CHAPTER II
EXPERIMENTAL

2.1 Instruments and equipment

Melting points were determined with a Fishers-Johns melting point apparatus and are uncorrected. Thin layer chromatography (TLC) was performed on aluminium sheets precoated with silica gel (Merck Kieselgel 60 F_{254}). Column chromatography was performed on silica gel (Merck Kieselgel 60 G).

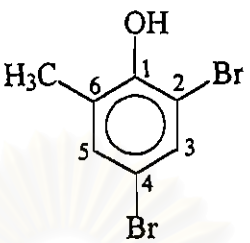
The FT-IR spectra were recorded on a Nicolet Fourier Transformed Infrared Spectrophotometer model Impact 410. Solid samples were incorporated into a pellet of potassium bromide. Liquid samples were dropped on a sodium chloride cell. The ^{1}H-NMR and ^{13}C-NMR spectra were obtained with a Bruker model ACF200 spectrometer and a Jeol, model JNM-A500 FTNMR which operated at 200.13 MHz for ^{1}H and 50.32 MHz for ^{13}C nuclei and 500.00 MHz for ^{1}H and 125.00 MHz for ^{13}C nuclei, respectively. The chemical shifts (δ) were assigned by comparison with residue protons. Elemental analysis (EA) was carried out on a Perkin Elmer PE 2400 Series II: option CHN on.

2.2 Chemicals

All solvents used in this research were purified prior to use by standard methodology except for those which were reagent grades. The reagents used for synthesizing the precursors, benzoquinones and hydroquinones were purchased from Fluka Chemical Company or otherwise stated and were used without further purification.

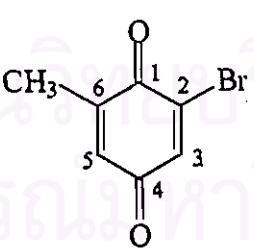
2.3 Synthesis

2.3.1 *2,4-dibromo-6- methylphenol* (Compound **1a**)[25]



Bromine (32 g, 0.20 mol) was dissolved in 100 mL acetic acid. *o*-Cresol (10.8 g, 0.10 mol, in 20 mL of AcOH) was added dropwise to above solution within 10 min with stirring at room temperature and continued to stir for another 20 min. The solution was diluted with 200 mL of water. The white crystals of product was precipitated and collected. The product was recrystallized from EtOH:H_2O to give 24.65 g (93%), m.p. 54-55 °C (lit.[25] 57 °C), R_f 0.75 (CH_2Cl_2). IR(KBr) 3500-3250, 3083, 1465, 1398 and 1137 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 2.26 (3H, s), 5.51 (1H, s), 7.19 (1H, d, J = 1.83 Hz) and 7.42 (1H, d, J = 2.14 Hz); ^{13}C-NMR ($CDCl_3$) δ (ppm): 16.5 (1C, CH_3), 110.4 (1C, C-2), 112.0 (1C, C-4), 127.7 (1C, C-6), 131.3 (1C, C-5), 133.1 (1C, C-3) and 149.8 (1C, C-1).

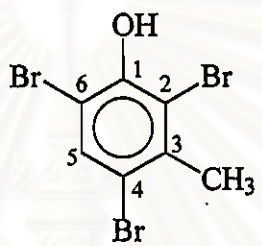
2.3.2 *2-bromo-6-methyl-1,4-benzoquinone* (Compound **1b**)[25]



Compound **1a** (15.30 g, 0.06 mol) was dissolved in 600 mL of acetic acid: acetonitrile:water (5:1:1). The solution was then heated at 60 °C. Chromic acid (CrO_3, 6 g, 0.06 mol) was added within 30 min with stirring and continued to stir for another 1 h at 60-65 °C. The solution was then cooled and extracted with $CHCl_3$. The $CHCl_3$ layer was washed with 1 M $NaHCO_3$ and water, and then dried over anhydrous Na_2SO_4. After removing solvent, the residue was applied to a silica gel flash column which eluted by a mixture of hexane-ether (2:1). A yellow eluate was collected. The

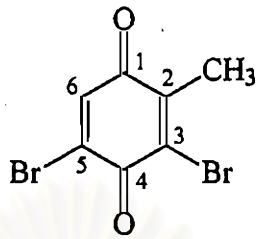
product 6.93 g (90% yield) as orange needles, m.p. 97 °C (lit.[25] 94-95 °C), R_f 0.54 (hexane:ether (1:1)) was obtained. IR(KBr) 3047, 1669, 1557 and 681 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 2.17 (3H, d, J = 1.53 Hz), 6.86 (1H, q, J = 1.53 Hz) and 7.45 (1H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 16.6 (1C, CH_3), 132.7 (1C, C-3), 137.0 (1C, C-6), 139.6 (1C, C-2), 146.1 (1C, C-5), 177.2 (1C, C-1) and 177.8 (1C, C-4).

2.3.3 *2,4,6-tribromo-3-methylphenol* (Compound **2a**)[25]



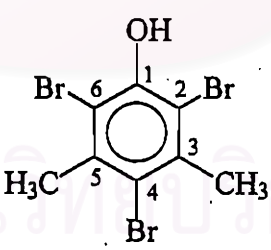
m-Cresol (5.4 g, 0.05 mol) was treated with bromine (24 g, 0.15 mol) by a similar procedure to that employed for the synthesis of Compound **1a**. The crude product was purified by recrystallization from hexane to give white needles, 17.01 g (99%), m.p. 80-81 °C (lit.[25] 81.5-82 °C), R_f 0.64 (CH_2Cl_2). IR(KBr) 3500-3200, 3073, 1572 and 1444 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 2.53 (3H, s), 5.93 (1H, s) and 7.67 (1H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 24.0 (1C, CH_3), 106.8 (1C, C-6), 113.1 (1C, C-2), 115.0 (1C, C-4), 134.1 (1C, C-5), 137.7 (1C, C-3) and 148.9 (1C, C-1).

2.3.4 *3,5-dibromo-2-methyl-1,4-benzoquinone* (Compound **2b**)[25]



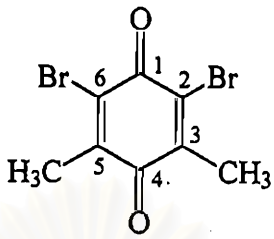
Compound **2a** (6.9 g, 0.02 mol) was dissolved in 250 mL of AcOH:H_2O (7:3), which was then heated to 70 °C. CrO_3 (2.05 g, 0.02 mol) was added within 10 min with stirring and continued to stir for another 30 min. The mixture was diluted with water 600 mL. The yellow solid was precipitated and collected, which was then recrystallized from a mixture of hexane:ether (1:1) to give yellow plates 4.25 g (60%), m.p. 115-116 °C (lit.[25] 117 °C), R_f 0.40 (hexane:ether (1:1)). IR(KBr) 1690, 1659, 1588 and 681 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 2.25 (3H, s) and 7.33 (1H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 17.1 (1C, CH_3), 134.3 (1C, C-5), 135.6 (1C, C-3), 137.9 (1C, C-6), 146.5 (1C, C-2), 172.3 (1C, C-1) and 181.8 (1C, C-4).

2.3.5 *2,4,6-tribromo-3,5-dimethylphenol* (Compound **3a**)



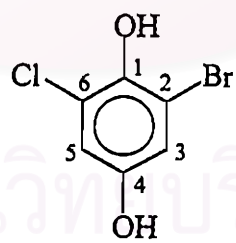
3,5-Dimethylphenol (5.40 g, 0.05 mol) was treated with bromine (16 g, 0.10 mol) by a similar procedure to that utilized for the preparation of Compound **1a**. The crude product was purified by recrystallization from hexane to give white needles, 10.13 g (64%), m.p. 120 °C (lit.[32] 116 °C), R_f 0.74 ($CHCl_3$). IR(KBr) 3400-3200, 2940 and 1367 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 2.60 (6H, s) and 6.02 (1H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 25.2 (2C, CH_3), 109.7 (2C, C-2 and C-6), 117.9 (1C, C-4), 137.3 (2C, C-3 and C-5) and 148.4 (1C, C-1).

2.3.6 *2,6-dibromo-3,5-dimethyl-1,4-benzoquinone* (Compound **3b**)



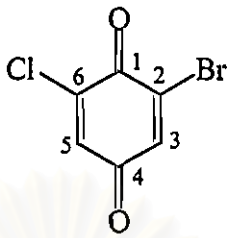
Compound **3a** (4.09 g, 0.02 mol) was dissolved in 200 mL of AcOH:CH_3CN:H_2O (5:1:1) and then treated with CrO_3 (2 g, 0.02 mol) by a similar procedure to that used for the synthesis of Compound **1b**. The crude product was purified by flash column which eluted by a mixture of hexane-ether (2:1). A yellow eluate was collected. After removing the solvent, yellow crystals 6.75 g (94%) was gained, m.p. 151-152 °C, R_f 0.51 ($CHCl_3$). IR(KBr) 1685, 1665 and 702 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 2.27 (6H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 17.4 (2C, CH_3), 134.2 (2C, C-2 and C-6), 146.1 (2C, C-3 and C-5), 172.1 (1C, C-1) and 181.4 (1C, C-4).

2.3.7 *2-bromo-6-chlorohydroquinone* (Compound **4a**)



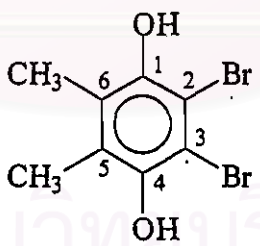
Chlorohydroquinone (7.26 g, 0.05 mol) was treated with bromine (16 g, 0.10 mol) using a similar fashion to that for the preparation of Compound **1a**. The crude product was purified by recrystallization from hexane to give yellow plates, 7.14 g (63%), m.p. 210 °C (dec.), R_f 0.70 ($CHCl_3$). IR(KBr) 3656-3375, 1567, 1265 and 881 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 7.47 (2H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 112.4 (1C, C-2), 115.8 (1C, C-5), 120.2 (1C, C-3), 126.5 (1C, C-6), 150.8 (1C, C-1) and 153.2 (1C, C-4).

2.3.8 *2-bromo-6-chloro-1,4-benzoquinone* (Compound **4b**)



Compound **4a** (4.19 g, 0.02 mol) was dissolved in 200 mL of AcOH:CH_3CN:H_2O (5:1:1) and then treated with CrO_3 (2 g, 0.02 mol) by a similar procedure to that employed for the synthesis of Compound **1b**. The crude product was purified by recrystallization from a mixture of hexane-ether (1:1) to give yellow powder, 2.01 g (48%), m.p. 223 °C (dec.), R_f 0.79 ($CHCl_3$). IR(KBr) 1660, 1675, 1055 and 886 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 7.26 (2H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 110.5 (1C, C-2), 132.3 (1C, C-6), 134.2 (1C, C-3), 136.2 (1C, C-5), 169.0 (1C, C-1) and 171.1 (1C, C-4).

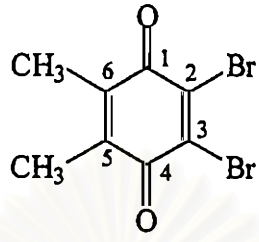
2.3.9 *2,3-dibromo-5,6-dimethylhydroquionone* (Compound **5a**)



2,3-Dimethylhydroquinone (3.45 g, 0.025 mol) was treated with bromine (8 g, 0.05 mol) by utilizing a similar procedure to that for the synthesis of Compound **1a**. The crude product was purified by recrystallization with hexane to give white needles, 5.34 g (98%), m.p. 165°C, R_f 0.53 ($CHCl_3$). IR(KBr) 3600-3200, 1434 and 1388 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 2.19 (6H, s) and 5.24 (2H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 13.1 (2C, CH_3), 108.3 (2C, C-5 and C-6), 124.8 (2C, C-2 and C-3) and 145.1 (2C, C-1 and C-4). Elemental analysis found %C 32.53 and %H 2.65; calcd. for $C_8H_8Br_2O_2$ (MW 295.96): %C 32.47 and %H 2.72.

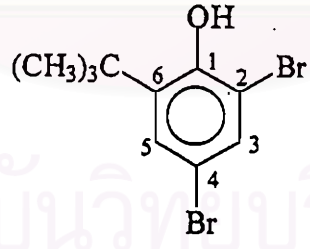
The IR, ^{1}H-NMR and ^{13}C-NMR spectra of this compound are exhibited in Figs. **A1-A3**.

2.3.10 *2,3-dibromo-5,6-dimethylbenzoquinone* (Compound **5b**)



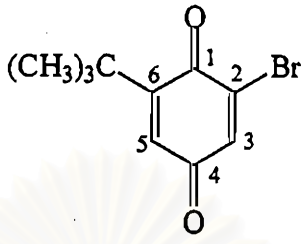
Compound **5a** (4.37g, 0.02 mol) was dissolved in 200 mL of AcOH:CH_3CN:H_2O (5:1:1) and then treated with CrO_3 (2 g, 0.02 mol) by a similar procedure to that used for the synthesis of Compound **1b**. The crude product was purified by recrystallization from a mixture of hexane-ether (1:1) to give yellow plates, 3.93 g (91%), m.p. 150-152°C, R_f 0.56 ($CHCl_3$). IR(KBr) 1670, 1583 and 702 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 2.13 (6H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 13.5 (2C, CH_3), 139.2 (2C, C-5 and C-6), 141.3 (2C, C-2 and C-3), and 177.7 (2C, C-1 and C-4).

2.3.11 *2,4-dibromo-6-tert-butylphenol* (Compound **6a**)



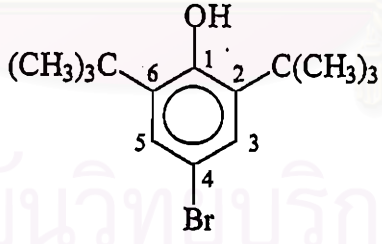
2-*tert*-Butylphenol (7.51g, 0.05 mol) was treated with bromine (16 g, 0.10 mol) by a similar procedure to that employed in Compound **1a** synthesis. The crude product 14.39 g, (94% yield) was precipitated and collected. After recrystallization with hexane, the product as white crystal, m.p. 50-51°C, R_f 0.57 (30% $CHCl_3$-hexane) was obtained. IR(KBr) 3500-3400, 2960 and 1440 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 1.38 (9H, s), 5.78 (1H, s), 7.31 (1H, d, J = 2.44 Hz) and 7.47 (1H, d, J = 2.44 Hz); ^{13}C-NMR ($CDCl_3$) δ (ppm): 29.1 (3C, CH_3), 35.6 (1C, $\underline{C}$-CH_3), 112.2 (1C, C-2), 112.5 (1C, C-4), 129.8 (1C, C-5), 131.5 (1C, C-3), 139.4 (1C, C-6) and 149.7 (1C, C-1).

2.3.12 *2-bromo-6-tert-butylbenzoquinone* (Compound **6b**)



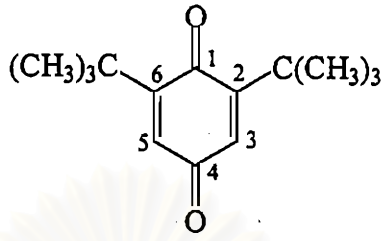
Compound **6a** (6.16 g, 0.02 mol) was dissolved in 200 mL of AcOH:CH_3CN:H_2O (5:1:1) and then treated with CrO_3 (2 g, 0.02 mol) by a similar manner to that used for the synthesis of Compound **1b**. The crude product was purified by recrystallization to give yellow plates 3.83 g (79%), m.p. 215°C, R_f 0.66 (20% EtOAc-hexane). IR(KBr) 1624, 1076 and 738 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 1.39 (9H, s), 7.68 (1H, d, J = 2.44 Hz) and 8.26 (1H, d, J = 2.75 Hz); ^{13}C-NMR ($CDCl_3$) δ (ppm): 29.4 (3C, CH_3), 36.7 (1C, C-CH_3), 127.5 (1C, C-2), 132.3 (1C, C-3), 133.1 (1C, C-5), 136.0 (1C, C-6), 150.8 (1C, C-1) and 178.8 (1C, C-4).

2.3.13 *4-bromo-2,6-di-tert-butylphenol* (Compound **7a**)



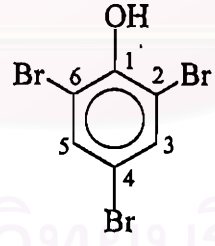
2,6-Di-*tert*-butylphenol (10.36 g, 0.05 mol) was treated with bromine (16 g, 0.10 mol) by a similar procedure to that used for the synthesis of Compound **1a**. The crude product was precipitated and collected. After purification by recrystallization with hexane, to give white crystal 6.83 g (48%), m.p. 195°C, R_f 0.70 (20% EtOAc-hexane) was gained. IR(KBr) 3660-3200, 2955 and 1368 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 1.42 (18H, s), 5.79 (1H, s) and 7.71 (2H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 29.6 (6C, CH_3), 36.0 (2C, C-CH_3), 126.0 (1C, C-4), 129.8 (2C, C-3 and C-5), 138.2 (2C, C-2 and C-6) and 150.5 (1C, C-1).

2.3.14 *2,6-di-tert-butylbenzoquinone* (Compound **7b**)



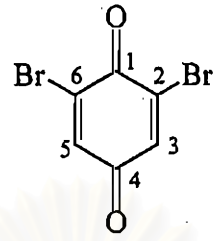
Compound **7a** (5.74 g, 0.02 mol) was dissolved in 200 mL of $AcOH:CH_3CN:H_2O$ (5:1:1) and then treated with CrO_3 (2 g, 0.02 mol) by a similar fashion to that used for the synthesis of Compound **1b**. The crude product was purified by recrystallization with a mixture of hexane-ether (1:1) to give reddish-brown powder 3.77 g (85%), m.p. 220°C (dec.), R_f 0.59 (20% EtOAc-hexane). IR (KBr) 2950 and 1603 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 1.37 (18H, s) and 7.71 (2H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 29.6 (6C, CH_3), 36.0 (2C, C-CH_3), 126.0 (2C, C-3 and C-5), 136.1 (2C, C-2 and C-6), 150.5 (1C, C-1) and 186.5 (1C, C-4).

2.3.15 *2,4,6-tribromophenol* (Compound **8a**)[28]



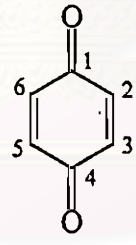
3-5% Bromine solution was added dropwise to 2% phenol (0.88 g in 40 mL of water) until the solution was turned to be yellow. The precipitate was collected and recrystallized from hexane to give pale yellow needles. The yield obtained was 2.11 g (68%), m.p. 88-89°C (lit.[32] 87-89°C), R_f 0.62 ($CHCl_3$). IR(KBr) 3493-3390, 1460 and 1235 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 5.90 (1H, s) and 7.59 (2H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 110.4 (2C, C-2 and C-6), 112.7 (1C, C-4), 134.2 (2C, C-3 and C-5) and 149.0 (1C, C-1).

2.3.16 *2,6-dibromo-1,4-benzoquinone* (Compound **8b**)[29]



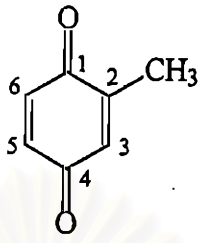
Compound **8a** (0.50 g, 0.0015 mol) was dissolved in ice-cold nitric acid (5 mL, 0.12 mol), the mixture was poured after 15 min stirring on ice. The yellow powder 0.22 g (50%) was precipitated and collected, m.p. 134-135°C (lit.[32] 131 °C), R_f 0.68 (50% $CHCl_3$-MeOH). IR(KBr) 1583, 1521 and 1327 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 7.25 (2H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 109.6 (2C, C-2 and C-6), 127.9 (2C, C-3 and C-5), 134.2 (1C, C-1) and 155.0 (1C, C-4).

2.3.17 *1,4-benzoquinone* (Compound **9**)[30]



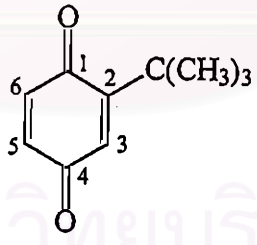
Cool a solution of 16 g (0.165 mol) of hydroquinone in 75 mL of 60 % acetic acid contained in a 250 mL flask to below 5 °C in an ice bath. Dissolve 21 g (0.21 mol) of chromium trioxide in 35 mL of water, and add 15 mL of glacial acetic acid. Add the chromium trioxide solution to the stirred hydroquinone solution at such a rate that the temperature does not rise above 10 °C; the addition takes about 2 h. Filter the mixture at once and wash the quinone several times with 5 mL portions of ice-cold water. The product was purified by recrystallization from hexane to give yellow needles 7.49 g (46%), m.p. 109-111°C (lit.[30] 115°C), R_f 0.56 ($CHCl_3$). IR(KBr) 1675 and 1590 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 6.67 (4H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 134.5 (4C, C-2, C-3, C-5 and C-6) and 186.2 (2C, C-1 and C-4).

2.3.18 *2-methylbenzoquinone* (Compound **10**)



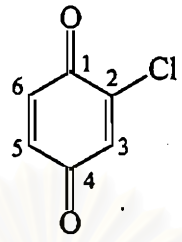
Cool a solution of toluhydroquinone (10.30 g, 0.10 mol) in 37.5 mL of 60% AcOH and then CrO_3 (10.50 g, 0.10 mol) in 17.5 mL of water and 7.5 mL of glacial AcOH was added using a similar procedure to that used for the synthesis of Compound **9**. The product was purified by recrystallization from hexane-EtOH (1:1) to give yellow plates, 3.47 g (34%), m.p. 65-66 °C (lit.[32] 69°C), R_f 0.56 ($CHCl_3$). IR (KBr) 3052, 1654 and 1603 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 2.00 (3H, s) and 6.57-6.69 (3H, m); ^{13}C-NMR ($CDCl_3$) δ (ppm): 15.8 (1C, CH_3), 133.3 (1C, C-3), 136.5 (1C, C-6), 136.6 (1C, C-5), 145.8 (1C, C-2), 187.5 (1C, C-1) and 187.7 (1C, C-4).

2.3.19 *2-tert-butyl-1,4-benzoquinone* (Compound **11**)



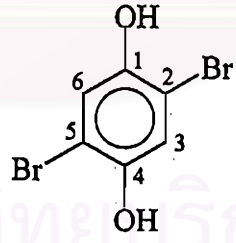
Cool a solution of *tert*-butylhydroquinone (13.71 g, 0.08 mol) in 37.5 mL of 60% AcOH and then CrO_3 (10.50 g, 0.10 mol) in 17.5 mL of water and 7.5 mL of glacial AcOH was added using a similar manner to that described for the synthesis of Compound **9**. The product was purified by recrystallization from hexane-EtOH (1:1) to give yellow plates, 13.20 g (98%), m.p. 54 °C, R_f 0.60 ($CHCl_3$). IR(KBr) 1660, 1337 and 933 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 1.34 (9H, s) and 6.65-6.73 (3H, m); ^{13}C-NMR ($CDCl_3$) δ (ppm): 29.1 (3C, CH_3), 35.2 (1C, $\underline{C}$-CH_3), 131.5 (1C, C-3), 135.0 (1C, C-6), 138.6 (1C, C-5), 156.0 (1C, C-2), 187.4 (1C, C-1) and 188.4 (1C, C-4).

2.3.20 *2-chloro-1,4-benzoquinone* (Compound **12**)



Cool a solution of chlorohydroquinone (11.93 g, 0.08 mol) in 37.5 mL of 60% AcOH and then CrO_3 (10.50 g, 0.10 mol) in 17.5 mL of water and 7.5 mL of glacial AcOH was added employing a similar procedure to that used for the synthesis of Compound 9. The product was purified by using a silica gel column eluting by chloroform to yield the yellow plate products 7.00 g (60%), m.p. 56-58 °C (lit.[32] 57 ° C), R_f 0.58 ($CHCl_3$). IR(KBr) 1670, 1540 and 714 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 6.78-7.00 (3H, m); ^{13}C-NMR ($CDCl_3$) δ (ppm): 133.7 (1C, C-2), 136.1 (1C, C-6), 136.8 (1C, C-5), 144.1 (1C, C-3), 179.2 (1C, C-1) and 184.9 (1C, C-4).

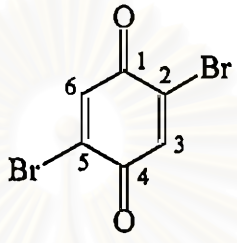
2.3.21 *2,5-dibromohydroquinone* (Compound **13a**)[31]



Bromine (32 g, 0.20 mol, 10.25 mL) in 10 mL of glacial acetic was added dropwise to a stirred suspension of hydroquinone (11.35 g, 0.10 mol) in 100 mL of glacial acetic acid. The temperature rised to 30 °C and a clear solution formed. After 5-10 min a colorless precipitate formed. Stirring was continued for another 1 h. The mixture was filtered and the solid was washed with a small amount of glacial acetic acid. The mother liquor was reduced to *ca.* 1/2 volumn and allowed to stand for 12 h to crystallize more product. The product was white crystal, 11.15 g (42%), m.p. 189-190 °C (lit.[31] 186 °C), R_f 0.27 (CH_2Cl_2). IR(KBr) 3498-3100, 1424, 1235 and 799 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 5.17 (2H, s) and 7.17 (2H, s); ^{13}C-NMR ($CDCl_3$)

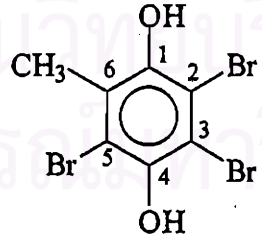
δ (ppm): 109.8 (2C, C-2 and C-5), 118.5 (2C, C-3 and C-6) and 146.7 (2C, C-1 and C-4).

2.3.22 *2,5-dibromo-p-benzoquinone* (Compound **13b**)[31]



A solution of iron (III) chloride hexahydrate (21.68 g, 0.08 mol) in 46 mL of water was added dropwise to a stirred, refluxing solution of **13a** (9.02 g, 0.03 mol) in 263 mL of water over a period of 15 min. The *p*-quinone which was crystallized immediately, was collected by filtration after cooling to RT, washed with water, and recrystallized from EtOH to give reddish-brown needles. The yield is 2.63 g (33%), m.p. 188-189 °C (lit.[31] 188 °C), R_f 0.62 (CH_2Cl_2). IR(KBr) 3303, 3057, 1675, 1588 and 681 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 7.47 (2H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 137.1 (2C, C-3 and C-6), 137.8 (2C, C-2 and C-5) and 176.9 (2C, C-1 and C-4).

2.3.23 *2,3,5,-tribromo-6-methylhydroquinone* (Compound **14a**)

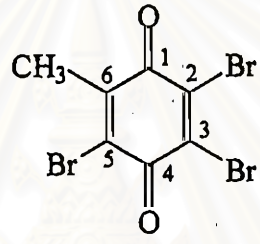


Bromine (32 g, 0.20 mol, 10.25 mL) in 10 mL of glacial acetic acid was added dropwise to a stirred suspension of toluhydroquinone (12.53 g, 0.10 mol) in 100 mL of glacial acetic acid by using a similar procedure to that mentioned for the preparation of Compound **13a**. The white solid was precipitated and collected, which then recrystallized from a mixture of hexane:ether (1:1) to give white powder, 8.48 g (30%), m.p. 205-206 °C, R_f 0.24 (hexane:ether (1:1)). IR(KBr) 3500-3100, 1393, 1296 and 676 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 2.40 (3H, s), 5.44 (1H, s) and 5.69

(1H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 17.2 (1C, CH_3), 108.2 (1C, C-6), 112.2 (1C, C-3), 113.1 (1C, C-2), 128.8 (1C, C-5), 144.4 (1C, C-4) and 145.4 (1C, C-1). Elemental analysis found %C 23.46 and %H 1.27; calcd. for $C_7H_5Br_3O_2$ (MW 360.83): %C 23.30 and %H 1.40.

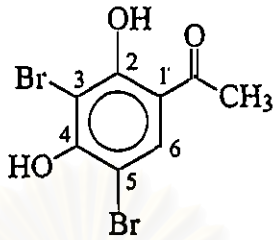
The IR, ^{1}H-NMR and ^{13}C-NMR spectra of this compound are exhibited in Figs. **A4-A6.**

2.3.24 *2,3,5-tribromo-6-methylbenzoquinone* (Compound **14b**)



Compound **14a** (5.15 g, 0.02 mol) was treated with a solution of iron (III) chloride hexahydrate (11.27 g, 0.04 mol) by employing a similar procedure to that described in Compound **13b** synthesis. The product was purified by recrystallization from a mixture of hexane:ether (1:1) to give reddish-brown needles, 4.21 g (82%), m.p. 201-202 °C, R_f 0.68 ($CHCl_3$). IR(KBr) 1685, 1659, 1577 and 676 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 2.33 (3H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 18.2 (1C, CH_3), 134.7 (1C, C-2), 137.8-139.5 (2C, C-5 and C-6), 146.1 (1C, C-3), 170.6 (1C, C-4) and 175.1 (1C, C-1).

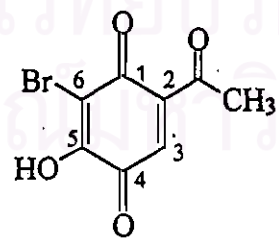
2.3.25 *3,5-dibromo-2,4-dihydroxyacetophenone* (Compound **15a**)



Bromine (16 g, 0.10 mol) in 5 mL of glacial acetic acid was added dropwise to a stirred suspension of 2,4-dihydroxyacetophenone (7.57 g, 0.05 mol) in 50 mL of glacial acetic acid using a similar procedure to that described for the preparation of Compound **13a**. The white solid was precipitated and collected, which was recrystallized from AcOH to yield white crystals, 8.68 g (76%), m.p. 164-165°C, R_f 0.62 (50% $CHCl_3$-MeOH). IR(KBr) 3500-3100, 2970 and 1630 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 2.61 (3H, s), 7.88 (1H, s) and 13.32 (1H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 26.3 (1C, CH_3), 98.9 (1C, C-3), 99.4 (1C, C-5), 115.2 (1C, C-1), 133.3 (1C, C-6), 155.6 (1C, C-2), 160.3 (1C, C-4) and 202.0 (1C, C=O). Elemental analysis found %C 31.52 and %H 2.05; calcd. for $C_8H_7BrO_3$ (MW 309.94): %C 31.00 and %H 1.95.

The IR, ^{1}H-NMR and ^{13}C-NMR spectra of this compound are exhibited in Figs. **A7-A9**.

2.3.26 *2-acetyl-6-bromo-5-hydroxybenzoquinone* (Compound **15b**)

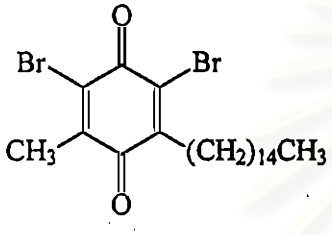


Compound **15a** (5.86g, 0.025 mol) was treated with a solution of iron (III) chloride hexahydrate (16.39 g, 0.06 mol) by using a similar procedure to that mentioned for the synthesis of Compound **13b**. The product was purified by recrystallization from ethanol to give rectangle crystals 4.99 g (86%), m.p. 173-174°C, R_f 0.57 (50% $CHCl_3$-MeOH). IR(KBr) 3500-3200 and 1650 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 2.61 (3H, s), 7.89 (1H, s) and 13.32 (1H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 26.3

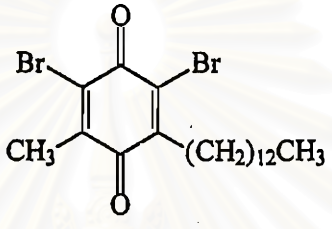
(1C, CH_3), 98.9 (1C, C-6), 99.4 (1C, C-2), 115.3 (1C, C-3), 133.4 (1C, C-5), 155.6 (1C, C-4), 160.3 (1C, C-1) and 202.0 (1C, C=O).

The IR, ^{1}H-NMR and ^{13}C-NMR spectra of this compound are exhibited in Figs. **A10-A12.**

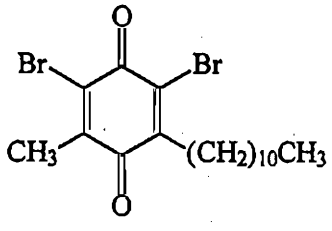
General procedure for the synthesis of Compounds **16 - 21** [25]



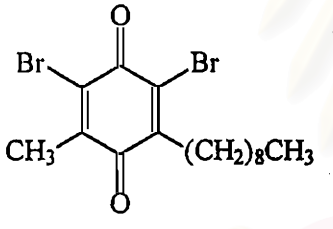
(16)



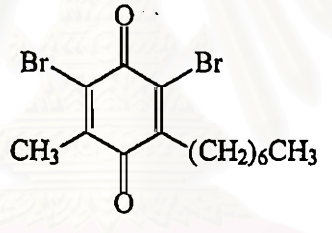
(17)



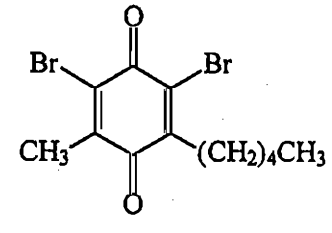
(18)



(19)



(20)



(21)

3,5-Dibromo-2-methyl-1,4-benzoquinone (0.56 g, 2 mmol) was placed in 30 mL of CH_3CN-H_2O (1:1). The fatty acid (4 mmol) and $AgNO_3$ (0.34 g, 2 mmol) were added to the solution, which then heated to 70-80°C and stirred. $(NH_4)_2S_2O_8$ (0.91 g, 4 mmol, in 10 mL H_2O) was added dropwise with stirring and continued to stir for another 1 h at 70-80°C, which then cooled and extracted with ether. The ether layer was washed with dilute $NaHCO_3$ solution and water, then dried over anhydrous Na_2SO_4 and concentrated in *vacuo*. The concentrate was purified by silica gel column chromatography, which was developed with a suitable solvent system. A yellow solution was collected and gave designed products.

2.3.27 *2,6-dibromo-3-methyl-5-pentadecyl-benzoquinone* (**16**): reddish-brown semisolid, 0.12 g (25%), R_f 0.70 (20% EtOAc-hexane), IR(KBr) 1654 and 1583 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 0.89 (3H, t, J = 6.72 Hz), 1.17-1.46 (26H, m) and 2.26 (3H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 14.1-37.8 (16C, $14CH_2$ and $2CH_3$), 133.8-134.0 (2C, C-2 and C-6), 146.1-149.9 (2C, C-3 and C-5), 172.4 (1C, C-1) and 181.2 (1C, C-4). Elemental analysis found %C 23.46 and %H 1.27; calcd. for $C_{22}H_{34}Br_2O_2$ (MW 490.32): %C 23.30 and %H 1.40.

The IR, ^{1}H-NMR and ^{13}C-NMR spectra of this compound are exhibited in Figs. **A13-A15**.

2.3.28 *2,6-dibromo-3-methyl-5-tridecyl-benzoquinone* (**17**): brown semisolid, 0.80 g (86%), R_f 0.56 (40% $CHCl_3$-hexane), IR(KBr) 1650 and 1589 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 0.90 (3H, t, J = 6.01 Hz), 1.18-1.24 (22H, m) and 2.30 (3H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 14.1-31.9 (14C, $12CH_2$ and $2CH_3$), 133.8-134.0 (2C, C-2 and C-6), 146.0-149.9 (2C, C-3 and C-5), 175.5 (1C, C-1) and 181.0 (1C, C-4).

2.3.29 *2,6-dibromo-3-methyl-5-undecyl-benzoquinone* (**18**): brown semisolid, 0.60 g (69%), R_f 0.58 (40% $CHCl_3$-hexane), IR(KBr) 1656 and 1590 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 0.90 (3H, t, J = 6.94 Hz), 1.18-1.25 (18H, m) and 2.30 (3H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 14.2-36.8 (12C, $10CH_2$ and $2CH_3$), 132.3-135.2 (2C, C-2 and C-6), 145.9-149.9 (2C, C-3 and C-5), 172.4 (1C, C-1) and 180.5 (1C, C-4).

2.3.30 *2,6-dibromo-3-methyl-5-nonyl-benzoquinone* (**19**): brown semisolid, 0.41 g (51%), R_f 0.54 (40% $CHCl_3$-hexane), IR(KBr) 1655 and 1598 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 0.90 (3H, t, J = 7.02 Hz), 1.17-1.30 (14H, m) and 2.25 (3H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 14.1-34.9 (10C, $8CH_2$ and $2CH_3$), 131.6-134.0 (2C, C-2 and C-6), 146.0-150.0 (2C, C-3 and C-5), 169.0 (1C, C-1) and 180.5 (1C, C-4).

2.3.31 *2,6-dibromo-3-methyl-5-heptyl-benzoquinone* (**20**): reddish-brown semisolid, 0.80 g (10%), R_f 0.48 (30% $CHCl_3$-hexane), IR(KBr) 1659 and 1587 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 0.90 (3H, t, J= 6.23 Hz), 1.20-1.46 (10H, m) and 2.30 (3H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 14.0-37.8 (8C, $6CH_2$ and $2CH_3$), 131.5-133.9 (2C, C-2 and C-6), 146.0-148.9 (2C, C-3 and C-5), 176.2 (1C, C-1) and 180.8 (1C, C-4).

2.3.32 *2,6-dibromo-3-methyl-5-pentyl-benzoquinone* (**21**): reddish-brown semisolid, 0.11 g (15%), R_f 0.46 (30% $CHCl_3$-hexane), IR(KBr) 1660 and 1592 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ (ppm): 0.90 (3H, t, J = 6.58 Hz), 1.18-1.26 (6H, m) and 2.28 (3H, s); ^{13}C-NMR ($CDCl_3$) δ (ppm): 14.2-35.0 (6C, $4CH_2$ and $2CH_3$), 131.5-134.0 (2C, C-2 and C-6), 145.9-150.0 (2C, C-3 and C-5), 172.6 (1C, C-1) and 179.7 (1C, C-4).

2.4 Bioassay experiments

2.4.1 Hatching the shrimp [27]

Brine shrimp eggs were hatched in a shallow rectangular dish (13x18 cm) filled with artificial sea water which was prepared by dissolving about 38 g of a commercial sodium chloride in 1 liter of distilled water. The shallow rectangular dish had two unequal compartments and had two little pores that made two parts joined. The eggs (*ca.* 30 mg) were sprinkled into the larger compartment which was darkened, while the smaller compartment was illuminated. After 24 hours the phototropic nauplii were collected by pipette from the light side, having been separated by the divider from their shells.

2.4.2 Preparation of test solution

Samples were prepared by dissolving 4 mg of interested compound in 80 μL of solvent and added artificial sea water to make 4000 μL (Solution **A**). Solution **B** was prepared by diluting 400 μL of Solution **A** with 3600 μL of artificial sea water. Then diluting 400 μL of Solution **B** with 3600 μL of artificial sea water to obtain Solution **C**. Concentrations of Solutions **A**, **B** and **C** were 1000, 100 and 10 μg/mL, respectively. Control solution was prepared by using 80 μL of solvent and make the volume of the solution to be 4000 μL by using artificial sea water.

2.4.3 Bioassay procedure[33]

Five shrimps in 100 μL artificial sea water were transferred to each well containing 400 μL of a tested solution. Each compound with 3 concentrations (Solutions **A**, **B** and **C**) was tested with six replications for each solution. All tests had control solution. The plates were covered and incubated at 22-29 °C for 24 hours. Plates were then examined under a binocular microscope and the numbers of dead (non-motile) nauplii in each well were counted. LC_{50} values at 24 hours were then calculated by Probit analysis, which showed in μg/mL in brine shrimp medium. The results of brine shrimp lethality tests of synthesized compounds are presented in Table 2.1.

Table 2.1 The LC_{50} value at 24 h of synthesized compounds

Compound	LC_{50} (μg/mL)	Bioactivity
1b	0.39994	High
2b	1.19669	High
3b	3.00263	High
4a	23.4510	Medium
4b	12.6080	Medium
5a	3.56320	High
5b	0.24241	High
6b	1478.99	No activity
7b	12003.4	No activity
8b	17.4008	Medium
9	6.84589	High
10	8.78579	High
11	20.4979	Medium
12	-	-
13a	18.8579	Medium
13b	19.6714	Medium

Table 2.1 (cont.)

Compound	LC_{50} (μg/mL)	Bioactivity
14a	1.96083	High
14b	1.52098	High
15a	1.50140	High
15b	2.14170	High
16	14.7906	Medium
17	64.6224	Medium
18	151.218	Low
19	239.684	Low
20	*	*
21	*	*

Note: * not test for BSLT

- High activity (LC_{50} < 10 μg/mL)
- Medium activity (LC_{50} < 100 μg/mL)
- Low activity (LC_{50} < 1000 μg/mL)
- No activity (LC_{50} > 1000 μg/mL)