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Iodine-catalyzed one-pot synthesis and antimalarial activity evaluation of symmetrically and asymmetrically substituted 3,6-diphenyl[1,2,4,5]tetraoxanes

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ABSTRACT

A novel iodine-catalyzed one-pot synthesis of symmetrically and asymmetrically substituted 3,6-diphenyl-[1,2,4,5]tetraoxanes is described. The synthetic protocol is general with substituted benzaldehydes and proceeds well under acidic conditions. Total 17 tetraoxanes have been prepared during this study and some of these compounds exhibit promising antimalarial activity. None of the compounds shows any toxicity against vero cells.

Malaria affects over 40% of world's population, causing deaths of 1-3 million people every year.[1–4] The incidence of malaria is increasing because of many *Plasmodium falciparum* strains have become resistant to most of the drugs. Over the last two decades endoperoxide class of compounds have received considerable attention of chemist and biologist due to their potent activity against *P. falciparum*.[5] Artemisinin, a 1,2,4-trioxane compound isolated from Chinese plant *Artemisia annua*, has been one of the most effective antimalarial against *P. falciparum*. However limited availability, high cost, and poor bioavailability have been the major drawback of artemisinin.[6] Artesunate and artemether, a semi-synthetic derivatives of artemisinin, also shows poor pharmacokinetic properties.[7] Structure-activity relationship studies conducted on artemisinin and its semi-synthetic derivatives revealed that the peroxide linkage is the most crucial pharmacophore in these molecules.[2,8] This discovery was the beginning of a significant effort to identify synthetically accessible antimalarial peroxides.[9,10] Dispiro-tetraoxane is one such class of compounds which was found to be equally potent as artemisinin.[11] However, structural diversity of this important class of compound is not available.[12] Numerous methods have been reported for the synthesis of symmetrical tetraoxane derivatives.[13] The acid-catalyzed cyclocondensation of hydrogen peroxide with ketones or aldehydes,[13–16] ozonolysis of olefins,[17] enol-ethers,[18] *O*-ether oxime,[19] and cyclocondensation of bis(trimethylsilyl) peroxide with carbonyl compounds catalyzed by trimethylsilyl trifluoromethanesulfonate (TMSOTf)[20] have been the most commonly used methods. The unsymmetrical tetraoxanes have been synthesized by cyclocondensation of ketones and aldehydes with steroidal gem-bis-hydroperoxides (H_2SO_4 catalyst),[21] aliphatic and alicyclic gem-hydroperoxides ($MeReO_3$–HBF_4 catalyst),[21] and gem-bis(trimethylsilyldioxy)alkanes (TMSOTf catalyst).[22] Most of these methods are highly dependent on several factors, such as the structure of the carbonyl compounds, temperature, concentration, pH, mode of addition, solvent, and the equilibrium between ketone and the precursors of cyclic peroxides,[23] which lead to variable yields of the tetraoxanes. Most of these procedures involve cyclic ketones as a starting material, which provides limited opportunity for functionalization. So there is always a need for the development of an easy, cost effective, and efficient route to symmetrical and unsymmetrical tetraxoanes with functional group diversity. Structural diversity in this class of compounds can be generated if one uses aromatic aldehydes as a starting material rather than cyclic ketones. Careful literature survey revealed that little over 25 tetraoxanes having aromatic ring as a part of the active pharmacophore have been evaluated for their antimalarial activity. As part of our ongoing efforts towards the synthesis of biologically active compounds,[24–26] we report herein an efficient, cost effective, iodine-catalyzed simple one-pot synthesis of symmetrically and asymmetrically substituted tetraoxanes, and evaluation of their antimalarial activity.

Due to the possibility of selective incorporation of various functional groups in the phenyl ring, we decided to use substituted benzaldehydes as a starting material for the preparation of tetraoxanes. Use of substituted benzaldehydes will not only generate wide range of tetraoxanes, but this will help to study the structure activity relationship in this class of compounds. Recently, we have reported synthesis and antimalarial activity of symmetrically and

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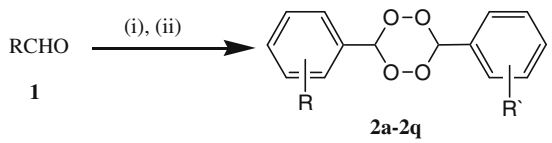

Scheme 1. Reagents and conditions: (i) I_2, H_2O_2, CH_3CN; (ii) R′CHO, $HBF_4{\cdot}Et_2O$, rt.

Table 1
Symmetrical and asymmetrical tetraoxanes via Scheme 1

Ent	R	R′	Mp	Yield (%)	Reference
2a	*o*-Me	*o*-Me	165–167	42	26,31–33
2b	*m*-Me	*m*-Me	148–150	51	26
2c	*p*-Me	*p*-Me	233–234	54	26
2d	*p*-Cl	*p*-Cl	240 (dec)	25	26,31–33
2e	*p*-Br	*p*-Br	>280	22	26
2f	*p*-F	*p*-F	212–213	29	26,31–33
2g	*p*-OMe	*p*-OMe	195–196	27	33
2h	*p*-Et	*p*-Et	190–192	44	
2i	*p*-*n*-Pr	*p*-*n*-Pr	155	38	
2j	*p*-*i*-Pr	*p*-*i*-Pr	156–157	41	
2k	*p*-*n*-But	*p*-*n*-But	142–145	40	
2l	*p*-*t*-But	*p*-*t*-But	222–224	53	
2m	*p*-Et	*p*-Me	205	41	
2n	*p*-Me	*p*-*i*-Pr	215–216	33	
2o	*p*-Me	*p*-*t*-But	188–190	46	
2p	*p*-Me	*p*-OMe	206	25	
2q	*p*-Me	*p*-CO_2Me	207–208	37	

asymmetrically substituted tetraoxanes which have been prepared under MTO/TFE reaction conditions[21] using substituted benzaldehydes as a starting material.[26] Some of the compounds, especially tolyl based tetraoxanes exhibit good antimalarial activity.[26] During the course of this study we observed that the MTO-catalyzed reaction has many limitations as MTO/TFE system is uneconomical, non-selective for various functional groups, and yield was poor in some cases.[26] In continuation to this study, we developed a novel cost effective method for the preparation of tetraoxanes and their antimalarial activity was determined in an in vitro assay. Bis-hydroperoxide has been used as an intermediate for the preparation of tetraoxanes in most of the cases, and it has been prepared via various methods.[22,27,28] Encouraged by a recent publication by Iskra and co-workers[29,30] that deals with the iodine-catalyzed synthesis of bis-hydroperoxides, we speculated iodine may catalyze the reaction of bis-hydroperxides and aromatic aldehydes that may lead to tetraoxanes.

In a typical reaction conditions, substituted benzaldehyde was added to a stirred solution of H_2O_2 (6 equiv) and I_2 (0.1 equiv) in 10 mL of acetonitrile. Bis-hydroperoxide formation was confirmed by TLC and ^{1}H NMR. After this, same or different substituted benzaldehyde (1.0 equiv) was added followed by the addition of 1.0 mL of $HBF_4{\cdot}Et_2O$ (Scheme 1).

Asymmetrical tetraoxanes (**2m–q**) have also been prepared by this method, which are useful for the structure activity relationship studies. It is important to mention here that during the preparation of asymmetrical tetraoxanes, symmetrical tetraoxanes formation was not observed by ^{1}H NMR. This synthetic protocol is of particular interest, as the functional groups can be further manipulated chemically, if required. Notably, electron withdrawing OMe group favors the formation of tetraoxane (Table 1,[26,31–33] entry **2g**, literature yield 2%) while electron withdrawing NO_2 group inhibit the reaction, as bis-hydroperoxide formation was not observed in this case.[29,30] Using same reaction conditions cyclic ketones did not react. All of the compounds were purified over SiO_2 column, and characterized spectroscopically.[34]

In vitro antimalarial activity and cytotoxicity. In vitro antimalarial activity and cytotoxicity of these tetraoxanes were determined against chloroquine sensitive (D6), and chloroquine resistant (W2) strains of *P. falciparum* by literature methods.[26,35–37] Among the series, compounds **2h–j**, and **2m–p,** exhibited the most potent antimalarial activity with IC_{50} values ranging from 0.38 to 0.99 μM for D6 and 0.45 to 1.08 μM for W2 strain as shown in Table 2. Other compounds (entries **2k**, and **2l**) also showed moderate to mild antimalarial activities (IC_{50} 2.19–3.79 μM). None of these tetraoxanes showed any cytotoxicity (Table 2) to mammalian kidney fibroblasts (vero cells). A higher selectivity index (S.I. > 20) for antimalarial activity was observed for compounds **2i, 2j,** and **2m–p**. Among the series of 10 unknown compounds, symmetrical tetraoxanes having *para n*-butyl or *p*-*tert*-butyl group have shown poor antimalarial activity (entries **2k** and **2l**). While in case of asymmetrical tetraoxanes *p*-ethyl group at one end and *p*-methyl or *m*-methyl or *p*-isopropyl group at the other end of the tetraoxane, were found to be most active compounds (entries **2m–o**) in terms of IC_{50} value and selectivity index. These three compounds (entries **2m–o**) were effective against both strains of *P. falciparum* (D6 and W2) with IC_{50} values in the range of 0.38–0.79 μM and selectivity index of 25.1–43.6. However, ester functionality has negative effect on the antimalarial activity (entry **2q**). This study indicates that substitution of methyl group in the previously reported tetraoxanes[26] by ethyl, *n*-propyl, *i*-propyl groups not only improves antimalarial activity, but selectivity index of the resulting tetraoxanes was also better.

In conclusion, we have developed a new, convenient, simple and efficient method for the synthesis of symmetrical and asymmetrical tetraoxanes. Use of readily accessible and inexpensive substituted aromatic aldehydes, iodine as a catalyst, and acetoni-

Table 2
Antimalarial activity and cytotoxicity of symmetrical and asymmetrical tetraoxanes via Scheme 1

Ent	R	R′	*P. falciparum* (D6 clone)		*P. falciparum* (W2 clone)		Cytotoxicity (vero cells)
			IC_{50} (μM)	S.I.	IC_{50} (μM)	S.I.	
2h	*p*-Et	*p*-Et	0.99	>15.9	0.99	>15.9	NC
2i	*p*-*n*-Pr	*p*-*n*-Pr	0.61	>23.8	0.76	>19	NC
2j	*p*-*i*-Pr	*p*-*i*-Pr	0.67	>21.6	1.03	>14	NC
2k	*p*-*n*-But	*p*-*n*-But	3.65	>3.7	4.77	>2.8	NC
2l	*p*-*t*-But	*p*-*t*-But	2.19	>6.1	3.93	>3.4	NC
2m	*p*-Me	*p*-Et	0.38	>43.6	0.45	>36.7	NC
2n	*p*-Me	*p*-*i*-Pr	0.59	>26.4	0.77	>20.7	NC
2o	*p*-Me	*p*-*t*-But	0.60	>25.1	0.79	>19.0	NC
2p	*p*-Me	*p*-OMe	0.76	>21.6	1.08	>15.4	NC
2q	*p*-Me	*p*-CO_2Me	3.79	>4.0	6.01	>2.5	NC
CQ			0.05	>298	0.41	>42	NC
Art			0.035	>476	0.015	>1400	NC

NC: no cytotoxicity upto 16.72 μM.
SI: selectivity index (IC_{50} for cytotoxicity/IC_{50} for antimalarial activity).
CQ: chloroquine; Art: artemisinin.

trile as a reaction medium, makes this synthetic methodology more practical and reliable method for the synthesis of medicinally important tetraoxane class of compounds. The use of I_2 as a catalyst and acetonitrile as a solvent offers many advantages as they are environmentally benign, easily available, and excess of H_2O_2, and aldehydes are not required in this reaction which has been one of the major drawback of the existing synthetic methods. Some of these compounds have shown very promising antimalarial activity with no toxicity against vero cells.

Caution. We have not encountered any difficulties in working with these compounds, routine precautions such as shields, fume hoods, and avoidance of transition metal salts should be observed whenever possible, as organic peroxides are explosive in nature.

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References and notes

1. Haynes, K.; Vonwiller, S. C. *Acc. Chem. Res.* **1997**, *30*, 73.
2. O'Neill, P. M.; Posner, G. H. *J. Med. Chem.* **2004**, *47*, 2945.
3. Tang, Y.; Dong, Y.; Vennerstrom, J. L. *Med. Res. Rev.* **2004**, *24*, 425.
4. Biagini, G. A.; O'Neill, P. M.; Bray, P. G.; Ward, S. A. *Curr. Opin. Pharmacol.* **2005**, *5*, 473.
5. Haynes, R. K. *Curr. Top. Med. Chem.* **2006**, *6*, 509.
6. White, N. J. *Science* **2008**, *320*, 330.
7. Klayman, D. L. *Science* **1985**, *228*, 1049.
8. Meshnick, S. R. *Int. J. Parasitol.* **2002**, *32*, 1655.
9. Singh, C.; Kanchan, R.; Sharma, U.; Puri, S. K. *J. Med. Chem.* **2007**, *50*, 521.
10. Vennerstrom, J. L.; Arbe-Barnes, S.; Brun, R.; Charman, S. A.; Chiu, F. C. K.; Chollet, J.; Dong, Y.; Dorn, A.; Hunziker, D.; Matile, H.; Mclntosh, K.; Padmanilayam, M.; Tomas, J. S.; Scheurer, C.; Scorneaux, B.; Tang, Y.; Urwyler, H.; Wittin, S.; Charman, W. N. *Nature* **2004**, *430*, 900.
11. Vennerstrom, J. L.; Fu, H.-N.; Ellis, W. Y.; Ager, A. L.; Wood, J. K.; Andersen, S. L.; Gerena, L.; Milhous, W. K. *J. Med. Chem.* **1992**, *35*, 3023.
12. Terentèv, A. O.; Kutkin, A. V.; Starikova, Z. A.; Antipin, M. Y.; Ogibin; Yu, N.; Nikishin, G. I. *Synthesis* **2004**, 2356.
13. Kim, H.-S.; Nagai, Y.; Ono, K.; Begum, K.; Wataya, Y.; Hamada, Y.; Tsuchiya, K.; Masuyama, A.; Nojima, M.; McCullough, K. *J. Med. Chem.* **2001**, *44*, 2357.
14. Solaja, B. A.; Terzic, N.; Pocsfalvi, G.; Genena, L.; Tinant, B.; Opsenica, D.; Milhous, W. K. *J. Med. Chem.* **2002**, *45*, 3331.
15. Brune, H. A.; Wulz, K.; Hetz, W. *Tetrahedron* **1971**, *27*, 3629.
16. Adam, W.; Asensio, G.; Curci, R.; Marco, J. A.; Gonzalez-Nunez, M. E.; Melio, R. *Tetrahedron Lett.* **1992**, *33*, 5833.
17. Keul, H. *Chem. Ber.* **1975**, *108*, 1198.
18. Wojchiechowski, B. J.; Person, W. H.; Kuczkowski, R. L. *J. Org. Chem.* **1989**, *54*, 115.
19. Yokaya, H.; Umehara, Y.; Matsuura, T. *Bull. Chem. Soc. Jpn.* **1980**, *53*, 2407.
20. Jefford, C. W.; Boukouvalas, A. J. J. *Synthesis* **1988**, 391.
21. Iskra, J.; Bonnet-Delpan, D.; Begue, J.-P. *Tetrahedron Lett.* **2003**, *44*, 6309.
22. Kim, H.-S.; Tsuchiya, K.; Shibata, Y.; Wataya, Y.; Ushigoe, Y.; Masuyama, A.; Nojima, A.; McCullough, K. *J. Chem. Soc., Perkin Trans. 1* **1999**, 1867.
23. McCullough, K. J. M.; Nonhebel, A. R.; Pauson, D. C.; White, P. L. *J. Chem. Res. (S)* **1980**, *42*, 601.
24. Joshi, M. C.; Bisht, G. S.; Rawat, D. S. *Bioorg. Med. Chem. Lett.* **2007**, *17*, 3226.
25. Sharma, M.; Agarwal, N.; Rawat, D. S. *J. Heterocycl. Chem.* **2008**, *45*, 737.
26. Atheaya, H.; Khan, S. I.; Mamgain, R.; Rawat, D. S. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 1446.
27. Tevent'ev, A. O.; Kutkin, A. V. N.; Troizky, A.; Ogibin, Y. N.; Nikishin, G. I. *Synthesis* **2005**, 2215.
28. Todorovic, N. M.; Stefanovic, M.; Tinant, B.; Declercq, J. P.; Makler, M. T.; Solaja, B. A. *Steroids* **1996**, *61*, 688.
29. Zmitek, K.; Zupan, M.; Stavber, S.; Iskra, J. *Org. Lett.* **2006**, *8*, 2491.
30. Zmitek, K.; Zupan, M.; Stavber, S.; Iskra, J. *Org. Chem.* **2007**, *72*, 6534.
31. Miura, M.; Ikegami, A.; Nojima, M.; Kusabayashi, S. *Chem. Commun.* **1980**, 1279.
32. Bertrand, M.; Fliszar, S.; Rousseau, Y. *J. Org. Chem.* **1968**, *33*, 1931.
33. Kim, H.-S.; Shibata, Y.; Wataya, Y.; Tsuchiya, K.; Masuyama, A.; Nojima, M. *J. Med. Chem.* **1999**, *42*, 2604.
34. *Spectral data of selected compounds*: *3,6-Bis-(4-ethyl-phenyl)-[1,2,4,5]tetroxane* (**2h**): Yield: 44%; mp: 190–192 °C; IR (KBr, cm^{-1}): 2969, 1610, 1512, 1458, 1422, 1362, 1181, 1117, 1022, 1003, 909, 840; ^{1}H NMR (300 MHz, $CDCl_3$): 1.26 (t, J = 6 Hz, 6H), 2.66–2.94 (m, 4H), 6.91 (s, 2H), 7.28 (m, J = 8 Hz, 4H), 7.53 (m, J = 8 Hz, 4H); MS–ESI (m/z): 300.2 (M^+); Anal. Calcd for $C_{18}H_{20}O_4$: C, 71.98; H, 6.71. Found: C, 71.77; H, 6.53. *3,6-Bis-(4-tert-butyl-phenyl)-[1,2,4,5]tetroxane* (**2l**): Yield: 53%; mp: 222–224 °C; IR (KBr, cm^{-1}): 2924, 1614, 1461, 1370, 1312, 1267, 1187, 1021, 1003, 911, 838, 803; ^{1}H NMR (300 MHz, $CDCl_3$): 1.31 (s, 18H), 6.92 (s, 2H), 7.50–7.62 (m, 8H); ^{13}C NMR (75.5 MHz, $CDCl_3$): 31.16 (CH_3), 34.91 (CH_3), 108.10 (CH), 125.76 (CH), 127.57 (CH), 128.06 (C), 154.67 (C); MS–ESI (m/z): 356.4 (M^+); Anal. Calcd for $C_{22}H_{28}O_4$: C, 74.13; H, 7.92. Found: C, 74.37; H, 7.73. *3-(4-Ethyl-phenyl)-6-p-tolyl-[1,2,4,5]tetroxane* (**2m**): Yield: 41%; mp: 205 °C; IR (KBr, cm^{-1}): 2949, 1610, 1361, 1180, 1021, 909, 840; ^{1}H NMR (300 MHz, $CDCl_3$): 1.26 (t, J = 6 Hz, 3H), 2.40 (s, s, 3H), 2.69 (q, J = 6 Hz, 2H), 6.90 (s, 1H), 6.91 (s, 1H), 7.27–7.30 (m, 4H), 7.41–7.47 (m, 4H); MS–ESI (m/z): 286.1 (M^+); Anal. Calcd for $C_{17}H_{18}O_4$: C, 71.31; H, 6.34. Found: C, 71.61; H, 6.54.
35. Jain, M.; Khan, S. I.; Tekwani, B. L.; Jacob, M. R.; Singh, S.; Singh, P. P.; Jain, R. *Bioorg. Med. Chem.* **2005**, *13*, 4458.
36. Makler, M. T.; Hinrichs, D. *J. Am. J. Trop. Med. Hyg.* **1993**, *48*, 205.
37. Mustafa, J.; Khan, S. I.; Ma, G.; Walker, L. A.; Khan, I. A. *Lipids* **2004**, *39*, 167.