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Synthesis of Pyrrolidine Oxazoles as Thromboxane A_2/Endoperoxide Receptor Antagonists

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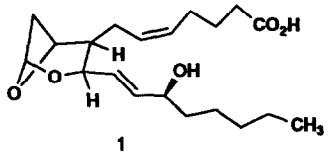
Abstract: The synthesis and antiplatelet activity of a series of bis-heterocyclic thromboxane A_2 receptor antagonists is described. The L-proline analog **9** was the most potent ligand in this class with a K_D = 7.9±0.71 nM in washed human platelets.

Thromboxane A_2 (**1**, TxA_2), an unstable metabolite of arachidonic acid, is a potent stimulator of platelet aggregation and elicits contraction of smooth muscle.[1] A diverse group of structures have been identified which display TxA_2/endoperoxide antagonistic activity in platelets, bronchial and vascular smooth muscle.[2,3] Recent reports from these laboratories have described potent TxA_2 antagonists related to BMS 180,291(**2**) which display an outstanding duration of action *in vivo*.[4,5] In conjunction with these studies, we have examined BMS 180,291 analogs in which the 7-oxabicycloheptane moiety has been replaced with a pyrrolidine or substituted pyrrolidine ring. Some of these targets were particularly attractive since optically active heterocycles could be derived from readily available amino acids such as proline. Disclosed here in preliminary form are the synthesis and the antiplatelet activity of these pyrrolidine oxazoles.



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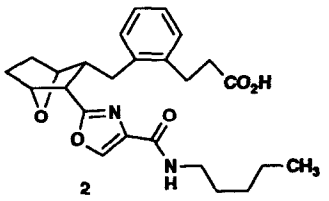

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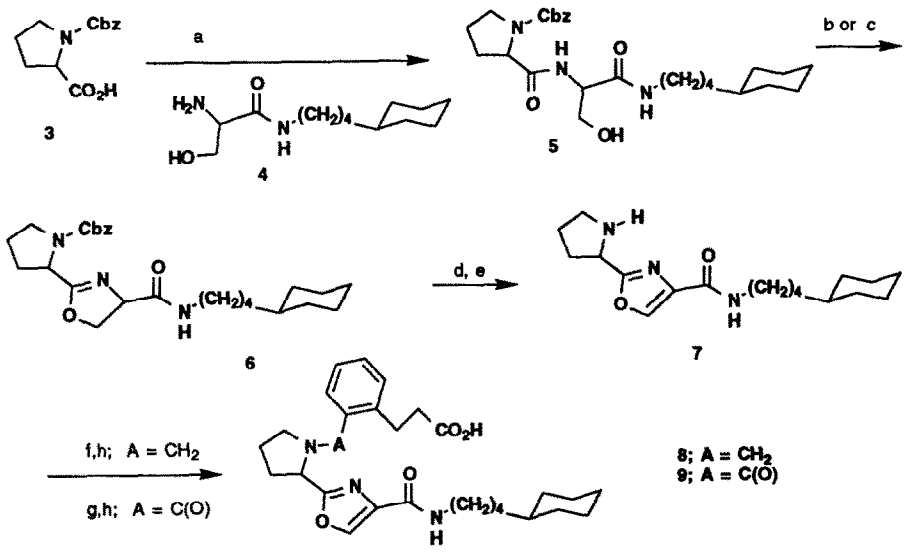
Synthesis

The targets of this investigation were prepared as shown in Schemes I - III. As illustrated in Scheme I, standard peptide coupling of proline **3** and serine amide **4**[6] afforded dipeptide **5**. Cyclization and subsequent oxidation of oxazoline **6** was accomplished using the methodology described previously for BMS 180,291.[4] The choice of cyclization conditions were dependent on the substrate; the mesylation/K_2CO_3 method was only used with racemic amino acids due to concern about epimerization. Although we anticipated that the Ph_3P conditions would not lead to racemization of the amino acid, this possibility was addressed directly at the stage of the oxazole-amine **7**. Oxidation of oxazoline **6** followed by hydrogenolysis provided oxazole-amine **7**. The stereochemical purity of this intermediate was assessed by HPLC on a Chiralcel OD column. The enantiomeric excess of intermediates **7**, derived from D- and L-proline, were 97.5% and 100% respectively[7]. Elaboration of **7** proceeded by either alkylation or acylation to give the target compounds **8**[8] and **9**.

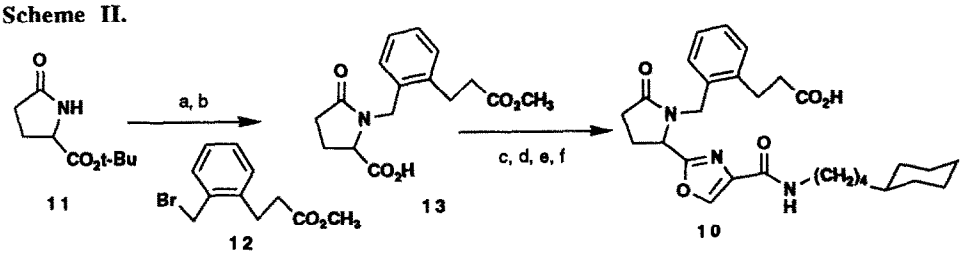
The related analog **10**, derived from pyroglutamic acid, was prepared using a different strategy in which the acid side chain was attached via alkylation of pyroglutamic acid-t-butyl ester. Condensation of the sodium salt of **11** with benzyl bromide **12** provided lactam **13** in 48% yield. Homologation of the oxazole side-chain followed the route outlined in Scheme I to provide oxazole **10**.

Scheme I: Preparation of Proline-Based TxA_2 Antagonists



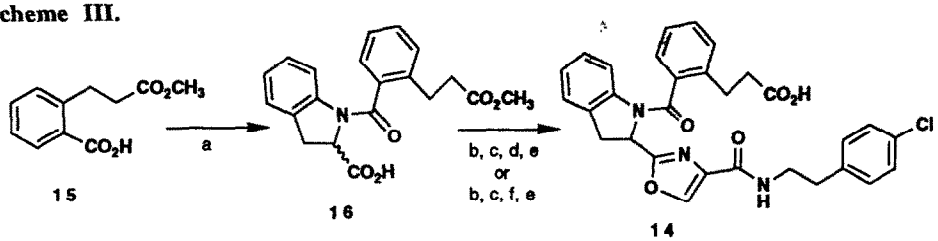
(a) RNH_2, Et_3N, HOBt, ethyl-3-(3-dimethylamino)propylcarbodiimide • HCl (WSC) (84%); (b) PhP_3, CCl_4, CH_3CN, iPr_2EtN, 23°C (50%); (c) MsCl, Et_3N, 0°C; then K_2CO_3, acetone, reflux (96%); (d) $CuBr_2$, DBU, $CHCl_3$, EtOAc, 23°C (21-68%); (e) 10% Pd/C, H_2, CH_3OH, 23°C (99%); (f) $NaCNBH_3$, HOAc, 2-(CHO)-$PhCH_2CH_2CO_2CH_3$, CH_3OH, 23°C (77-87%); (g) 2-(CO_2H)-$PhCH_2CH_2CO_2CH_3$, oxalyl chloride, Et_3N; then 7 (36%); (h) LiOH, H_2O, THF (69-87%)

Scheme II.



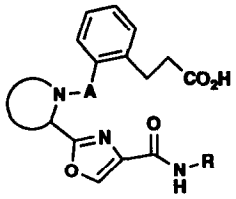
(a) NaH, THF, RBr, -78°-23°C (48%); (b) 4N HCl/dioxane, 0-23°C (67%); (c) RNH_2, Et_3N, HOBt, ethyl-3-(3-dimethylamino)-propylcarbodiimide • HCl (WSC), (52%); (d) MsCl, Et_3N, 0°C; then K_2CO_3, acetone, reflux (98%); (e) $CuBr_2$, DBU, $CHCl_3$, EtOAc, 23°C (41%); (f) LiOH, H_2O, THF (76%)

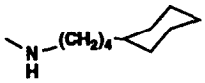
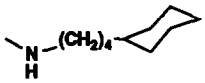
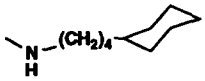
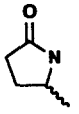
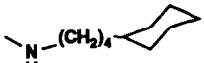
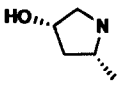
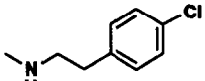
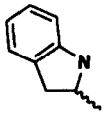
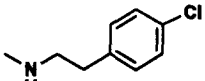
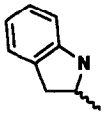
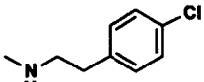
Scheme III.



(a) oxalyl chloride, CH_2Cl_2, DMF, 23°C; then indoline-2-carboxylic acid (60%); (b) RNH_2, Et_3N, HOBt, ethyl-3-(3-dimethylamino)-propylcarbodiimide • HCl (WSC), (72%); (c) MsCl, Et_3N, 0°C; then K_2CO_3, acetone, reflux (66, 93%); (d) $CuBr_2$, DBU, $CHCl_3$, EtOAc, 23°C (45%); (e) LiOH, H_2O, THF (32, 73%); (f) NiO_2, CH_2Cl_2, 23°C (38%).

Table 1.



Compound	Heterocycle	A	HNR	AAIPA I_{50} (μM)
(-)**8**		CH_2		0.019
(+)**8**		CH_2		1.7
(-)**9**		C(O)		0.019
10		CH_2		23
17		C(O)		3.7
(-)**14**		C(O)		3.7
(+)**14**		C(O)		0.25

The indoline analog **14** was synthesized as outlined in Scheme III. Condensation of excess acid chloride derived from **15** with indoline-2-carboxylic acid provided amide **16** in 60% yield. Elaboration of the oxazole side-chain used the previously described methods. Of interest is the observation that condensation of (±)**16** with L-serine amide **4** provided diastereoisomers which could be separated readily by flash chromatography. The individual amides were then carried on to yield (+)**14** and (-)**14**. Amide **17** was prepared by a route analogous to that used for **9** starting with 4-*cis*-t-butyldimethylsilyloxyproline.

Pharmacology

All compounds were tested for their ability to inhibit arachidonic acid-induced (800 μM) and ADP-induced (20 μM) platelet aggregation of human platelet rich plasma and the results are shown in Table 1[9]. Consistent with these compounds as selective TxA_2 receptor antagonists, none were effective in inhibiting ADP-induced platelet

aggregation. Both antagonists derived from proline, (+)**8** and (-)**8**, inhibited AA-induced aggregation; however this inhibition was enantioselective as (-)**8** was 100-fold more active than (+)**8**. The basic nitrogen present in (-)**8** was not necessary for potent TxA_2 antagonism as the acylated derivative **9** possessed identical activity *in vitro*. In contrast to the equivalent potency of (-)**8** and **9**, the related lactam **10** was 1,000-fold less active. The hydroxylated analog **17** was also weakly active. The poor activity of **10** and **17** can not be explained by increased steric bulk in this region of the molecule since indoline (+)**14** was only 10-fold less active than (-)**8**. An alternative explanation is that the polar functionality present in **10** and **17** binds to an apolar site on the receptor, thereby destabilizing the binding interaction. Although somewhat less potent than the corresponding oxabicycloheptane analog (SQ 33,961; AAIPA I_{50} = 2 nM)[5], pyrrolidines **8** and **9** were more active than a well-characterized antagonist BM 13.505[10] (AAIPA I_{50} = 730 nM).

The most potent analogs were studied for their ability to displace [^{3}H]-SQ 29,548 from its specific binding sites in human platelet membranes[11]. Oxazoles **8** and **9** displayed binding affinities at the TxA_2 receptor consistent with their potent antiplatelet activity; K_d = 11 ± 0.35 nM (slope = 1.9 ± 0.17) and 7.9 ± 0.71 nM (slope = 1.0 ± 0.11), respectively. The binding interaction appears to be of the competitive nature for **9** but given the steep slope, non-competitive for pyrrolidine **8**.

We have demonstrated that the 7-oxabicycloheptane nucleus of potent TxA_2 antagonists such as BMS 180,291 can be replaced with a pyrrolidine while maintaining potent TxA_2 antagonistic activity *in vitro*.

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Notes and References

1. Hamberg, M.; Svensson, J.; Samuelsson, B. *Proc. Natl. Acad. Sci. U.S.A.* **1975**, *72*, 2994-2998.
2. (a) Halushka, P. V.; Mais, D. E. *Drugs of Today* **1989**, *25* (6), 383-393. (b) Smith, E. F. III *Eicosanoids* **1989**, *2* (4), 199-212. (c) Ogletree, M. L. *Federation Proc.* **1987**, *46* (1), 133-138.
3. For a comprehensive review of TxA_2 antagonists see: Hall, S. E. *Med. Res. Rev.* **1991**, *11* (5), 503-579.
4. Misra, R.N.; Brown, B. R.; Sher, P. M.; Patel, M. M.; Hall, S. E.; Han, W.-C.; Barrish, J. C.; Floyd, D. M.; Sprague, P. W.; Morrison, R. E.; Ridgewell, R. E.; White, R. E.; DiDonato, G. C.; Harris, D. N.; Hedberg, A.; Schumacher, W. A.; Webb, M. L.; Ogletree, M. L. *BioMed. Chem. Lett.* **1992**, *2* (1), 73-76.
5. Misra, R.N.; Brown, B. R.; Sher. P. M.; Patel, M. M.; Goldenberg, H. J.; Michel, I. M.; Harris, D. N. *BioMed. Chem. Lett.* **1991**, *1* (9), 461-464.
6. Serine amide **7** (white solid, mp 77-78°) was prepared in 2 steps from CBZ-L-serine by coupling with amylamine (WSC/HOBT/DMF, 0 to 25°, 69%) followed by CBZ-deprotection (20% $Pd(OH)_2$-C/CH_3OH/H_2 (1 atm), 85%).
7. The enantiomeric purity of the starting D-proline was not checked; therefore it is not clear whether there was a small degree of racemization during the synthesis of (+)**8**. Conditions for the assay of **7**: 10μ Chiralcel OD, 0.46 x 25 cm, 93% hexane/7%(98:2 IPA:n-PrOH).
8. Purification of this compound was most easily effected by conversion to the lithium salt followed by chromatography on HP-20. This resulted in the compound being isolated as a stable mono-hydrate of the lithium salt; m.p. = 92-94°C.
9. (a) Assay as described by Harris, D. N.; Phillips, M. B.; Michel, I. M.; Goldenberg, H. J.; Heikes, J. E.; Sprague, P. W.; Antonaccio, M. J. *Prostaglandins* **1981**, *22* (2), 295-307; the I_{50} for BM13.505 and GR 32,191 were 730 nM and 33 nM, respectively, under identical assay conditions.
(b) Ogletree, M. L.; Harris, D. N.; Schumacher, W. A.; Hall. S. E.; Brown, B. R.; Misra, R. N. *Circulation*, **1991**, *84*, II-79.
10. Lefer, A. M. *Drugs Future* **1988**, *13*, 999-1005.
11. Assay as described by Hedberg, A.; Hall, S. E.; Ogletree, M. L.; Harris, D. N.; Liu, E. C.-K. *J. Pharmacol. Exp. Ther.* **1988**, *245* (3), 786-792.