Supplementary Material (ESI) for Chemical Communications

Phase-transfer of porphyrins by polypeptide- containing hyperbranched polymers and the novel iron(III) porphyrin biomimetic catalyst

Qi-Zhi Ren, Yuan Yao, Xiao-Jian Ding, and De-Yue Yan

1. Synthesis of porphyrins

(1) The preparation of 5, 10, 15, 20-tetrakis(sulphophenyl)- porphyrin (**H_2TPPSH**)

4.00 g of meso-tetraphenylporphyrin (H_2TPP) and 40.0 mL of 98.0% H_2SO_4 were ground into a homogeneous paste in a 100mL beaker. The paste was transferred to a 250 mL flask and 60 ml of concentrated H_2S0_4 was added. The mixture was stirred 6 hours and then allowed to stand at room temperature for 48 hours. Then the mixture was filtered through a G5 sintered glass filter to remove unreacted H_2TPP, and the filtrate was cautiously diluted with two volumes of distilled water. The resulting bright green precipitate was washed several times with acetone, and purified by six successive reprecipitations from methanol solution with acetone. Yield (5.3g, 88%); Elemental Analysis: Found: C, 56.69; H, 3.32; N 5.79. Calc. for $C_{44}H_{30}N_4O_{12}S_4$: C, 56.51; H, 3.23; N, 5.95%; UV-vis: $\lambda_{max}(H_2O)$/nm 418 (ε/dm^3 mol^{-1} cm^{-1} 221 000), 512 (6 010), 549 (1 840) and 594 (2 310); ^{1}H NMR: δ_H(500 MHz, D_2O, Me_4Si) 8.75(8H, s, βH), 8.31 (8H, d, Ph-2, 6H), 7.20(8H, d, Ph-3, 5H); MS: *m/z* 933.1.

(2) The preparation of tetra sodium meso-tetra(sulfonatophenyl)-porphyrin (**H_2TPPSNa**).

4.00 g of H_2TPP and 40.0 mL of 98.0 % H_2SO_4 were added in a 100 mL open beaker, and ground into homogeneous paste. The paste was transferred in a 250mL flask, and then 60mL of 98% H_2SO_4 was also added, The mixture was heated on 105°C oil bath and was stirred for 6 hours. The mixture was stood for 48 hours in a 400 mL beaker at room temperature. Then about 300 mL deionized water was slowly added with stirring. After cooled, the mixture was filtered by G5 sintered glass filter, and the filtrate was washed by acetone. The solid product was dissolved in 150 mL deionized water, and neutralized with saturated $NaHCO_3$ until the solution turned to purplish red. After adjusted the pH value to 6.0, the solution was dialyzed and concentrated, and recrystallized twice by dehydrated methanol/ethanol solvent mixture. After filtered and dried under vacuum, the resulting H_2TPPSNa was obtained. Yield (5.8g, 90%); Elemental Analysis: Found: C, 59.69; H, 2.72; N, 5.39. calc. for $C_{44}H_{26}N_4O_{12}S_4Na_4{\cdot}2H_2O$: C, 49.91; H, 2.86; N, 5.29%; UV-vis: $\lambda_{max}(H_2O)$/nm 419 (ε/dm^3 mol^{-1} cm^{-1} 246 000), 516 (6 050), 551 (1 980), 592 (2 500) and 648 (1 960); FT- IR (KBr)(ν_{max}/cm^{-1}) 3430, 1590, 1460, 1389, 1190, 968, 741 and 714; ^{1}H NMR: δ_H(500 MHz, DMSO-d6, Me_4Si) 8.84(8H, s, βH), 8.18 (8H, d, Ph-2, 6H), 8.50(8H, d, Ph-3, 5H), -2.955(2H, s, NH pyrrole); MS: *m/z* 999.0.

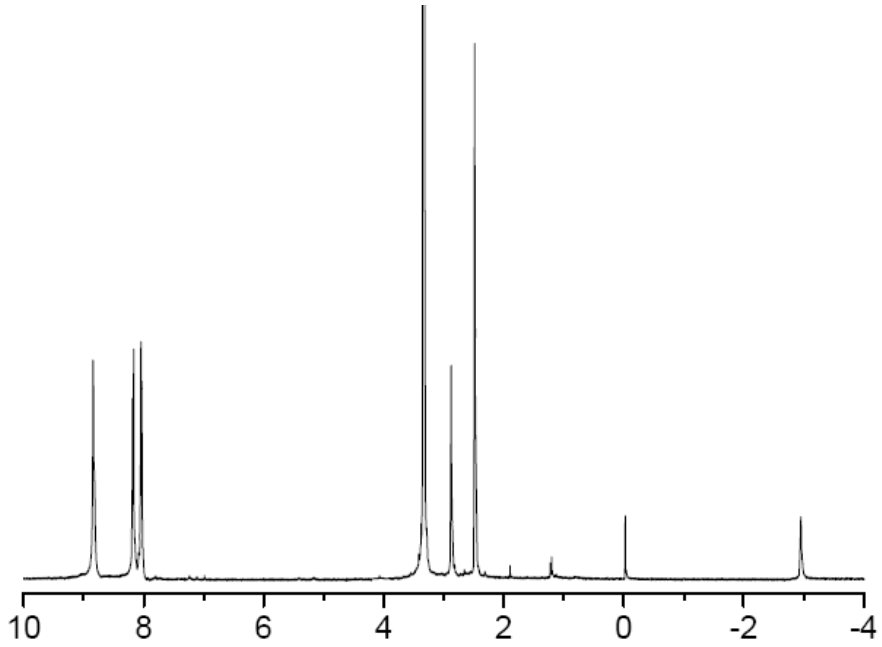

Fig. 1 ^{1}H NMR spectrum of **H_2TPPSNa**

(3) The preparation of Tetra sodium meso-tetra(sulfonatophenyl)-porphyrin iron(III) (**FeTPPSNa**)

0.40 g of H_2TPPSNa was dissolved by 80.0 mL of deionized water in a 250 mL flask, and 1.32 g of $FeSO_4 \cdot 7H_2O$ was added at 100°C , the pH value of the mixture was adjusted to 6.0 with saturated $NaHCO_3$. After 30 minutes, another 0.24g of $FeSO_4 \cdot 7H_2O$ was added, and the reaction continued until free H_2TPPSNa could not detected by Uv-vis. The result solution was purified by acidic cation exchange resin column, and dialyzed after concentrated to 40 mL. Then the solution was dried out to obtain resulting FeTPPSNa. Yield (0.47g, 67%); Elemental Analysis: Found: C, 47.15; H, 2.59; N, 4.92. Calc. for $C_{44}H_{24}N_4O_{12}S_4Na_4$ FeCl·$2H_2O$: C, 47.49; H, 2.54; N, 5.03%; UV-vis: $\lambda_{max}(H_2O)$/nm 394 (ε/dm^3 mol^{-1} cm^{-1} 102 000), 527 (10 800) and 580 (7 060); FT- IR (KBr)(ν_{max}/cm^{-1}) 1640, 1490, 1390, 1190 and 719; MS: *m/z* 1054.9.

(4) The preparation of 4-N-methylpyridyl)porphyrin (**H_2TMPyP**)

Propionic acid, acetic anhydride and corresponding aldehyde were mixed in a 250 mL flask, and heated to reflux at 130 °C with stirring. 4-pyridinecarboxaldehyde and pyrrole were then dropped into the flask separately. The mixture was continually refluxed for 1.5 h. Then the resulting solution was subsequently dried under vacuum. The residue was purified by column

chromatography to obtain the resulting H_2TMPyP. Methylation by an excess amount of methyl iodide afforded target compound. UV-vis: λ_{max}(10 μM in Tris buffer)/nm 423 (ε/dm^3 mol^{-1} cm^{-1} (42 700), 517(2 455), 562(3 020), 585(1 000), and 643(977); ^{1}H NMR: δ_H(300 MHz, DMSO-d6, Me_4Si) 9.45(6H, d, 2, 6-pyridinium), 8.99-9.01(8H, s,β-pyrrole), 8.27(6H, d, 3,5-pyridinium), 4.69(9H, s, N^+–Me), -3.01(2H, s, NH pyrrole); MS: *m/z* 678.

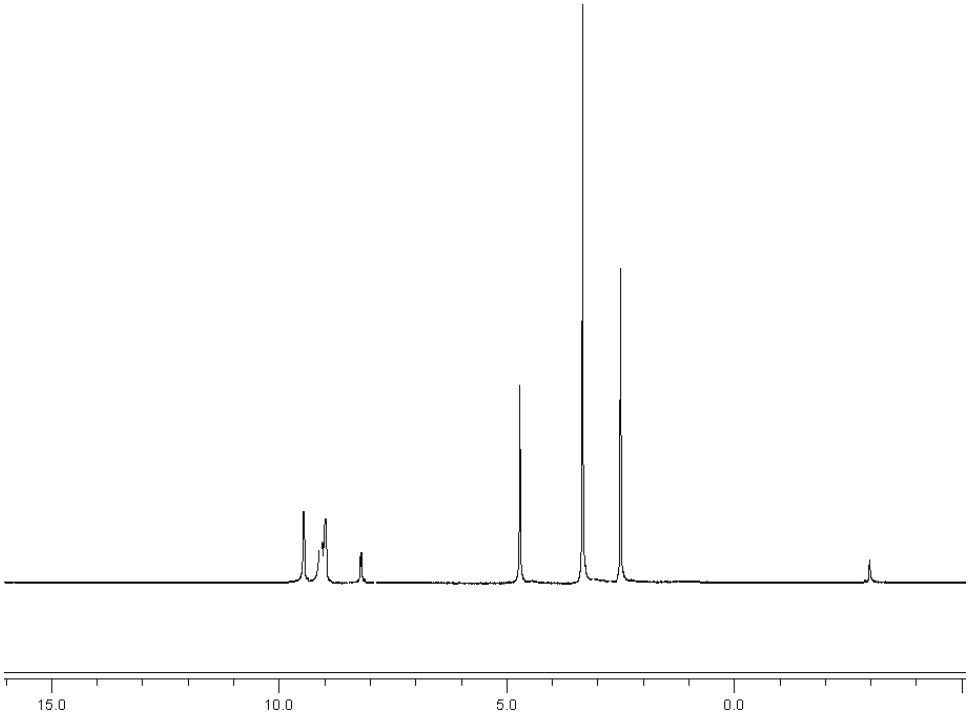

Fig. 2 ^{1}H NMR spectrum of **H_2TMPyP**

2. Synthesis of polymer

(1) Preparation of **ZLys-NCA**

10 g of ZLys, 5 g of BTC and 100 mL of anhydrous THF was added into a 250 mL three-necked flask under N_2 atmosphere, and then stirred at 50 ^{o}C with N_2 bubbling for about 1 h. After turning transparent, the reaction solution was continuously bubbled by N_2 for 30 min, followed by pouring into 500 mL petroleum ether. The mixture was filtered to obtain the crude ZLys-NCA，and the product was recrystallized for three times from the mixture solvent of anhydrous ethyl acetate and petroleum ether (2:1). Yield (70%). ^{1}H NMR: δ_H(400 MHz, $CDCl_3$, Me_4Si) 7.34(4H, m, **1**), 5.08(2H, s, **2**), 4.26(1H, t, **8**), 3.18(2H, t, **4**), 1.67~2.00(2H, d, **7**), 1.48(2H, m, **5**) and 1.39(2H, m, **6**).

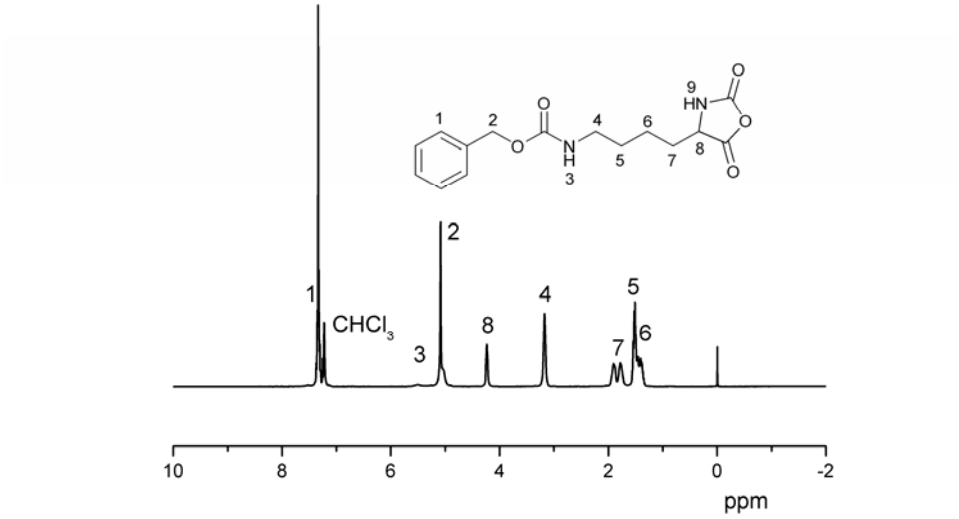

Fig. 3 ^{1}H NMR spectrum of **ZLys-NCA**

(2) Preparation of PEI-PZLys hyperbrabched copolymer

2g of ZLys-NCA was dissolved in 60mL of chloroform in an enclosed airtight reaction bottom, and then 10 mL of 0.4g PEI chloroform solution was injected in the bottom. After stirred for 3 days, the solution was dropped into 300mL ethyl ether, and the deposits was filtered and dried under vaccum. The molecular weight was characterized by ^{1}H-NMR (Fig. 4) referring paper [1, 2] . The result *Mn* was about 5.87×10^{4}. ^{1}H NMR: δ_H(400 MHz, $CDCl_3$, Me_4Si) 8.00 (0.2H, **8**), 7.13~7.35 (2.9H, **7**), 5.01 (1.0H, **6**), 2.33~3.43 (4.0H, **1**, **5** and **PEI**), 1.13~1.95 (3.1H, **2**, **3**, **4**).

References:

[1] H. Tian, X. Chen, H. Lin, et al. *Chem. Euro. J.*, 2006, **12**, 4305-4312.

[2] K. Kunath, A. von Harpe, D. Fischer, et al. *J. Controlled Release*, 2003, 88, 159-172.

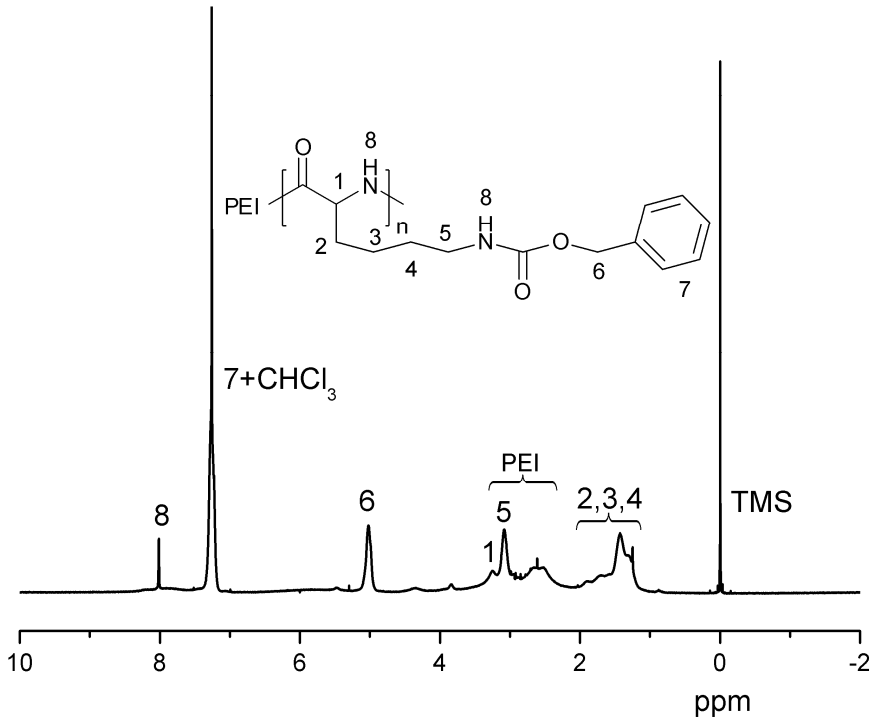

Fig. 4 ^{1}H NMR spectrum of **PEI-ZLys**

3. Reagents and instruments

ε-carbobenzoxy-L-lysine (ZLys) is purchased from GL Biochem (Shanghai) Co. Ltd. Polyethylenimine is obtained from Aldrich, and use as received. Bis(trichlormethyl)carbonate (BTC) is purchased from Wenshui chemicals Co. Ltd., and recrystalized using chloroform for three times. All solvents, including ethyl acetate, chloroform, tetrahydrofuran(THF) and petroleum ether (boiling point 60-90 ^{o}C) are obtained from Sinopharm Chemical Reagent Co. Ltd., and dehydrate before using.

Uv-vis spectra are characterized by Spectrum Co. Ltd. SP-1902 instrument. ^{1}H-NMR is characterized by VARIAN 400 instrument. Fluorescent emitting spectra are characterized using Perkin-Elmer LS50B instrument. Circular dichroism(CD) spectra are characterized by Jasco-J815 CD spectrometer. Electron paramagnetic resonance measurements (EPR) of the frozen solution of the porphyrins in a 3-mm EPR quartz tube were performed with a Bruker EMX-8-2.7 spectrometer at the X-band (ca. 9.450 GHz) at 100K using liquid N_2.

4. Phase transfer experiment

Definite concentration PEI-PZLys chloroform solutions and porphyrins water solutions were

prepared firstly, and filtrated by 0.45μm filter. 5 mL PEI-PZLys chloroform solution and 5 mL porphyrin water solution were added in a 25 mL bottle, and shacked sufficiently, then stood for 3hours. After the two phase separated entirely, the chloroform phase was carefully extracted by a syringe, and prepared for Uv-vis, Fluorescent emitting and CD spectra characterization.

5. Circular dichroism spectra

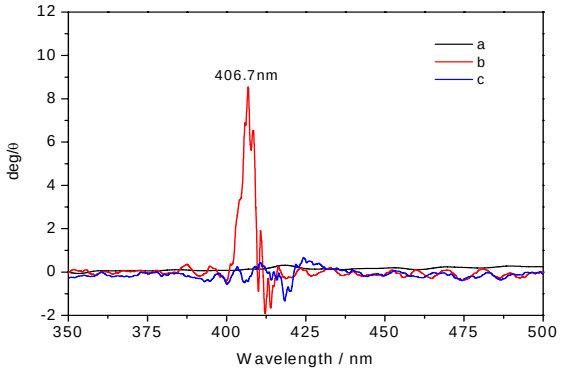

Fig. 5 Circular dichroism spectra of (a): 2.0mg/mL **PZLys-PEI** in chloroform; (b) and (c): 100μmol/L **H_2TPPSNa** in water and phase-transferred by 2.0mg/mL **PEI -PZLys** in chloroform respectively.

6. EPR spectra

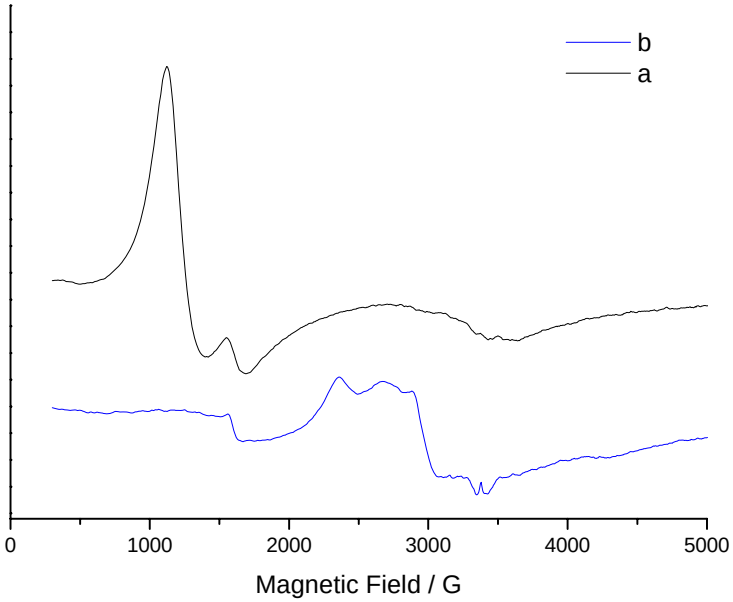

Fig. 6 EPR spectra of the encapsulated **FeTPPSNa** in chloroform, in the absence of Imidazole (a) and in the presence of excess Imidazole (b).

6. **General procedure for styrene epoxidation and ethylbenzene hydroxylation:**

oxidations including epoxidation and hydroxylation were carried out using the following standard conditions: A mixture of the catalyst FeTPPSNa or PEI-PzLys/FeTPPSNa system (1.0 μmol), styrene or ethylbenzene (30 mmol) and 1,2,4-trichlorobenzene (170 μmol, as an internal standard) in freshly distilled and degassed $CHCl_3$ (5 mL) were stirred under N_2 in a dry 10 ml vial at 20°C. PhIO was added continuously. After the first addition of PhIO (165 μmol, 36 mg), aliquots were taken, purified over a short silica-gel column chromatography and monitored by GC at appropriate intervals. After the end of the cycle1, FeTPSNa was released by pH triggered method, the trigger pH value was 12.0. The retention times were compared to the retention times of standard racemates. The turnover numbers for epoxidation of styrene is the molar ratio of styrene oxide to FeTPPSNa, the turnover numbers for hydroxidation of ethylbenzene is the total molar ratio of sec-phenethyl alcohol and acetophenone to FeTPPSNa. The yields of the reactions were calculated on the base on the consumed PhIO on capillary column.

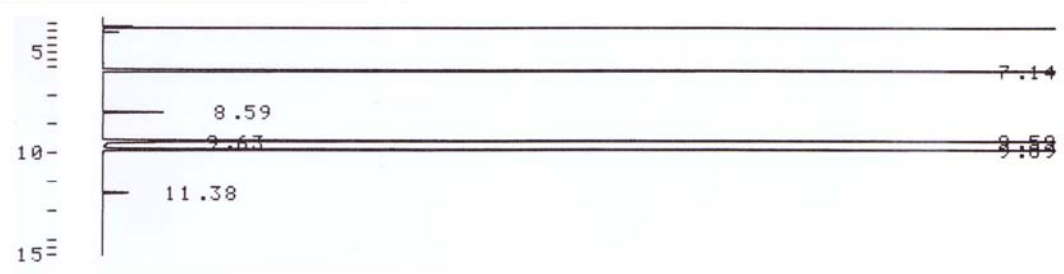

Fig. 5 Epoxodation of styrene catalyzed by **PEI-PzLys/FeTPPSNa** in cycle 1

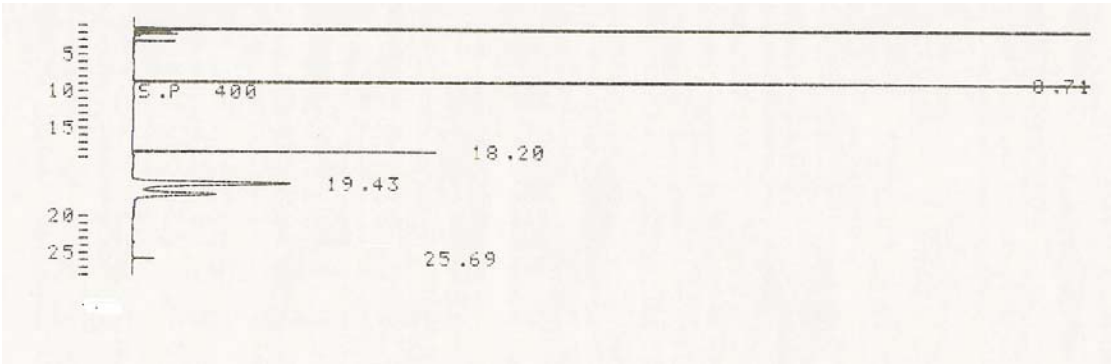

Fig. 6 Hydroxylation of ethylbenzene catalyzed by **PEI-PzLys/FeTPPSNa** in cycle 1

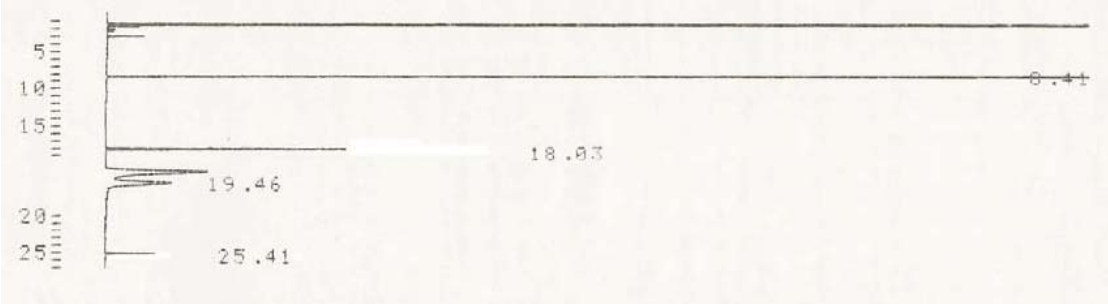

Fig. 7 Hydroxylation of ethylbenzene catalyzed by **PEI-PzLys/FeTPPSNa** in cycle 3

Table 1 the retention times of the catalytic system

Oxidation	No.	RT	Assignment
Epoxidation	1	7.14	styrene
	2	9.59	PhI
	3	9.89	Styrene oxide
	4	11.38	1,2,4-trichlorobenzene
Hydroxylation	1	8.71	ethylbenzene
	2	18.20	PhI
	3	19.43	phenethyl alcohol
	4	19.62	acetophenone
	5	25.69	1,2,4-trichlorobenzene