Bioorganic & Medicinal Chemistry Letters, Vol.1, No.6, pp. 327-332, 1991
Printed in Great Britain

0960-894X/91 $3.00 + .00
© 1991 Pergamon Press plc

SYNTHESIS AND BIOLOGICAL ACTIVITY OF MK 287 (L-680,573): A POTENT, SPECIFIC AND ORALLY ACTIVE PAF RECEPTOR ANTAGONIST

Soumya P. Sahoo*, Donald W. Graham, John Acton, Tesfaye Biftu, Robert L. Bugianesi, Narindar N. Girotra, Chan-Hwa Kuo, Mitree M. Ponpipom, Thomas W. Doebber, Margaret S. Wu, San-Bao Hwang, My-Hanh Lam, D. Euan MacIntyre, Thomas J. Bach, Silvi Luell, Roger Meurer, Philip Davies, Alfred W. Alberts and John C. Chabala

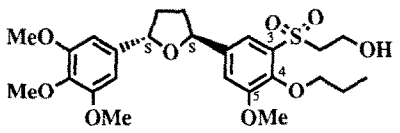
Merck Sharp & Dohme Research Laboratories, P.O.Box 2000, Rahway, NJ 07065

(*Received* 30 *May* 1991)

Abstract: An enantioselective synthesis of MK 287 (L-680,573), a member of a family of *trans*-2,5-diaryltetrahydrofurans, and its biological activity are described.

Platelet-activating factor (PAF)[1], a family of endogenous phospholipids chemically identified as 1-*O*-alkyl-2-*O*-acetyl-*sn*-glycero-3-phosphocholines, is considered to play a major role in pathophysiological conditions in several human diseases[2]. PAF is synthesized and secreted by a wide variety of cells involved in inflammatory responses such as basophils, eosinophils, neutrophils, macrophages, endothelial cells and IgE-sensitized bone marrow mast cells[2,3] and it induces a wide range of biological and pharmacological responses[4] including smooth muscle contraction, bronchoconstriction, neutrophil and platelet aggregation, eosinophil chemotaxis, hypotension and acute renal failure. PAF has also been postulated to play a major role in the pathogenesis of asthma, particularly in the late phase responses[5,6].

There is strong evidence to suggest that PAF elicits its biological functions both *in vitro* and *in vivo* by binding to specific receptors on target cells. A number of PAF antagonists encompassing a wide variety of structural types have been reported[7] including PAF based structural analogs[8], complex natural products such as kadsurenone[9] and other lignans, ginkgolides[10], as well as totally synthetic entities like WEB-2086[11] and L-652,731[12]. In this communication, we report the synthesis and biological activity of MK 287 (L-680,573)[13], a highly potent, specific, orally active PAF receptor antagonist which is undergoing clinical trials.



MK 287 (L-680,573)

MK 287 (L-680,573) is a member of a family of *trans*-2,5-diaryl tetrahydrofurans which exhibit PAF receptor binding antagonism. This compound evolved from a screening program designed to discover novel natural products with PAF antagonist activity. Using the inhibition of PAF binding to rabbit platelet membrane preparation as a screening assay, two such compounds were discovered: kadsurenone[9] (IC_{50} = 150 nM) and veraguensin (IC_{50} = 1000 nM). Initial structural optimization of veraguensin provided the simplified but more potent PAF antagonist L-652,731 (K_i = 103 nM)[12].

The weak *in vivo* activity of L-652,731[14] coupled with its relatively short half-life in rhesus monkeys ($t_{1/2}$ = 41 min, *i.v.*) due to rapid metabolism precluded its clinical development. Further structural modification of L-652,731 revealed that incorporation of an electron-withdrawing substituent into the 3-position of one of the aryl rings and replacement of 4-methoxy group of L-652,731with other longer alkyl ethers in the same aryl ring led to a marked potency enhancement with concomitant improvement of metabolic profile as exemplified by L-659,989[15] (human platelet membranes, K_i = 9 nM). In order to find a compound with further improved metabolic stability as well as a better pharmacokinetic profile than L-659,989[14], a number of polar group modifications were investigated, from which MK 287, which contains a 5-hydroxyethylsulfone group was identified. The detailed structure - activity relationship leading up to the discovery of MK 287 will be the subject of future publications.

CHEMISTRY

The synthetic strategy described in this communication for the synthesis of MK 287 is a modified version of the strategy developed and published from these laboratories for the preparation of L-659,989[16] and related compounds. Access to most of the *trans*-2(S),5(S)-diaryl tetrahydrofurans prepared by this general route involved regioselective reduction of a diketone to the corresponding hydroxy-ketone, followed by resolution after esterification with (-)-(R)-*O*-methylmandelic acid and subsequent reduction to the diol which was shown to cyclize without racemization with 5% TFA in $CHCl_3$. Early during this investigation, the synthesis of MK 287 and its enantiomer, L-680,574, was accomplished, using the above mentioned synthetic sequence with minor modification. Moreover, in keeping with our earlier observations with L-652,731[14] and L-659,989[16], the (-)-isomer MK 287 was found to be the active enantiomer responsible for most of the PAF receptor binding inhibitory activity.

Since one can regioselectively reduce the keto group adjacent to the aryl ring having electron withdrawing substituents at position 3, it seemed reasonable that one might also be able to reduce it in an enantioselective manner as well, providing access to a chiral synthesis of the 2,5-diaryl tetrahydrofurans.

In this communication, we report such a reduction of our diketone intermediate in a regio- and enantioselective manner using (S)-BINAL-H (developed by Noyori[17] *et al.*) as the reducing agent. To the best of our knowledge, this reduction has not previously been reported on a diketone substrate.

The diketone sulfone (**2**) (Scheme-1) was prepared in three high yield steps from the reported intermediate (**1**)[15], which in turn was prepared in three steps from 5-iodovanillin and 3,4,5-trimethoxyacetophenone. The diketone sulfone (**2**) was reduced with (S)-BINAL-H using a modification of Noyori's conditions[18] to the hydroxy-ketone (**3**) and 5-7% of the regio-isomeric hydroxy-ketone. The pure hydroxy-ketone (**3**) was obtained by crystallization (ether, hexane) in 70-76% yield ($[\alpha]_D$ = -12.6°, c=1, $CHCl_3$). The hydroxy-ketone (**3**) was further reduced to the diol by $NaBH_4$ in methanol at 0°C. The crude diol upon exposure to 5% TFA in $CHCl_3$ at 0°C yielded 1:4 mixture of *cis* and *trans* tetrahydrofurans. The 2(S),5(R)- *cis* isomer was reequilibrated (10% TFA in $CHCl_3$, 20°C, 2 hrs) to a 1:1 mixture of *cis* and *trans* isomers, enhancing the overall yield of the *trans* isomer. The electron deficient hydroxyethylsulfonylphenyl group as expected suppresses the ionization and racemization at the chiral benzylic center and there by helps maintain the stereochemical integrity during both the cyclization and equilibration processes, which was well demonstrated in the synthesis of L-659,989[16]. The desired 2(S),5(S)-*trans* isomer was treated with n-$Bu_4N^+F^-$ in THF which, after usual workup and crystallization (ethyl acetate, hexane) yielded MK 287[19] in 62% yield over three steps in an enantiomeric excess[20] of >98%.

BIOLOGICAL RESULTS AND DISCUSSION

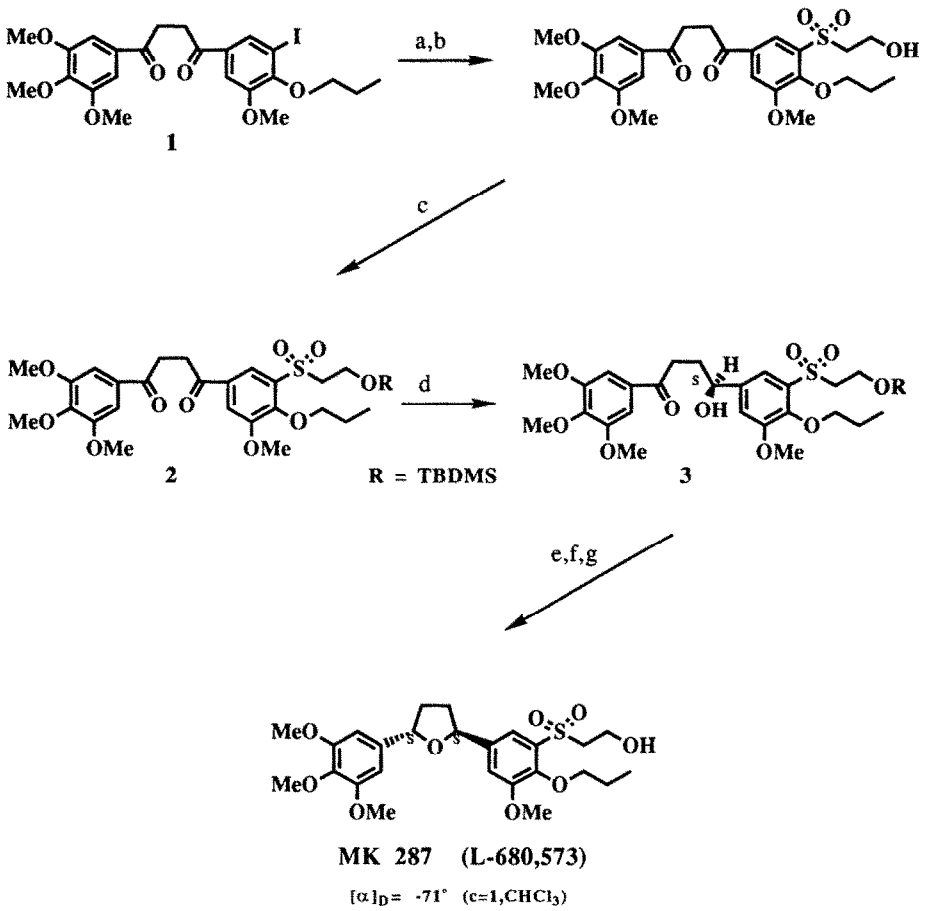
In vitro studies:

MK 287 potently inhibited [^{3}H]-C_{18}PAF binding to human platelet and PMN membrane receptors[21] with K_i values of 6.1 nM and 3.2 nM, respectively, while the K_i values of the (+)-enantiomer L-680,574 were found to be 116 nM and 59 nM, respectively. The above results indicate that most of the biological activity resides in the (-)-enantiomer MK 287, which is 20-fold more potent than the (+)-enantiomer, L-680,574. This is consistent with our earlier finding of the potency observed for (+)- and (-)-enantiomers of L-659,989[21].

Cellular Studies:

MK 287 was found to be a potent and specific inhibitor of PAF-induced human platelet aggregation. The IC_{50} values for inhibition of PAF-induced aggregation by MK 287 were 56 nM in plasma-rich platelets and 1.5 nM in washed platelets. This difference in potency can be attributed to a high degree of binding to plasma proteins. In addition, preincubation of isolated human neutrophils with MK 287 potently suppressed PAF-induced degranulation with an IC_{50} value of 4.0 nM.

SCHEME - 1



a: Cu, $(HOCH_2CH_2S)_2$, DMF, N_2, 130°C 2hr, then diketone iodide, 16hr, 83%

b: mCPBA, CH_2Cl_2, RT 2hr, 88% c: TBDMSCl, imidazole, DMF, RT, 4hr, 87%

d: (S)-BINAL-H, THF, -78°C, 6hr, 73% e: $NaBH_4$, MeOH, quant.

f: 5% TFA in $CHCl_3$, 5°C, 6hr g: n-$Bu_4N^+F^-$, THF, RT, 2hr, 62%

Specificity Studies:

MK 287 was found to be a specific PAF receptor antagonist as it showed no significant inhibition of specific binding of several radioligands such as [^{3}H]-LTB$_4$, [^{3}H]-LTC$_4$, [^{3}H]-LTD$_4$, [^{3}H]-fMLP, or [^{125}I]-C5a to their respective receptors at concentrations from 1to10 μM. MK 287 also showed no appreciable affinity for guinea pig heart β_1, guinea pig lung β_2 and calf brain α_1 or α_2 receptors at a concentration of 10 μM. The PAF-induced platelet aggregation by MK 287 was found to be specific as it had no effect on human platelet aggregation induced by ADP, vasopressin or the thromboxane A$_2$ agonist U44069 at concentrations from 1to10 μM.

In vivo studies:

The *in vivo* efficacy of MK 287 was demonstrated in two different animal models. MK 287, when administered orally, potently inhibited PAF-induced (10nM/kg) extravasation and increased plasma *N*-acetyl-β-D-glucosaminidase (NAGA) activity[22] in male and female rats in a dose-dependent manner with ED$_{50}$ values of 2.3 and 0.1 mg/kg, respectively. The observed gender difference could be due to differences in bioavailability and/or metabolism. The above gender difference was less pronounced in PAF-induced bronchoconstriction in guinea pigs. MK 287, when administered intravenously, potently inhibited PAF-induced bronchoconstriction in a dose-dependent manner in both male and female guinea pigs with ED$_{50}$ values of 0.19 and 0.10 mg/kg, respectively.

In summary, we have described an efficient enantioselective synthesis of MK 287 (L-680,573) which has been shown to be a potent, specific and orally active PAF receptor antagonist and is currently undergoing clinical trials.

References and Notes

1. C. A. Demopoulos, D. J. Hanahan, *J.Biol. Chem.*, **1979**, *254*, 9355. J. Benveniste, M. Tence, J. Bidault, C. Boullet, P. Varence and J. Polonsky C. *R. Seances Acad. Sci., Ser. D*, **1979**, *289*, 1037.
2. D. J. Hanahan, *Ann. Rev. Biochem.*, **1986**, *55*, 483. P. Braquet, L. Touqui, T. Y. Shen, B. B. Vargaftig, *Phamacol. Rev.*, **1987**, *39*, 97.
3. J. Benveniste, A. Arnoux, ed. Platelet-Activating Factor and Structurally Related Ether-Lipids. *Inserm Symposium no. 23*. Elsevier Science, Amsterdam, **1983**.
4. M.C. Venuti, *Ann. Reports Med. Chem.*, **1985**, *20*, 1935.
5. C.P. Page, J. Morley, *J. Pharmacol. Research Commun.*, **1986**, *18*, Suppl., 217.
6. J. Morley, *Agents and Actions*, **1986**, *19*, 100.

7. K. Cooper, M. J. Parry, *Ann. Reports Med. Chem.*, **1989**, *24*, 81.

8. Z.-I. Terashita, S. Tsushima, Y. Yoshioka, H. Nomura, Y. Inada, K. Nishikawa, *Life Sci*, **1983**, *32*, 1975.

9. T. Y. Shen, S.-B. Hwang, M. N. Chang, T. W. Doebber, M. -H. Lam, M. S. Wu, X. Wang, G. Q. Han, R. Z. Li, *Proc. Natl. Acad. Sci., U.S. A.* **1985**, *82*, 672. M. M. Ponpipom, R. L. Bugianesi, D. R. Brooker, B. -Q.Yue, S. -B. Hwang, T. Y. Shen. *J. Med. Chem.*, **1987**, *30*, 136.

10. L. Dupont, O. Dideberg, G. Germain, P. Braquet, *L.Acta Cristallogr.* **1986**, *C42*, 1759. P.G. Braquet, B. Spinnewyn, M. Braquet, R. H. Bourgain, J. E. Taylor, A. Etienne, K. Drieu, *Blood Vessels*, **1985**, *16*, 558.

11. J. Casals-Stenzel, G. Muacevic, K. H. Weber, *J. Pharmacol. Exp. Ther.*, **1987**, *241*, 974.

12. T. Biftu, N.F. Gamble, T. Doebber, S.-B. Hwang, T.Y. Shen, J. Snyder, J.P. Springer, R. Stevenson, *J. Med. Chem.*, **1986**, *29*, 1917.

13. Part of this work was presented at the Medicinal Chemistry Division of the American Chemical Society, 199th meeting, 1990, Boston.

14. T. Biftu, J. C. Chabala, J. Acton, C.-H. Kuo, *Drugs of the Future*, **1989**, *14*, 359 and references cited therein.

15. M.M. Ponpipom, S.-B. Hwang, T.W. Doebber, J.J. Acton, A.W. Alberts, T. Biftu, D.R. Brooker, R.L. Bugianesi, J.C. Chabala, N.L. Gamble, D.W. Graham, M.-H. Lam, M.S. Wu, *Biochem and Biophys. Res. Comm.*, **1988**, *150*, 1213.

16. M.M. Ponpipom, R.L. Bugianesi, J.C. Chabala, *Tetrahedron Lett.*, **1988**, *29*, 6211.

17. R. Noyori, I. Tomino, Y. Tanimoto, M. Nishizawa, *J. Am. Chem. Soc.*, **1984**,*106*, 6709.

18. The reaction was run at -78°C instead of -100°C for 6-8 hr. without any loss of optical purity.

19. All compounds reported in this communication gave correct microanalyses and exhibited ^{1}H NMR spectral characteristics that were in agreement with their structures. MK 287: NMR ($CDCl_3$) δ 1.04 (t, J = 7.1 Hz, CH_3), 1.87 (sext. J = 7.1 Hz, $CH_2\underline{CH_2}CH_3$), 1.99 & 2.50 (2m, 4H, H-3 & H-4), 3.66 (m, $\underline{CH_2}SO_2$), 3.83, 3.88, 3.92 (4 s, 4 OCH_3), 3.92 (m, $\underline{CH_2}OH$), 4.11 (t, J = 7.1 Hz, $CH_3CH_2\underline{CH_2}O$), 5.22 & 5.24 (m, H-5 & H-2), 6.61 (s, 2H, C_5 ArH), 7.29 & 7.50 (d, J = 2.0 Hz, 2H, C_2ArH).

20. The optical purity of MK 287 was determined by High Performance Liquid Chromatography using Pirkle Type 1A D-Phenylglycine column.

21. S.-B. Hwang, M. -H. Lam, T. Biftu, T. R. Beattie, T. Y. Shen, *J. Biol. Chem.*, **1985**, *260*, 5639.

22. M. S. Wu, T. Biftu, T. W. Doebber, *J. Pharmacol Exp. Therap.* **1986**, *239*, 841.