New copper porphyrins as functional models of catechol oxidase

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Abstract
In recent years, many research groups, including ours, have focused their attention on the development of synthetic and functional models of biological systems based on copper. In this work, we report the synthetic strategy, characterization and catalytic activity of new copper porphyrins (CuP2 and CuP3) obtained by metallation of the corresponding tetra-ethylenediamine and tetra-N-tosylethylenediamine derivatives of 5,10,15,20-tetrakis(pentafluorophenyl)porphyrin (H2P1). CuP2 allows the formation of structured and very insoluble solid (CuP2S) after metallation with an excess (10 equiv.) of Cu(II) ions, which were explored in oxidative heterogeneous catalysis of 3,5-di-tert-butylcatechol and catechol. The catalytic activity of this copper coordination polymer...
was compared with their precursor CuP2 and CuP3, not only in heterogeneous phase, but also under homogeneous conditions, using dioxygen or hydrogen peroxide as oxidants. The obtained results show that the new copper porphyrins (CuP2 and CuP3) and the CuP2S material are able to efficiently mimic the activity of catecholase, being the last one easily reused and active form more than 3 catalytic cycles.

1. Introduction

Green chemistry approaches to obtain products of commercial interest represent an important challenge for a sustainable development. A well-known pillar of green chemistry is based on the use of catalysts, generally resulting in significant gains to a chemical reaction in terms of overall efficiency [1]. In this area, metalloporphyrins are efficient catalysts for the oxidative insertion of oxygen in hydrocarbons and also in a wide variety of organic compounds with a great efficacy [2-4]. In this context, several studies have already been performed with iron and manganese porphyrins [2, 5, 6] showing the great efficacy of these compounds as models of cytochrome P-450 [5, 7-11]. Besides of that they have been also used for the preparation of oxidized drug metabolites in the context of the pharmaceutical industry. Although effective in homogeneous systems, particularly for epoxidation and hydroxylation of hydrocarbons, some problems are frequently reported with the use of these complexes in solution, such as deactivation of the catalytic species by secondary reactions which often leads to a low catalytic efficiency or, depending on the structure of the porphyrin ligand, destructive oxidation [11]. To minimize these problems, there has been much effort invested in the synthesis of new porphyrins that do not suffer such effects [12, 13]. Halogenated metalloporphyrin catalysts have been shown to be more robust, and more resistant to degradation by free radical attack than those
substituents containing electron donors. Because of that, porphyrin H₂TPPF₂₀ (H₂P₁, Scheme 1) is a suitable platform for the synthesis of new robust porphyrins, by simple nucleophilic aromatic substitution of the para fluorine atoms of the pentafluorophenyl groups by appropriate nucleophiles.

Metalloporphyrins containing other metals, such as vanadium, nickel, zinc, copper, among others have also been investigated for different purposes over the last few years [REF]. The biological essentiality of copper element is long recognized, however, only in recent decades has been studied by many research groups in the development of synthetic/functional models of biological systems important for the maintenance of life [14-16]. Proteins containing copper as active center are usually involved as redox catalyst in many biological processes in electron transfer or oxidation of a variety of organic substrates. One of them is the Catechol oxidase (COx), where the active center is a binuclear copper complex, which catalyzes the oxidation of a variety of o-diphenols (catechols), such as caffeic acid and their derivatives to the corresponding o-quinone, in a process known as catecholase. The resulting quinones are highly reactive to auto-polymerization leading to the formation of a brown polyphenolic pigment, i.e. melanin, a process thought to protect the skin cells from UV radiation damage, reducing the risk of skin cancer against pathogens or insects.

The importance of oxidation of catechol is associated with the respective quinones, since several quinones have great importance in several scientific and technological areas like in materials chemistry. In medicine, they present several biological activities, such as antitumor, anti-inflammatory, anti-oxidant, among others [37].

Over the years different functional and structural models, based on mono- and binuclear copper complexes, have been reported, however there are few reports of
copper porphyrins and phthalocyanines as catalysts for oxidation of catechols. Meunier et al. showed that iron phthalocyanines are active in the oxidation of catechol while Cu(II) phthalocyanine are inactive [45]. Some of us [13] reported recently that the copper porphyrins are also able to oxidize catechol very efficiently, in the presence of hydrogen peroxide and under homogeneous or heterogeneous conditions.

Also, a new classes of materials, like coordination polymers (CPs), have been recently emerging, as an attractive approach to prepare robust and efficient catalysts [24-27]. In this area, porphyrins with appropriate functional groups on the periphery of the porphyrin ring, can constitute efficient building blocks for the design and synthesis of novel CPs. The connectivity is driven by coordinative chemistry between the functional group of the linker and metal centers, and can extend the structures into one, two or three dimensions [28] [24]. In fact, porphyrin derivatives containing amine, carboxylate, pyridine, phosphonate and sulfonate substituents tend to form CPs [9, 24, 26, 29, 30]. Such behavior is attributed to the affinity of these substituents of the porphyrin ring to metal ions, forming supramolecular insoluble clusters [24].

Based on the efficiency and selectivity of metalloporphyrins and corresponding hybrid materials for oxidation reactions associated with the presence of copper in catechol oxidase, which is the responsible for the catalysis process, we report here the synthetic strategy, characterization and catalytic activity of new copper materials based on the porphyrin $H_2TPPF_{20} (H_2P1$, Scheme 1) scaffold. The nucleophilic aromatic substitution of the para fluorine atoms of the pentafluorophenyl groups by ethylenediamine and $N$-tosylethlenediamine and subsequent insertion of copper(II), afforded the corresponding CuP2 and CuP3 porphyrins, respectively, and the insoluble solid material CuP2S. The two new metalloporphyrins and the hybrid material are able
to mimic the activity of catecholase using hydrogen peroxide or oxygen, from the air, as oxidants.

![Scheme 1](image)

i) NMP, 800 W, 1 bar, 200 °C; ii) Cu(AcO)₂ (1 equiv.), DMF, 120 °C, 24 h; iii) Cu(AcO)₂ (10 equiv.), DMF, 120 °C, 24 h.

**Scheme 1**

2. Experimental

All chemicals used in this study were purchased from Sigma-Aldrich or Merck and were of analytical grade. HRMS were recorded on an APEXqe FT-ICR mass spectrometer (Bruker Daltonics, Billerica, MA). Electronic spectra (UV-VIS) were obtained on a Shimadzu UV-2501PC spectrophotometer, in the 350-800 nm range. For the solids diffuse reflectance characterization an UV-VIS Jasco V560 spectrophotometer was used. Attenuated total reflectance Transmission Fourier Transform Infrared (ATR-FTIR) spectra were registered on a FT Mattson 7000 galaxy series spectrophotometer in the 400-4000 cm⁻¹ range, the spectra were collected with a resolution of 4 cm⁻¹ and accumulation of 32 scans. For the X-ray diffraction (PXRD) measurements, self-oriented solids were placed on neutral glass sample holders. The measurements were performed in the
reflection mode using X'Pert MPD Philips diffractometer (Cu Kα1,2 X-radiation, λ1 = 1.540598 Å and λ2 = 1.544426 Å), equipped with an X'Celerator detector and a flat-plate sample holder in a Bragg–Brentano para-focusing optics configuration (40 kV, 50 mA). Intensity data were collected by the step counting method (step 0.02), in continuous mode. Electron paramagnetic resonance (EPR) spectra of the solid materials were recorded on an EPR Bruker EMX micro X spectrometer operating in the X band (approximately 9.5 GHz) at 77 K using liquid N2. Dynamic Light Scattering (DLS) measurements were performed in a Zeta Sizer Nano Series (Malvern).

2.1 Synthesis and characterization

2.1.1 Synthesis of copper porphyrins CuP2 and CuP3

The copper porphyrins CuP2 and CuP3 (Scheme 1) were obtained from the respective free-bases H2P2 and H2P3 already described in the literature [17], using the modified Kobayashi method [18]. The metallation reactions were performed with copper acetate in dimethylformamide (DMF) using stoichiometric ratio of 1:1 (mol:mol) for CuP2 and an excess for CuP3. The reactions were performed at 120 °C for 24 hours under magnetic stirring, and followed by recording the electronic spectra at different reaction times. Then, the solvent was removed under reduced pressure and the desired copper porphyrins CuP2 and CuP3 were purified by column chromatography (silica gel) using dichloromethane: methanol as eluent. After isolation, they were characterized by UV-VIS, infrared (ATR-FTIR) and electron paramagnetic resonance (EPR) spectroscopies and HRMS.

CuP2 (C52H36F16N12Cu): UV-VIS (DMF) λmax, nm (log e): 417 (4.54) and 540 (4.03), HRMS-ESI: calcd. for C52H37F16N12Cu [M+H]+ 1196.22600 found 1196.22591;
CuP3 \((C_{80}H_{60}F_{16}N_{12}O_{8}S_{4}Cu)\) UV-VIS (DMF) \(\lambda_{\text{max}}\), nm (log \(\varepsilon\)): 419 (5.10) and 539 (4.32).

HRMS-ESI: calcd. for \(C_{80}H_{61}F_{16}N_{12}O_{8}S_{4}Cu\) [M+H]\(^{+}\) 1812.26140 found 1809.51772;

ESI-MS/MS: calcd. for \(C_{80}H_{61}F_{16}N_{12}O_{8}S_{4}Cu\) [M+H+Na]\(^{+}\) 1835.25 found 1835.15; (see Figures SI1-4).

2.1.2 Synthesis of the coordination polymer CuP2S

The insoluble CuP2S hybrid was prepared by reacting directly \(H_2P2\) with 10 equivalents of copper(II) acetate using the previous conditions. At the end of the reactions, it was observed the precipitation of the desired solid CuP2S that was washed with different solvents (chloroform, dichloromethane, DMF, THF, water and acetone) in order to remove/prevent any contamination from free-base porphyrin \(H_2P2\) or copper(II) acetate. Subsequently, the solid was dried in air at room temperature and was characterized using different techniques.

2.1.3 Single-Crystal X-ray Diffraction Studies of CuP3

Purple crystals of porphyrin CuP3 were obtained by dissolving the porphyrin in a small amount of chloroform and methanol (1:1) and leaving the resulting mixture under slow evaporation at ambient temperature over a period of four days.

2.2 Catalytic Studies

The catalytic activity of the new copper porphyrins was investigated in the oxidation of catechol and 3,5-di-\textit{tert}-butylcatechol (3,5-DTBC) using \(O_2\) or \(H_2O_2\) as oxidants, under homogeneous (CuP2 or CuP3) and heterogeneous (CuP2S) conditions.
2.2.1 Oxidation of 3,5-di-tert-butylcatechol under O$_2$ from the air

In a typical reaction, the solid catalyst and the substrate (3,5-DTBC) were placed in a 10 mL reaction flask, dissolved in 9.0 mL of methanol. Then, 1.0 mL of phosphate buffer (pH = 8.0) was added to the flask. The reaction was carried out in the dark for different reaction times (between 15 min to 2 h) under constant stirring and in a thermostatic bath at 30 °C. The kinetics of the oxidation of 3,5-DTBC was determined by the growing band at 400 nm of the product 3,5-di-tert-butyl-o-benzoquinone (3,5-DTBQ), monitored by UV-VIS spectroscopy at different time intervals.

2.2.2 Oxidation of Catechol under H$_2$O$_2$ and O$_2$ from the air

The catalytic activity of CuP2, CuP2S and CuP3 was evaluated through the oxidation of the substrate catechol in water. In a typical reaction, the solid catalyst and the substrate (catechol) were placed in a 10 mL volumetric flask. Then, 7.1 mL of phosphate buffer (pH = 8.0) and 30 µL of hydrogen peroxide (30% solution v/v) were added to the flask (CuPx:H$_2$O$_2$:catechol molar ratio = 1:500:100) The reaction was carried out in the dark for different reaction times (between 15 min to 24 h) under constant stirring and in a thermostatic bath (30 °C).

Similar procedure was performed in absence of hydrogen peroxide using oxygen from the air. In this case, the reactions were made in open reaction flask to ensure the presence of oxygen.

The progress of the catalytic reactions were monitored by UV-VIS spectroscopy and quantified using the nitrite method (band at 508 nm, $\varepsilon = 13.536$ L mol$^{-1}$ cm$^{-1}$) [19]. Nitrite method is based on the fact that catechol (incolor substance), when combined with nitrous acid becomes yellow. The yellow color changes to an intense pink-red in the presence of excess sodium hydroxide (hydrogen ions from the phenolic OH groups
react with nitrite ions from the sodium nitrite, the molecular nitrous acid then forming NO compounds in presence of phenols), being the absorbance intensity directly correlated to the present catechol concentration. The same reactions were also performed using chloroform as solvent (2.0 mL) and phosphate buffer (8.0 mL) in a two phase experiments.

3. Results and Discussion

**Synthesis of copper porphyrins CuP2 and CuP3**

The new **CuP2** and **CuP3** metalloporphyrins were prepared from the reaction of the corresponding free-bases **H2P2** and **H2P3** derivatives [17] and copper acetate (Scheme 1). After the workup they were isolated, almost in quantitative yields, and fully characterized. The UV-VIS spectra of **CuP2** and **CuP3** present the characteristic Soret band at 418 and 420 nm respectively, with a small red shift in comparison to the Soret band of the free-base porphyrins and two Q-bands at 540 and 616 or 620 nm, resulting from the alteration in the micro symmetry of the porphyrin macrocycle from D2h to D4h, due to the presence of the Cu(II) ion in the porphyrin core (Figures SI1 and SI2).

The FTIR spectra exhibit the typical bands of metalloporphyrins, with bands at 3405 and 3313 cm⁻¹, ascribed to symmetric and asymmetric N-H stretching, and at 2955 and 2812 cm⁻¹, belonging to CH (phenyl) and CH (pyrrole) stretching (Figure SI5). The bands at 1600 cm⁻¹ were attributed to symmetrical angular deformation in the N-H plane of the pyrrole ring; δ N–H (in planarity) and δ N–H (out of planarity) appear at 960 cm⁻¹ and 752 cm⁻¹, among other bands related with skeletal ring vibrations. The bands in **H2P3** spectrum at 1320 and 1153 cm⁻¹ can be related to the S=O groups [20].

Since EPR analysis is a good tool to investigate copper complexes, providing valuable information with respect to it oxidation state, ligand type and symmetry, we
performed the EPR spectra of CuP2 and CuP3 (Figure 1). As expected, they show typical EPR spectra of Cu(II), with a multiple lines pattern in the particular spectrum region, being the expected deployment of each signal in nine lines. This pattern is attributed to a super-hyperfine interaction of the copper ion unpaired electron with the four magnetically equivalent 14N nucleus (I=1) [21]. In fact, g∥ is higher than g⊥ for both CuP derivatives, indicating that the unpaired electron of copper is occupying the orbital dx^2-2, corresponding to the equatorial plane of Cu(II) with elongated tetragonal symmetry [22]. The g∥ and g⊥ values, along with other parameters, are in conformity with a square planar geometry for copper(II) porphyrins (CuP2 (g∥ = 2.20; g⊥ = 2.06, A∥/Cu = 206 and A⊥ = 17.8) and CuP3 (g∥ = 2.24; g⊥ = 2.05, A∥/Cu = 200 and A⊥ = 17.1) [23].

![EPR spectra](image)

**Figure 1.** EPR spectra in acetone solution at 77 K: a) CuP2 and b) CuP3.

**Synthesis of the solid (CuP2S)**

The CuP2S was prepared from the reaction of H2P2 with 10 equivalents of copper acetate in DMF at 120 ºC. Under these conditions CuP2S precipitated as a dark solid, being insoluble in solvents where H2P2 and CuP2 are very soluble or partially soluble (e.g., methanol, chloroform, dichloromethane, acetone, acetonitrile
and dimethylformamide). Here, the fact that solid **CuP2S** emerged during metallation of **H2P2**, by the using of an excess of copper, might be related with the ability of the peripheral amine groups to form solid structures, like CPs, by copper self-assembly mediation. The novel **CuP2S** hybrid solid was characterized by UV-VIS, ATR-FTIR, XRD and EPR techniques.

Diffuse reflectance UV-VIS spectra (DR UV-VIS) for free-base porphyrins and metalloporphyrins are shown in Figure 2. Usually, porphyrins in solid state have an extended Soret band and four bands of lower intensity attributed to the Q bands [31]. In our case, the free-base porphyrins **H2P2** and **H2P3** solids present a characteristic Soret band in the region of 400 nm and a set of four Q bands between 500 and 700 nm.

The UV-VIS spectra of the new complexes **CuP2** and **CuP3** have one maximum in the Soret region (428 nm), showing a hypsochromic shift in comparison to the Soret band of the corresponding free-base porphyrins (**H2P2** at 431 nm and **H2P3** at 435 nm), resulting from the alteration in the micro symmetry of the porphyrin macrocycle due to the presence of the Cu(II) ion in the porphyrin core [13, 33]. The spectrum of the **CuP2S** shows a broad Soret band at 428 nm, the same position observed for **CuP2**, and two Q-band at 552 and 626 nm. The two Q-bands, different from the three and four Q-bands of **CuP2** and **H2P2**, respectively, indicate that **H2P2** was metallated during the self-assembly process and that CuP2S is different from **CuP2**.
Figure 2. UV-VIS spectra of the solid sample: (a) CuP$_2$S, (b) CuP$_2$, (c) CuP$_3$, (d) H$_2$P$_2$ and (e) H$_2$P$_3$.

The ATR-FTIR spectrum of CuP$_2$S (Figure S15) shows the characteristic bands of porphyrin CuP$_2$. The bands at 1544 and 1387 cm$^{-1}$ are assigned to the symmetric and asymmetric vibrations of the C=O group belonging to the cluster acetate [20, 29].

The crystallinity of CuP$_2$S was verified by PXRD (Figure 3). The PXRD pattern of CuP$_2$S was different from the diffraction pattern of the starting materials (H$_2$P$_2$ and Cu(OAc)$_2$, Figure S16). The PXRD of CuP$_2$S showed characteristic feature of nanomaterials, such as high dispersion and disorder of phases; diffraction patterns are characterized by significant broadening of peaks [35].

The particle size, measured from dynamic light scattering (DLS) analyses of CuP$_2$S showed an average nanosized diameter of 292.9 nm (Figure S17).
Figure 3. PXRD of the solid material CuP2S.

The EPR spectrum of solid CuP2 presents the characteristic anisotropic signal expected for Cu(II) in axial symmetry, showing the four lines expected for Cu(II) EPR species, with $g_\parallel > g_\perp$ and with hyperfine splitting ($A_\parallel = \sim 210$ G) (Figure SI8) [21]. In case of CuP2S, it shows a similar spectrum of those of the CuP2, but with appreciable differences in the values for the $g_\parallel$ and $A_\parallel$ parameters. CuP2S ($g_\parallel = 2.227$ and $A_\parallel = 192$ G) shows a greater $g_\parallel$ and a lower $A_\parallel$ than CuP2 ($g_\parallel = 2.191$ and $A_\parallel = 210$ G) (Figure 4a and Figure SI8). A greater values of $g_\parallel$ and lower values of $A_\parallel$ are associated to a larger distortion of the porphyrin macrocycle [21]. Comparing the measured values of g-tensor and A, CuP2S has a greater distortion. This difference may indicate the presence of the copper atom as linker in the solid CuP2S, since changes in the EPR parameters are observed.

CuP2S stability studies to acidic condition

In general, PCs are not stable in acidic media, since that may occur protonation of the ligands and subsequent metal de-coordination [13]. In order to verify the stability of CuP2S to not mild acidic condition it was dissolved in a solution of hydrochloric acid.
(4 mol/L) and evaporated to dryness. UV-VIS, ESI-MS and EPR results suggested that CuP2S was dissolved in this conditions resulting in CuP2 in solution that can be isolated by dryness. The EPR spectrum of this acid treated material, dissolved in acetone, displays similar EPR of the free CuP2 (Figure 4b and 1).

![EPR spectra](image)

**Figure 4.** EPR spectra: a) solid sample CuP2S at 77 K and b) CuP2S dissolved in hydrochloric acid solution, dried, and redissolved in acetone (frozen solution EPR spectrum at 77 K).

From the combined results of all spectroscopic analyses, seems that, most probably, the solid CuP2S is a structured solid, like a CP, obtained by self-assembly mediated by the copper ions and the amine groups of H2P2.

**Catalytic Studies**

The catalytic activity of the prepared copper complexes was investigated in the oxidation of 3,5-DTBC and catechol under homogeneous (CuP2 and CuP3) and heterogeneous (CuP2S) conditions, using dioxygen or hydrogen peroxide as oxidants.
Oxidation catalysis of 3,5-Di-tert-butylcatechol (3,5-DTBC) with O₂ from air in methanol and phosphate buffer (pH = 8.0)

As a preliminary experiment, in order to evaluate if the new complexes were catalytic active, 3,5-DTBC was used as model substrate. The choice of this substrate is due to the low redox potential for catechol-quinone in comparison to catechol, which makes it easy to oxidize into quinone [38].

The first oxidation reactions of 3,5-DTBC catalyzed by CuP² and CuP³ under O₂ from the air where done in methanol (homogeneous catalysis). This solvent was chosen because both reactants (3,5-DTBC and catalysts) are fairly soluble in it. CuP²S was investigated in heterogeneous conditions since it is insoluble in methanol or in the mixture methanol/substrate.

The reactions were monitored by UV-VIS, the appearance of a band at 400 nm is an indicative of the oxidation of 3,5-DTBC to 3,5-di-tert-butyl-o-quinone (3,5-DTBQ). Additionally, the broad band between 500 and 600 nm can also be assigned to the quinone (Figure S19).

In general, it was observed that the obtained conversions rates of 3,5-DTBQ with CuP² and CuP³ were similar for all studied reaction times (Figure 5), being however slightly higher with CuP³. This small and steady, but not negligible, difference can be directly related to the structural differences of both metalloporphyrins. The presence of bulkier substituents on the periphery of the porphyrin macrocycle can affect the ring symmetry and, consequently, influences the formation and stabilization of the catalytic intermediate. The presence of N-tosylethlenediamine bulky groups on CuP³ can provide additional protection to the effects of oxidative destruction in homogeneous catalysis [39].
For example, Nappa et al. have shown that subtle alterations in the catalyst structure markedly change the selectivity of the reaction products, i.e., the metal catalytic activity can be modulate by the structure of the ligands, which in our case are the porphyrins [40].

Krebs et al., working with the unsymmetrical binuclear compounds containing N₄O as donor ligand, with different coordination surroundings for two copper ions in close proximity, showed that small differences of the binuclear complex structures resulted in different catalytic performances. The authors justified such behavior due of ability coordination sites for substrate binding [41].

Nakagaki et al. showed that it is possible modulate catalyst selectivity and efficiency by choosing the appropriate porphyrin substituent and metal. Different metalloporphyrins were prepared via structural modification of H₂P1 with 4-mercaptobenzoic acid. The substituent groups present in the structure of the resulting porphyrins furnished structured solids. The metalloporphyrins, including the structured solids provided good to excellent yields during catalysis of (Z)-cyclooctene, cyclohexane, and heptane oxidation [12].

The conversions observed for CuP2 and CuP3 were slightly higher than those observed by He et al. using as catalyst the copper porphyrins meso-tetrakis[4-(nicotinoyloxy)phenyl]porphyrinato copper(II) (21.9%) and meso-tetrakis[4-(methoxyl)phenyl]porphyrinato copper(II) (23.3%) in the 3,5-DTBC oxidation, using peroxide hydrogen as oxidant [42]. The authors observed similar results for both CuP in homogeneous catalysis and concluded that the catalytic activity was not influenced by nicotinoyl group.

The efficiency of a catalyst can be also analyzed in terms of turnover frequency (TOF). The catalytic rate can be conveniently given in terms of TOF measured in
turnovers per unit time. The turnover rate for CuP3 (30.3 h⁻¹) is almost two times higher that observed for CuP2 (17.7 h⁻¹)[13].

Comparing these results with the ones obtained for other copper complexes reported in the literature, it is possible to conclude that the observed catalytic activities of CuP2 and CuP3 are comparable to those reported[38, 43].

It was noted that with increasing the reaction time of 1 h to 2 h, the catalytic performance is almost maintained for CuP2 and CuP3. On the other hand, the solid CuP2S shows a progressive increase of the product conversion with the increase of the reaction time and after 120 minutes of reaction, it is observed 26% of conversion (Figure 6).

The higher conversion values observed in homogeneous catalysis using CuP2 (24.2%) and CuP3 (26.7%) at 90 min, in comparison to the conversion achieved by CuP2S (120 min., 26%), clearly show that to achieve similar conversion rate in heterogeneous catalysis an extra time is necessary, which can be directly related with the less access of the substrates to the metal center of the catalytic active species present in the structured solid CuP2S in comparison to the complexes in solution. As consequence, longer reaction times are needed to improve the efficiency of CuP2S.

The possible presence of channels in the micro- or mesopores of the solid CuP2S can improve the catalytic activity, and at the same time provide the benefits of the heterogeneous catalysis with reused capability. Besides of that, effects frequently observed in homogeneous systems are minimized, such as, aggregation and oxidative self-degradation.
Figure 5. 3,5-DTBC oxidation in presence of O$_2$ from the air and catalyzed by CuP$_2$, CuP$_3$ and CuP$_2$S in methanol with phosphate buffer (pH = 8.0). Conversions were calculated based on the amount of substrate. Results represent reactions performed in duplicate or triplicate. Under these conditions the conversion values were in the range of ± 0.7%.

**Oxidation catalysis of Catechol with O$_2$ from air in PBF (pH = 8.0)**

Considering the capacity of the prepared compounds to catalyse efficiently the conversion of 3,5-DTBC to 3,5-DTBQ (Figure 5), the following studies were focused on their catalytic behaviours when catechol was employed as substrate. However, if compared to 3,5-DTBC, catechol is harder to oxidize [42, 45]. This substrate was used in order to check if the prepared compounds are also catalytic active in this oxidation reaction and if there is some preferential selectivity of each catalyst compounds against different catechols.

Also, the search for efficient catalysts for the catechol oxidation that are functional in water is a major challenge [44]. For this purpose the oxidation of this substrate was investigated in phosphate buffer (PBF) and organic solvent using O$_2$
from the air as oxidant. Under these conditions, **CuP2** and **CuP3** are sparingly soluble and **CuP2S** is insoluble. The reaction rates were followed using the absorbance decrease at 508 nm, the characteristic band of the presence of catechol by nitrite method (Figure SI10,).

For both **CuP**, an increase of quinone yield was observed compared to 3,5-DTBBQ at the same reaction time (**CuP2**: 21.7% vs 30.6 and **CuP3**: 34.5 vs 40.4%). Although 3,5-DTBC is more susceptible to oxidation, steric hindrance promoted by the bulky groups on the substrate affects the catalytic efficiency of both **CuP**, resulting in a slower kinetics.

The quinone conversion yield was slightly lower for **CuP2** than for **CuP3**. These results suggesting again that the difference of substituents present on the copper porphyrins affects the catalytic activity. These results can be related to the macrocycle distortions, caused by the nature of the substituents present on the porphyrin ring. For example, the structure of [Cu(OETPP)] is highly distorted in comparison to [Cu(TPP)] due to the steric crowding of the multiple peripheral substituents [46].

**CuP3** has greater steric hindrance provided by substituents compared to **CuP2** as discussed before. This fact may prevent the formation of inactive species and the porphyrin ring auto-oxidative destruction [47]. The best result for **CuP3** can also be partly explained by the different solubility compared to **CuP2**.

By monitoring the, a dependence of the conversion versus time was observed for both **CuP**. The catalytic conversion values versus reaction time show that **CuP2** has a slower kinetic (Figure 7). After 24 h of the reaction time (**CuP2**: 57.5% and **CuP3**: 62.2%), the difference in catalytic performance observed is lower compared to 1 h of reaction (**CuP2**: 30.6% and **CuP3**: 40.4%). This study confirms the importance of the proper choice of substituents in the periphery of the macrocycle.
The results obtained here (Figure 7) using CuP2 and CuP3 are very promising in comparison to previous reports and, particularly, for using oxygen from air as the oxidant, showing that the solids are catalytically active under mild conditions. For example, Constantino et al. reported that catechol conversion obtained for FePcTs in homogeneous conditions is 21%, being by lower than that obtained for both CuP [48].

CuP2S also showed better catalytic activity for the catechol (22.9%) compared to 3,5-DTBC (9.1%) at the same reaction time (1 h). To understand if under heterogeneous reaction conditions the access of the reactants to the catalyst metal center may be limited, longer reaction times could be used [49]. In general, this improves the efficiency of the hetero-catalytic system. In fact, conversion values increase considerably with the increase of the reaction time for CuP2S, being at 24 h even better than the ones obtained with CuP2 and CuP3. The best result observed for CuP2S can be justified by the structure of this solid compound, that can minimize the deactivation effects, sometimes observed in homogeneous catalysis, as the oxidative destruction of the catalyst during the catalytic reactions, with consequently diminish of the catalytic performance.
Figure 6. Catechol oxidation in presence of O$_2$ from the air catalysed by CuP$_2$, CuP$_3$ and CuP$_2$S using water and phosphate buffer (pH = 8.0). Conversions were calculated based on the amount of substrate remaining. Results represent reactions performed in duplicate or triplicate. Under these conditions the conversion values were in the range of ± 2.0%.

**Oxidation catalysis of Catechol with O$_2$ from air** in a biphasic system

In order to verify the influence of the solvent in the reactions, the oxidation reactions were performed using biphasic system. The biphasic system could be applied to the oxidation of organic substrates, wherein the catalyst is fully soluble in one phase while the substrates and/or products are soluble in the other phase. In order to ensure solubility of the catalysts was used chloroform as solvent. Catechol is poorly soluble in this solvent, to ensure its solubility it was used water and phosphate buffer, making the reaction medium a biphasic system.

The reaction at interface depends on the reactants meeting at the interface boundary, the diffusion rate across the bulk of the phase. Diffusion across in the interface is more important for reaction proceeded in one of the phases. Catechol oxidation by CuP$_2$ and CuP$_3$ in presence of oxygen probably occurs at the interface phase. This assumption is based on the catalytic results obtained (Figure 7). Catalysis processed in a biphasic system, in general, has a slower kinetics, since diffusion processes are crucial for the reaction. Diffusion across the interface is more difficult than the diffusion across. If the reaction takes place in aqueous phase it is expected conversion values similar those obtained in aqueous system but with larger reaction time. On the other hand if the reaction occurs at the interface, lower conversion results are expected in the same time reaction. In fact, the conversions
values observed for **CuP2** and **CuP3** in biphasic system were lower than those observed for aqueous system in the same time reaction (**CuP2**: 18.3% vs 30.6% and **CuP3**: 23.5% vs 40.4%) suggesting that the reaction probably occurs at the interface.

In spite of the CuP catalyst are not completely soluble in this biphasic systems (it is not a true homogeneous catalytic system), the advantage of using biphasic system is that the catalyst can be easily separable, and when stable can be reused. In fact, at the end of the reaction the catalyst was easily separated by decanting and liquid-liquid extraction process.

The same trend catalytic from for 3,5-DTBC was observed for both CuP, the metalloporphyrins in solution (in fact biphasic system) showed differences in the catalytic performance.

As observed for the oxidation of 3,5-DTBC (Figure 6), the conversion results obtained using **CuP2** and **CuP3** as catalyst (“homogeneous catalysis”) varied between 18.3% to 35.8% depending on the reaction time. For this substrate, again the compound **CuP3** presented best catalytic results than **CuP2** confirming that the structure influence the catalytic activity.

For both CuP, after about 3 h the catalytic reaction seems to reached a plateau and no significative catalyst conversion modification (21.6% and 35.6%) was observed for the catechol oxidation by **CuP2** and **CuP3**, respectively (Figure 8). This behaviour can be explained by diffusion across of the reactants from one phase to another which can be affected by the concentration of relation to saturation of solution. Such behaviour was not observed 3,5-DTBC oxidation.
Gamez et. al [44] investigated the catecholase activity in biphasic system water/toluene solvent mixture in the presence of dioxygen atmosphere and good conversion results were obtained in the oxidation of 3,5-DTBC.

For both CuP, better results were obtained in aqueous system (Figure 7). The explanation for these results is due to the fact the catalyst and substrate are in the same phase, since the conditions the access of the reactants to the metal center is more easy, although CuP are slightly soluble in the reaction medium, the small amount of CuP is able to catalyze the oxidation of catechol. Oxidation rates of catechol were dependent on the reaction time. In general, longer contact time between the catalyst and the substrate can facilitate access of both species promoting catalysis.

**Figure 7.** Catechol oxidation in presence of O$_2$ from the air catalyzed by CuP2 and CuP3 using a biphasic system. Conversions were calculated based on the amount of substrate remaining. Results represent reactions performed in duplicate or triplicate.

**Oxidation catalysis of Catechol with H$_2$O$_2$ in PBF (pH = 8.0)**

In order to investigate the influence of oxidant in the catechol oxidation reaction, the same reactions were also performed in the presence of hydrogen peroxide. Under
these conditions, CuP2 and CuP3 show similar results (Figure 8). However, comparing these results, in the presence of hydrogen peroxide to the ones under O2, similar results are achieved but with less reaction times. In both cases after about 6 h the catalytic reaction seems to reached a plateau. The plateau in the absence of peroxide was achieved using 6 h of reaction time to CuP3 and 24 h for CuP2 in the presence of oxygen of the air and without using H2O2 (Figure 7).

Meunier et. al [45] studied the oxidation of catechol using a series of MPcs as catalysts with different oxygen donor compounds. In this study it was found an oxidizing dependence for the oxidation of catechol. Depending on the oxygen compound, pH and reaction time it was observed complete conversion of the catechol using iron(III) Pcs. In the presence of peroxide hydrogen, the conversions values observed for iron(III) Pcs varies between 15% and 40% at 1 h.

Comparing the results obtained here for the CuP investigated with those reported by Meunier [45] for iron(III) Pcs at the same reaction time, it is noted that CuP2 showed a similar result (37.9%) and CuP3 (58.3%) a better catalytic performance.

Beside of that, CuP2 and CuP3 show a better catalytic performance than the parent CuP1 (63.4% and 60.9% vs 41.5%) suggesting that both catalysts are more stable than CuP1. However, CuP2S was more effective than CuP2 and CuP3 under the similar reaction conditions. CuP2 shows a conversion rate of 63.7% after 24 hours of reaction, while the solid CuP2S give complete conversion in the same time of reaction. CuP3 shows a conversion rate of 63%.

Good linear plots of ln A/A0 vs time were obtained under the pseudo-first order conditions, for a constant H2O2 concentration. The rates of the non-catalyzed reactions ($k_{obs}$ = 6.85 x 10⁻⁴ min⁻¹) were smaller than those observed for the catalyzed reactions.
(CuP2 $k_{obs} = 1.78 \times 10^{-2}$ min$^{-1}$, CuP3 $k_{obs} = 1.77 \times 10^{-2}$ min$^{-1}$ and CuP2S $k_{obs} = 1.00 \times 10^{-2}$ min$^{-1}$). The $k_{obs}$ for CuP2S is about fifteen times greater than those observed for non-catalyzed reaction and similar to observed for CuP2 and CuP3. The $k_{obs}$ observed for all the catalyzed reactions were similar to that observed by us for the material Cu₄CuP₃S using a lower concentration of hydrogen peroxide and higher that observed for meso-tetra(2,6-dichloro-3-sulfonatophenyl)porphyrinate of iron(III) chloride ($k_{obs} = 3.95 \times 10^{-4}$ min$^{-1}$) [50].

Iron PCs immobilized in LDH (HT-FePcTs) have been used as catalysts in the oxidation of catechol; the conversion observed for this solid catalyst varied between 63% and 88% after 2 h using hydrogen peroxide [48]. This study showed higher conversions for this material than those observed for the free FePc complex in solution (21%).

For CuP2S solid catalyst lower result was observed in comparison to HT-FePcTs. However, as shown by Meunier [45], the increase in pH increases the conversion. The lower result observed can be justified by the difference in pH. Furthermore, total conversion was attained for CuP2S by increasing the reaction time.

Structured solids, such as coordination polymers associate the catalytic process heterogeneous with the benefits of homogeneous catalysis; in this case, each part of the solid catalyst contains the metalloporphyrin. This situation does not occur for the solids prepared by immobilization of metalloporphyrins in supports as the example cited using LDH as support [48]. One of the great advantage of using CuP2S as heterogeneous catalyst is the possibility of easy reuse. For this purpose, the stability of the solid CuP2S after reaction was evaluated by powder XRD and UV-VIS.

The great advantage of the solid catalyst CuP2S is its insolubility in the reaction media. Because of that it acting as a solid catalyst for heterogeneous processes and
the reuse capacity was comproved after each reaction since no evidences of soluble metalloporphyrin was observed by the monitoring the reaction solution using UV-Vis spectroscopy.

In both catalyzed reactions a large enhancement in the conversion percentages was observed for the catalyzed reactions when compared to the control reaction.

**Figure 8.** Catechol oxidation in presence of H₂O₂ (30%) catalyzed by CuP₂, CuP₃ and CuP₂S using phosphate buffer (pH = 8.0) as solvent. Conversions were calculated based on the amount of substrate remaining. Results represent reactions performed in duplicate or triplicate. Under these conditions the conversion values were in the range of ± 1.2%.

**Reused studies of CuP₂S in chatecol oxidation with H₂O₂ in PBS (pH =8)**

In order to investigate the recyclability capability of CuP₂S, reuse tests were performed using hydrogen peroxide as oxidant. After recovery and reuse of the solid catalyst CuP₂S, the catechol conversion remains similar to the first use (Figure 9).
was observed that after the first use, the kinetics is only slightly slower. Therefore, it was possible to efficiently recycle this catalyst, at least three times, without loss of catalytic efficiency.

![Figure 9](image)

**Figure 9.** Catechol oxidation conversion results in presence of H$_2$O$_2$ (30%) catalyzed by CuP2S at different times under reuse conditions. Conversions were calculated based on the amount of substrate remaining. Results represent reactions performed in duplicate.

### 4. Conclusion

The new synthesized metalloporphyrins CuP2, CuP3 and the insoluble solid CuP2S were prepared, characterized and evaluated as catalysts in oxidation reactions. The obtained results show that they are able to mimic the catalytic activity of catecholase, affording good conversions of catechol and 3,5-di-tert-butylcatechol to the correspond quinone derivatives.
In this study it was verified that appropriate substituents at the \( p \)-positions of the \textit{meso}-aryl groups of a porphyrin macrocycle can fine-tune the catalytic efficiency of the resulted compounds/materials. For example, it was observed better results using \textbf{CuP3}, that presents appropriate number of bulky groups on the parent H\textsubscript{2}TPPF\textsubscript{20}(H\textsubscript{2}P\textsubscript{1}), when compared to \textbf{CuP2}.

The steric effect of the substrate 3,5-DTBC used in the catalytic reaction was also important for the best catalytic performance of the compounds used as catalyst. It was observed that when 3,5-DTBC was used instead catechol, the catalytic results for both CuP were lower for the same time reaction.

The influence of the solvent and the oxygen donor agent was also studied in the oxidation reactions of catechol. The catalytic results for both catalysts are promising, since those are active under mild conditions. The conversions obtained using both CuP suggested that introduction of appropriate bulky groups into the structure of H\textsubscript{2}P\textsubscript{1}, created a suitable environment to obtain more efficient biomimetic catalysts.

In the presence of the solid catalyst \textbf{CuP2S} a full conversion of catechol was observed after 24 hours of reaction. The high insolubility of this solid in the reaction medium can be considered a great advantage since it can be reused. \textbf{CuP2S} showed the ability to be reused in the catechol oxidation for at least three cycles without loss of catalytic efficiency, proving to be indeed a strong and efficient reusable catalyst.

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