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# Synthesis and Antiprotozoal Activity of Naphthofuranquinones and Naphthothiophenequinones Containing a Fused Thiazole Ring

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**Abstract**—The synthesis of tetracyclic quinones **10a**,**b**, **14a**,**b**, **19a**,**b** and **20a**,**b** is described. The preparations involve regioselective Diels–Alder reactions via trapping the thiazole *o*-quinodimethane **9** with several benzofuranquinones and benzothiophenequinones. The structure of the regioisomers was assigned through 2D NMR $^{1}H$–$^{13}C$ HMBC experiments performed on **10a** and **14a**. Compounds **10a**,**b**, **14a** as well as phenol **1** and the starting quinones **2**, **5**, **7** and **15** are evaluated against *Leishmania* sp., *Toxoplasma gondii* and THP-1 cells. Almost all the tested compounds exhibit significant antiprotozoal activities with lower cytotoxicities than the reference compounds. Among them, quinones **2** and **14a** possess the best activities towards *L. donovani* and *T. gondii* with the lowest toxicities.


## Introduction

Toxoplasmosis, a common zoonosis asymptomatic in many adults, is due to the protozoan *Toxoplasma gondii*. The latter can develop brain cysts which are activated in immunodeficient patients and may lead to an encephalic toxoplasmosis.[1] On the other hand, leishmaniasis is a group of tropical diseases which are caused by protozoa belonging to *Leishmania* species. Among them, *Leishmania donovani* is responsible for visceral diseases which are amongst the most severe clinical lesions. Frequent resistance to the initial treatment with antimonial derivatives led to the use of amphotericin B and pentamidine. But, administration of such compounds was accompanied with an unacceptable toxicity at effective therapeutic doses. Likewise, toxoplasmosis and leishmaniasis are also opportunistic diseases in AIDS. At the present time there is still a need for safe and effective drugs against these diseases.

Natural or synthetic naphthoquinones have been described for many years for their antiprotozoal activity.[2–5] In addition, some anthraquinones and pyranonaphthoquinones[6] were found active in vitro against virulent strains of *T. gondii* and *L. donovani* while the trypanocidal or leishmanicidal activities of some naphthofuranquinones or naphthothiophenequinones of types **I**, **II** and **III** were reported for a few years (Scheme 1).[7,8] More recently, thienoquinolinedione and furoquinolinedione derivatives **IV** and **V** were also found active in vitro against virulent strains of *Leishmania* sp. and *T. gondii*, respectively.[9,10] On the other hand, some benzimidazole-4,5-diones were described as inhibitors of purine nucleoside phosphorylase of *T. gondii*.[11] Among the tested quinones, that substituted by a thiazole ring **VI** was found a better inhibitor than the reference compound 8-aminoguanosine.

These observations prompted us for investigations with the aim to synthesize new tetracyclic quinones containing the biologically active naphthofuran and naphthothiophene quinonic framework fused to a thiazole ring. Recently, we reported an efficient and mild method to

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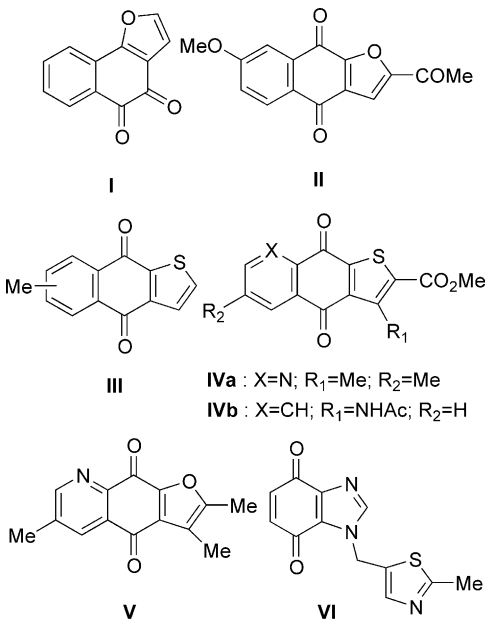



Scheme 1.

obtain anthra[2,3-*b*]thiazole-5,10-diones through a Diels–Alder trapping of thiazole *o*-quinodimethanes (*o*-QDMs) with naphthoquinones.[12,13] Applying this methodology to benzofuranquinone and benzothiophenequinone derivatives may constitute a direct access to the target compounds.

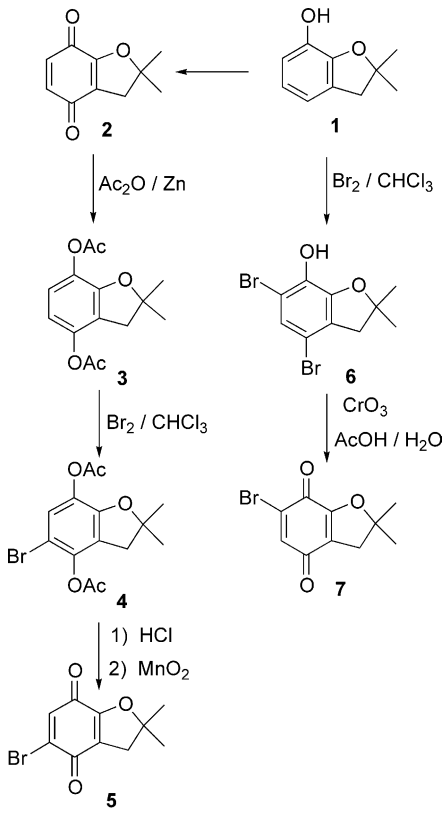
## Results and Discussion

### Chemistry

Since it is well established that the regiochemistry of Diels–Alder reactions between *o*-QDM **9** and brominated naphthoquinones is under control of the bromine atom position on the dienophilic double bond,[12] we decided to prepare the unknown 5- and 6-bromoquinones **5** and **7** (Scheme 2).

2,3-Dihydrobenzo[*b*]furan-4,7-dione **2** was previously obtained in 70% yield through oxidation of benzofuranol **1** with Frémy's salt.[14] But, carrying out the reaction in a one-gram scale improved the yield to 95%. Then, a reductive acetylation of quinone **2** into **3** followed by a selective bromination of **3** gave the 5-bromo derivative **4**. Its acid hydrolysis and oxidation with manganese dioxide provided the brominated quinone **5** in 42% overall yield. On the other hand, treatment of phenol **1** with two equivalents of bromine in chloroform at 0 °C afforded the dibromo compound **6** which was oxidized into 6-bromoquinone **7** with chromium(VI) oxide in 51% overall yield from **1**.

Then, treatment of 4-(bromomethyl)-5-(dibromomethyl)thiazole **8**[12] with sodium iodide in dimethylformamide afforded 4-methylene-5-(bromomethylene)-4,5-dihydrothiazole **9** which was reacted in situ with the dihydrobenzofuran-4,7-diones **2**, **5** or **7** (Scheme 3). Starting with **2** the reaction provided a mixture of the

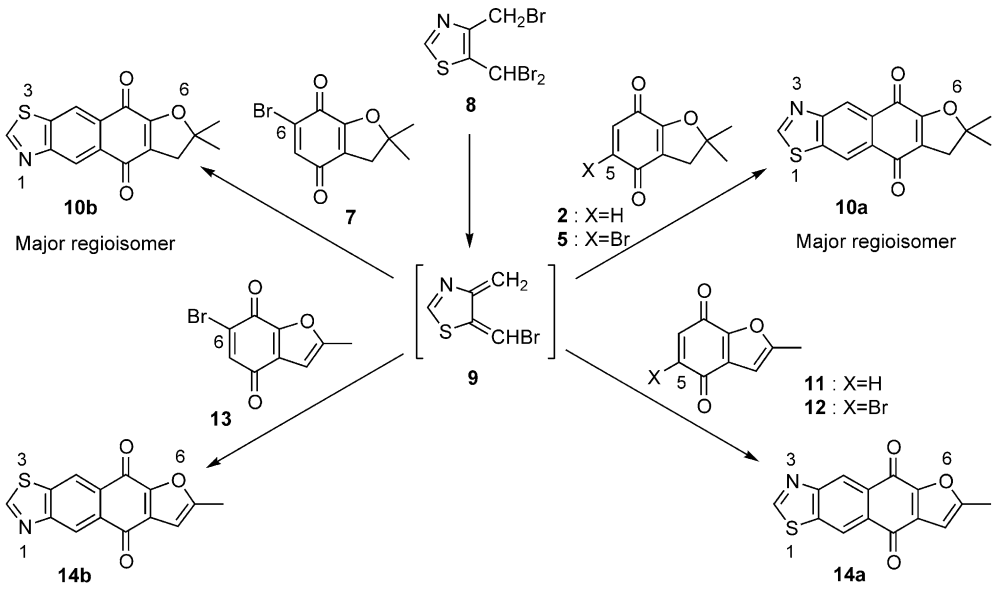



Scheme 2.

tetracyclic quinone **10a** + **10b** where **10a** is the major regioisomer (Table 1, entry 1). The use of bromoquinones **5** or **7** afforded **10a** or the mixture of regioisomers **10a** + **10b**, respectively with a reversed regiochemistry (Table 1, entries 2 and 3).

The thiazole *o*-QDM strategy was then applied to the previously described[15] benzo[*b*]furan-4,7-dione **11** and its 5- or 6-bromo derivatives **12** or **13** (Scheme 3). Thus, trapping **9** with quinone **11** led to a mixture of tetracyclic quinones **14a** + **14b** with a weak regioselectivity (Table 1, entry 4). Better results were obtained by the use of 5- and 6-bromoquinones **12** and **13**. Indeed, their reactions with **9** afforded **14a** or **14b**, respectively as a single regioisomer (Table 1, entries 5 and 6). Therefore, starting with bromoquinones **12** and **13** as dienophiles, the Diels–Alder reactions were regiospecific.

In order to study the cycloaddition reactions of benzo[*b*]thiophenedione derivatives, we selected the biologically active quinone **15**[9a] and benzo[*b*]thiophenequinone carbaldehyde **18** (Scheme 4). Quinone **15** was prepared by an oxidative demethylation of methyl 4,7-dimethoxybenzo[*b*]thiophen-2-carboxylate following our reported procedure.[16] The new quinone **18** was obtained as described in Scheme 4. Thus, oxidation of the benzo[*b*]thiophene methanol derivative **16**[16] with pyridinium chlorochromate (PCC) afforded the aldehyde **17**. Subsequent oxidative demethylation of the latter with cerium(IV) ammonium nitrate (CAN) provided quinone **18** in 65% overall yield. Attempts to prepare bromo derivatives of these quinones through selective bromination of their dimethoxy precursors was reported unsuccessful.[17]




**Scheme 3.**

**Table 1.** Trapping of *o*-quinodimethane **9** with heterocyclic quinones

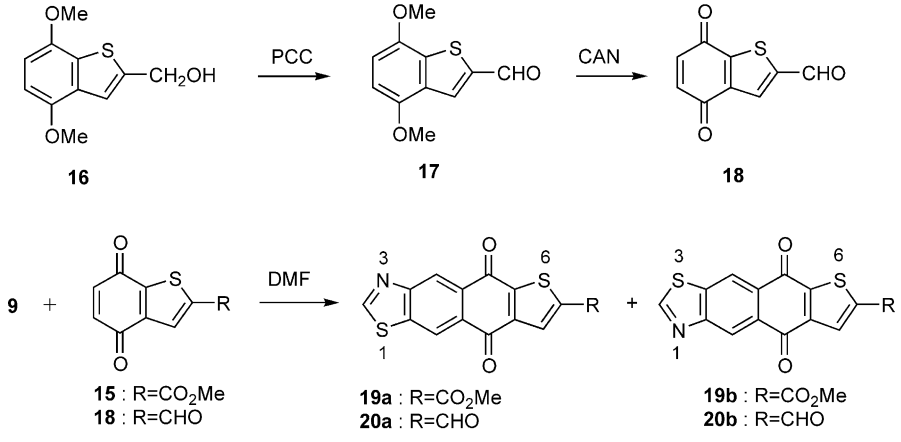
| Entry | Quinone | Product[a] | Yield (%) | Ratio **a/b** |
|---|---|---|---|---|
| 1 | **2** | **10a** + **10b** | 47 | 88/12 |
| 2 | **5** | **10a** | 45 | — |
| 3 | **7** | **10a** + **10b** | 47 | 14/86 |
| 4 | **11** | **14a** + **14b** | 40 | 55/45 |
| 5 | **12** | **14a** | 45 | — |
| 6 | **13** | **14b** | 40 | — |
| 7 | **15** | **19a** + **19b** | 42 | 70/30 |
| 8 | **18** | **20a** + **20b** | 35 | 55/45 |

[a]Attempts to separate the regioisomers from **19a** + **19b** and **20a** + **20b** were unsuccessful.

The trapping of *o*-QDM **9** with quinones **15** and **18** afforded unseparable mixtures of the regioisomers **19a** + **19b** and **20a** + **20b**, respectively (Table 1, entries 7 and 8).

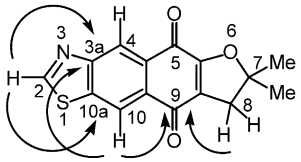
The regiochemical assignment for compounds **10** and **14** was confirmed by NMR studies. Thus, 2D NMR $^{1}H$–$^{13}C$ HMBC experiments were performed on both **10a** and **14a** (Tables 2 and 3). We used the characteristic long range $^{3}J$ coupling of the benzothiazole nucleus.[18] Thus, the H-2 proton has two $^{3}J$ couplings with C-3a and C-10a. But, the former appears as a doublet typical of a cross peak through the nitrogen atom of the thiazole ring ($^{3}JC_{3a\text{-}H2} = 15$ Hz) while the latter gives an unresolved singlet indicating a coupling of $< 5$ Hz for $^{3}JC_{10a\text{-}H2}$. This marked C-3a and C-10a. Then, the three $^{3}J$ couplings, H-10 with C-3a and C-9, H-8 with C-9 led us to assign the structure for **10a** and **14a**. Therefore, their regioisomers **10b** and **14b** would have the opposite regiochemistry.

The results shown in Scheme 3 indicate that the regiochemistry of the cycloadditions is under control of the bromine atom position on both the diene and dienophiles. Thus, the reaction of *o*-QDM **9** with the 5-brominated quinones **5** and **12** led to the same type of regioisomer (**10a** and **14a** as a single regioisomer). On the other hand, **9** reacted with the 6-bromoquinones **7** and **13** to afford the opposite regioisomers **10b** and **14b** (**10b** as the major product and **14b** as a single one),




**Scheme 4.**

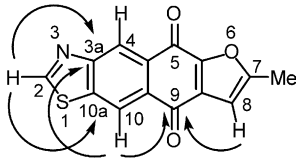
**Table 2.** 2D $^{1}H$–$^{13}C$ HMBC correlations for **10a** ($CDCl_3$, 500.13 and 125.78 MHz, δ ppm)



**10a**

| Atom | $^{13}C$ | $^{1}H$ | HMBC [J(C,H)] | | | | |
|---|---|---|---|---|---|---|---|
| | | | $^{1}J$ | $^{2}J$ | $^{3}J$ | $^{4}J$ | $^{5}J$ |
| 2 | 159.8 | 9.24 | 2-H | | | | |
| 3a | 156.0 | | | 4-H | 2-H, 10-H | | |
| 4 | 122.7 | 8.77 | 4-H | | | 10-H | |
| 4a | 130.5 | | | | 10-H | | 2-H |
| 5 | 178.1 | | | | 4-H | 8-H, 10-H | |
| 5a | 160.2 | | | | 8-H | | |
| 7 | 92.8 | | | 8-H | | | |
| 7-$CH_3$ | 28.8 | 1.62 | | | 8-H | | |
| 8 | 40.6 | 3.06 | 8-H | | | | |
| 8a | 124.5 | | | 8-H | | | |
| 9 | 182.0 | | | | 10-H, 8-H | 4-H | |
| 9a | 130.4 | | | | 4-H | | |
| 10 | 121.3 | 8.71 | 10-H | | | 2-H, 4-H | |
| 10a | 139.8 | | | | 2-H, 4-H | | |

**Table 3.** 2D $^{1}H$–$^{13}C$ HMBC correlations for **14a** ($CDCl_3$, 500.13 and 125.78 MHz, δ ppm)



**14a**

| Atom | $^{13}C$ | $^{1}H$ | HMBC [J(C,H)] | | | | |
|---|---|---|---|---|---|---|---|
| | | | $^{1}J$ | $^{2}J$ | $^{3}J$ | $^{4}J$ | $^{5}J$ |
| 2 | 161.2 | 9.25 | | | | | |
| 3a | 156.7 | | | | 2-H, 10-H | | |
| 4 | 123.2 | 8.96 | 4-H | | | 10-H | |
| 4a | 131.8 | | | | 10-H | | 2-H |
| 5 | 172.8 | | | | 4-H | | |
| 5a | 152.7 | | | | 8-H | 7-$CH_3$ | |
| 7 | 161.2 | | | 8-H, 7-$CH_3$ | | | |
| 7-$CH_3$ | 14.5 | 2.56 | 7-$CH_3$ | | 8-H | | |
| 8 | 105.5 | 6.67 | 8-H | | | | |
| 8a | 132.8 | | | 8-H | 7-$CH_3$ | | |
| 9 | 180.3 | | | | 10-H | 4-H | |
| 9a | 130.6 | | | | 4-H | | |
| 10 | 122.4 | 8.82 | 10-H | | | | |
| 10a | 139.3 | | | | 2-H, 4-H | | |

respectively. These observations are in good agreement with our previous results where the brominated carbon atom of *o*-QDM **9** attacks the quinonic dienophile at the carbon bearing the halogen atom.[12,19]

Using quinones **15** and **18**, we assume that the regiochemistry of their reactions with **9** could be that predicted by the frontier molecular orbital (FMO) theory.

**Table 4.** In vitro inhibitory activity against *Leishmania* sp., *Toxoplasma gondii* and cytotoxicity against THP-1 cells of the synthetized compounds. The values are the means ± SD of triplicate experiments

| Compounds | *Leishmania donovani* $IC_{50}$ (μM) | *Leishmania major* $IC_{50}$ (μM) | *Toxoplasma gondii* $IC_{50}$ (μM) | THP-1 cells $IC_{50}$ (μM) |
|---|---|---|---|---|
| **1** | 0.006 | 0.005 | 0.0041 | 0.010 |
| **2** | 0.010 | 0.012 | 0.0007 | 0.123 |
| **5** | 0.010 | 0.013 | 0.0032 | 0.053 |
| **7** | 0.006 | 0.198 | 0.0010 | 0.085 |
| **10a** | 0.014 | 0.011 | 0.0009 | 0.007 |
| **10b** | 0.008 | 0.010 | 0.0041 | 0.007 |
| **14a** | 0.009 | 2.384 | 0.0007 | 1.466 |
| **15** | 0.006 | 0.052 | 0.0010 | 0.052 |
| Pentamidine | 0.0012 | 0.0018 | | 0.0043 |
| Pyrimethamine | | | 0.0034 | 0.010 |
| Spiramycin | | | 0.0042 | 0.013 |
| Sulfadiazine | | | 0.0034 | 0.012 |

Indeed, calculations of the HOMO and LUMO orbital coefficients by the semi-empirical PM3 method show that for *o*-QDM **9**, the largest HOMO coefficient is located at the carbon bearing the bromine atom (0.441 for CHBr and 0.354 for $CH_2$) while for quinones **15** and **18**, the largest LUMO ones are always situated at 5-C (**15**: 5-C = 0.237 and 6-C = 0.215; **18**: 5-C = 0.242 and 6-C = 0.221). Thus, following FMO calculations, the Diels–Alder trapping of **9** with these quinones would provide **19a** and **20a** as the major regioisomers.

## Biology

The in vitro antileishmanial activity of compounds **1**, **2**, **5**, **7**, **10a**, **10b**, **14a** and **15** was evaluated against promastigote forms of *L. donovani* and *Leishmania major*. The potential toxicity of these derivatives was also determined against a THP-1 cell line. Pentamidine was used as the reference drug (Table 4). Except **7** and **14a** which are more active against *L. donovani* than *L. major*, the tested compounds exhibit significant activities against both *Leishmania* sp. It is noteworthy that 7-hydroxy-2,3-dihydro-2,2-dimethylbenzofuran **1** and the tetracyclic dihydrobenzofurandiones **10** were found the most active compounds towards both *Leishmania* sp. Most of these compounds are less cytotoxic than the reference pentamidine. Among them, quinones **2** and **14a** possess the lowest cytotoxicity against THP-1 cells. The fact that **14a** shows an activity similar to that of pentamidine but, with a much lower toxicity indicates a selectivity of the action against the *Leishmania* sp.

In vitro assays against *T. gondii* were performed using an infected human myelomonocytic cell line THP-1 and well-known effective drugs as positive controls (Table 4). Almost all the tested compounds possess a better potency against *T. gondii* than the reference compounds. They are also less cytotoxic, particularly **2** and **14a** which are the more active.

Unfortunately, the very low solubility of compounds **19** and **20** in usual solvents was a limitation for a valuable estimation of their biological activity.

## Conclusion

This work describes an efficient way to reach unusual bis-heterocyclic tetracyclic quinones through the trapping of the thiazole *o*-QDM **9** with benzo[*b*]furan-4,7-diones or benzo[*b*]thiophene-4,7-diones. Starting with 5-bromoquinones **5** and **12** and the 6-bromo derivatives **7** and **13**, we obtain regioselectively or regiospecifically the tetracyclic compounds **10a**, **14a**, on one hand, and **10b**, **14b**, on the other hand. The regiochemistry observed is under control of the position of bromine atom on both *o*-QDM **9** and these quinones. Several of the synthesized compounds are evaluated for their antiprotozoal properties against *Leishmania* sp. and *T. gondii*. Almost all of them show significant inhibitory activities towards *L. donovani* and *T. gondii* with lower cytotoxicities against TPH-1 cells than the reference drugs. The best results are observed with quinones **2** and **14a**.

## Experimental

### Chemical synthesis

Melting points were measured with a Stuart Scientific SMP3 apparatus and are uncorrected. IR spectra were recorded on a Bruker Vector 22 spectrophotometer using KBr discs. $^1$H and $^{13}$C NMR spectra were obtained on Bruker AM-200, AM-300 and DRX-500 spectrometers using tetramethylsilane as an internal reference. Column chromatography was performed on silica gel Merck 60 (70–230 mesh). Thin layer chromatography separations were performed on Merck Kieselgel 60 (70–230 mesh). Elemental analyses were carried out on a FISONS EA 1108 CHNS-O analyzer.

4-(Bromomethyl)-5-(dibromomethyl)thiazole **8**, the precursor of *o*-QDM **9** was prepared by selective bromination of the commercially available 4,5-dimethylthiazole as previously reported.[12] Benzofurandiones **11**, **12** and **13** were obtained according to the literature.[15] Benzothiophenequinone **15** was prepared following our previously reported procedure.[16]

**2,3-Dihydro-2,2-dimethylbenzofuran-4,7-dione (2).** A solution of potassium nitrosodisulfonate (2.04 g, 7.6 mmol) in water (105 mL) was added dropwise to a stirred solution of phenol **1** (500 mg, 3.05 mmol) in ethanol (30 mL) at room temperature. After 1 h the solution was extracted with dichloromethane (4×50 mL). The combined extracts were dried over $MgSO_4$ and evaporated under reduced pressure to give an orange solid which was purified by column chromatography on silica gel using dichloromethane as the eluent to afford quinone **2** (0.515 g, 95%), mp 133–134 °C (dichloromethane–petroleum ether, lit.[14] 133–134 °C).

**2,3-Dihydro-2,2-dimethyl-4,7-diacetoxybenzofuran (3).** A mixture of quinone **2** (0.48 g, 2.7 mmol), zinc powder (2.64 g, 40.5 mmol), sodium acetate (0.86 g, 10.5 mmol) and acetic anhydride (61 mL) is refluxed for 1 h 30 min. The mixture was filtered and the solution evaporated under vacuum. Then, cold water was added to the residue cooled at 0 °C. The precipitate formed was separated by filtration and washed with water and then, with ether. Compound **3** was obtained as a white solid (0.34 g, 48%), mp 143 °C. IR (KBr) ν cm$^{-1}$ 1760 (CO). $^1$H NMR (200 MHz, $CDCl_3$) δ: 1.48 (s, 6H, 2 $CH_3$), 2.27 (s, 3H, $COCH_3$), 2.30 (s, 3H, $COCH_3$), 2.93 (s, 2H, $CH_2$), 6.54 (d, 1H, $J$=8.8 Hz, 5-H), 6.87 (d, 1H, $J$=8.8 Hz, 6-H); $^{13}$C NMR (50 MHz, $CDCl_3$) δ: 20.7, 20.8, 28.1, 41.3, 89.4, 113.0, 121.8, 122.1, 132.2, 144.9, 151.1, 168.3, 168.5. Anal. calcd for $C_{14}H_{16}O_5$: C, 63.63, H, 6.10. Found: C, 63.68, H, 6.06.

**5-Bromo-2,3-dihydro-2,2-dimethyl-4,7-diacetoxybenzofuran (4).** A solution of bromine (60 mg, 0.38 mmol) in chloroform (2 mL) was added dropwise to a stirred solution of the 2,3-dihydro-2,2-dimethyl-4,7-diacetoxybenzofuran **3**[5] (100 mg, 0.38 mmol) in chloroform (10 mL) at 0 °C. The mixture was stirred for 2 h and then was treated with a 10% aqueous sodium thiosulfate (10 mL). The organic phase was washed with brine and dried over $MgSO_4$. Evaporation of the solvent gave a white solid, which was purified by column chromatography on silica gel using dichloromethane as the eluent to afford benzofuran **4** (105 mg, 81%), mp 124–125 °C (methanol). IR (KBr) ν cm$^{-1}$ 1760 (CO). $^1$H NMR (200 MHz, $CDCl_3$) δ: 1.48 (s, 6H, 2 $CH_3$), 2.30 (s, 3H, $COCH_3$), 2.33 (s, 3H, $COCH_3$), 2.95 (s, 2H, $CH_2$), 7.14 (s, 1H, 6-H); $^{13}$C NMR (50 MHz, $CDCl_3$) δ: 20.8, 28.4, 42.1, 91.5, 125.5, 126.8, 133.9, 143.7, 152.1, 153.3, 168.3, 168.8. Anal. calcd for $C_{14}H_{15}BrO_5$: C, 49.00, H, 4.41. Found: C, 49.20, H, 4.30.

**5-Bromo-2,3-dihydro-2,2-dimethylbenzofuran-4,7-dione (5).** A mixture of bromodiacetate **4** (170 mg, 0.50 mmol), methanol (30 mL) and 10% aqueous hydrochloric acid (1 mL) was refluxed for 3 h. The reaction mixture was neutralized by careful addition of a saturated solution of $NaHCO_3$ and extracted with ethyl acetate. The organic phase was dried over $MgSO_4$ and concentrated under vacuum. The residue was dissolved in diethyl ether (50 mL) and after the addition of $MnO_2$, the mixture was stirred at room temperature for 1 h. The suspension was filtered through Celite, and the solvent was removed at reduced pressure. The residue was purified by column chromatography on silica gel using dichloromethane as the eluent to afford quinone **5** (67.2 mg, 53%), mp 101–102 °C (methanol). IR (KBr) ν cm$^{-1}$ 1680 and 1650 (CO), 1625 (C=C). $^1$H NMR (200 MHz; $CDCl_3$) δ: 1.53 (s, 6H, 2 $CH_3$), 2.89 (s, 2H, $CH_2$), 6.76 (s, 1H, 6-H); $^{13}$C NMR (50 MHz, $CDCl_3$) δ: 28.7, 39.9, 119.8, 130.2, 145.9, 157.6, 176.1, 177.8. Anal. calcd for $C_{10}H_9BrO_3$: C, 46.72, H, 3.53. Found: C, 46.80, H, 3.66.

**5,7-Dibromo-2,3-dihydro-2,2-dimethyl-7-hydroxybenzofuran (6).** A solution of bromine (7.6 mmol, 0.39 mL) in chloroform (10 mL) was added dropwise to a stirred solution of benzofuran **1** (500 mg, 3.04 mmol) in chloroform (50 mL) at 0 °C. The mixture was stirred at room temperature for 1.5 h. Evaporation of the solvent gave a brown solid, which was purified by column chromatography on silica gel using dichloromethane as

the eluent to afford dibromobenzofuran **6** (810 mg, 83%), mp 101–102 °C (methanol). IR (KBr) ν $cm^{-1}$ 3390 (OH); $^1H$ NMR (200 MHz, $CDCl_3$) δ: 1.50 (s, 6H, 2 $CH_3$), 3.01 (s, 2H, $CH_2$), 5.06 (br s, 1H, OH), 7.05 (s, 1H, 5-H); $^{13}C$ NMR (53.3 MHz, $CDCl_3$) δ: 28.2, 46.2, 89.3, 111.6, 113.7, 120.0, 130.2, 140.0, 145.1. Anal. calcd for $C_{10}H_{10}Br_2O_2$: C, 37.30, H, 3.13. Found: C, 37.28, H, 2.99.

**6-Bromo-2,3-dihydro-2,2-dimethylbenzofuran-4,7-dione (7).** A solution of chromium(VI) oxide (93 mg, 0.93 mmol) in water (1.0 mL) was added dropwise to a stirred solution of dibromobenzofuran **6** (100 mg, 0.31 mmol) in acetic acid–water (7:2, 4.5 mL). The mixture was stirred at room temperature for 1 h. Then, the reaction mixture was diluted with water (15 mL) and extracted with dichloromethane. The organic phase was washed with $NaHCO_3$ and dried over $MgSO_4$. After evaporation of the solvent, the residue was purified by column chromatography on silica gel using dichloromethane as the eluent to afford quinone **7** (40 mg, 51%), mp 109–110 °C (methanol). IR (KBr) ν $cm^{-1}$ 1675 and 1650 (CO), 1630 (C=C). $^1H$ NMR (200 MHz, $CDCl_3$) δ: 1.48 (s, 6H, 2 $CH_3$), 2.83 (s, 2H, $CH_2$), 6.99 (s, 1H, 5-H); $^{13}C$ NMR (50 MHz, $CDCl_3$) δ: 28.2, 40.1, 93.2, 119.6, 134.5, 139.8, 156.8, 176.4, 177.4. Anal. calcd for $C_{10}H_9BrO_3$: C, 46.72, H, 3.53. Found: C, 46.61, H, 3.42.

**Generation of *o*-QDM 9 and its trapping with benzofurandiones 2, 5, 7, 11, 12 and 13. General procedure.** A solution of 4-(bromomethyl)-5-(dibromomethyl)thiazole **8**[12] (0.216 g, 0.6 mmol) in dry DMF (2.0 mL) was slowly added to a stirred and heated solution, at 60 °C, of the corresponding quinone (0.5 mmol) and NaI (5 equiv) in DMF (3 mL). Stirring and heating were maintained for 1 h. After cooling, the precipitate was filtered off and washed with water and then with ethyl acetate.

**7,7-Dimethyl-8-dihydro-6-oxa-1-thia-3-azadicyclopenta[*b*,*g*]naphthalene-5,9-dione (10a).** Compound **10a** was obtained from quinone **5** as a single regioisomer (64 mg, 45% yield) or from quinone **2** admixed with **10b** (67 mg, 47% overall yield, ratio **10a**/**10b**: 88/12). Compound **10a** was separated from the mixture by column chromatograhy on silica gel using a mixture of dichloromethane/petroleum ether: 5/95; mp 243–244 °C. IR (KBr) ν $cm^{-1}$ 1675, 1635 (CO). $^1H$ NMR (500.13 MHz; $CDCl_3$) δ: 1.62 (s, 6H, 7-$CH_3$), 3.06 (s, 2H, 8-H), 8.71 (s, 1H, 10-H), 8.77 (s, 1H, 4-H), 9.24 (s, 1H, 2-H); $^{13}C$ NMR (125.78 MHz, $CDCl_3$) δ: 28.4 (7-$CH_3$), 40.6 (8-C), 92.8 (7-C), 121.3 (10-C), 122.7 (4-C), 124.5 (8a-C), 130.4 (9a-C), 130.5 (4a-C), 139.8 (10a-C), 156.0 (3a-C), 159.8 (2-C), 160.2 (5a-C), 178.1 (5-C), 182.0 (9-C). Anal. calcd for $C_{15}H_{11}NO_3S$: C, 63.14, H, 3.89, N, 4.91, S, 11.24. Found: C, 63.13, H, 3.98, N, 5.09, S, 11.51.

**7,7-Dimethyl-8-dihydro-6-oxa-3-thia-1-azadicyclopenta[*b*,*g*]naphthalene-5,9-dione (10b).** Compound **10b** was obtained from quinone **7** admixed with the regioisomer **10a** (67, 47% overall yield, ratio **10b**/**10a**: 14/86). Compound **10b** was separated from the mixture by column chromatograhy on silica gel using a mixture of dichloromethane/petroleum ether: 5/95; mp 236–237 °C. IR (KBr) ν $cm^{-1}$ 1677, 1635 (CO). $^1H$ NMR (300.13 MHz; $CDCl_3$) δ: 1.63 (s, 6H, 7-$CH_3$), 3.06 (s, 2H, 8-H), 8.67 (s, 1H, 4-H), 8.78 (s, 1H, 10-H), 9.23 (s, 1H, 2-H); $^{13}C$ NMR (75.47 MHz, $CDCl_3$) δ: 28.4, 40.1, 92.4, 120.9, 122.2, 124.1, 130.1, 130.3, 139.4, 155.6, 158.2, 159.8, 177.6, 181.6.

**7-Methyl-6-oxa-1-thia-3-azadicyclopenta[*b*,*g*]naphthalene-5,9-dione (14a).** Compound **14a** was obtained from quinone **12** (61 mg, 45% yield), mp > 300 °C. IR (KBr) ν $cm^{-1}$ 1667 (CO). $^1H$ NMR (500.13 MHz, $CDCl_3$) δ: 2.56 (s, 3H, 7-$CH_3$), 6.67 (s, 1H, 8-H), 8.82 (s, 1H, 10-H), 8.96 (s, 1H, 4-H), 9.25 (s, 1H, 2-H); $^{13}C$ NMR (125.78 MHz, $CDCl_3$) δ: 14.5 ($CH_3$), 105.5 (8-C), 122.4 (10-C), 123.2 (4-C), 130.6 (9a-C), 131.8 (4a-C), 132.8 (8a-C), 139.3 (10a-C), 152.7 (5a-C), 156.7 (3a-C), 161.2 (2-C, 7-C), 172.8 (5-C), 180.3 (9-C). Anal. calcd for $C_{14}H_7NO_3S$, $0.5H_2O$: C, 60.42, H, 2.89, N, 5.03, S, 11.52. Found: C, 60.33, H, 2.82, N, 5.20, S, 11.90.

**7-Methyl-6-oxa-1-aza-3-thiadicyclopenta[*b*,*g*]naphthalene-5,9-dione (14b).** Compound **14b** was prepared from quinone **13** (54 mg, 40% yield), mp > 300 °C. IR (KBr) ν $cm^{-1}$ 1670 (CO). $^1H$ NMR (300.13 MHz, $CDCl_3$) δ: 2.54 (s, 3H, 7-$CH_3$), 6.70 (s, 1H, 8-H), 8.83 (s, 1H, 4-H), 8.90 (s, 1H, 10-H), 9.27 (s, 1H, 2-H); $^{13}C$ NMR: (75.47 MHz, $CDCl_3$) δ: 14.5, 105.0, 122.0, 123.0, 129.1, 130.0, 131.8, 139.2, 151.3, 156.2, 158.5, 171.8, 178.5. Anal. calcd for $C_{14}H_7NO_3S$, $0.5H_2O$: C, 60.42, H, 2.89, N, 5.03, S, 11.52. Found: C, 60.32, H, 2.85, N, 5.18, S, 11.80.

**4,7-Dimethoxybenzo[*b*]thiophene-2-carbaldehyde (17).** To a solution of (4,7-dimethoxybenzo[*b*]thiophen-2-yl)-methanol **16**[16] (134 mg, 0.60 mmol) in dichloromethane (20 mL), pyridinium chlorochromate (193 mg, 0.90 mmol) was added while stirring at room temperature. After 3 h, the reaction mixture was filtered through silica gel. The filtrate was evaporated to afford compound **17** (109 mg, 83%) as yellow crystals (ether–hexane), mp 131–132 °C. IR ν $cm^{-1}$ 1667 (C=O). $^1H$ NMR (200 MHz, $CDCl_3$) δ: 3.90 (s, 3H, $OCH_3$), 3.92 (s, 3H, $OCH_3$), 6.63 (d, 1H, $J$ = 8.5 Hz, 6-H or 5-H), 6.77 (d, 1H, $J$ = 8.5 Hz, 5-H or 6-H), 8.08 (s, 1H, 3-H), 10.00 (s, 1H, CHO); $^{13}C$ NMR (50 MHz, $CDCl_3$) δ: 55.74, 56.01, 104.79, 108.21, 131.01, 132.07, 133.38, 142.39, 148.48, 150.89, 184.57. Anal. calcd for $C_{11}H_{10}O_3S$: C, 59.44, H, 4.53, S, 14.43. Found: C, 59,08, H, 4.47, S, 14.18.

**4,7-Dioxo-4,7-dihydrobenzo[*b*]thiophene-2-carbaldehyde (18).** A solution of CAN (1.19 g, 2.17 mmol) in water (3.4 mL) was added dropwise to a stirred solution of 4,7-dimethoxybenzo[*b*]thiophene-2-carbaldehyde **17** (200 mg, 0.9 mmol) in acetonitrile (10 mL). The mixture was stirred at room temperature for 30 min, diluted with water and extracted with ethyl acetate (3×15 mL). The extract was dried under $MgSO_4$ and evaporated. The residue was column chromatographed on silica gel to afford compound **18** (140 mg, 81%) as yellow crystals (hexane–ethanol), mp 133–134 °C. IR ν $cm^{-1}$ 1678, 1666, 1657 (CO). $^1H$ NMR (200 MHz, $CDCl_3$) δ: 6.93

(d, 1H, $J = 10.3$ Hz, 6-H or 5-H), 7.00 (d, 1H, $J = 10.3$ Hz, 5-H or 6-H), 8.17 (s, 1H, 3-H), 10.08 (s, 1H, CHO). $^{13}$C NMR (50 MHz, $CDCl_3$) δ: 132.5, 138.4, 138.5, 140.8, 147.7, 148.4, 179.84, 180.4, 183.1 (CHO). Anal. calcd for $C_9H_4O_3S$: C, 56.24, H, 2.10, S, 16.68. Found: C, 56.30, H, 2.09, S, 16.44.

**Methyl 5,9-dioxo-5,9-dihydro-1,6-dithia-3-azadicyclopenta[*b*,*g*]naphthalene-7-carboxylate (19a) and methyl 5,9-dioxo-5,9-dihydro-3,6-dithia-1-azabicyclopenta[*b*,*g*]naphthalene-7-carboxylate (19b).** A solution of the tribromo derivative **8** (0.43 g, 0.82 mmol) in DMF (2.5 mL) was added over 20 min to a stirred, heated (70 °C) mixture of NaI (0.62 g, 4.1 mmol) and quinone **15** (0.15 g, 0.68 mmol) in DMF (3 mL). The reaction mixture was heated at 70 °C for 2.5 h. After cooling, the precipitate formed was filtrated, washed with water, acetone and AcOEt. Addition of acetone to the filtrate allows to obtain another amount of precipitate. Compounds **19**, in regioisomeric mixture, were obtained as a pale yellow solid (31 mg, 42% yield, ratio **19a**/**19b**: 70/30), mp > 300 °C. IR (KBr) ν cm$^{-1}$ 1730, 1710, 1665 (CO). $^1$H NMR (300.13 MHz, DMSO-$d_6$) δ: 3.94 (s, 1H, $CH_3$), 8.20 (s, 1H, 8-H), 8.71 (s, 1H, 10-H or 4-H), 9.10 (s, 1H, 4-H or 10-H), 9.76 (s, 1H, 2-H). Anal. calcd for $C_{15}H_7NO_4S_2$: C, 54.70, H, 2.14, N, 4.25, S, 19.47. Found: C, 54.52, H, 2.16, N, 4.01, S, 19.12.

**5,9-Dioxo-5,9-dihydro-1,6-dithia-3-azadicyclopenta[*b*,*g*]-naphthalene-7-carbaldehyde (20a) and 5,9-Dioxo-5,9-dihydro-3,6-dithia-1-azadicyclopenta[*b*,*g*]naphthalene-7-carbaldehyde (20b).** A solution of the tribromo derivative **8** (0.50 g, 0.95 mmol) in DMF (2.5 mL) was added over 20 min to a stirred, heated (70 °C) mixture of NaI (0.71 g, 4.75 mmol) and quinone **18** (0.15 g, 0.79 mmol) in DMF (3 mL). The reaction mixture was heated for 1 h. After the same work up as above, compounds **20**, in regioisomeric mixture, were obtained as a pale yellow solid (27 mg, 35% yield, ratio **20a**/**20b**: 55/45), mp > 300 °C. IR (KBr) ν cm$^{-1}$ 1680, 1660, 1650 (CO). $^1$H NMR (300.13 MHz, $CF_3COOD$) δ: 8.70 (s, 1H, 8-H), 9.33 (s, 1H, 10-H or 4-H), 9.36 (s, 1H, 4-H or 10-H), 10.19 (s, 1H, CHO), 10.61 (s, 1H, 2-H); $^{13}$C NMR (75.47 MHz, $CF_3COOD$) δ: 126.0, 131.4, 139.2, 141.2, 142.2, 142.8, 149.3, 150.7, 155.9, 158.4, 173.7, 184.4, 184.6, 194.1. Anal. calcd for $C_{14}H_5NO_3S_2$: C, 56.18, H, 1.68, N, 4.68, S, 21.43. Found: C, 56.02, H, 1.91, N, 4.43, S, 21.74.

Antileishmanial activity against promastigotes of *L. donovani* MHOM/ET/67/L82 and *Leishmania major* MHOM/PT/92/CRE26: LV9 was assessed in 96-well plates (Falcon) at 27 °C using CellTiter 96® Aqueous Non-Radioactive Cell Proliferation Assay (Promega), colorimetric method. $10^5$ parasites were resuspended in fresh medium/well. The compound was dissolved in DMSO and then diluted at the appropriate concentration in the standard culture medium [RPMI 1640 medium (sigma) containing 20% foetal calf serum]. Median inhibitory concentrations ($IC_{50}$) were determined after 48 h of culture, the drug being tested as a serial fourfold dilution from 0.01 to 1 μM and six replicate cultures being set up at each concentration.

Growth inhibition of *T. gondii*: the virulent RH strain of *T. gondii* was maintained in culture with the human myelomonocytic cell line THP-1 (ECACC number 88081201, Sophia-Antipolis, France) as previously described.[20] Three different experiments were performed in quadruplicate.

Assays of cytotoxicity of the drugs were conducted on a human myelomonocytic cell line THP-1 (European collection of animal cell culture number 88081201: Sophia-Antipolis, France). These non-adherent cells were suspended in RPMI 1640 medium (DAP, Vogelgrun, France) supplemented with 100 U/mL of penicillin, 100 μg/mL of streptomycin and 10% fetal calf serum (DAP). The growth of THP-1 cells was assessed in 96-well plates at 37 °C using the method described above for parasites.

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