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Synthesis and in vitro DMPK profiling of a 1,2-dioxolane-based library with activity against *Plasmodium falciparum*

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ABSTRACT

A 43-member 1,2-dioxolane library was synthesized by coupling a 1,2-dioxolane-3-acetic acid derivative to a range of amines. Ten compounds had EC_{50}s $\leqslant$ 30 nM against *Plasmodium falciparum* 3D7 and Dd2 strains, and another 15 compounds had EC_{50}s $\leqslant$ 50 nM against both 3D7 and Dd2. The library was then subjected to a range of in vitro DMPK assays, which revealed that side chains with a heteroatom were required for favorable solubility, Log *D* and membrane permeability. CYP450 inhibition was isoform dependent, with 2C19 and 3A4 particularly susceptible, and the majority of compounds tested against rat and human microsomes were metabolized rapidly.

Malaria is a major parasitic disease in many areas of Africa, Asia and South America. *Plasmodium falciparum* causes the majority of fatalities, and its resistance to many contemporary antimalarials has compounded the problem of disease control.[1] First line malaria treatments are combination therapies containing at least one of the endoperoxides (**1a**–**c**) displayed in Figure 1. These compounds are derived in a semi-synthetic fashion from the natural occurring compound artemisinin (**1d**). As *P. falciparum* has developed resistance to every other clinically deployed agent, it seems prudent to investigate alternative artemisinin-inspired templates.

Several groups have pursued the development of antimalarial endoperoxides produced by fully synthetic methods,[2] of which the most well known is OZ277 (**2**, Fig. 1). Its central trioxolane core resembles the endoperoxide of artemisinin, and the amino side group was introduced to improve pharmacokinetics.[2a]

Previous work in our laboratories[3] utilized a short synthetic route, with a $SnCl_4$-mediated annulation[4] producing the 1,2-dioxolane core, to afford novel endoperoxides that displayed antiplasmodial activity. The most potent compound (**3a**, Fig. 2) displayed potent activity against all five *P. falciparum* strains against which it was tested. Dioxolane **3b**, which possessed the same 2-amino-2-methyl propyl auxiliary group as **2**, was less active against 3D7 and Dd2 than **3a**.

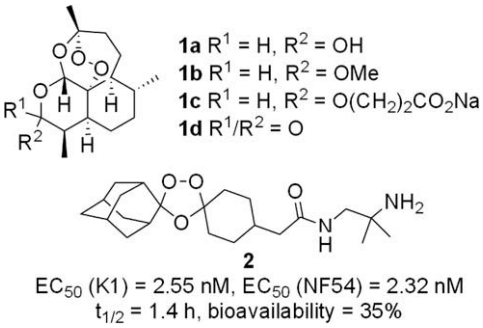

Figure 1. Artemisinin derivatives (**1a**–**c**), artemisinin (**1d**), and OZ277 (**2**).

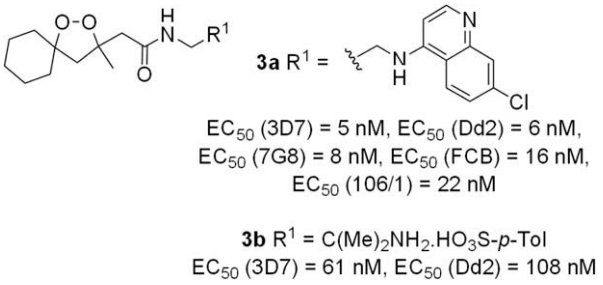

Figure 2. Activity of dioxolanes **3a**–**b** against selected parasite strains.

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Table 1
DMPK assay data for dioxolane **3a**

Solubility[a] (μg/mL)	Log *D*[a]	PAMPA ($\times 10^{-6}$ cm/s)[b]	Metabolic stability (Cl_{int}, μL/min/mg)		CYP450 IC_{50} (μM)				
			Rat	Human	1A2	2C9	2C19	2D6	3A4
>56	3.4	66	>119	>72	>5	>5	3.4	3.9	1.0

[a] Assay performed at pH 7.4.
[b] Assay performed at pH 6.5–7.4.

The basic in vitro drug metabolism and pharmacokinetic (DMPK) properties of **3a** revealed that it would be a poor lead for the development of a potentially useful therapeutic agent (Table 1). Dioxolane **3a** was metabolized rapidly by both rat and human microsomes, and it inhibited three of five CYP450 isozymes with IC_{50}s <5 μM. However, **3a** displayed high solubility, high passive permeability in a parallel artificial membrane permeability assay (PAMPA), and its Log *D* value was within the range of most orally administered drugs.[5] The DMPK properties of **3b** could not be determined as a result of the free amine lacking a suitable chromophore for detection in the DMPK assays.

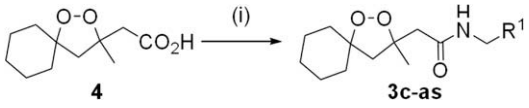
The next logical step was to synthesize a library of compounds that would ideally have improved in vitro DMPK properties compared to **3a** while maintaining its potent antiplasmodial activity. This letter describes the synthesis of a library of 1,2-dioxolanes made by introducing a range of auxiliary groups in the final coupling step, their activity against *P. falciparum* strains 3D7 and Dd2, and the subsequent profiling of the library in a panel of in vitro DMPK assays.

Synthesis of the library is outlined in Scheme 1. The 1,2-dioxolane-3-acetic acid **4** (see Ref. 3 for the synthesis of this intermediate) was reacted with a range of primary amines, using PyBOP to mediate the coupling,[6] to afford amides **3c–as**. We restricted our amine building blocks to aromatic methylamines for both ease of synthesis and compatibility with the DMPK assays. The crude material was submitted for automated mass-directed purification[7] to give material that was >90% pure by LCMS (assessed at 210 and 254 nm). The side groups, product yields and activity against *P. falciparum* strains 3D7 and Dd2 are displayed in Table 2. The antiplasmodial activity of **3c–as** was measured by a DAPI staining-based method for determining *P. falciparum* cell viability after exposure to the compound for 72 h.[8†]

The 1,2-dioxolane library displayed generally high activity against *P. falciparum*. Except for **3v** and **3aa**, the compounds were slightly more active against the laboratory strain 3D7 than the more robust field isolate Dd2. Of the 43 compounds in this library, ten afforded EC_{50}s ⩽ 30 nM against both 3D7 and Dd2, and another 15 compounds gave EC_{50}s ⩽ 50 nM against both strains. Only the benzoic acid derivative **3u** gave EC_{50}s >500 nM, which was in agreement with our previous study[3] showing that a carboxylic acid moiety significantly lowers activity compared to other analogs. The flat SAR of these compounds was also a feature of our previous study, where the majority of compounds gave EC_{50}s between 50 and 250 nM against 3D7 and Dd2.

All compounds were then subjected to the in vitro DMPK assays listed in Table 3. Compounds that displayed poor physicochemical properties (solubility ⩽ 1 mg/mL and/or Log *D* >4 and/or PAMPA ⩽ 5×10^{-6} cm/s) were typically not subjected to further assaying. The five CYP450 isozymes were selected to gain a preliminary understanding of the interaction between the test compounds and the CYP450 superfamily. Isozymes 1A2, 2C9, 2D6 and 3A4 are known to metabolize the majority of drug substrates,[9] with 3A4 alone estimated to metabolize up to 50% of drugs on the market.[10]

† Chloroquine was used as a positive control for all live-dead assay experiments.



Scheme 1. Reagents: (i) Amine, PyBOP, Et_3N.

Table 2
Reaction data and antiplasmodial activity of **3c–as**

Compd	R^1	% Yield	EC_{50} *Pf* 3D7 (nM)	EC_{50} *Pf* Dd2 (nM)
3c	Phenyl	65	29	39
3d	4-^{t}Butylphenyl	44	33	56
3e	4-Chloro-2-methylphenyl	53	22	31
3f	3-Methoxyphenyl	43	34	48
3g	2-Ethoxyphenyl	15	26	61
3h	3,5-Dimethoxyphenyl	55	36	58
3i	3,4,5-Trimethoxyphenyl	56	81	100
3j	3-Trifluoromethylphenyl	55	26	40
3k	4-Difluoromethoxyphenyl	34	16	25
3l	2-Trifluoromethoxyphenyl	18	19	34
3m	4-Trifluoromethoxyphenyl	30	20	29
3n	3-Chlorophenyl	54	26	29
3o	3-Bromophenyl	59	22	29
3p	3-Iodophenyl	63	24	30
3q	2,3-Dichlorophenyl	45	22	29
3r	2,4-Dichlorophenyl	20	23	31
3s	4-Bromo-2-fluorophenyl	58	26	37
3t	^{t}Butyl 3-benzylcarbamate	44	59	96
3u	4-Benzoic acid	16	814	1120
3v	Methyl 4-benzoate	47	51	51
3w	2-Aminophenyl	32	25	26
3x	4-(Dimethylamino)phenyl	14	24	33
3y	4-Nitrophenyl	53	22	26
3z	4-(Methylthio)phenyl	59	27	53
3aa	4-Sulfamoylphenyl	35	81	80
3ab	3-Biphenyl-3-yl	59	56	67
3ac	4′-Fluorobiphenyl-4-yl	58	26	30
3ad	3′,5′-Dichlorobiphenyl-4-yl	37	42	100
3ae	2-(Piperidin-1-yl)phenyl	61	39	50
3af	4-Morpholinophenyl	26	39	41
3ag	4-(1*H*-Pyrrol-1-yl)phenyl	55	27	44
3ah	4-(Thiophen-2-yl)phenyl	37	28	41
3ai	4-(4-Fluorophenoxy)phenyl	54	28	68
3aj	4-(3,5-*bis*(Trifluoromethyl) phenoxy)phenyl	49	62	106
3ak	3-(4-Chlorophenoxy)phenyl	53	18	30
3al	2-(2-(Hydroxymethyl) phenylthio)phenyl	62	28	44
3am	2,3-Dihydrobenzofuran-6-yl	34	40	89
3an	2,3-Dihydrobenzo[*b*] [1,4]dioxin-6-yl	60	40	41
3ao	3,4-Dihydro-2*H*-benzo[*b*][1,4]dioxepin-7-yl	39	76	148
3ap	Picolinyl	50	42	48
3aq	2-Pyridyl	66	118	201
3ar	4-Pyridyl	23	73	99
3as	Isoquinolyl	5	42	69

The physicochemical properties displayed a number of marked trends. High solubility generally correlated with the presence of a heteroatom in the aromatic side chain, whether it is exocyclic (**3f–i**, **3k**, **3u–aa**) or part of a heterocyclic ring system (**3af**, **3am–as**). Low solubility was determined for the biphenyls **3ab–ad** and a

Table 3
DMPK data for dioxolanes **3c–az**

Compd	R^1	Solubility (μg/mL)	Log D	PAMPA ($\times 10^{-6}$ cm/s)	CYP450 IC_{50}[a] (μM)					Cytoxicity EC_{50} (μM)			
					1A2	2C9	2C19	2D6	3A4	Dermal fibroblast	SI[b]	Kidney epithelial	SI[b]
3c	Phenyl	ND	ND	ND	ND	ND	ND	ND	ND	23	793	9	310
3d	4-^{t}Butylphenyl	4	3.5	ND	>5	>5	>5	>5	>5	1	33	0.8	24
3e	4-Chloro-2-methylphenyl	<1	4.2	ND	ND	ND	ND	ND	ND	1	45	>62	>2800
3f	3-Methoxyphenyl	>42	3.1	65	>5	4.2	2.6	>5	3.7	23	676	8	235
3g	2-Ethoxyphenyl	>43	3.1	74	>5	>5	2.3	>5	>5	18	692	6	231
3h	3,5-Dimethoxyphenyl	>45	3.3	55	>5	>5	1.9	>5	1.7	18	500	10	278
3i	3,4,5-Trimethoxyphenyl	>49	2.3	54	>5	>5	>5	>5	1.9	33	407	23	284
3j	3-Trifluoromethylphenyl	7	4	ND	>5	3.3	1.5	>5	3.2	7	269	4	154
3k	4-Difluoromethoxyphenyl	>46	3.1	ND	>5	>5	4.6	>5	>5	11	688	8	500
3l	2-Trifluoromethoxyphenyl	5	3.3	ND	>5	>5	3.7	>5	4.2	9	474	5	263
3m	4-Trifluoromethoxyphenyl	13	3.2	ND	>5	4	3.9	>5	2.9	4	200	1	50
3n	3-Chlorophenyl	>42	3.7	ND	>5	3.4	2.1	>5	4.9	8	308	3	115
3o	3-Bromophenyl	7	3.3	ND	>5	3.7	1.8	>5	>5	8	364	3	136
3p	3-Iodophenyl	1	3.5	ND	ND	ND	ND	ND	ND	2	83	2	83
3q	2,3-Dichlorophenyl	<1	3.4	ND	ND	ND	ND	ND	ND	2	91	0.3	14
3r	2,4-Dichlorophenyl	1	>3.5	ND	ND	ND	ND	ND	ND	2	87	0.5	22
3s	4-Bromo-2-fluorophenyl	3	4.1	ND	>5	3.9	2.8	>5	4.4	5	192	2	77
3t	^{t}Butyl 3-benzylcarbamate	38	3	ND	>5	>5	4.6	>5	1	2	34	>62	1050
3u	4-Benzoic acid	>43	0.3	0	>5	>5	>5	>5	>5	62	76	8	10
3v	Methyl 4-benzoate	>45	3.4	57	>5	>5	3.9	>5	>5	27	529	14	275
3w	2-Aminophenyl	>40	2.5	37	>5	>5	3.8	>5	1.9	9	360	7	280
3x	4-(Dimethylamino)phenyl	>43	3.1	90	3.8	>5	2.3	>5	4.8	31	1290	27	1130
3y	4-Nitrophenyl	>44	3.4	49	>5	>5	3.2	>5	>5	17	773	5	227
3z	4-(Methylthio)phenyl	>44	3.7	97	>5	>5	4	>5	>5	3	111	3	111
3aa	4-Sulfamoylphenyl	>48	1.4	ND	>5	>5	>5	>5	>5	24	296	>62	765
3ab	3-Biphenyl-3-yl	<1	3.8	ND	ND	ND	ND	ND	ND	2	36	0.3	5
3ac	4′-Fluorobiphenyl-4-yl	<1	4.8	ND	ND	ND	ND	ND	ND	1	38	0.5	19
3ad	3′,5′-Dichlorobiphenyl-4-yl	<1	>3.5	ND	ND	ND	ND	ND	ND	2	48	0.4	10
3ae	2-(Piperidin-1-yl)phenyl	7	>3.5	62	>5	>5	2.2	>5	1.6	2	51	>62	1590
3af	4-Morpholinophenyl	>49	2.7	ND	>5	>5	4.4	>5	>5	25	641	13	333
3ag	4-(1*H*-Pyrrol-1-yl)phenyl	0.6	4.3	70	ND	ND	ND	ND	ND	1	37	>62	2300
3ah	4-(Thiophen-2-yl)phenyl	<1	3.7	ND	ND	ND	ND	ND	ND	2	71	1	36
3ai	4-(4-Fluorophenoxy)phenyl	<1	>3.5	ND	ND	ND	ND	ND	ND	2	71	0.5	18
3aj	4-(3,5-*bis*(Trifluoromethyl) phenoxy)phenyl	<1	3.2	ND	ND	ND	ND	ND	ND	0.8	13	>62	1000
3ak	3-(4-Chlorophenoxy)phenyl	<1	3.2	ND	ND	ND	ND	ND	ND	1	56	0.2	11
3al	2-(2-(Hydroxymethyl) phenylthio)phenyl	11	4.3	77	>5	2.2	2.9	>5	1.2	6	214	2	71
3am	2,3-Dihydrobenzofuran-6-yl	>43	2.8	69	>5	>5	2.9	>5	2.6	29	725	7	175
3an	2,3-Dihydrobenzo[*b*] [1,4]dioxin-6-yl	>45	2.7	56	>5	>5	>5	>5	>5	22	550	11	275
3ao	3,4-Dihydro-2*H*-benzo[*b*][1,4]dioxepin-7-yl	>47	3.1	55	>5	>5	>5	>5	>5	6	79	27	355
3ap	Picolinyl	>43	3	74	>5	0.3	1.5	4.8	0.1	23	548	6	143
3aq	2-Pyridyl	>38	2.3	33	>5	>5	>5	>5	>5	24	203	22	186
3ar	4-Pyridyl	>38	2.1	18	>5	>5	0.1	>5	>5	30	411	3	41
3as	Isoquinolyl	>56	3.4	66	>5	>5	3.4	3.9	0.1	4	95	22	524

[a] SI = selectivity index, that is, cytotoxicity EC_{50}/*Pf* 3D7 EC_{50}.
[b] ND = not determined.

number of the halogenated derivatives (**3o–s**, **3ai–ak**). Log *D* values were generally in an acceptable range (between 1 and 4) with a small number of outliers, the most noticeable being the benzoic acid analog **3u**. PAMPA values tended to correlate with solubility, exemplified by the heteroatom-containing compounds **3f–i**, **3v–z** and **3am–as**, which all displayed high membrane permeability. Compound **3u** displayed no membrane permeability, which supports the hypothesis in our previous study[3] that the carboxylic acid moiety prevents passage of the compound to its site of action in the plasmodial cell.

The CYP450 assay results showed that enzyme inhibition by the dioxolanes was isoform dependent. 1A2 and 2D6 displayed low levels of inhibition, with 1A2 only inhibited by **3x**, and 2D6 inhibited by **3ap** and **3as**. Moderate levels of inhibition were observed for 2C9, whereas the two other isoforms were inhibited at <5 μM by a significant proportion of tested compounds. 2C19 was inhibited by 23 of 30 compounds at this threshold, and 3A4 was inhibited by 16 of 30 compounds. The results obtained for isoforms 2C9, 2C19 and 3A4 concur with a recent analysis that described 2C9 as less promiscuous with respect to the range of substrates it metabolized compared to 2C19 and 3A4.[11]

The dioxolanes were generally less active against human dermal fibroblast and kidney epithelial cell lines than *P. falciparum*. Selective toxicity towards *P. falciparum* versus non-cancerous human cell lines has been previously observed for the synthetic endoperoxide OZ277[12] and artemisinin-based dimers.[13] The majority of the compounds were slightly more cytotoxic towards kidney epithelial cells rather than dermal fibroblasts, and in the select occasions when the compounds were less toxic towards kidney epithelials, generally they displayed no toxicity (EC_{50} >62 μM).

The in vitro metabolic stability of 28 selected dioxolanes is displayed in Table 4. The vast majority of compounds displayed the rapid metabolism observed for **3a**. The only compounds that displayed any reduction from the maximum rate of metabolism were the benzoic acid **3u** (the dioxolane with the weakest antiplasmodial activity), the sulfamoyl analog **3aa**, and the pyridyl derivatives **3aq** and **3ar**. The lower metabolism of the pyridine derivatives suggests that incorporating nitrogenous aromatic heterocycles might be one method to decrease microsomal instability.

Table 4
In vitro metabolic stability for selected dioxolanes

Compd	R[1]	Metabolic stability (Cl_{int}, μL/min/mg)	
		Rat	Human
3d	4-^{t}Butylphenyl	>119	>72
3f	3-Methoxyphenyl	>119	>72
3g	2-Ethoxyphenyl	>119	>72
3h	3,5-Dimethoxyphenyl	>119	>72
3i	3,4,5-Trimethoxyphenyl	>119	>72
3j	3-Trifluoromethylphenyl	>119	>72
3k	4-Difluoromethoxyphenyl	>119	>72
3l	2-Trifluoromethoxyphenyl	>119	>72
3m	4-Trifluoromethoxyphenyl	>119	>72
3n	3-Chlorophenyl	>119	>72
3o	3-Bromophenyl	>119	>72
3s	4-Bromo-2-fluorophenyl	>119	>72
3t	^{t}Butyl 3-benzylcarbamate	>119	>72
3u	4-Benzoic acid	7.7	11
3w	2-Aminophenyl	>119	>72
3x	4-(Dimethylamino)phenyl	>119	>72
3y	4-Nitrophenyl	>119	>72
3z	4-(Methylthio)phenyl	>119	>72
3aa	4-Sulfamoylphenyl	56	>72
3ae	2-(Piperidin-1-yl)phenyl	>119	>72
3ah	4-(Thiophen-2-yl)phenyl	>119	>72
3al	2-(2-(Hydroxymethyl) phenylthio)phenyl	>119	>72
3am	2,3-Dihydrobenzofuran-6-yl	>119	>72
3an	2,3-Dihydrobenzo[*b*] [1,4]dioxin-6-yl	>119	>72
3ao	3,4-Dihydro-2*H*-benzo[*b*][1,4]dioxepin-7-yl	>119	>72
3ap	Picolinyl	>119	>72
3aq	2-Pyridyl	>119	68
3ar	4-Pyridyl	109	69
3as	Isoquinolyl	>119	>72

In summary, a 43-member library of dioxolanes was synthesized by coupling a range of aromatic methylamines to the 1,2-dioxolane-3-acetic acid **4**. After purification, the library was assayed against *P. falciparum* 3D7 and Dd2, and a high percentage of compounds displayed EC_{50}s ⩽ 50 nM against both strains. A range of in vitro DMPK assays revealed that side chains with a heteroatom were required for favorable solubility, Log *D* and membrane permeability. CYP450 inhibition was isoform dependent, with 2C19 and 3A4 particularly susceptible, and the majority of compounds tested against rat and human microsomes were metabolized rapidly. The high rates of 2C19 and 3A4 inhibition and microsomal metabolism reveal that significant optimization of the groups appended to the dioxolane core is needed before a viable candidate for drug development is identified.

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Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bmcl.2009.08.024.

References and notes

1. Daily, J. P. *J. Clin. Pharmacol.* **2006**, *46*, 1487.
2. (a) Vennerstrom, J. L.; Arbe-Barnes, S.; Brun, R.; Charman, S. A.; Chiu, F. C.; Chollet, J.; Dong, Y.; Dorn, A.; Hunziker, D.; Matile, H.; McIntosh, K.; Padmanilayam, M.; Santo, T. J.; Scheurer, C.; Scorneaux, B.; Tang, Y.; Urwyler, H.; Wittlin, S.; Charman, W. N. *Nature* **2004**, *430*, 900; (b) Singh, C.; Sharma, U.; Saxena, G.; Puri, S. K. *Bioorg. Med. Chem. Lett.* **2007**, *17*, 4097; (c) Singh, C.; Kanchan, R.; Sharma, U.; Puri, S. K. *J. Med. Chem.* **2007**, *50*, 521; (d) Dong, Y.; Creek, D.; Chollet, J.; Matile, H.; Charman, S. A.; Wittlin, S.; Wood, J. K.; Vennerstrom, J. L. *Antimicrob. Agents Chemother.* **2007**, *51*, 3033; (e) Terzic, N.; Opsenica, D.; Milic, D.; Tinant, B.; Smith, K. S.; Milhous, W. K.; Solaja, B. A. *J. Med. Chem.* **2007**, *50*, 5118; (f) O'Neill, P. M.; Rawe, S. L.; Borstnik, K.; Miller, A.; Ward, S. A.; Bray, P. G.; Davies, J.; Oh, C. H.; Posner, G. H. *ChemBioChem* **2005**, *6*, 2048; (g) Benoit-Vical, F.; Lelievre, J.; Berry, A.; Deymier, C.; Dechy-Cabaret, O.; Cazelles, J.; Loup, C.; Robert, A.; Magnaval, J.-F.; Meunier, B. *Antimicrob. Agents Chemother.* **2007**, *51*, 1463.
3. Martyn, D. C.; Ramirez, A. P.; Beattie, M. J.; Cortese, J. F.; Patel, V.; Rush, M. A.; Woerpel, K. A.; Clardy, J. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 6521.
4. Ramirez, A.; Woerpel, K. A. *Org. Lett.* **2005**, *7*, 4617.
5. Lipinski, C. A.; Lombardo, F.; Dominy, B. W.; Feeney, P. J. *Adv. Drug Delivery Rev.* **2001**, *46*, 3.
6. To the amine (0.075 mmol) at rt under an inert atmosphere in a 6 mL reaction vial was added **4** (16 mg, 0.075 mmol) in dry CH_2Cl_2 (1.5 mL), PyBOP (42 mg, 0.083 mmol) in dry CH_2Cl_2 (1.5 mL), and Et_3N (15 mg, 0.15 mmol). After 20 h water (2 mL) was added, and the mixture was shaken vigorously. The CH_2Cl_2 layer was withdrawn, dried ($MgSO_4$), and the liquid was decanted and dried under reduced pressure to give the crude dioxolane. Formation of the desired dioxolane was confirmed by LCMS, and in selected examples by ^{1}H NMR.
7. Mass-directed purification was performed on the Autopurification system from Waters Co. (Milford, MA), operated by FractionLynx 4.0 software. Column: Xterra C18, 10 μm, 19 × 50 mm, OBD by Waters Co. Flow rate = 44 mL/min. Method: 5–95% MeCN/Water with 0.1% formic acid. Run time = 5 min.
8. Baniecki, M. L.; Wirth, D. F.; Clardy, J. *Antimicrob. Agents Chemother.* **2007**, *51*, 716.
9. Rendic, S. *Drug. Metab. Rev.* **2002**, *34*, 83.
10. Thummel, K. E.; Wilkinson, G. R. *Ann. Rev. Pharmacol. Toxicol.* **1998**, *38*, 389.
11. Nath, A.; Atkins, W. M. *Biochemistry* **2008**, *47*, 157.
12. Vennerstrom, J. L.; Dong, Y.; Chollet, J.; Matile, H. US Patent 6,486,199, 2002.
13. Rosenthal, A. S.; Chen, X.; Liu, J. O.; West, D. C.; Hergenrother, P. J.; Shapiro, T. A.; Posner, G. A. *J. Med. Chem.* **2009**, *52*, 1198.