blakemore).

0960-894X/$ - see front matter © 2009 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2009.11.118

levels of steric hindrance on attack of the *exo* and *endo* faces of the bicyclic system).

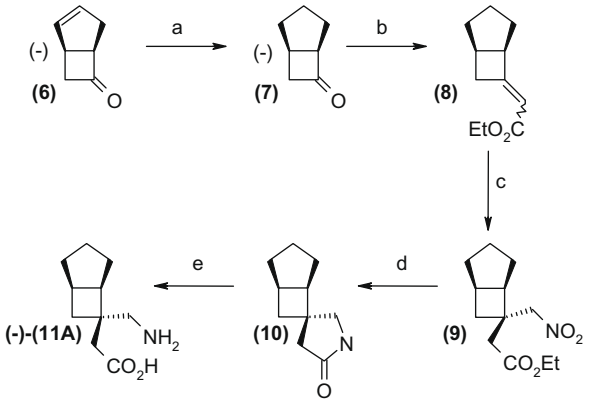


Bicyclic analogues without a symmetry element required both an enantiopure ketone precursor and control of the stereogenic quaternary centre. This is illustrated in the synthesis of (−)-(**11A**) from the known chiral ketone (**7**) (Scheme 1).

Enantiopure bicyclo[3.2.0]hept-2-en-6-one[10] (**6**) was hydrogenated to give ketone (**7**) which was transformed in good yield to the α,β-unsaturated ester (**8**) via a Horner–Emmons reaction. Conjugate addition of the anion of nitromethane to the unsaturated ester (**8**) resulted in the formation of the nitroester (**9**). The nitromethane anion attacks the bicyclic system exclusively from the less hindered *exo* face (as confirmed by NOE analysis) to give (**9**) as a single diastereoisomer. Reduction of (**9**) by hydrogenation in the presence of a nickel catalyst gave lactam (**10**) as a result of the in situ cyclisation of the intermediate amino ester generated. Hydrolysis of lactam (**10**) with 6 N HCl produced the target amino acid (−)-(**11A**) as a hydrochloride salt (chiral HPLC showed the enantiomeric excess to be 98%).

The diastereoisomeric bicyclic *cis* analogues (**20A**) and (**20B**) both possess a plane of symmetry and so are achiral compounds. Analogue (**20B**), where the aminomethyl group is on the more hindered *endo* face of the bicyclic system, can be formed via an intermediate cyanide and this is illustrated in Scheme 2.

Reduction of the commercially available cyclobutane-1,2-dicarboxylic acid (**12**) gave a diol which was converted to the bis-mesylate (**13**). Displacement of the mesyl groups with lithium bromide in acetone occurred in high yield to give dibromide (**14**). Ring closure of dibromide (**14**) with ethyl cyanoacetate gave cyanoester (**15**) as a mixture of diastereoisomers. Krapcho reaction of the cyanoester with lithium chloride, water and dimethylsulfoxide yielded cyanide (**16**) via hydrolysis and decarboxylation. The anion of cyanide (**16**) was generated by deprotonation with lithium hexamethyldisilazide and this was quenched at low temperature with dimethylallyl bromide to give cyanoalkene (**17**). Attack occurred on the less hindered *exo* side of the bicyclic system (as confirmed by NOE studies) to give (**17**) as a single diastereoisomer (99% diastereoisomeric excess as confirmed by HPLC). Ozonolysis of alkene (**17**) in the presence of sodium hydroxide and methanol allowed the generation of cyanoester (**18**). Reduction of the cyanide group of (**18**) yielded the lactam (**19**) which was hydrolysed to the amino acid (**20B**) with hydrochloric acid.

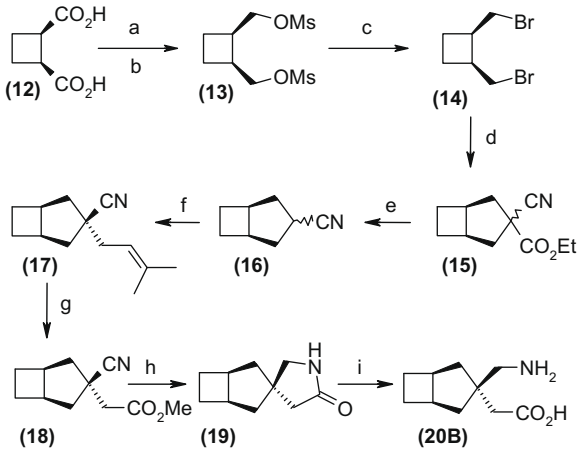


Scheme 1. Reagents and conditions: (a) H_2, Pd/C, EtOAc (b) $(EtO)_2P(O)CH_2CO_2Et$, NaH, THF (72% over 2 steps); (c) $MeNO_2$, $Bu_4N^+F^-$, THF, 70 °C (87%); (d) Ni sponge, H_2, MeOH (54%); (e) 6 N HCl (72%).

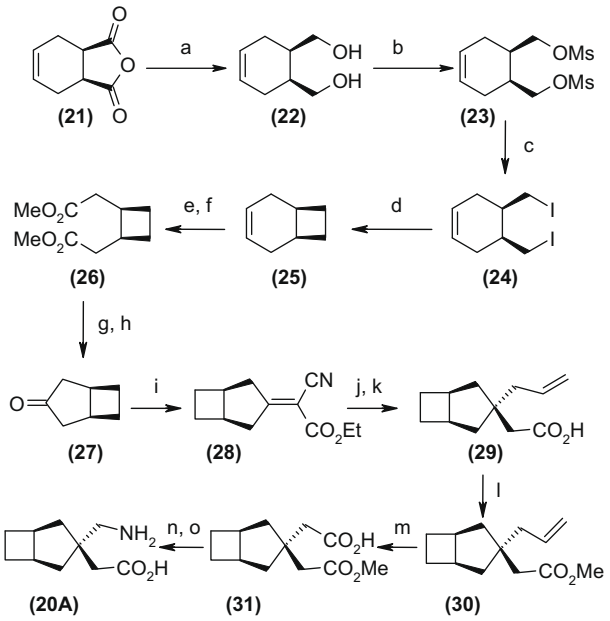
Analogue (**20A**), where the aminomethyl group is on the less hindered *exo* face of the bicyclic system, can be formed via an intermediate ketone and this is illustrated in Scheme 3. Commercially available anhydride (**21**) was reduced to *cis*-diol (**22**). Following its conversion to di-iodide (**24**), ring-closure to generate the *cis*-cyclobutyl ring was carried out using *tert*-butyllithium (or, alternatively, with *n*-butyllithium). Oxidative cleavage of the cyclohexene ring gave a diacid which was converted to diester (**26**). Dieckmann cyclisation followed by decarboxylation gave the desired *meso* ketone (**27**). This could be converted to amino acid (**20A**) using the nitromethane addition method used in Scheme 1 (although the nitromethane addition gave 10–15% of the undesired diastereoisomeric product) or via the allyl addition route shown below. Knoevenagel condensation on ketone (**27**) gave unsaturated cyanoester (**28**). Allylmagnesium bromide added to the unsaturated cyanoester in a conjugate manner with addition occurring exclusively from the less hindered *exo* face of the molecule (a diastereoisomeric excess of 99% was determined by HPLC). Hydrolysis of the addition product gave acid (**29**) as a single diastereoisomer. Following conversion of acid (**29**) to ester (**30**), oxidative cleavage of the allyl group gave acid (**31**) which was Curtius rearranged and hydrolysed to give desired amino acid (**20A**) as a single diastereoisomer (NOE studies confirmed that the aminomethyl group was on the *exo* face of the bicyclic system).

In the case of the *trans*-bicyclic analogues, the target molecules were made as racemates from the precursor ketone using the nitromethane addition route.

It has been a recurring theme with the gababutins that small changes in structure can dramatically change binding affinity at the receptor and that the receptor is very sensitive to chirality. Binding affinity data for the *trans*-bicyclic gababutins are shown in Table 1 (a radioligand binding assay incorporating [^{3}H]gabapentin at the $\alpha_2\delta$ subunit of a calcium channel was utilised as previously described[5]).



Scheme 2. Reagents and conditions: (a) $LiAlH_4$, Et_2O, 0 °C to rt (98%); (b) MsCl, NEt_3, DCM, −40 °C to rt (73%); (c) LiBr, acetone, reflux (91%); (d) $NCCH_2CO_2Et$, K_2CO_3, DMF (99%); (e) LiCl, H_2O, DMSO, 150 °C (44%); (f) LHMDS, THF, −40 °C; dimethylallylbromide, −78 °C (72%); (g) O_3, NaOH, MeOH, DCM (71%); (h) H_2, Ni, MeOH; (i) 6 N HCl, 1,4-dioxane (80% from cyanoester).



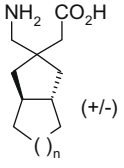
Scheme 3. Reagents and conditions: (a) $LiAlH_4$, THF, reflux (80%); (b) MsCl, NEt_3, DCM, −40 °C to rt (80%); (c) NaI, acetone, reflux (70%); (d) *t*-BuLi, pentane–ether (3:2), −25 °C; (e) $NaIO_4$, $RuCl_3{\cdot}H_2O$, MeCN, EtOAc, H_2O; (f) MeOH, concd H_2SO_4 (85% from di-iodide); (g) KO*t*-Bu, THF, reflux (95%); (h) DMSO, H_2O, 155 °C (97%); (i) ethyl cyanoacetate, NH_4OAc, AcOH, toluene, reflux (78%); (j) allylmagnesium bromide, dimethylzinc, THF, −78 °C; (k) KOH, ethylene glycol, 165 °C (89% over the two steps); (l) $TMSCHN_2$, toluene (97% from cyanoester); (m) $RuCl_3$, $NaIO_4$, CCl_4, MeCN, H_2O (83%); (n) DPPA, NEt_3, toluene, reflux; (o) 6 N HCl, reflux (72% over the two steps).

The effect of constraint is to dramatically reduce binding affinity at $\alpha_2\delta$ as compared to the *trans*-dimethyl gababutin (**5**). While this can be rationalised for compounds (**33**) and (**34**) where the extra methylene groups might make the molecules too large to fit in the receptor it is surprising that the direct bicyclic analogue of (**5**), compound (**32**), has poor binding affinity.

The tight SAR around these bicyclic gababutins is further illustrated with the *cis* compounds (Table 2). All the compounds with the aminomethyl group on the more hindered *endo* face of the molecule have poor binding affinity. Of the compounds where the aminomethyl is on the *exo* face of the molecule, only compound (**20A**) has high affinity for $\alpha_2\delta$.

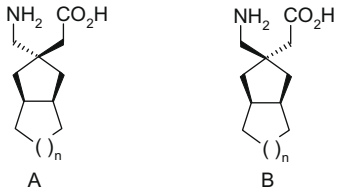
Due to the limited scope for change in the size of the bicyclic skeleton, we hypothesised that (**20A**) fitted the binding pocket optimally. Keeping the bicyclic skeleton the same, we synthesised regioisomers of (**20A**) with the amino acid portion of the molecule directly attached to the four-membered ring; two diastereoisomers (**11A**) and (**11B**) are possible, and we were pleased to find that

Table 1



n	Compound	IC_{50} (nM)
0	**(32)**	2900
1	**(33)**	>10,000
2	**(34)**	>10,000

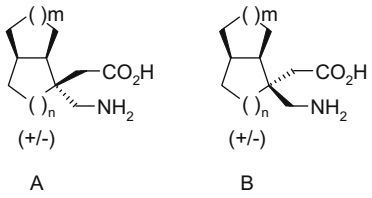
Table 2



n	Compound	IC_{50} (nM)
0	(**20A**)	38
0	(**20B**)	332
1	(**35A**)	2600
1	(**35B**)	>10,000
2	(**36A**)	720
2	(**36B**)	>10,000

(**11A**) has good binding affinity at the receptor. Again, increasing the size of either ring of the bicyclic system reduced binding affinity (Table 3) reinforcing our belief that the bicyclo[3.2.0]heptyl system was optimally sized.

Table 3



m	*n*	Compound	IC_{50} (nM)
1	0	(**11A**)	25
1	0	(**11B**)	2800
1	1	(**37A**)	7181
2	0	(**38A**)	600
2	0	(**38B**)	3088

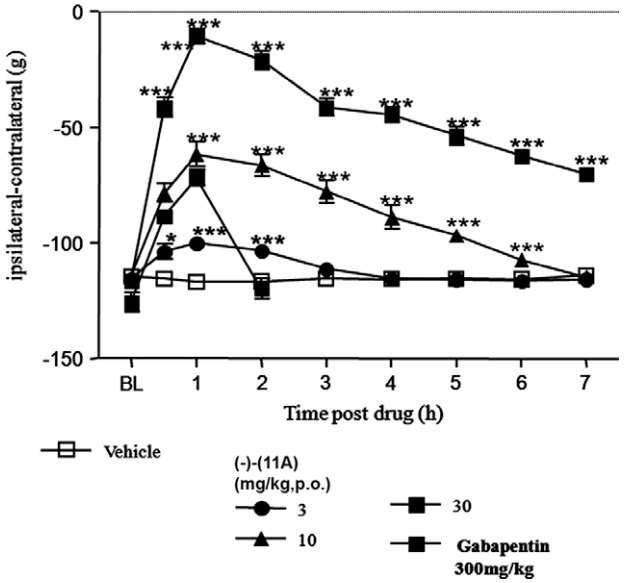


Figure 1. (* = P <0.05, *** = P <0.01, significantly different compared to vehicle.

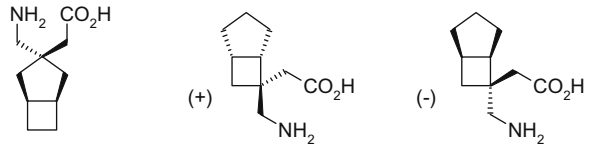
Table 4

Compound	System L binding IC_{50}* (μM)
(**4**)	7
(**5**)	10
(**20A**)	38
(−)-(**11A**)	>10,000
(+)-(**11A**)	6931

* Prevention of uptake of ^{3}H-Leucine into CHO cells.[11]

Table 5

Compound	F (%)	Elimination half-life (h)	Clearance (mL/min/kg)
(−)-(**11A**)	100	2.3	4.5
(**20A**)	84	2.4	4.3



(20A) IC_{50} = 38 nM

(+)-(11A) IC_{50} = 19 nM

(-)-(11A) IC_{50} = 46 nM

Both enantiomers of (**11A**) have good binding affinities at $\alpha_2\delta$ and these two compounds and (**20A**) were dosed orally in an in vivo model of neuropathic pain, the CCI (chronic constriction injury) weight-bearing model. In this model, loose ligatures are placed around the sciatic nerve of a rat causing hypersensitivity in one hind paw. The ability of the rat to place weight on each hind paw is measured and the differential between ipsilateral (affected) and contralateral (unaffected) paw is calculated.

All potent gababutins (IC_{50} values of less than 150 nM at $\alpha_2\delta$) tested in this model to date have achieved similar efficacy to gabapentin. Both (**20A**) and (−)-(**11A**) proved to be more efficacious than gabapentin in this model, with (−)-(**11A**) having the most effective profile yet seen in this model giving a complete blockade of the pain effect (Fig. 1).

It is noteworthy that (+)-(**11A**) was completely inactive in this model unless dosed *intrathecally*. It would, therefore, appear that the compound is not penetrating the blood–brain barrier. Gabapentin itself is a substrate for the Large amino acid transporter (System-L amino acid transporter) and it has been assumed that it is this transporter that conveys the compounds into the CNS.[7] Interestingly, while (**20A**) binds to the transporter, neither (+)-(**11A**) or (−)-(**11A**) appear to have any affinity for it (Table 4). It is, however, possible that (−)-(**11A**) is entering the CNS via another mechanism.

(−)-(**11A**) and (**20A**) have very similar rat pharmacokinetic profiles with high bioavailabilities and a short half-lives (Table 5). As expected for such polar molecules ($\log D_{7.4} = -1.0$ in both cases), the compounds are not metabolised but are renally cleared at glomerular filtration rate.

Compounds (−)-(**11A**) and (**20A**) have both been progressed for further studies.

Acknowledgements

Ti-Zhi Su is thanked for the System L binding data. Alison Betts is thanked for the pharmacokinetic data. We thank the staff of the Structural and Analytical Services Department, Sandwich, for analytical data.

References and notes

1. Bryans, J. S.; Wustrow, D. J. *Med. Res. Rev.* **1999**, *19*, 149.
2. Dworkin, R. H.; Kirkpatrick, P. *Nat. Rev. Drug Disc.* **2005**, *4*, 455.
3. Zareba, G. *Drugs Today* **2005**, *41*, 509.
4. Rice, A. S. C.; Maton, S. *Pain* **2001**, *94*, 215.
5. Gee, N. S.; Brown, J. P.; Dissanayake, V. U. K.; Offord, J.; Thurlow, R.; Woodruff, G. N. *J. Biol. Chem.* **1996**, *271*, 5768.
6. Stahl, S. M. *J. Clin. Psychiatry* **2004**, *65*, 1033. and references cited therein.
7. Field, M. J.; Li, Z.; Schwarz, J. B. *J. Med. Chem.* **2007**, *50*, 2569. and references cited therein.
8. Blakemore, D. C.; Bryans, J. S.; Carnell, P.; Chessum, N. E. A.; Field, M. J.; Kinsella, N.; Kinsora, J. K.; Osborne, S. A.; Williams, S. C.; *Bioorg. Med. Chem. Lett.*, **2009**. doi:10.1016/j.bmcl.2009.10.089.
9. Blakemore, D. C.; Bryans, J. S.; Carnell, P.; Field, M. J.; Kinsella, N.; Kinsora, J. K.; Meltzer, L. T.; Osborne, S. A.; Thompson, L. R.; Williams, S. C.; *Bioorg. Med. Chem. Lett.*, **2009**. doi:10.1016/j.bmcl.2009.10.121.
10. Klempier, N.; Geymayer, P.; Stadler, P.; Faber, K.; Griengl, H. *Tetrahedron: Asymmetry* **1990**, *1*, 111.
11. Belliotti, T. R.; Caparis, T.; Ekhato, I. V.; Kinsora, J. J.; Field, M. J.; Heffner, T. J.; Meltzer, L. T.; Schwarz, J. B.; Taylor, C. P.; Thorpe, A. J.; Vartanian, M. G.; Wise, L. D.; Su, T.-Z.; Weber, M. L.; Wustrow, D. J. *J. Med. Chem.* **2005**, *48*, 2294.