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Synthesis and Antiprotozoal Properties of Pentamidine Congeners Bearing the Benzofuran Motif

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Forty-eight cationically substituted pentamidine congeners possessing benzofuran rings were synthesized by a copper mediated heteroannulation of substituted *o*-iodophenols with phenyl acetylenes. Activities of compounds **1**−**48** against *Trypanosoma brucei rhodesiense*, *Plasmodium falciparum*, and *Leishmania donovani* and cytotoxicities for mammalian cells were influenced by the nature of cationic substituents, placement of the benzofuran fragment, and the length of the carbon linker between aromatic moieties. Several dications exhibited superior antiplasmodial and antileishmanial potencies compared to pentamidine.

Introduction

In recent years, the emergence of drug resistant pathogens has led to treatment failures for many infectious diseases, including malaria, human African trypanosomiasis (HAT[a] or sleeping sickness), and leishmaniasis. The aromatic diamidine 1,5-bis(4-amidinophenoxy)pentane (pentamidine) has been used against early stage *T. b. gambiense* HAT,[1,2] antimony-resistant leishmaniasis,[3] and *Pneumocystis jiroveci* (formerly *P. carinii*) pneumonia.[4] Additionally, the drug exhibited some antimalarial potency,[5,6] although it was never approved to treat the disease. Despite its extensive use as an HAT prophylactic, widespread resistance of trypanosomes to pentamidine in the field has not been observed.[6,7] However, there is growing pentamidine resistance of leishmaniasis. Thus, while pentamidine may be a viable starting point for novel antimicrobial agents, there is an urgent need for novel classes of compounds that are effective against these re-emerging infections.

Recently, we discovered several cationic bisbenzofurans[8] and 2-phenyl benzofurans[9] exhibiting excellent antitrypanosomal and antiplasmodial activities. To combine the rigidity of 2-phenyl benzofurans with the flexibility of pentamidine congeners, we incorporated the benzofuran ring into molecules of pentamidine-related analogues. Here we report an investigation of structure−activity relationships of novel pentamidine congeners **1**−**48** bearing benzofuran motifs against *T. b. rhodesiense*, *P. falciparum*, *L. donovani*, and for cytotoxicity against mammalian cells.

Chemistry

4-Hydroxy-3-iodobenzonitrile **49** and methyl 3-hydroxy-4-iodobenzoate **50** were prepared from 4-cyanophenol and 3-hydroxybenzoic acid, respectively, as previously described.[8] Synthesis of isomeric 1-ethynyl-3-methoxybenzene **51** and 1-ethynyl-4-methoxybenzene **52** was carried out following the literature procedure.[10]

The synthesis of the benzofurans **59**−**62** is outlined in Scheme 1. Copper-mediated coupling of *o*-iodophenols **49** and **50** with phenylacetylenes **51** and **52** using the modified Castro reaction[11] afforded benzofurans **53**−**56** in 72−81% yields. Esters **55** and **56** were directly converted to the corresponding nitriles **57** and **58** in 68 and 69% yields, respectively, by treatment with two-fold excess of Me_2AlNH_2 in hot xylene.[12] Deprotection of the compounds **53**, **54**, **57**, and **58** with melted pyridine hydrochloride[9] afforded cyanophenols **59**−**62** in 69−80% yields.

4-Bromoalkoxybenzonitriles[13] **63**−**66** were prepared by alkylation of 4-cyanophenol with excess of appropriate dibromoalkane. Alkylation of the compounds **59**−**62** with 4-bromoalkoxybenzonitriles **63**−**66** by treatment with K_2CO_3 in hot DMF afforded isomeric dinitriles **67**−**82** in 65−94% yields (Scheme 2). Compounds **67**−**82** were converted to imidate esters by the modified Pinner method,[14] followed by reaction with ethanolic solutions of ammonia, isopropylamine, or ethylenediamine at ambient temperature to yield dicationic benzofurans **1**−**48**.

Results and Discussion

This study investigates the effects of the substitution on the amidine groups, varying the placement of cationic moieties, altering the overall geometry of molecules by changing the position of the benzofuran fragment, and the elongation of the aliphatic linker between aromatic rings from three to six carbon atoms on antiprotozoal properties of novel pentamidine analogues **1**−**48**. The results of the in vitro evaluation of dications **1**−**48** against bloodstream form trypomastigotes of *T. b. rhodesiense* (STIB900), chloroquine resistant *P. falciparum* (K1), axenic amastigotes of *L. donovani* (MHOM/SD/62/1S−$CL2_D$), and their cytotoxicities against rat myoblast cells

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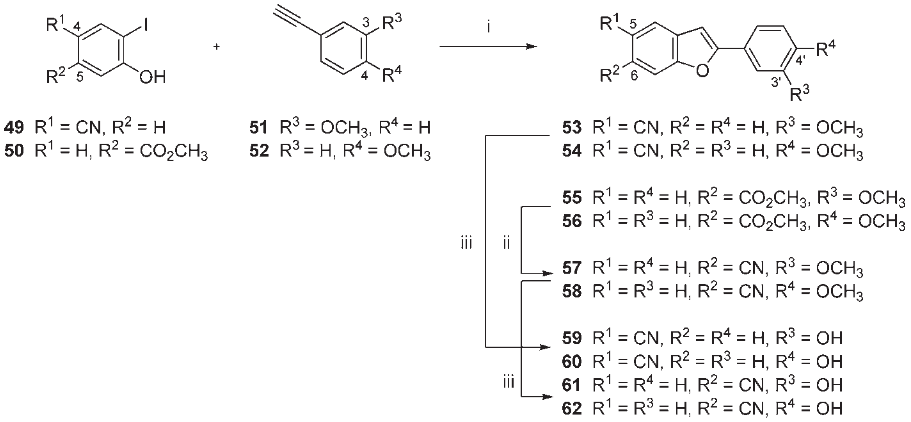
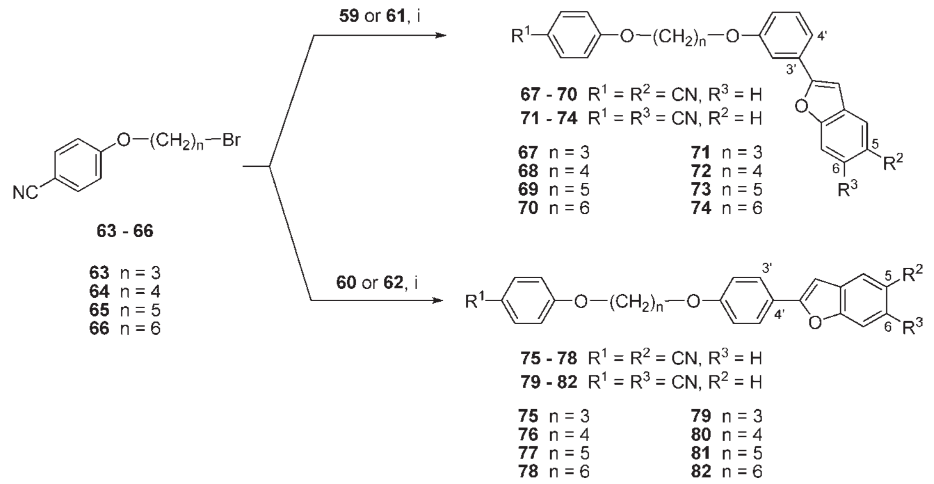
[a] Abbreviations: HAT, human African trypanosomiasis; PMD, pentamidine.

Scheme 1[a]



[a] Reagents and conditions: (i) Cu_2O, pyridine, 100 °C, overnight; (ii) Me_2AlNH_2, *o*-xylene, 110−120 °C, 3 h; (iii) pyridine hydrochloride, 170−180 °C, 3 h.

Scheme 2[a]



[a] Reagents and conditions: (i) K_2CO_3, DMF, 80−100 °C, overnight.

(L6) are summarized in Tables 1 and 2 and compared to the activity of pentamidine. To demonstrate the degree of selectivity between parasite and mammalian cells, selectivity indexes (SI) expressed as the ratio [IC_{50} (L6)/IC_{50} (*parasite*)] were calculated for each tested parasite.

Biology

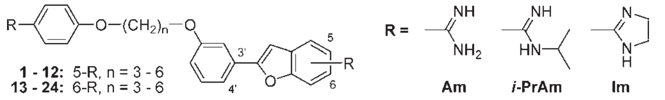
Cytotoxicity Study. Pentamidine congeners containing benzofuran motifs exhibited cytotoxicities versus L6 cells ranging from 0.8 to 82 μM. Bis(*N*-isopropyl)amidines were generally less cytotoxic than diamidines and diimidazolines apart from 5,4′-disubstituted congener **35** and 6,4′-disubstituted dications **41** and **47**. Compounds bearing cationic groups in the 6-position displayed lower cytotoxicity than their 5-substituted counterparts in all cases except diamidine **4**, bis(*N*-isopropyl)amidine **32**, and diimidazolines **6** and **12**, which were more cytotoxic than the corresponding isomers **16**, **44**, **18**, and **24**. The elongation of the carbon linker increased cytotoxicity of dications, especially those possessing the benzofuran ring in the 3′-position.

Antitrypanosomal Activity. All compounds **1**−**48** displayed antitrypanosomal activities in vitro with IC_{50} values varying from 25 nM to 1.74 μM. Dications **26**, **27**, **37**, **38** possessing the trimethylene linker and the benzofuran ring attached to the 4′-position exhibited antitrypanosomal IC_{50} values less than 100 nM. None of the tested compounds demonstrated a level of activity comparable to that of the standard trypanocidal medications. Thus, the most potent diamidine **37** (IC_{50} = 25 nM) was 8-fold less active against *T. b. rhodesiense* than pentamidine.

The antitrypanosomal properties of congeners **1**−**48** depended on the nature and the placement of cationic moieties, the position of the benzofuran ring, and the length of the carbon linkers. Thus, bis(*N*-isopropyl)amidines possessing one cationic group in the 5-position were more active against the pathogen than diamidines and diimidazolines in all cases except dications **11** and **29**. There was no correlation between the type of cationic moieties and activity of 6,3′-disubstituted congeners **13**−**24** versus *T. b. rhodesiense*. The antitrypanosomal potency of 6,4′-disubstituted dications **40**−**48** connected by 4−6-carbon chains against the pathogen reduced in the order Im > Am > *i*-PrAm, while the activity of the dications with the propylene linker decreased as the substitution on the amidine group increased (Table 2).

Among the compounds with the benzofuran ring attached to the 3′-position (Table 1), 6,3′-disubstituted cations connected by the 3-, 5-, and 6-carbon chains were more active versus *T. b. rhodesiense* than the corresponding 5,3′-disubstituted congeners, except for bis(*N*-isopropyl)amidine **20**. At the same time, 5,3′-disubstituted isomers possessing the

Table 1. Cytotoxicity and in Vitro Antiprotozoal Activity of Congeners **1**−**24**



compd	R	*n*	cytotoxicity[b] IC_{50} (μM)	*T. b. rhodesiense*[c]		*P. falciparum*[e]		*L. donovani*[g]	
				IC_{50} (μM)	SI_T[d]	IC_{50} (μM)	SI_P[f]	IC_{50} (μM)	SI_L[h]
1	Am	3	10.3	0.907	11	0.040	258	3.4	3
2	*i*-PrAm	3	26.5	0.547	48	0.061	434	4.5	6
3	Im	3	6.5	1.17	6	0.109	60	5.9	1
4	Am	4	8.1	0.779	10	0.076	107	4.5	2
5	*i*-PrAm	4	36.5	0.537	68	0.269	136	11	3
6	Im	4	9.5	1.17	8	0.099	96	12	< 1
7	Am	5	3.0	0.863	3	0.428	7	11	< 1
8	*i*-PrAm	5	10.1	0.452	22	0.123	82	6.9	1
9	Im	5	2.6	0.597	4	0.099	26	17	< 1
10	Am	6	3.1	1.61	2	0.430	7	23	< 1
11	*i*-PrAm	6	7.4	1.16	6	0.183	40	7.0	1
12	Im	6	2.5	0.996	26	0.049	51	1.9	1
13	Am	3	19.8	0.327	61	0.084	236	11	2
14	*i*-PrAm	3	82.3	0.483	170	0.202	407	11	7
15	Im	3	10.1	1.08	9	0.095	106	4.7	2
16	Am	4	7.6	1.33	6	0.321	24	5.9	1
17	*i*-PrAm	4	46.5	0.936	50	0.178	261	12	4
18	Im	4	5.8	2.86	2	0.098	59	7.4	< 1
19	Am	5	4.1	0.531	8	0.282	15	5.7	< 1
20	*i*-PrAm	5	20.1	0.911	22	0.294	68	8.9	2
21	Im	5	3.7	0.442	8	0.047	79	17	< 1
22	Am	6	4.3	1.22	4	0.479	9	24	< 1
23	*i*-PrAm	6	10.1	0.421	24	0.347	29	6.2	2
24	Im	6	1.8	0.681	3	0.041	44	44	< 1
PMD[a]			46.6	0.003	15533	0.058	803	1.8	25

[a] PMD, pentamidine. [b] Cytotoxicity (L6). [c] *T. b. rhodesiense* (STIB900). [d] Selectivity index for *T. b. rhodesiense* (SI_T) [IC_{50} (L6)/IC_{50} (*T. b. rhodesiense*)]. [e] *P. falciparum* (K1). [f] Selectivity index for *P. falciparum* (SI_P) [IC_{50} (L6)/IC_{50} (*P. falciparum*)]. [g] *L. donovani* (MHOM/SD/62/1S-CL2$_D$) axenic amastigotes. [h] Selectivity index for *L. donovani* (SI_L) [IC_{50} (L6)/IC_{50} (*L. donovani*)].

butylene linker displayed better antitrypanosomal properties compared to the 6,3′-disubstituted analogues. In the series of dications bearing benzofuran motifs in the 4′-position, 5,4′-disubstituted congeners were more active against the pathogen than the corresponding 6,4′-disubstituted isomers apart from diamidines **25** and **31** and diimidazolines **33** and **36**.

The geometry of the molecules defined by the placement of the benzofuran ring also affected antitrypanosomal activities of dications **1**−**48**. For example, 4′-substituted analogues were more active against *T. b. rhodesiense* than the 3′-substituted congeners in all cases except bis(*N*-isopropyl)amidines **29**, **41**, and **47**, which were less potent than the corresponding isomers **5**, **17**, and **23**.

Dications **1**−**48** were selective for the *Trypanosoma* parasite although none of the tested analogues reached the selectivity of pentamidine.

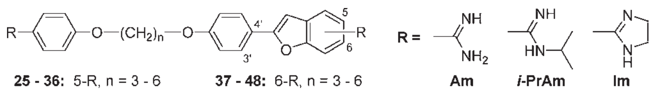
Congeners possessing the propylene linker, except for bis(*N*-isopropyl)amidine **2** and diimidazoline **3**, exhibited higher antitrypanosomal selectivity than those connected by the 4- to 6-carbon chains. 5,4′-Disubstituted bis(*N*-isopropyl)amidine **26**, showing antitrypanosomal IC_{50} value of 41 nM, was the most selective compound in the group for *T. b. rhodesiense* (SI_T = 1112).

Compounds **26**−**28**, **37**, and **39** have been evaluated in vivo in the STIB900 acute mouse model of trypanosomiasis. Administered for 4 days at 20 mg/kg daily, all of the tested dications increased survival time compared to the untreated controls, although none provided cures to the infected mice (data not shown).

Antiplasmodial Activity. Dications **1**−**48** displayed activities against *P. falciparum* with IC_{50} values ranging from 28 to 479 nM. Thirteen compounds (**1**, **12**, **21**, **24**−**27**, **37**−**39**, **42**, **45**, and **48**) exhibited antiplasmodial properties superior to those of pentamidine (IC_{50} = 58 nM). This group of congeners included three diamidines (**1**, **25**, and **37**), two bis(*N*-isopropyl)amidines (**26** and **38**), and eight diimidazolines (**12**, **21**, **24**, **27**, **39**, **42**, **45**, and **48**). Five compounds (**1**, **12**, **25**−**27**) had one cationic group attached to the 5-position of the benzofuran motif, and eight were 6-substituted isomers (**21**, **24**, **37**−**39**, **42**, **45**, and **48**). Nine dications (**12**, **25**−**27**, **37**−**39**, **42**, **45**, and **48**) possessed benzofuran fragment connected to the 4′-position of the phenyl ring and four (**1**, **12**, **21**, and **24**) had it attached to the 3′-position. Seven of the 13 analogues (**1**, **25**−**27**, **37**−**39**), including all diamidines and bis(*N*-isopropyl)amidines, possessed the propylene linker. 6,4′-Disubstituted diimidazoline **45** connected by the 5-carbon chain was the most potent compound in the series showing the antiplasmodial IC_{50} value of 28 nM.

The type and the placement of cationic groups, the position of the benzofuran ring, and the length of the carbon linkers influenced antiplasmodial properties of congeners **1**−**48**. Diamidines joined by the propylene linkers exhibited higher activities versus *P. falciparum* than bis(*N*-isopropyl)amidines and diimidazolines. However, among the congeners

Table 2. Cytotoxicity and in Vitro Antiprotozoal Activity of Congeners **25**−**48**



				T. b. rhodesiense[c]		*P. falciparum*[e]		*L. donovani*[g]	
compd	R	*n*	cytotoxicity[b] IC_{50} (μM)	IC_{50} (μM)	SI_T[d]	IC_{50} (μM)	SI_P[f]	IC_{50} (μM)	SI_L[h]
25	Am	3	7.3	0.451	16	0.041	178	0.78	9
26	*i*-PrAm	3	45.6	0.041	1112	0.042	1086	1.6	29
27	Im	3	10.3	0.061	169	0.043	240	NT[i]	NT[i]
28	Am	4	1.3	0.164	8	0.141	9	0.61	2
29	*i*-PrAm	4	6.9	1.11	6	0.302	23	3.0	2
30	Im	4	2.9	0.374	8	0.064	45	9.2	< 1
31	Am	5	4.2	0.369	11	0.313	13	5.1	< 1
32	*i*-PrAm	5	20.5	0.302	68	0.193	106	2.1	10
33	Im	5	0.9	0.338	3	0.060	15	39	< 1
34	Am	6	3.3	0.368	9	0.263	13	0.65	5
35	*i*-PrAm	6	0.8	0.211	4	0.063	13	5.4	< 1
36	Im	6	1.5	0.360	4	0.116	13	3.3	< 1
37	Am	3	8.6	0.025	344	0.044	195	0.78	11
38	*i*-PrAm	3	49.9	0.088	567	0.037	1349	3.2	16
39	Im	3	16.8	0.144	117	0.042	400	0.48	35
40	Am	4	14.1	1.06	13	0.166	85	3.2	4
41	*i*-PrAm	4	15.9	1.74	9	0.139	114	4.5	4
42	Im	4	16.9	0.942	18	0.046	367	13	1
43	Am	5	9.0	0.313	29	0.268	34	2.9	3
44	*i*-PrAm	5	10.7	0.557	19	0.074	145	1.4	8
45	Im	5	3.2	0.262	12	0.028	114	10	< 1
46	Am	6	8.0	0.416	19	0.280	29	NT[i]	NT[i]
47	*i*-PrAm	6	4.9	0.884	6	0.052	94	2.6	2
48	Im	6	3.4	0.224	15	0.032	106	15	< 1
PMD[a]			46.6	0.003	15533	0.058	803	1.8	25

[a] PMD, pentamidine. [b] Cytotoxicity (L6). [c] *T. b. rhodesiense* (STIB900). [d] Selectivity index for *T. b. rhodesiense* (SI_T) [IC_{50} (L6)/IC_{50} (*T. b. rhodesiense*)]. [e] *P. falciparum* (K1). [f] Selectivity index for *P. falciparum* (SI_P) [IC_{50} (L6)/IC_{50} (*P. falciparum*)]. [g] *L. donovani* (MHOM/SD/62/1S-$CL2_D$) axenic amastigotes. [h] Selectivity index for *L. donovani* (SI_L) [IC_{50} (L6)/IC_{50} (*L. donovani*)]. [i] NT, not tested.

with elongated carbon chains, diimidazolines displayed greater antiplasmodial potency.

5,3′-Disubstituted diamidines and bis(*N*-isopropyl)amidines were more active than the corresponding 6,3′-disubstituted isomers except for congeners **5** and **7**, while diimidazolines **15**, **18**, **21**, bearing cationic groups in the 6,3′-positions exhibited superior antiplasmodial properties compared to the 5,3′-disubstituted analogues **3**, **6**, **9**, and **12**. Among the dications possessing the benzofuran motif in the 4′-position, 5,4′-disubstituted diamidines were more potent against the pathogen than the corresponding 6,4′-disubstituted isomers except diamidine **31**. At the same time, 6,4′-disubstituted bis(*N*-isopropyl)amidines and diimidazolines were more active versus *P. falciparum* than the comparable 5,4′-disubstituted isomers.

All 6,4′-disubstituted analogues **37**−**48** displayed higher antimalarial activities than the corresponding 6,3′-disubstituted congeners **13**−**24** while no correlation between the position of the benzofuran ring and antiplasmodial properties of 5-substituted analogues was observed. All dications **1**−**48** were selective for *P. falciparum* and displayed selectivity indexes ranging from 7 to nearly 1350. Regardless of the position of the cationic substituents and the length of the carbon chains between the aromatic moieties, bis(*N*-isopropyl)amidines and diimidazolines were more selective for the parasite compared to diamidines.

Elongation of the alkyl chains usually reduced the antiplasmodial selectivity of the congeners **1**−**48**. Thus, bis(*N*-isopropyl)amidines **26** and **38**, both connected by the propylene linker, were more selective for *P. falciparum* than pentamidine.

Antileishmanial Activity. All tested compounds were active in the *L. donovani* axenic amastigote assay demonstrating activities ranging from 0.48 to 44 μM. Four diamidines (**25**, **28**, **34**, and **37**), three bis(*N*-isopropyl)amidines (**26**, **32**, and **44**), and two diimidazolines (**12** and **39**) exhibited antileishmanial IC_{50} values comparable to or lower than that of pentamidine (IC_{50} = 1.8 μM). Out of nine dications, only diimidazoline **12** had the benzofuran motif in the 3′-position. Among the eight congeners with the benzofuran fragment connected to the 4′-position of the phenyl ring, five compounds (**25**, **26**, **28**, **32**, and **34**) contained their substituents in the 5,4′-positions and three (**37**, **39**, and **44**) were 6,4′-disubstituted isomers. Diamidine **28** and diimidazoline **39** were the most potent compounds versus *L. donovani* among 5- and 6-substituted congeners, respectively.

There was no obvious correlation between the position of attachment of the cationic moieties to the benzofuran fragment and the antileishmanial properties of 5,4′- and 6,4′-disubstituted congeners. However, in the series of dications bearing the benzofuran motif in the 3′-position, 5,3′-disubstituted diamidines and bis(*N*-isopropyl)amidines were more active against *L. donovani* than 6,3′-disubstituted analogues apart from congeners **7** and **11**. Concurrently, 6,3′-disubstituted diimidazolines exhibited higher antileishmanial potency than their 5,3′-disubstituted counterparts, except for congener **24**. The placement of the benzofuran motif influenced the antileishmanial properties of dications **1−48**. Thus, regardless of the position of the cationic moieties, 4′-substituted congeners displayed higher activities versus *L. donovani* than their analogues with the benzofuran fragment connected to the 3′-position of the phenyl ring in all cases except for diimidazolines **33**, **36**, and **42**.

The antileishmanial selectivity of dications **1−48** depended on the substitution on the amidine groups, the placement of the benzofuran fragment, and the length of the carbon linkers in the molecules. Bis(*N*-isopropyl)amidines were more selective for *L. donovani* than diimidazolines and diamidines. Compounds bearing the benzofuran motif connected to the 4′-position generally exhibited higher selectivity for the pathogen compared to the 3′-substituted isomers. Elongation of the carbon linkers usually reduced the antileishmanial selectivity of congeners **1−48**. Diimidazoline **39** was not only the most potent compound in the series exhibiting antileishmanial IC_{50} value of 0.48 μM but also demonstrated the highest selectivity for *L. donovani* (SI_L = 35), which was greater than that of pentamidine.

Conclusions

A series of pentamidine congeners **1−48** containing benzofuran fragments have been synthesized and tested in vitro against *T. b. rhodesiense*, *P. falciparum*, and *L. donovani*, and for cytotoxicity versus mammalian cells. The antiprotozoal properties and cytotoxicities of dications **1−48** depended on the nature of the cationic substituents, the placement of the benzofuran motif, and the length of the carbon linker. The cytotoxicities of tested compounds decreased with the substitution on cationic groups and increased with the elongation of the carbon linker. Dications with the benzofuran motif in the 4′-position and connected by the propylene linker demonstrated higher potency and were more selective against *T. b. rhodesiense*, *P. falciparum* and *L. donovani*. Improved antileishmanial activities of select congeners compared to pentamidine warrant further investigation of these compounds as potential leads for antileishmanial chemotherapy.

Experimental Section

Full experimental details and characterization data are given in the Supporting Information. All tested compounds are >95% pure by elemental analysis and HPLC. Proton NMR spectra were recorded on a Varian Gemini 2000 spectrometer (300 MHz) with tetramethylsilane as an internal standard.

General Procedure for Syntheses of Cationic Benzofurans 1−48. 2-(3-(3-(4-Carbamimidoylphenoxy)propoxy)phenyl)benzofuran-5-carboxamidine Dihydrochloride (1). A mixture of anhydrous ethanol (10 mL) and 1,4-dioxane (30 mL) was saturated with gaseous HCl at 0−5 °C. 2-(3-(3-(4-Cyanophenoxy)propoxy)phenyl)benzofuran-5-carbonitrile (**67**) (1.00 g, 2.54 mmol) was added and the sealed mixture was stirred at room temperature. The reaction mixture was diluted with dry ether. A precipitate was separated under Ar and dried under high vacuum over KOH to give an imidate ester as a white solid (1.42 g, 100%).

The imidate (0.51 g, 0.90 mmol) was suspended in dry EtOH saturated with gaseous ammonia (11 mL) and was stirred at room temperature. The reaction mixture was diluted with diethyl ether (50 mL). A precipitate was separated, washed with ether, dried and recrystallized from 1 M HCl/EtOH to give **1** as a white solid (0.230 g, 51%): mp 175−178 °C. ^{1}H NMR (DMSO-d_6) δ 9.41 (br s, 2H), 9.22 (br s, 2H), 9.12 (br s, 2H), 8.94 (br s, 2H), 8.19 (d, J = 1.8 Hz, 1H), 7.90 (d, J = 8.6 Hz, 1H), 7.85 (d, J = 8.8 Hz, 2H), 7.78 (dd, J = 8.6 and 1.8 Hz, 1H), 7.70 (s, 1H), 7.57 (d, J = 7.8 Hz, 1H), 7.55 (m, 1H), 7.43 (dd, J = 7.8 and 7.8 Hz, 1H), 7.21 (d, J = 8.8 Hz, 2H), 7.07 (dd, J = 7.8 and 1.8 Hz, 1H), 4.36−4.16 (m, 4H), 2.30−2.18 (m, 2H). HPLC (method A) t_R = 7.91 min (100 area %). Anal. ($C_{25}H_{24}N_4O_3\cdot 2HCl\cdot 1.4H_2O$) C, H, N, Cl.

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Supporting Information Available: Experimental details and characterization data. This material is available free of charge via the Internet at http://pubs.acs.org.

References

(1) Barrett, M. P.; Boykin, D. W.; Brun, R.; Tidwell, R. R. Human African trypanosomiasis: pharmacological re-engagement with a neglected disease. *Br. J. Pharmacol.* **2007**, *152*, 1155–1171.

(2) Burri, C.; Brun, R. Human African Trypanosomiasis. In *Manson's Tropical Diseases*; Cook, G. C., Zumla, A., Eds.; W. B. Saunders: London, 2008; pp 1307−1325.

(3) Croft, S. L.; Yardley, V. Chemotherapy of leishmaniasis. *Curr. Pharm. Des* **2002**, *8*, 319–342.

(4) Goa, K. L.; Campoli-Richards, D. M. Pentamidine isethionate. A review of its antiprotozoal activity, pharmacokinetic properties and therapeutic use in *Pneumocystis carinii* pneumonia. *Drugs* **1987**, *33*, 242–258.

(5) Stead, A. M.; Bray, P. G.; Edwards, I. G.; DeKoning, H. P.; Elford, B. C.; Stocks, P. A.; Ward, S. A. Diamidine compounds: selective uptake and targeting in *Plasmodium falciparum*. *Mol. Pharmacol.* **2001**, *59*, 1298–1306.

(6) Bray, P. G.; Barrett, M. P.; Ward, S. A.; de Koning, H. P. Pentamidine uptake and resistance in pathogenic protozoa: past, present and future. *Trends Parasitol.* **2003**, *19*, 232–239.

(7) Fairlamb, A. H. Chemotherapy of human African trypanosomiasis: current and future prospects. *Trends Parasitol.* **2003**, *19*, 488–494.

(8) Bakunova, S. M.; Bakunov, S. A.; Wenzler, T.; Barszcz, T.; Werbovetz, K. A.; Brun, R.; Hall, J. E.; Tidwell, R. R. Synthesis and in vitro antiprotozoal activity of bisbenzofuran cations. *J. Med. Chem.* **2007**, *50*, 5807–5823.

(9) Bakunov, S. A.; Bakunova, S. M.; Wenzler, T.; Barszcz, T.; Werbovetz, K. A.; Brun, R.; Tidwell, R. R. Synthesis and antiprotozoal activity of cationic 2-phenylbenzofurans. *J. Med. Chem.* **2008**, *51*, 6927–6944.

(10) Dulog, L.; Korner, B.; Heinze, J.; Yang, J. Synthesis and electrochemical properties of 4-phenyl-1-buten-3-yne-1,1,2-tricarbonitriles and tricyanoacrylates. *Liebigs Ann. Chem* **1995**, 1663–1671.

(11) Doad, G. J. S.; Barltrop, J. A.; Petty, C. M.; Owen, T. C. A versatile and convenient synthesis of benzofurans. *Tetrahedron Lett.* **1989**, *30*, 1597–1598.

(12) Wood, J. L.; Khatri, N. A.; Weinreb, S. M. A direct conversion of esters to nitriles. *Tetrahedron Lett.* **1979**, *20*, 4907–4910.

(13) Berg, S. S.; Newbery, G. The search for chemotherapeutic amidines. Part X. Substituted 4,4'-diamidino-αω-diphenoxyalkanes and -diphenyl ethers. *J. Chem. Soc.* **1949**, 642–648.

(14) Dox, A. W.; Whitmore, F. C. Acetamidine hydrochloride. In *Organic Syntheses*, 2nd ed.; Blatt, A. H., Ed. John Wiley and Sons: New York, 1941; Vol. 1, pp 5−7.