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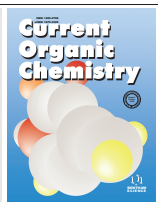
RESEARCH ARTICLE

A New Look at the Halogenation of Porphyrins



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Abstract: The present study describes the selective halogenation at the *β*- or *meso*-position of the porphyrin macrocycle. The reactions of deuteroporphyrin IX dimethyl ester, mesoporphyrin III dimethyl ester and their Cu(II) and Ni(II) complexes with *N*-bromosuccinimide or phenylselenyl chloride were investigated. It could be observed that when the bromination of deuteroporphyn IX dimethyl ester using NBS took place, the isolated products were the result of an electrophilic substitution in *β*-free porphyrin ring positions. Employing the same reagent for halogenating mesoporphyrin III dimethyl ester, which does not possess *β*-free position, different derivatives were obtained from the allylic bromination of the ethyl side chain. When phenylselenyl chloride was used as halogenating agent with deuteroporphyn IX dimethyl ester or its Cu(II) complex, in both cases the replacement of *β*-free hydrogen atoms by phenylselenyl group was afforded. When the Ni(II) mesoporphyrin III dimethyl ester was used the desired and selective replacement of *meso* hydrogen atoms of aromatic ring by chlorine atoms was obtained. The structural assignment of the porphyrin derivatives thus obtained was performed by high-resolution mass spectrometry and detailed analysis of the NMR spectra.

Keywords: Halogenation, deuteroporphyrin IX dimethyl ester, mesoporphyrin III dimethyl ester.

INTRODUCTION

In the general metabolism, endogenous heme is degraded by heme oxygenase (HO), which selectively oxidizes the *α* bridge position of the porphyrin ring to give biliverdin IX*α*, carbon monoxide and free iron [1]. Biliverdin is next reduced to bilirubin by biliverdin reductase. In adult organisms, this yellow pigment is conjugated with glucuronic acid to become water soluble and thus be excreted, into the intestinal tube together with other components of bile [2]. Although bilirubin is a powerful physiological antioxidant [3], at high concentrations it is neurotoxic, as it occurs in neonatal jaundice [4]. While the normal adult has the enzymes necessary to conjugate bilirubin, the newborn infant has not developed this mechanism completely [5].

To prevent the development of neonatal jaundice, the desirable task would be to achieve inhibition of HO. Several studies have shown that some metalloporphyrins are competitive inhibitors of HO. These metal complexes are derived from iron, zinc, chromium and tin [6–9]. The drawback is, that they are also inhibitors of other enzymes such as biliverdin reductase, the guanylyl cyclase and nitric oxide synthetase [10].

The aim of the present study was to synthesize a porphyrin that had, at least, the hydrogen position 5 of the porphyrin system replaced by a halogen (Scheme **1**) [11]. This porphyrin, recombined with the apoenzyme of the truncated human heme oxygenase-1(hHO-1) should act as an inhibitor rather than as substrate [11].

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To achieve this goal, we analyzed the halogenation of deuteroporphyrin IX dimethyl ester and mesoporphyrin III dimethyl ester. Both porphyrins were replaced with strings of propionic acid in positions 13 and 17 of the porphyrin macrocycle (Scheme **1**). This is an indispensable requirement to be substrate of the HO-1 from different origins [12-16].

Numerous research papers have described the bromination of the porphyrin ring at the *meso* positions using *N*-bromosuccinimide (NBS) [17-21]. However, the desired monobromo derivative is isolated in high yield if only three of the four *meso* carbons are substituted. When the halogenation takes place in porphyrins with two of their carbon substituted bridges with similar or identical residues, mono- and dibromo derivatives are obtained. However, when the bromination reaction is performed on *meso* monosubstituted porphyrins, mono-, di-, and tribromo derivatives are obtained and small amounts of the starting material are recovered.

RESULTS AND DISCUSSION

In the present study, the bromination of deuteroporphyrin IX dimethyl ester (**1**) was performed using NBS as reactant [20] under different experimental conditions. The results showed a mixture of mono-brominated compounds **2a**, **2b** and a dibrominated derivative **3**. In both cases the isolated products were the result of an electrophilic substitution in the *β*-free porphyrin ring positions (Scheme **1**).

To obtain the desired halogenated porphyrins at the *meso* positions, the bromination of a synthetic mesoporphyrin III dimethyl ester (**4**) [22] was performed using NBS under the same conditions described above. In this case, besides recovering the starting material (18%), different compounds **5** were obtained from the allylic bromination, being difficult to be isolated, since the release of hy-

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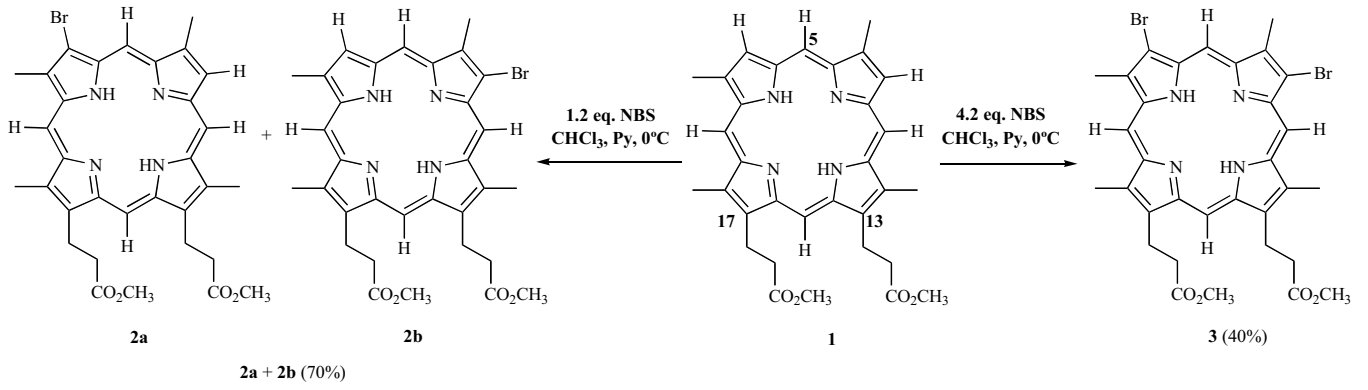
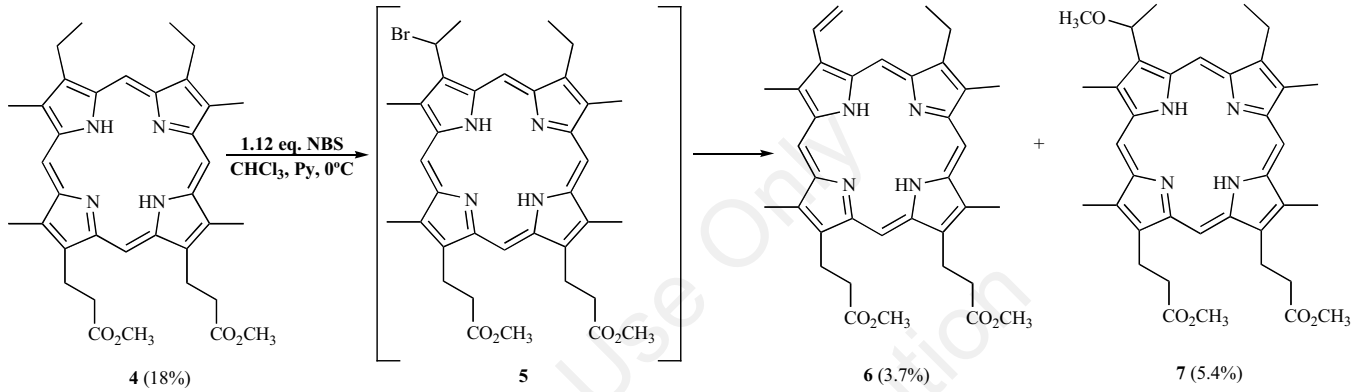


Scheme 1. Bromination of deuteroporphyrin dimethyl ester.



Scheme 2. Bromination of mesoporphyrin III dimethyl ester.

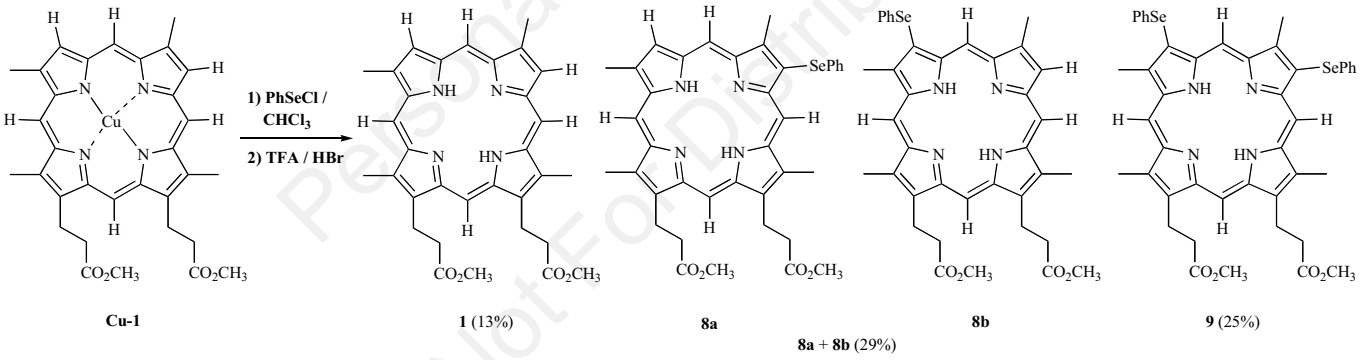


Fig. (1). Reaction of Cu(II) deuteroporphyrin IX dimethyl ester (1eq.) with PhSeCl (1.1 eq.).

drogen bromide or reaction with methanol during the isolation gave rise to vinyl **6** and methoxy **7** compounds, respectively (Scheme **2**).

Regarding these results, the reaction of Cu(II) and Ni(II) complexes obtained from porphyrin **4** was explored with phenylselenyl halides (PhSeX) [23]. These halogenating agents react by electrophilic aromatic substitution and are characterized by replacing the hydrogen at the *meso* carbons of the porphyrin ring at higher rates than those from the side chains [24]. The Cu(II) and Ni(II) complexes of the porphyrins studied were synthesized by the method previously described [25]. When the halogenation reaction was performed using Cu(II) deuteroporphyrins IX dimethyl ester (**Cu-1**) with PhSeCl (1:1.1), a mixture of porphyrins was obtained (Fig. **1**).

To analyze the structure of the Cu(II) porphyrins obtained, it was necessary to remove the paramagnetic ion with trifluoroacetic acid (TFA) and 48% aqueous HBr [26]. The isolated products were a mixture of 8-selenophenyl **8a,** 3-selenophenyl **8b** derivatives and 3,8-diselenophenyl deuteroporphyrin IX dimethyl ester (**9**) and deuteroporphyrin IX dimethyl ester **1**. When deuteroporphyrin IX dimethyl ester **1** was treated with PhSeCl (1:1.1) the same mixture of porphyrins was obtained. Also, the Ni(II) complex from compound **1** with PhSeCl in a relation 1.1:4 gave rise to a complex mixture of *β*-substituted porphyrins.

When the mesoporphyrin III dimethyl ester (**4**) was used with PhSeCl in a ratio 1:1.1, we isolated mostly starting material. However, the Ni(II)-complex of the mentioned porphyrin (**Ni-4**) was used with PhSeCl in a 1:4 ratio, the porphyrins thus obtained were purified by chromatography to give three fractions. The first band was a mixture of Ni(II) 5,10,15,20-tetrachloro mesoporphyrin III dimethyl ester **13** (66%) and Ni(II) 10,15,20-trichloro mesoporphyrin III dimethyl ester **12** (34%). The second band was Ni(II) dichloro mesoporphyrin III dimethyl ester **11a** or **11b** (75%) and the last band was Ni(II) 10-monochloro mesoporphyrin III dimethyl ester **10** (85%) (Fig. **2**). The structural assignment was carried out

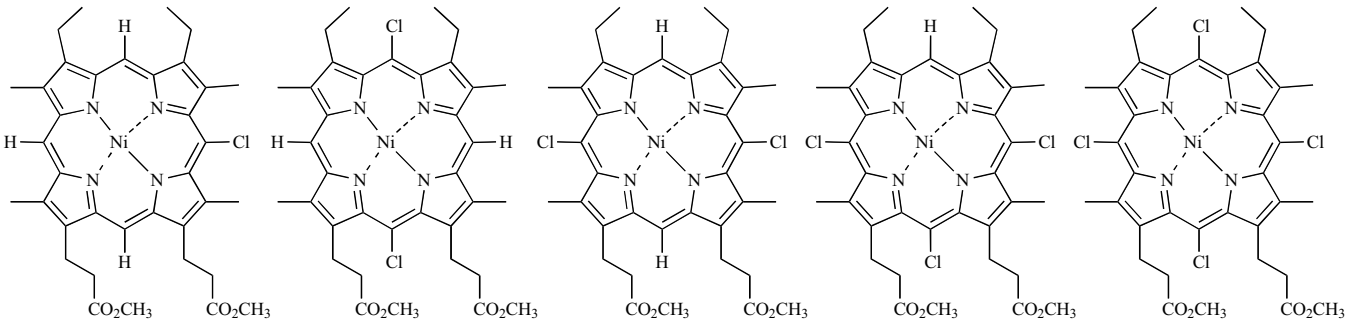
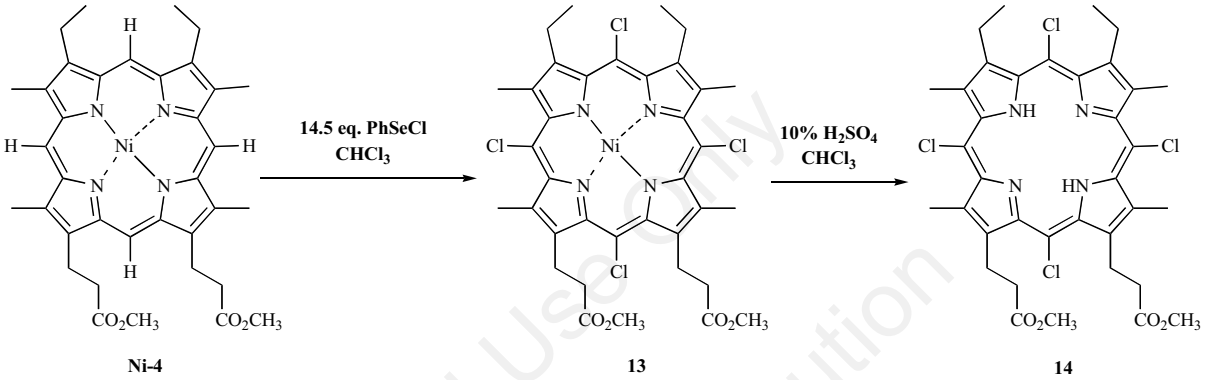


Fig. (2). Chlorination of Ni(II) mesoporphyrin III dimethyl ester (1 eq.) with PhSeCl (4 eq.).



Scheme 3. Synthesis of 5,10,15,20-tetrachloro mesoporphyrin III dimethyl ester.

by high-resolution mass spectrometry and detailed analysis of the 1D and 2D ^{1}HNMR spectra (Supporting information).

By using a halogenating agent: porphyrin ratio equal to 1:14.5, in anhydrous chloroform, we obtained a unique product, whose structure was assigned to the Ni(II) complex of a 5,10,15,20-tetrachloromesoporphyrin III dimethyl ester (**13)** as the main product. Furthermore, the treatment of **13** with 10% H_2SO_4 in dichloromethane [27], allowed obtaining the desired porphyrin 5,10,15,20-tetrachloromesoporphyrin III dimethyl ester **(14)** (Scheme **3**).

CONCLUSION

The present results allow us to conclude that the porphyrin structure, especially the methyl, ethyl and metoxycarbonylethyl β-substituted ones, leads the reaction mechanism of the halogenating reagent used and thus the halogenated positions as well as the product. It could be observed that when the bromination of deuteroporphyn IX dimethyl ester using NBS took place, the isolated products were the result of an electrophilic substitution in β-free porphyrin ring positions. Employing the same reagent for halogenating mesoporphyrin III dimethyl ester, which does not possess β-free position, different derivatives were obtained from the allylic bromination of the ethyl side chain. When phenylselenyl chloride was used as halogenating agent with deuteroporphyn IX dimethyl ester or its Cu(II) complex, in both cases the replacement of β-free hydrogen atoms by phenylselenyl group was afforded and this kind of derivatives were not reported in the literature. When the Ni(II) mesoporphyrin III dimethyl ester was used the desired and selective replacement of *meso* hydrogen atoms of aromatic ring by chlorine atoms was obtained. The experimental conditions determined the relative amounts of the synthesized porphyrins.

EXPERIMENTAL

1D and 2D NMR spectra were determined in $CDCl_3$ and were recorded at room temperature by means of a Bruker 300 MHz, Bruker 500 MHz and Bruker 600 MHz spectrometers. High-resolution mass spectra (HRMS) were obtained using the electrospray ionization (ESI) technique and Q-TOF detection (Bruker micrOTOF-Q II). Melting points were determined with a Fisher-Johns apparatus. All solvents and chemicals were of analytical grade and commercially available. Analytical TLC was performed on DC-Alufolien Kieselgel 60 F_{254} Merck.

1. Bromination of deuteroporphyrin dimethyl ester **1**.

1.1. Porphyrin **1** (26.93 mg, 0.050 mmol) was dissolved in 5 ml of chloroform at room temperature (25 °C). Pyridine (0.05 ml) was added to act as an acid scavenger. NBS (10.00 mg, 0.060 mmol) was added directly to the flask, and the reaction was followed by TLC. When the reaction reached completion, it was quenched with 0.5 ml of acetone. The solvents were evaporated under reduced pressure. Silica gel column chromatography (CH_2Cl_2-CH_3OH 0.5%) afforded the equimolecular mixture of **2a** (10.33 mg, 35.0 %) and **2b** (10.33 mg, 35.0%).

2a: ^{1}H NMR (500MHz, $CDCl_3$): δ = 3.22-3.28 [m, 4H, CH_2 (13^2, 17^2)]; 3.53-3.77 [m, 18H, CH_3 (2^1, 7^1, 12^1, 18^1), OCH_3 (13^5, 17^5)]; 4.31 [t, J = 7.5 Hz, 4H, CH_2 (13^1, 17^1)]; 9.11 [s, 1H, =CH (8)]; 9.92 [b, 1H, =CH (20)]; 9.92 [s, 1H, =CH (10)]; 9.99 [b, 1H, =CH (15)]; 10.17 [s,1H, =CH (5)].

2b: ^{1}H NMR (500MHz, $CDCl_3$): δ = 3.22-3.28 [m, 4H, CH_2 (13^2, 17^2)]; 3.53-3.77 [m, 18 H, CH_3 (2^1, 7^1, 12^1, 18^1), OCH_3 (13^5, 17^5)]; 4.44 [t, J = 7.5 Hz, 4H, CH_2 (13^1, 17^1)]; 9.09 [s, 1H, =CH (3)]; 9.92 [b, 1H,=CH (5)]; 9.99 [b, 1H, =CH (15)]; 10.02 [s,1H,=CH (20)]; 10.13 [s,1H, =CH (10)].

ESI-HRMS: Calcd for $C_{32}H_{34}BrN_4O_4$ $[M+H]^+$ 617.17579; found 617.17759.

1.2. Porphyrin **1** (26.93 mg, 0.050 mmol), was dissolved in 5 ml of chloroform at 25°C. NBS (35.00 mg, 0.210 mmol) and pyridine (0.05 ml) were added directly to the flask, and the reaction was followed by TLC. When the reaction reached completion it was quenched with 3ml of acetone. The solvents were evaporated under reduced pressure and the product was purified by column chromatography (silica gel, CH_2Cl_2-CH_3OH 10%) followed by crystallization (CH_2Cl_2-hexane) yielding the porphyrins **3** (13.90 mg, 40.0%); mp >270°C.

3: ^{1}H NMR (500MHz, $CDCl_3$): δ = 3.21 [t, *J* = 7.7 Hz, 4H, CH_2 (13^2, 17^2)]; 3.50 [s, 3H, CH_3 (7^1)], 3.52 [b, 6H, CH_3 (18^1, 2^1)]; 3.57 [s, 3H, CH_2 (12^1)]; 3.64 [s, 6H, OCH_3 (13^5, 17^5)]; 4.33 [t, *J* = 7.5 Hz, 4H, CH_2 (13^1, 17^1)]; 9.67 [s, 1H, =CH (20)]; 9.70 [s, 1H, =CH (5)]; 9.80 [s, 1H, =CH (15)]; 9.85 [s, 1H, =CH (10)].

ESI-HRMS: Calcd for $C_{32}H_{33}Br_2N_4O_4$ $[M+H]^+$ 695.08631; found 695.08434.

2. Bromination of mesoporphyrin III dimethyl ester **4.**

Porphyrin **4** (29.74 mg, 0.050 mmol) was dissolved in 25 ml of chloroform at 25°C. Pyridine (0.25 ml) was added. NBS (9.88 mg, 0.056 mmol) was added directly to the flask. The reaction was stirred at room temperature for 1.5 h. Then, it was quenched with 2.5 ml of acetone. The solvents were evaporated in vacuum. The product was purified by column chromatography (silica gel, CH_2Cl_2-CH_3OH 10%) to give three principal porphyrins, mesoporphyrin III dimethyl ester **4** (5.4 mg, 18.0 %), 3-(α-methoxyethyl)-7-ethyl-2,8,12,18-tetramethyl-14,17-(β-methoxycarbonylethyl) porphyrin **7** (1.7 mg, 5.4 %) and 3-vinyl-7-ethyl-2,8,12,18-tetramethyl-14,17-(β-methoxycarbonylethyl) porphyrin **6** (1.1 mg, 3.7 %).

4: ^{1}H NMR (500MHz, $CDCl_3$): δ = 1.86 [t, *J* = 8.2 Hz, 6H, CH_3 (3^2, 7^2)]; 3.29 [t, *J* = 7.9 Hz, 4H, CH_2 (13^2, 17^2)], 3.62 [s, 6H, OCH_3 (13^5, 17^5)]; 3.65 [s, 12H, CH_3 (2^1, 8^1, 12^1, 18^1)]; 4.09 [c, *J* = 8.0 Hz, 4H, CH_2 (3^1, 7^1)]; 4.43 [t, *J* = 7.9 Hz, 4H, CH_2 (13^1, 17^1)]; 10.08 [s, 1H, =CH (5)]; 10.09 [s, 1H, =CH (15)]; 10.10 [s, 2H, =CH (10, 20)].

ESI-HRMS: Calcd for $C_{36}H_{43}N_4O_4$ $[M+H]^+$595.32788; found 595.32903.

7: ^{1}H NMR (300MHz, $CDCl_3$): δ = 1.87 [t, *J* = 8.1 Hz, 3H, CH_3 (7^2)]; 2.26 [d, *J* = 6.6 Hz, 3H, CH_3 (3^2)]; 3.28-3.33 [m, 4H, CH_2 (13^2, 17^2)], 3.62-3.68 [m, 21H, OCH_3 (3^3, 13^5, 17^5), CH_3 (2^1, 8^1, 12^1, 18^1)]; 4.07 [c, *J* = 8.1 Hz, 2H, CH_2 (7^1)]; 4.37-4.48 [m, 4H, CH_2 (13^1, 17^1)]; 5.96-6.03 [m, 1H, -CH (3^1)]; 10.09 [b, 2H, =CH]; 10.14 [s, 1H, =CH]; 10.57 [s, 1H, =CH].

ESI-HRMS: Calcd for $C_{37}H_{45}N_4O_5$ $[M+H]^+$ 625.33845; found 625.33894.

6: ^{1}H NMR (500MHz, $CDCl_3$): δ = 1.89 [t, *J* = 7.4 Hz, 3H, CH_3 (7^2)]; 3.28-3.35 [m, 4H, CH_2 (13^2, 17^2)], 3.64-3.75 [m, 18H, OCH_3 (13^5, 17^5), CH_3 (2^1, 8^1, 12^1, 18^1)]; 4.13 [c, *J* = 7.4 Hz, 2H, CH_2 (7^1)]; 4.39-4.51 [m, 4H, CH_2 (13^1, 17^1)]; 6.20 [d, *J* = 12.4 Hz, 1H, CH_2 (3^2)]; 6.39 [d, *J* = 17.4 Hz, 1H, CH_2 ($3^{2'}$)]; 8.32 [dd, *J* = 11.2 Hz and 17.3 Hz, 1H, =CH (3^1)]; 10.11 [s, 2H, =CH]; 10.17 [s, 1H, =CH];10.25 [s, 1H, =CH].

ESI-HRMS: Calcd for $C_{36}H_{41}N_4O_4$ $[M+H]^+$ 593.31223; found 593.31123.

3. Chlorination of Cu(II) deuteroporphyrin IX dimethyl ester (**Cu-1**).

A mixture of Cu(II) deuteroporphyrin IX dimethyl ester (30.00 mg, 0.050 mmol) and PhSeCl (15.5 mg, 0.055 mmol) in 5 ml of $CHCl_3$ was stirred at room temperature for 50 h. The solvents were evaporated under reduced pressure. Silica gel column chromatography (CH_2Cl_2-CH_3OH 0.5%) followed by recrystallization (CH_2Cl_2-hexane) yielded 30 mg of a solid mixture of Cu(II) complex of porphyrins. Then, TFA (9.74 ml) was added to a well stirred solution of Cu(II) complex of porphyrins in $CHCl_3$ (9.74 ml) at room temperature. Then, three drops of 48% aqueous HBr were added and stirred for 5 min. The reaction mixture was washed five times with water and then with a saturated aqueous $NaHCO_3$ solution until the organic phase turned neutral. The organic layer was dried with anhydrous Na_2SO_4 and the chloroform evaporated. The product was purified by column chromatography (silica gel, CH_2Cl_2-CH_3OH 0.5%) to give three principals porphyrins, deuteroporphyrin IX dimethyl ester **1** (4.60 mg, 17.3%), a mixture of 8-selenophenyl and 3-selenophenyldeuteroporphyrin IX dimethyl ester (**8a** and **8b)** (5.90 mg, 17.0%) and 3,8-diselenophenyldeutero-porphyrin IX dimethyl ester **9** (4.00 mg, 9.4%).

9: ^{1}H NMR (300MHz, $CDCl_3$): δ = 3.72 [b, 2H, NH]; 3.29 [t, *J* = 7.3 Hz, 4H, CH_2 (13^2, 17^2)]; 3.55-3.76 [m, 18 H, CH_3 (2^1, 7^1, 12^1, 18^1), OCH_3 (13^5, 17^5)]; 4.37-4.46 [m, 4H, CH_2 (13^1, 17^1)]; 7.07-7.09 [b, 6H, =CH (3^3, 3^5, 8^3, 8^5)]; 7.07-7.09 [b, 4H, =CH (3^4, 8^4,)]; 10.03 [s, 1H, =CH]; 10.09 [s, 1H, =CH]; 10.37 [s, 1H,=CH]; 10.43 [s, 1H, =CH].

ESI-HRMS: Calcd for $C_{44}H_{43}N_4O_4Se_2$ $[M+H]^+$ 851.16093; found 851.16849.

8a: (58%): ^{1}H NMR (600MHz, $CDCl_3$): δ = 3.25-3.30 [m, 4H, CH_2 (13^2, 17^2)]; 3.55 [s, 3H, CH_3 (7^1)]; 3.64 [s, 6H, OCH_3 (13^5, 17^5)]; 3.66 [s, 3H, CH_3 (2^1)]; 3.67 [s, 3H, CH_3 (12^1)]; 3.69 [s, 3H, CH_3 (18^1)]; 4.33-4.36 [m, 4H, CH_2 (13^1,17^1)]; 7.06-7.07 [b, 3H, =CH (8^3, 8^5)]; 7.40-7.43 [b, 2H, =CH (8^4)]; 9.07 [s, 1H, =CH (3)]; 9.95 [s, 1H, =CH (5)]; 10.01 [s,1H, =CH (15)]; 10.06 [s, 1H, =CH (10)]; 10.42 [s, 1H, =CH (20)].

8b: (42%): ^{1}H NMR (600MHz, $CDCl_3$): δ = 3.25-3.30 [m, 4H, CH_2 (13^2, 17^2)]; 3.57, 3.58 [s, 3H, s 3H, CH_3 (18^1,12^1]; 3.67 [s, 3H, CH_3 (2^1)]; 3.68 [s, 6H, OCH_3 (13^5, 17^5)]; 3.76 [s, 3H, CH_3(7^1)]; 4.40-4.46 [m, 4H, CH_2 (13^1,17^1)]; 7.06-7.07 [b, 3H, =CH (3^3, 3^5)]; 7.40-7.43 [b, 2H, =CH (3^4)]; 9.11 [s, 1H, =CH (8)]; 10.01 [s, 1H, =CH (15)]; 10.03 [s,1H, =CH (20)]; 10.04 [s, 1H, =CH (5)]; 10.40 [s, 1H, =CH (10)].

ESI-HRMS: Calcd for $C_{38}H_{39}N_4O_4Se$ $[M+H]^+$ 695.21310 ; found 695.19863.

4. Chlorination of Ni(II) mesoporphyrin III dimethyl ester (**Ni-4**).

4.1. A mixture of Ni(II) mesoporphyrin III dimethyl ester (7.70 mg, 0.012 mmol) and PhSeCl (9.22 mg, 0.047 mmol) in anhydrous $CHCl_3$ (1.5 ml) was stirred at room temperature for 2.5 h. The mixture was diluted with CH_2Cl_2 (10 ml) and washed twice with water (10 ml). The solution was dried (anhydrous Na_2SO_4), filtered, and the solvent was evaporated under reduced pressure. The product was purified by column chromatography (silica gel, hexane-ethyl acetate 20%) to give three fractions. The first band was a mixture of Ni(II) 5,10,15,20-tetrachloro mesoporphyrin III dimethyl ester **13** (66%) and Ni(II) 10,15,20-trichloro mesoporphyrin III dimethyl ester **12** (34%). The second band was Ni(II) dichloro mesoporphyrin III dimethyl ester **11a** or **11b** (75%) and the last band was Ni(II) 10-monochloro mesoporphyrin III dimethyl ester **10** (85%).

13: ^{1}H NMR (600MHz, $CDCl_3$): δ = 1.54-1.63 [m, 6H, CH_3 (3^2,7^2)]; 2.93-2.96 [m, 4H, CH_2 (13^2, 17^2)], 3.25, 3.27 [s, 6H, s, 6H,

CH_3 (2^1, 8^1, 12^1, 18^1)]; 3.75-3.82 [m, 10H, CH_2 (3^1,7^1), OCH_3 (13^5, 17^5)]; 4.10-4.15 [m, 4H, CH_2 (13^1,17^1)].

ESI-HRMS: Calcd for $C_{36}H_{36}$ Cl_4N_4 NiO_4 $[M]^+$ 786.08387; found 786.08565.

12: ^{1}H NMR (500MHz, $CDCl_3$): δ = 1.54-1.63 [m, 6H, CH_3 (3^2, 7^2)]; 2.99-3.02 [m, 4H, CH_2 (13^2, 17^2)], 3.33, 3.37 [s, 6H, s, 6H, CH_3 (2^1, 8^1, 12^1, 18^1)]; 3.63 [c, *J* = 7.50 Hz, 4H, CH_2 (3^1, 7^1)]; 3.75-3.82 [m, 6H, OCH_3 (13^5, 17^5)]; 4.19-4.23 [m, 4H, CH_2 (13^1,17^1)]; 8.96 [s, 1H, =CH (1)].

ESI-HRMS: Calcd for $C_{36}H_{37}$ $Cl_3N_4NiO_4$ $[M]^+$ 752.12284; found 752.12356.

11: ^{1}H NMR (500MHz, $CDCl_3$): δ = 1.63-1.66 [m, 6H, CH_3 (3^2, 7^2)]; 3.00-3.03 [m, 4H, CH_2 (13^2, 17^2)], 3.42, 3.44 [s, 6H, s, 6H, CH_3 (2^1, 8^1, 12^1, 18^1)]; 3.71-3.74 [m, 10H, CH_2 (3^1, 7^1), OCH_3 (13^5, 17^5]; 4.06-4.09 [m, 4H, CH_2 (13^1, 17^1)]; 9.21, 9.28 [s, 1H, s,1H, =CH].

ESI-HRMS: Calcd for $C_{36}H_{38}$ Cl_2N_4 NiO_4 $[M]^+$ 718.16181; found 718.16148.

10: ^{1}H NMR (500MHz, $CDCl_3$): δ = 1.68-1.73 [m, 6H, CH_3 (3^2, 7^2)]; 3.05-3.13 [m, 4H, CH_2 (13^2, 17^2)], 3.36, 3.39, 3.51, 3.53 [s, 3H, s, 3H, s, 3H, s, 3H, CH_3 (2^1, 8^1, 12^1, 18^1)]; 3.69-3.71 [m, 6H, OCH_3 (13^5 , 17^5]; 3.80-3.83 [m, 4H, CH_2 (3^1,7^1)]; 4.13-4.19 [m, 4H, CH_2 (13^1,17^1)]; 9.45-9.50 [m, 3H, =CH].

ESI-HRMS: Calcd for $C_{36}H_{39}ClN_4NiO_4$ $[M]^+$ 684.20078; found 684.20026.

4.2. A mixture of Ni(II) mesoporphyrin III dimethyl ester (7.19 mg, 0.011 mmol) and PhSeCl (32.00 mg, 0.160 mmol) in $CHCl_3$ anhydrous (3.5 ml) was stirred at room temperature for 3 h.The mixture was diluted with CH_2Cl_2 (15 ml) and washed twice with water (15 ml). The solution was dried (anhydrous Na_2SO_4), filtered, and the solvent was evaporated under reduced pressure. The crude product was subjected to column chromatography (silica gel, hexane-ethyl acetate 20%) to give **13** which was crystallized from CH_2Cl_2-CH_3OH to yield 7.90 mg (91.3%); mp 174-176°C.

ESI-HRMS: Calcd for $C_{36}H_{36}$ $Cl_4N_4NiO_4$ $[M]^+$ 786.08387; found 786.08412.

The Ni(II) 5,10,15,20-tetrachloro mesoporphyrin III dimethyl ester **13** (7.00 mg, 0.010 mmol) was treated with 5 ml of a mixture of 10% H_2SO_4 in dichloromethane. The reaction mixture was stirred at room temperature for 15 min and then neutralized with a saturated solution of Na_2CO_3. The aqueous phase was extracted with CH_2Cl_2, and the organic phase was dried (anhydrous Na_2SO_4) and evaporated under vacuum to dryness. The crude product was subjected to column chromatography (silica gel, CH_2Cl_2-CH_3OH 1%) to give **14** which was crystallized from CH_2Cl_2-CH_3OH to yield 2.90 mg (44.5%); mp 137-139°C.

14: ^{1}H NMR (600MHz, $CDCl_3$): δ = -1.26 [s, 2H, NH]; 1.52 [t, *J* = 7.5Hz, 6H, CH_3 (3^2,7^2)]; 2.92-2.95 [m, 4H, CH_2 (13^2, 17^2)], 3.05 [s, 6H, CH_3 (2^1, 8^1)]; 3.09 [s, 6H, CH_3 (12^1,18^1)]; 3.55 [c, *J* = 7.4 Hz, 4H, CH_2 (3^1,7^1)]; 3.71 [s, 6H, OCH_3 (13^5, 17^5)]; 3.87-3.90[m, 4H, CH_2 (13^1,17^1)].

ESI-HRMS: Calcd for $C_{36}H_{39}Cl_4N_4O_4$ $[M+H]^+$ 731.17199; found 731.16937.

CONFLICT OF INTEREST

The authors confirm that this article content has no conflict of interest.

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SUPPLEMENTARY MATERIAL

Supplementary material is available on the publishers Web site along with the published article.

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