Pergamon

*Bioorganic & Medicinal Chemistry Letters*, Vol. 6, No. 9, pp. 1009-1014, 1996

0960-894X/96 $15.00 + 0.00

S0960-894X(96)00155-2

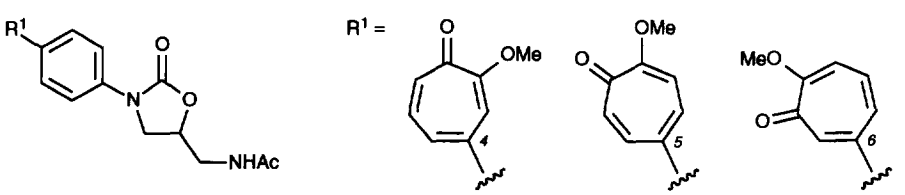
# SYNTHESIS AND ANTIBACTERIAL ACTIVITY OF NEW TROPONE-SUBSTITUTED PHENYLOXAZOLIDINONE ANTIBACTERIAL AGENTS. 2. MODIFICATION OF THE PHENYL RING - THE POTENTIATING EFFECT OF FLUORINE SUBSTITUTION ON *IN VIVO* ACTIVITY.

Michael R. Barbachyn,*,[†] Dana S. Toops,[†] Kevin C. Grega,[†] Susan K. Hendges,[†] Charles W. Ford,[‡] Gary E. Zurenko,[‡] Judith C. Hamel,[‡] Ronda D. Schaadt,[‡] Douglas Stapert,[‡] Betty H. Yagi,[‡] Jerry M. Buysse,[§] William F. Demyan,[§] James O. Kilburn,[⊥] and Suzanne E. Glickman[⊥]

[†]*Medicinal Chemistry Research,* [‡]*Cancer and Infectious Diseases Research,* [§]*Molecular Biology Research, Pharmacia & Upjohn, Inc., Kalamazoo, MI 49001 and* [⊥]*Centers for Disease Control, Atlanta, GA 30333*

**Abstract:** Various electron-withdrawing groups were incorporated into the *meta* position of tropone-substituted 3-phenyl-2-oxazolidinones and their influence on antibacterial activity examined. Consideration of *in vitro* and *in vivo* test results indicated that one or two fluorine atoms flanking the *para* tropone appendage is the optimum arrangement for these compounds. Synthetic routes to enantiomerically enriched analogues are reported.

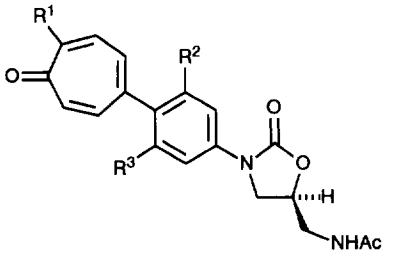

The oxazolidinones are an important new class of orally active, synthetic antibacterial agents.[1] In the preceding paper of this series we described the genesis of antibacterially active tropone-substituted phenyloxazolidinones, for example **1**.[2] Preliminary investigations into the effect of the tropone regioisomer on activity revealed that the 5- and 6-substituted analogues, **1b** and **1c**, respectively, were essentially equivalent in activity. Both **1b** and **1c** were found to be dramatically more active than the corresponding 4-substituted isomer **1a**.




**1** **a** **b** **c**

We were intrigued by the early oxazolidinone structure–activity relationships reported by workers at DuPont.[3] In particular, their finding that conjugated electron-withdrawing groups (e.g., acetyl) in the *para*

position of phenyloxazolidinones afforded the most active analogues suggested to us that further structural modifications leading to reduced electron density in the phenyl ring might provide compounds with increased potency. With this in mind, we contemplated the introduction of inductive electron-withdrawing substituents (e.g., F, Cl, and $CF_3$) into the *meta* position(s) of the phenyl ring.[4] Targets of generic structure **2** emerged after a decision to utilize the more readily accessible 5-bromo-2-methoxytropone as starting material.[5] Racemic analogues were judged adequate to explore preliminary structure–activity relationships in this series. The more active analogues were then prepared in enantiomerically enriched form.




**2**

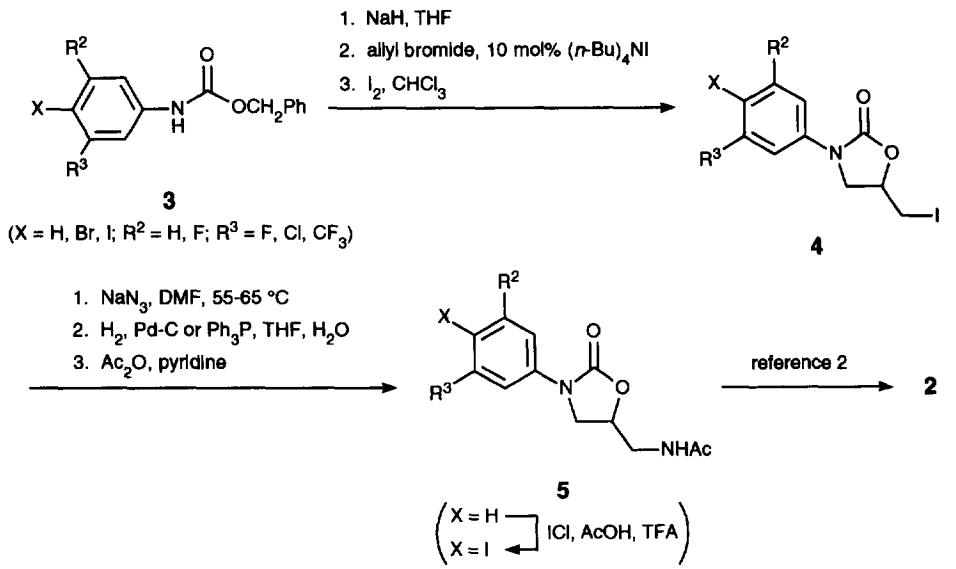
($R^1$ = alkoxy, amino; $R^2$ = H, F; $R^3$ = F, Cl, $CF_3$)

**Chemistry**

The synthetic procedures described in the preceding paper[2] for appending a methoxytropone moiety to an intermediate phenyloxazolidinone derivative were found to be applicable to the preparation of (±)-**2**. Therefore, preparation of the requisite *para* bromo- or iodophenyloxazolidinone precursors became a principal focus. As shown in Scheme 1, Cbz derivatives of substituted anilines (**3**) can be readily elaborated to the racemic 5-(iodomethyl)oxazolidinone intermediates **4** through an allylation/iodocyclocarbamation reaction sequence.[6-8] Treatment of **4** with sodium azide then affords a 5-azidomethyl derivative which, depending on the nature of the *para* X group, is reduced to the corresponding amine by one of two routes. When X = Br or I, reduction of the azide is achieved by formation and then hydrolysis of an intermediate iminophosphorane.[9] In cases where X = H, the desired 5-(aminomethyl)oxazolidinones were prepared either by the former route or by catalytic hydrogenation. The resultant amines were generally not isolated, but rather acetylated to afford intermediates **5** (X = H, Br, I). Examples of **5** with X = H were then iodinated to generate the corresponding *para* iodo derivatives. Further elaboration of **5** (X = Br, I), via a synthetic protocol involving a Stille coupling as its key step,[2] led to smooth generation of the targeted tropone-substituted phenyloxazolidinones **2**.

Since only the oxazolidinone (*S*)-enantiomer exhibits antibacterial activity,[1] the preparation of optically active **2** was an important goal. Two distinct synthetic approaches, exemplified by the monofluorophenyl series depicted in Scheme 2, were utilized to prepare enantiomerically enriched analogues. In the first of these, 3-fluorophenyl isocyanate (**6**) was reacted with (*R*)-glycidyl butyrate, followed by cleavage of the butyryl group

**Scheme 1**



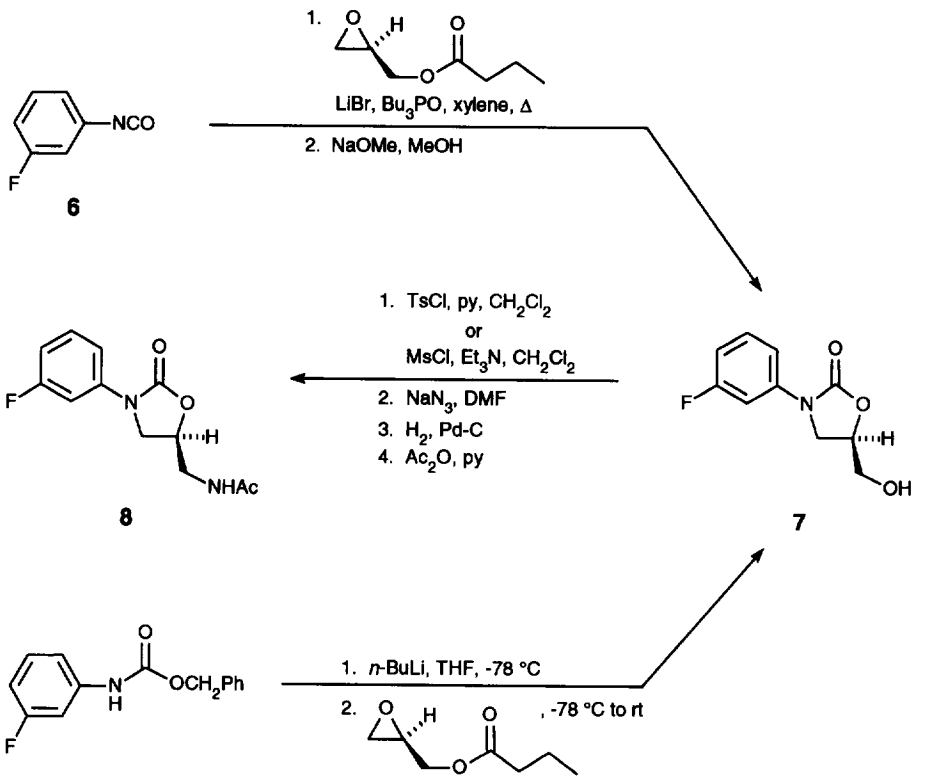

with sodium methoxide, to give the key (*R*)-5-(hydroxymethyl)oxazolidinone **7**.[1,10] A sequence of reactions involving mesylation or tosylation of **7**, azide displacement, reduction, and acetylation, then affords the (*S*)-5-(acetamidomethyl)oxazolidinone **8**. Intermediate **8** was then iodinated and elaborated via a Stille cross-coupling reaction[2] to generate optically active examples of structure **2**. The recent revelation that lithiated carbamate derivatives of anilines have general utility in preparing enantiomerically enriched 3-aryl-5-(hydroxymethyl)oxazolidinones[11] was also found to be applicable to the synthesis of intermediates **7**.

**Biological Evaluation and Discussion**

A preliminary evaluation of the antibacterial activity of selected tropone-substituted phenyloxazolidinones bearing one or two substituents in the *meta* position(s) of the phenyl ring was conducted. The *in vitro* activity was assessed by determination of minimum inhibitory concentration (MIC) values, utilizing standard agar dilution methods, and *via* a cell-free prokaryotic transcription coupled translation (T/T) assay, employing S-30 *Escherichia coli* extracts. In the latter test system, expression of plasmid encoded *lacZ* yields the functional reporter enzyme (β-galactosidase). *In vivo* efficacy was evaluated by effective dose$_{50}$ ($ED_{50}$) determinations, using standard lethal-systemic infection models in mice.

As shown in Table 1, the appended $R^2$ and $R^3$ groups on the phenyl ring of racemic analogues **9-13** have a profound effect on the antibacterial activity of these compounds. Using **11** as a reference point, it can be seen that the addition of a chlorine (**10**) results in reduced *in vitro* and *in vivo* antibacterial activity. The activity loss

**Scheme 2**

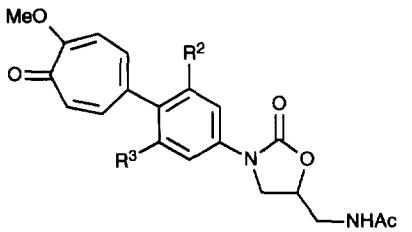



is even more pronounced for the corresponding trifluoromethyl analogue **9**. In contrast, the addition of one fluorine in the *meta* position affords a compound (**12**) with *in vitro* activity identical to that of **11**, but with significantly improved *in vivo* efficacy. Interestingly, the addition of a second fluorine substituent (**13**) imparts an additional two-fold improvement in the *in vivo* activity, relative to its monofluoro congener **12**. This trend is repeated in the potent *in vivo* activities (see Table 2) observed for the antibacterially active (*S*)-enantiomers[1] (*cf.* $ED_{50}$s of **14** and **15**). Compounds **14–17** demonstrate *in vitro* activity greater than that of the comparator, vancomycin, against aerobic gram-positive bacteria, including methicillin-resistant strains. *In vitro* activity against anaerobic bacteria (e.g., *Bacteroides fragilis*) and *Mycobacterium tuberculosis* is also indicated. Overall, the optically active compounds exhibit approximately twice the activity of their racemic counterparts (*cf.* MICs/$ED_{50}$s of **12** to **14** and **13** to **15**). Analogues **15** and **17** are particularly noteworthy in that their level of *in vivo* efficacy is comparable to or greater than, respectively, the current clinical benchmark vancomycin.

Based on the above results, the often significant potentiating effect of fluorine substitution is evident. The optimum arrangement in the tropone-substituted oxazolidinone subclass appears to be one or two fluorine

substituents flanking the appended *para* tropone moiety. Further evaluation of these agents is warranted.

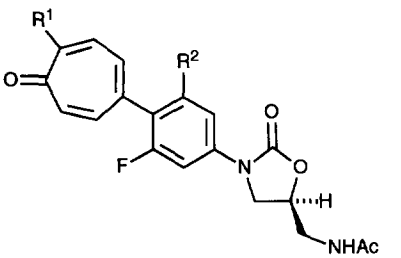
**Table 1.** Activity of Selected Racemic Tropone-Substituted Phenyloxazolidinones




| Compound | $R^2$ | $R^3$ | MIC (μg/mL)[1] | | | | $ED_{50}$, *S.a.*-1 (mg/kg)[2] | T/T (%)[3] |
|---|---|---|---|---|---|---|---|---|
| | | | *S.a.*-1 | *S.e.* | *E.f.* | *S.p.* | | |
| **9** | H | $CF_3$ | 32 | 4 | 16 | 4 | >20 (1.3) | 58 |
| **10** | H | Cl | 4 | 1 | 2 | 1 | >20 (2.2) | 71 |
| **11** | H | H | 2 | 0.5 | 1 | 0.25 | 12.2 (1.8) | 89 |
| **12** | H | F | 1 | 0.5 | 1 | 0.25 | 7.7 (1.7) | -- |
| **13** | F | F | 2 | 0.5 | 1 | 0.5 | 3.7 (2.2) | 85 |
| vancomycin | | | 1 | 2 | 4 | 0.5 | 1.3-2.2 | -- |

[1]Minimum inhibitory concentration: lowest concentration of drug (μg/mL) that inhibits visible growth of the organism. [2]Effective $dose_{50}$: amount of drug required (mg/kg body weight/dose) to cure 50% of infected mice subjected to a lethal systemic infection; oxazolidinones administered orally; vancomycin $ED_{50}$ (sc administration) in parentheses. [3]Transcription/translation assay; results reported as % inhibition, compound concentration of 2 μg/mL. *S.a.*-1, *Staphylococcus aureus* UC®9213 (methicillin-susceptible); *S.e.*, *Staphylococcus epidermidis* UC®12084 (methicillin-resistant); *E.f.*, *Enterococcus faecalis* UC®9217; *S.p.*, *Streptococcus pneumoniae* UC®9912.

**Table 2.** Activity of Optically Active Tropone-Substituted Mono- and Difluorophenyloxazolidinones



| Compound | $R^1$ | $R^2$ | MIC (μg/mL)[1] | | | | | | | $ED_{50}$, *S.a.*-1 (mg/kg)[2] |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | *S.a.*-1 | *S.a.*-2 | *S.e.* | *E.f.* | *S.p.* | *B.f.* | *M.tb.* | |
| **14** | MeO | H | 1 | 0.5 | 0.25 | 0.5 | 0.125 | 8 | ≤1[3] | 4.1 (1.7) |
| **15** | MeO | F | 1 | 0.5 | 0.5 | 1 | 0.25 | 8 | -- | 2.3 (2.0) |
| **16** | MeNH | H | 0.5 | 0.25 | 0.125 | 0.25 | 0.125 | 2 | -- | -- |
| **17** | propargylamino | H | 1 | 0.5 | 0.25 | 0.5 | 0.125 | 2 | -- | 1.3 (2.1) |
| vancomycin | | | 1 | 1 | 2 | 4 | 0.5 | 32 | -- | 1.7-2.1 |

[1]Minimum inhibitory concentration. [2]Effective $dose_{50}$; oxazolidinones administered orally; vancomycin $ED_{50}$ in parentheses (sc administration). [3]Isoniazid MIC = 0.2 μg/mL. *S.a.*-1, *Staphylococcus aureus* UC®9213 (methicillin-susceptible); *S.a.*-2, *Staphylococcus aureus* UC®6685 (methicillin-resistant); *S.e.*, *Staphylococcus epidermidis* UC®12084 (methicillin-resistant); *E.f.*, *Enterococcus faecalis* UC®9217; *S.p.*, *Streptococcus pneumoniae* UC®9912; *B.f.*, *Bacteroides fragilis* UC®12199; *M.tb.*, *Mycobacterium tuberculosis* H37Rv.

**Acknowledgment**

The authors thank Debra A. Ulanowicz and Steven J. Brickner for starting materials.

**References and Notes**

1. Gregory, W. A.; Brittelli, D. R.; Wang, C.-L. J.; Wuonola, M. A.; McRipley, R. J.; Eustice, D. C.; Eberly, V. S.; Bartholomew, P. T.; Slee, A. M.; Forbes, M. *J. Med. Chem.* **1989**, *32*, 1673.
2. Barbachyn, M. R.; Toops, D. S.; Ulanowicz, D. A.; Grega, K. C.; Brickner, S. J.; Ford, C. W.; Zurenko, G. E.; Hamel, J. C.; Schaadt, R. D.; Stapert, D.; Yagi, B. H.; Buysse, J. M.; Demyan, W. F.; Kilburn, J. O.; Glickman, S. E. *Bioorg. Med. Chem. Lett.* **1996**, *6*, preceding paper in this issue.
3. Gregory, W. A.; Brittelli, D. R.; Wang, C.-L. J.; Kezar, H. S., III; Carlson, R. K.; Park, C.-H.; Corless, P. F.; Miller, S. J.; Rajagopalan, P.; Wuonola, M. A.; McRipley, R. J.; Eberly, V. S.; Slee, A. M.; Forbes, M. *J. Med. Chem.* **1990**, *33*, 2569.
4. During the course of these investigations, oxazolidinones bearing multiple substituents on their phenyl ring, including fluorinated derivatives, were disclosed: Park, C.-H.; Brittelli, D. R.; Wang, C. L.-J.; Marsh, F. D.; Gregory, W. A.; Wuonola, M. A.; McRipley, R. J.; Eberly, V. S.; Slee, A. M.; Forbes, M. *J. Med. Chem.* **1992**, *35*, 1156. The analogues described are distinct from those shown in this paper. Interestingly, a blanket statement by Park and co-workers that 3,4,5-trisubstituted phenyloxazolidinones "are devoid of antibacterial activity" has been shown to be erroneous for the 3,4,5-trisubstituted phenyloxazolidinones (e.g., **13** and **15**) described herein.
5. Banwell, M. G.; Lambert, J. N.; Reum, M. E.; Onrust, R. *Org. Prep. Proc. Intl.* **1988**, 393.
6. Satisfactory spectra (IR, $^1H$ NMR, low and high resolution MS) were obtained for all new compounds.
7. Grega, K. C.; Barbachyn, M. R.; Brickner, S. J.; Mizsak, S. A. *J. Org. Chem.* **1995**, *60*, 5255.
8. A manuscript describing a comprehensive study of the iodocyclocarbamation reaction is under preparation: Brickner, S. J. *et al.*
9. Vaultier, M.; Knouzi, N.; Carrie, R. *Tetrahedron Lett.* **1983**, *24*, 763.
10. Herweh, J. E.; Kauffmann, W. J. *Tetrahedron Lett.* **1971**, 809.
11. Brickner, S. J.; Manninen, P. R.; Ulanowicz, D. A.; Lovasz, K. D.; Rohrer, D. C. *Abstracts of Papers*, 206th National Meeting of the American Chemical Society, Chicago, IL, August, 1993; American Chemical Society: Washington, DC, 1993; ORGN 089.

(*Received in USA* 9 *February* 1996; *accepted* 19 *March* 1996)