Pergamon

Bioorganic & Medicinal Chemistry Letters 12 (2002) 2121–2123

BIOORGANIC & MEDICINAL CHEMISTRY LETTERS

The Synthesis and Biological Evaluation of a Novel Series of Antimicrobials of the Oxazolidinone Class

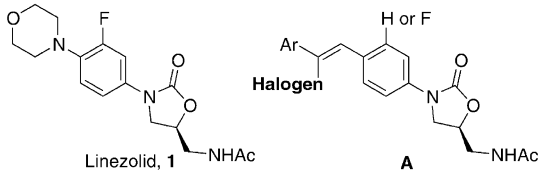
Richard J. Sciotti,* Marina Pliushchev, Paul E. Wiedeman, Darlene Balli, Robert Flamm, Angela M. Nilius, Kennan Marsh, DeAnne Stolarik, Robert Jolly, Roger Ulrich and Stevan W. Djuric*

Abbott Laboratories, Infectious Disease Research, Department 47N, Abbott Park, IL 60064, USA

Received 25 February 2002; accepted 8 May 2002

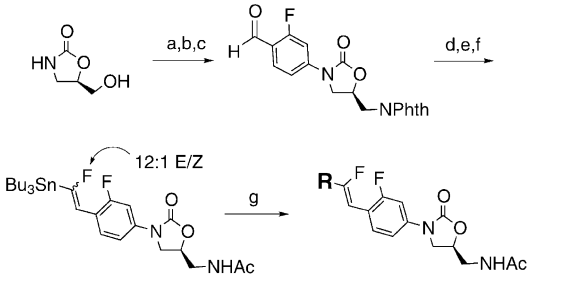
Abstract—A novel series of antimicrobials of the oxazolidinone class is disclosed. These compounds are characterized relative to previously described analogues by a 'halostilbene-derived' pharmacophore and demonstrate enhanced antimicrobial activity against key Gram-positive pathogens when compared to Linezolid.

The oxazolidinones represent a novel class of antimicrobial agent that possess excellent activity against antibiotic-sensitive and MIC_{90} resistant Gram-positive bacteria.[1] The prototype member of this class, Linezolid, has recently been approved for a number of clinical applications in man including the treatment of nosocomial and community-acquired pneumonia and skin infections caused by *Staphylococcus aureus*/Methicillin resistant *S. aureus*, Vancomycin resistant *Enterococci*, and *Streptococcus pneumoniae* (Pen-S). We were interested in identifying second generation compounds that had greater antimicrobial potency and less potential for side effects such as myelosuppression, including anaemia, and thrombocytopenia.[2] To this end, we pursued a series of stilbene/'heterostilbene'-based analogues of general structure **A** whose conception was based on the structure and activity of a previously disclosed α,β unsaturated ester[3] and a series of amide based phenyloxazolidinone derivatives.[4] In this context, it should be remembered that vinyl fluorides are a well-known class of amide bond mimetics.[5]



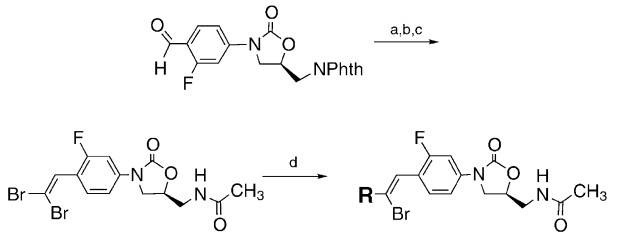
*Corresponding author. Fax: +1-847-938-1674. E-mail: richard.sciotti@pfizer.com (R. J. Sciotti); stevan.w.djuric@abbott.com (S. W. Djuric).

The compounds wherein the halogen group is fluoro were constructed as outlined in Scheme 1. Synthesis of the corresponding bromo analogues is shown in Scheme 2. The sequence to access the target vinyl fluorides featured as key components the palladium(0) catalyzed oxazolidinone/aryl halide coupling reaction recently disclosed by us[6,7] and the known but critical aldehyde to α-fluoro vinyl stannane conversion introduced by McCarthy and co-workers.[8–10] The α-fluoro vinyl stannane could be subsequently coupled under Pd(0) catalyzed conditions with the requisite aryl halide. This reaction proved to be especially capricious with only 11 out of 45 reactions providing product in modest yield (range 2–40%).



Scheme 1. Synthesis of fluorostilbenes: (a) *p*-TsCl, pyridine, −5 °C, 2 h, 95%; (b) potassium phthalimide, DMF, 80 °C, 5 h, 75%; (c) 4-bromo-2-fluorobenzaldehyde, Pd_2dba_3, Cs_2CO_3, BINAP, toluene, 100 °C, 16 h, 77%; (d) fluoromethyl phenyl sulfone, $(EtO)_2P(O)Cl$, $LiN(TMS)_2$, THF, 93%; (e) (i) N_2H_2–H_2O, EtOH; (ii) Ac_2O, pyridine/CH_2Cl_2, 67%; (f) *n*-Bu_3SnH (4 equiv), AIBN, benzene, 80 °C, 81%; (g) R-X (1.01 equiv), CuI (0.95 equiv), Pd_2dba_3 (0.05 equiv), P*t*-Bu_3 (0.2 equiv), dioxane (0.2 M), 2–8 h, 75 °C.

0960-894X/02/$ - see front matter
PII: S0960-894X(02)00352-9

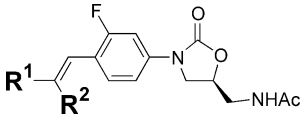


Scheme 2. Synthesis of bromostilbenes: (a) polystyrene-PPh_3 (6 equiv), CBr_4 (3 equiv), CH_2Cl_2, −5 °C, 1 h, 93%; (b) N_2H_4–H_2O (3 equiv), THF/EtOH, 70 °C, 3 h; (c) Ac_2O, pyridine, rt, 5 min, 77% total for (b) + (c); (d) R-$B(OH)_2$, $Pd(PPh_3)_4$ (0.1 equiv), 10% Na_2CO_3/H_2O, DME, 70 °C, 16 h.

Scheme 2 featured a new and useful version of the Corey–Fuchs[11] aldehyde to dibromoolefin conversion using polymer bound triphenylphosphine. This modification avoids the tedious removal of triphenyphosphine that is associated with the parent procedure. The 1,1-dibromoolefins coupled effectively with a variety of aryl halides to provide target analogues in poor to good yields (range 12–82%).

The compounds were evaluated for antimicrobial activity against a panel of representative Gram-positive pathogens including clinically relevant strains. Many compounds demonstrated significantly better activity than Linezolid against several of these organisms. In

Table 1. Structure–activity relationships in halostilbene oxazolidinones



Compd	R^1	R^2	*S. aureus* MIC[a] (μg/mL)	*S. epidermidis* MIC[a] (μg/mL)	*E. faecium* MIC[a] (μg/mL)	*M. cat.* MIC[a] (μg/mL)	*S. pneumo.* MIC[a] (μg/mL)	MPS[b] IC_{50} (μM)	MAO B[c] IC_{50} (μM)
Linezolid **1**			4	1	2	4	1	12 (7)	17 (5)
2	5-CN-3-Pyridyl	H	4	2	2	16	16	9.4±1.4	>300 (2)
3	5-CN-3-Pyridyl	F	1	0.5	1	4	≤0.125	<0.1 (2)	>300 (2)
4	5-CN-3-Pyridyl	Br	1	0.25	<0.125	2	≤0.125	1.3±0.2	>300 (2)
5	3-Pyridyl	H	4	2	2	16	16	7.8±1.0	59 (2)
6	3-Pyridyl	F	4	0.25	1	8	0.5	1.6±0.1	98 (2)
7	3-Pyridyl	Br	2	1	2	8	1	12 (2)	121 (4)
8	F	3-Pyridyl	8	4	4	32	4		
9	4-Pyridyl	Br	8	2	1	16	1	14(3)	186 (2)
10	4-MeO-Pyridin-3-yl	Br	8	4	2	32	2		
11	2-CN-Ph	Br	16	4	4	128	2	36 (2)	>150 (2)
12	3-CN-Ph	F	>128	128	>128	>128	2	>300 (2)	>300 (2)
13	3-CN-Ph	Br	0.25	0.125	0.125	1	0.25	24 (2)	>150 (4)
14	4-CN-Ph	Br	4	1	2	>128	0.5	76 (2)	>150 (4)
15	3-Acetyl-Ph	F	0.5	1	2	>128	1	69±12	>300 (2)
16	3-Acetyl-Ph	Br	2	1	1	4	1		
17	4-Acetyl-Ph	Br	64	64	32	128	1		
18	3-CHO-Ph	F	1	0.5	1	64	4	9.9±0.8	>300 (2)
19	3-CHO-Ph	Br	0.5	0.25	0.5	1	1	33 (2)	>300 (2)
20	3-$CONH_2$-Ph	Br	4	4	2	16	4		
21	3-CO_2Me-Ph	Br	8	8	4	>128	32	150 (2)	>300 (2)
22	3-CH_2OH-Ph	Br	2	1	0.5	4	1	8 (2)	>150 (4)
23	3-NH_2-Ph	Br	2	1	2	8	2		
24	3-N-Acetyl-Ph	Br	4	2	2	64	2		
25	5-*i*Pr-2-MeO-Phenyl	Br	>128	>128	>128	32	64		
26	Ph	Br	16	16	16	128	2	>300 (2)	<9.4 (2)
27	H	F	8	8	8	32	2		
28	$PhSO_2$–	F	8	8	8	64	8		
29	2-Furyl	Br	1	0.25	0.5	1	0.5		
30	2-Thiazolyl	F	2	2	2	8	2		<9.4 (1)
31	2-CN-Thiophen-3-yl	F	>128	16	16	>128	8	>300 (2)	285 (2)
32	5-Ac-Thiophen-2-yl	Br	1	0.5	0.5	2	0.5		
33	2-Me-2*H*-Tetrazol-5-yl	F	8	1	4	32	2	4.9±0.5	>300 (2)
34	5-Pyrimidinyl	F	2	1	2	4	1		475 (1)
35	2-Amino-5-pyrimidinyl	F	16	4	8	32	8		
36	3,5-di-Me-Oxazol-4-yl	Br	32	16	32	64	8	54 (2)	21 (2)

[a]National Committee for Clinical Laboratory Standards. 1997. Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria that Grow Aerobically: Approved Standard M7-A5. National Committee for Clinical Laboratory Standards, Wayne, PA, USA.
[b]Mitochondrial Protein Synthesis (MPS) IC_{50} expressed as a mean ±STD or as a mean of (*n*) experiments. See ref 12 for assay protocol.
[c]Monoaminoxidase B IC_{50} expressed as a mean of (*n*) experiments. Assay adapted from a Molecular Probes kit. The assay was carried out using rat primary hepatocytes because the rat was the primary model for toxicity used in assessing these compounds. We have reported the MAO B data because, in all cases, the compounds were significantly more inhibitory using substrate for this isozyme than for the MAO A isozyme, where little if any inhibition was noted.

addition, the compounds were evaluated in two key toxicity/selectivity assays (a) inhibition of mitochondrial protein synthesis (this effect may be, at least in part, responsible for the hematologic toxicities associated with Linezolid)[12] and (b) inhibition of monoamine oxidase. Although this is not a significant clinical problem with Linezolid, it is a class effect for oxazolidinones[13] and needs to be avoided. The biological SAR data is highlighted in Table 1. Several compounds from either series, gratifyingly, showed increased potency, reduced MPS and MAO inhibitory activity compared to Linezolid. Although it was difficult to draw SAR conclusions on the series in a general sense (no clear cut patterns or direct comparison between the bromo and fluoro series being impossible due to the relative inaccessibility of target compounds) some general comments are instructive. Compounds containing halogen-substituted double bonds were as a rule more potent than their corresponding des-halo counterparts (**2**–**4** and **5**–**7**). Within a halogenated series, the bromo analogues were generally more potent than the corresponding fluoro compounds (**3**–**4**, **6**–**7**, **12**–**13**, **15**–**16**, and **18**–**19**). Activity of compounds containing the (*Z*)-fluorostilbene unit was greater than that of the corresponding (*E*)-isomer (**6** and **8**). A variety of R^1 groups were well tolerated with a general trend that heterocycles bearing electron-withdrawing groups (higher sigma values) were the most active in the antimicrobial screens. There was a potency enhancement associated with substituents on the *meta* position of phenyl groups (**13** and **14**, **16** and **17**) or with a 3-pyridyl moiety to form a 'heterostilbene' (**7** and **9**). This was not *generally* true for analogues that bore a phenyl group substituted with an electron-withdrawing group in the para position, for example **17**.

Representative compounds with potency comparable to **1** were selected for pharmacokinetic evaluation in cassette dosing format in the rat. Compound **6** displayed characteristics that were considered to be appropriate for in vivo evaluation in animal infection models. For example, the PK profile of **6** following a 5 mpk iv dose was characterized by a low plasma clearance (CLp = 0.25 L/h kg), with a volume of distribution of 0.42 L/kg and an apparent elimination half life of $\sim$1.2 h. When administered orally at the same dose, the bioavailability of the compound was found to be 65%, $n = 6$). The outcome of these in vivo studies will be reported upon in due course.

References and Notes

1. Ford, C.; Hamel, J.; Stapert, D.; Moerman, J.; Hutchinson, D.; Barbachyn, M.; Zurenko, G. *Infect. Med.* **1999**, *16*, 435.
2. *SCRIP—World Pharmaceutical News*, 7th March 2001.
3. Brittelli, D. R.; Gregory, W. A.; Corless, P. F.; Park, C. H. US Patent 4, 977, 173, 1990; *Chem. Abstr.* **1989**, *111*, 232783.
4. Gordeev, M. F.; Luehr, G. W.; Patel, D.; Gordon, E. PCT Patent Application WO 99/37630; *Chem. Abstr.* **1999**, *131*, 116228.
5. Allmendinger, T.; Furet, P.; Hungerbuhler, E. *Tetrahedron Lett.* **1990**, *31*, 7279.
6. Madar, D. J.; Kopecka, H.; Pireh, D.; Pease, J.; Pliushchev, M.; Sciotti, R. J.; Wiedeman, P. E.; Djuric, S. W. *Tetrahedron Lett.* **2001**, *42*, 3681.
7. Danielmeier, K.; Steckhan, E. *Tetrahedron: Asymmetry* **1995**, *6*, 1181.
8. Matthews, D. P.; Persichetti, R. A.; McCarthy, J. R. *Org. Prep. Proced. Int.* **1994**, *26*, 605.
9. Chen, C.; Wilcoxen, K.; Zhu, Y.-F.; Kim, K.; McCarthy, J. R. *J. Org. Chem.* **1999**, *64*, 3476.
10. Huber, E. W.; Le, T.-B.; Laskovics, F. M.; Matthews, D. P.; McCarthy, J. R. *Tetrahedron* **1996**, *52*, 45.
11. Corey, E. J.; Fuchs, P. L. *Tetrahedron Lett.* **1972**, 3769.
12. **Mitochondrial Protein Synthesis (MPS) Assay**. Rat heart mitochondria were isolated as described by McKee et al. (*Am. J. Physiol.* **1990**, *258*, E492). Mitochondria were incubated at 1 mg/mL protein as determined by the Bradford assay in buffer containing constituents essential for translation (ADP, amino acids, etc.) in a total volume of 50 μL. Protein synthesis was measured by following incorporation of S^{35} methionine at a known specific activity for one h at 30 °C. The reaction was quenched by addition of 50 μL of 5% cold TCA/5 mM methionine and transferring it to ice. 20 μL aliquots (in triplicate) of each reaction were transferred to Millipore multiscreen plates. The plates were washed five times using 5% TCA/5 mM methionine. Plates were dried overnight, 50 μL of Optimax scintillant was added and the plates were counted using a Wallac Microbeta 1450 (Perkin-Elmer). Abbott compounds as well as positive and negative controls were prepared in DMSO as 100× stocks and assessed at 6–7 varying concentrations by including them in the incubation and then determining S^{35} incorporation relative to control. The percent organic in the reaction did not exceed 1% of the total volume. An IC_{50} based on percent of control was calculated from the linear portion of the inhibition/concentration curve. Compounds were analyzed in at least two separate experiments and these data were summarized to give a mean IC_{50}. We hypothesized that the hematosuppression of linezolid was due to an inhibition of mitochondrial function and protein synthesis similar to what has been described with chloramphenicol (see Holt, D.; Harvey, D.; Hurley, R. *Adverse Drug React. Toxicol. Rev.* **1993**, *12*, 83. Yunis, A. A. *Am. J. Med.* **1989**, *87*, 44N), and we used the MPS assay as an index of mitochondrial function in these studies. Both chloramphenicol and linezolid inhibit MPS at low to sub-micromolar concentrations in vitro (IC_{50}'s less than 1 and 12 μM, respectively). Both compounds have been shown to inhibit MPS and reduce both cytochrome C oxidase activity and mitochondrial protein synthesis in bone marrow of treated rats implicating the loss of mitochondrial function in the toxicity (data not shown).
13. Gates, K. S.; Silverman, R. B. *J. Am. Chem. Soc.* **1989**, *111*, 8891.