SELECTIVE OXYGENATION OF α-OLEFINS BY MEANS OF METALLOPORPHYRIN CATALYSTS MIMICKING CYTOCHROME P-450

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A significant problem associated with synthetic metalloporphyrin catalysts used to mimic oxygenation function of cytochrome P-450 is their deactivation due to the formation of inactive μ-oxo dimer. We prepared a series of porphyrin-based catalysts mimicking cytochrome P-450 potentially resistant to deactivation by varying the central metal ion, porphyrin ring substitution, and catalyst support. The influence of the nature of a solid support, the type of central cation and the chemical modification of the porphyrin ring on the catalytic efficiency was studied. The oxygenation ability of the synthesized catalysts was tested using 1-hexene as a substrate and *tert*-butyl hydroperoxide as an oxidant under various reaction conditions. Identification of the oxygenation products was performed with gas chromatography-mass spectrometry (GC-MS). Quantification of the products formed was based on GC with flame ionization detection (FID). It was found that the application of low GC injection temperature (150 °C) was necessary to detect primary products of the oxygenation, peroxo alkenes, from which secondary oxygenation products, mainly epoxides, alcohols, aldehydes and ketones, were formed slowly. The efficiency of the individual catalysts depended on their structure and varied significantly. The most active catalysts were Fe(II)-tetraphenylporphyrin (TPP) immobilized on aminopolystyrene (aminoPS) and Mn(II)-TPP immobilized on aminoPS.

Keywords: Cytochromes; Oxygenations; Porphyrinoids; Cytochrome P-450; Enzyme mimicking; Oxygenation; Metalloporphyrin; Product distribution.

Cytochrome P-450 is a member of the monooxygenase family of the heme enzymes that play an important role in metabolism of biomolecules. Cytochrome P-450 catalyzes the oxidation processes taking place mainly in the liver where numerous substrates of endogeneous and exogeneous origin are degraded. Because of its importance, cytochrome P-450 is intensively studied by many biologists, chemists and physicists. Despite of this effort a detailed mechanism of its action still remains unknown.

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A significant problem associated with synthetic metalloporphyrin catalysts is their deactivation due to the formation of inactive μ-oxo dimers (Metal–O–Metal). There are two ways to prevent the deactivation: first, the application of a homogeneous catalysis – modification of the porphyrin skeleton with various bulky electronegative substituents and an axial bond with an appropriate ligand of the central atom[1–9]; second, the use of a heterogeneous catalysis – an attachment of the catalysts to a suitable support. As a support cyclodextrin, cyclofan, molecular sieves, Merrifield resin or synthetic polymers can be used[10–12].

In our previous work[13] we have used the homogeneous catalysis with different metallocomplexes than porphyrin. The synthesis of such type of metallocomplexes based on novel ligands, specifically on arylmethyl substituted bis-β-aminoketones, has been described for instance in ref.[14]. The efficacy of the catalysts for 1-hexene oxygenation has been tested as function of M^{n+} and ligand type and reaction conditions. Our results have revealed that the best performance for conversion of 1-hexene to oxygenation products has been obtained for Co(II) metallocomplexes of ligand *N*,*N*′-ethylenebis[4-amino-1,5-di-(2-naphthyl)]pent-3-en-2-one. The analysis of the product distribution has been performed by GC-MS. Mild conditions of the analysis allowed for the detection of primary products of the oxygenation reaction.

Here, we present novel metalloporphyrins immobilized on a solid support (heterogeneous catalysis). We investigated the influence of the nature of a solid support, the type of a central cation and the chemical modification of the porphyrin ring on the catalytic efficiency.

EXPERIMENTAL

Catalytic activity of a group of catalysts mimicking cytochrome P-450 structure was studied. Experiments were carried out as heterogeneous catalysis where given complexes were fixed on the following supports: aminopropyl-silica gel (aminoSil), aminopolystyrene (aminoPS), and benzhydrylamine polystyrene (BHA-PS). As the catalysts we used a group of porphyrin metallocomplexes with various central cations (Fe^{2+}, Mn^{2+}, Co^{2+}) and porphyrin rings with two substituents, phenyl (P), tolyl (Tol), and several types of linkage to the support (Fig. 1).

Binding of the porphyrins to the appropriate support was realized by several different procedures.

4-Carboxyphenyl-triphenyl porphyrin or 4-carboxyphenyl-tritolyl porphyrin was converted to the acid chloride and reacted with the amino-substituted support by a standard procedure. The reaction product was filtrated out the reaction mixture and washed with dichloromethane, methanol, water, and methanol (Fig. 1; catalysts 1–5, 9–11)[15].

The application of the pyridyl-triphenyl porphyrin was based on the following reaction sequence: the pyridine ring on the porphyrin skeleton was quaternized with bromoacetic

acid and after conversion to chloride reacted with the amino support. The resulting product was filtered off, washed with methanol, dichloromethane and water (Fig. 1; catalyst 12).

Amine linkage between porphyrin derivative and the support was obtained by alkylation of the amino-substituted support with bromomethylphenyl-triphenyl porphyrin (catalysts Fig. 1; 6–8).

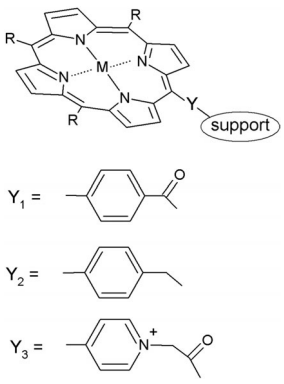
Metallation (incorporation of the central metal into the porphyrin skeleton) was carried out by a standard Macac strategy (heating of the porphyrin covalently immobilized on the support in the presence of the appropriate acetylacetone metallocomplex (Fe, Mn, Co) in DMF (80 °C, 8 h)). The resulting catalysts were washed with dichloromethane and methanol and after drying used in the subsequent experiments.

The experiments were carried out with *tert*-butyl hydroperoxide (*t*-BuOOH) as the oxygen donor and pyridine as the metallo-axial ligand, where pyridine was also used as an internal standard for the evaluation of the conversion and the product distribution measurement. In the present work 1-hexene was used as a substrate for α-olefin oxygenation.

The following aspects were studied and evaluated: (i) the efficiency of the individual catalysts, (ii) the reaction kinetics of the catalysis and (iii) the influence of the amount of the oxidant (*t*-BuOOH) on the quantity of the products.

A typical experimental setup:

The reaction mixture consisted of 30 mg of a catalyst, 500 µl of 1-hexene (substrate), 100 µl of *t*-BuOOH (oxidant) in dichloromethane (commercial aqueous 70% solution of *t*-BuOOH was extracted (ratio 1:1, v/v) with dichloromethane), 500 µl of dichlormethane (solvent) and 10 µl of pyridine (axial ligand and internal standard).



Catalyst-support	M	Catalyst No.
M-TrPP-aminoSil	Fe^{2+}	1
R = C_6H_5; Y_1	Mn^{2+}	2
	Co^{2+}	3
M-TrPP-aminoPS	Fe^{2+}	4
R = C_6H_5; Y_1	Mn^{2+}	5
M-TrPP-aminoPS	Fe^{2+}	6
R = C_6H_5; Y_2	Mn^{2+}	7
	Co^{2+}	8
M-TrPP-BHA-PS	Fe^{2+}	9
R = C_6H_5; Y_1	Co^{2+}	10
M-TrTolP-aminoSil	Co^{2+}	11
R = *p*-CH_3-C_6H_4; Y_1		
M-TrPP-aminoSil	Co^{2+}	12
R = C_6H_5; Y_3		

FIG. 1
Structures and description of the studied catalysts (TrPP, triphenyl porphyrin; TrTolP, tritolyl porphyrin)

The mixture was left to react for 7 days at the laboratory temperature in a closed vial. Then 1 µl of the supernatant (free of the catalyst) was injected into the gas chromatograph for the analysis.

In the case of the reaction kinetic study the supernatant in the reaction vial was sampled at specific times for a given time period.

The identification of the products in the reaction mixture was carried out by means of GC-MS with a Hewlett–Packard 5890 gas chromatograph equipped with a quadrupole mass detector 5971A (electron ionization at 70 eV), a capillary column used was BPX5 (25 m, 220 µm, film thickness 0.25 µm) and carrier gas, helium.

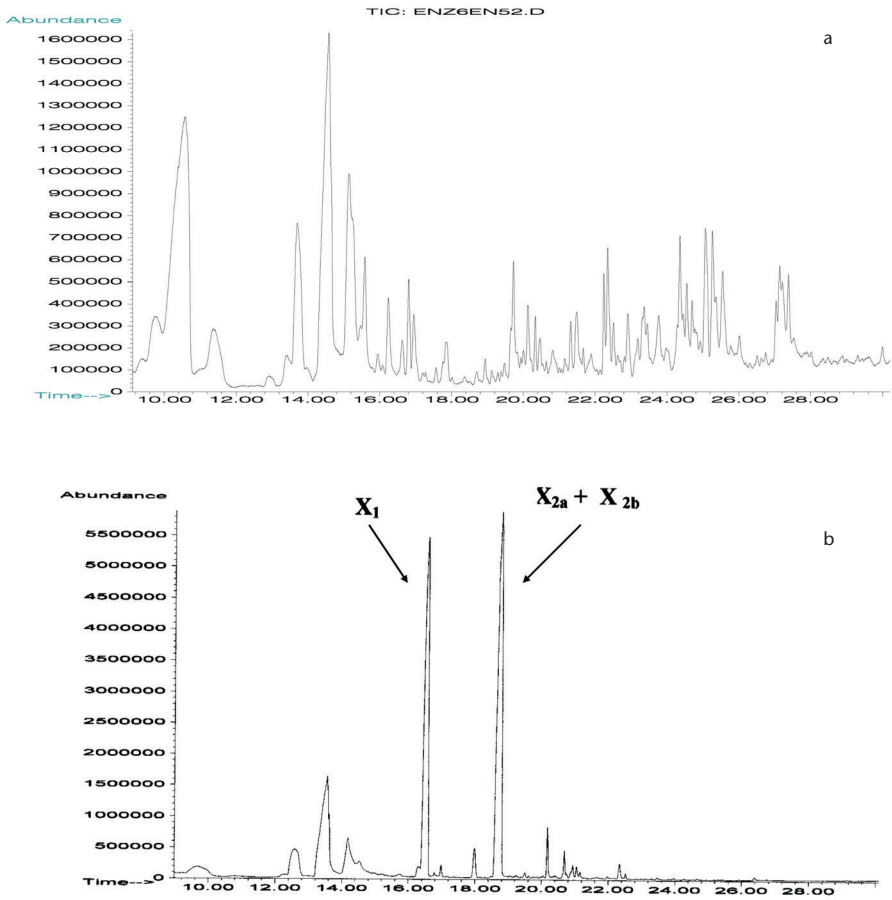


FIG. 2
Product distribution for oxygenation of 1-hexene with catalyst No. 12. GC-MS analysis, injection at: 250 °C (a), 150 °C (b)

Quantitative measurements were accomplished with a GC instrument Varian 3350 with a flame ionization detector, a capillary column was DB5 (20 m, 320 μm, film thickness 0.22 μm) and carrier gas, hydrogen.

At the beginning of our work a suitable chromatographic method allowing for a reliable monitoring of the progress of the reactions studied had to be developed. We found previously that the GC injection temperature had an essential influence on the appearance of the chromatogram and that this effect was independent on the presence/absence of the oxidant (*t*-BuOOH) in the injected sample[13]. The injection was carried out in the temperature range from 150 to 250 °C. The GC chromatograms obtained at 150 and 250 °C, respectively, are shown in Fig. 2.

As can be clearly seen, two dominant peaks found at 150 °C and marked X_1 and X_{2a}+X_{2b} nearly vanished at 250 °C (Fig. 2). In agreement with our previously published data[13] we found that three compounds X_1, X_{2a} and X_{2b} are thermally labile primary reaction products of the oxygenation and as such they represented precursors for consecutively forming secondary products identified in Fig. 3. Thus, it was determined experimentally that the injection temperatures above 150 °C led to a significant degradation of the primary reaction products X_1, X_{2a} and X_{2b}, and for that reason in all following experiments the injection temperature 150 °C was employed. It should be also mentioned that in some cases depending on chromatographic conditions and the composition of the injected sample the compounds X_{2a} and X_{2b} could be partially resolved (Fig. 3).

Our effort to identify unknowns X_1, X_{2a} and X_{2b} led into the proposal of their tentative structures (Fig. 4).

For the interpretation of our measured MS spectra for X_1 and X_{2a}+X_{2b} we inspired by ref.[17], where a similar structure was proposed and verified by NMR spectra. Based on that, we proposed structures for X_1 and X_{2a}+X_{2b}. We did not have any corresponding standards

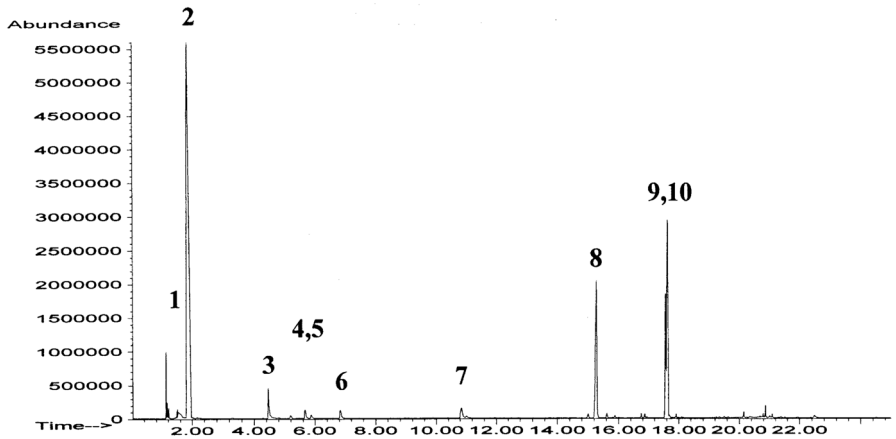


FIG. 3

Product distribution for oxygenation of 1-hexene (catalyst No. 5, after 7 days of the reaction). GC-MS analysis, injection at 150 °C: 1 dichlormethane, 2 1-hexene, 3 pyridine, 4 1-hexen-3-one, 5 1-hexen-3-ol, 6 butyloxirane, 7 2-hexenal, 8 X_1, 9 X_{2a}, 10 X_{2b}

or tabulated MS. Thus, to further support our proposal we took advantage of the mass fragment prediction software MassFrontier, ver. 4 (HighChem, USA), for fragment modeling. The thorough comparison of the software generated and the measured MS spectra (Fig. 5) showed a good agreement with the proposed structures for the compound X_1, X_{2a} and X_{2b}. The spectra of all three compounds contain characteristic ions *m*/*z* 57 $C_4H_9^+$ (*tert*-butyl), 67 $C_5H_7^+$, 73 $C_3H_5O_2^+$, 82 $C_6H_{10}^{\bullet+}$ (the loss of *t*-BuOOH from the molecule), and 83 $C_6H_{11}^+$ (the loss of *t*-BuOO$^\bullet$ from the molecule). In the spectra of X_{2a} and X_{2b} is present ion *m*/*z* 69 $C_5H_9^+$, which, in agreement with the MassFrontier's prediction, is missing in the MS spectrum of X_1 (Fig. 5).

RESULTS AND DISCUSSION

The majority of our catalysts were based on TrPP skeleton. The activity of these catalysts was expressed as a sum of peak areas of the primary reaction products X_1 and X_2 in % with respect to the peak area of pyridine used as the internal standard (Fig. 6). The efficiency of the catalysts was not intentionally expressed in % of the substrate (1-hexene) conversion because of its high volatility leading to losses during the manipulation with the samples and, consequently to inaccuracy in data evaluation.

It follows from Fig. 6 that the studied M-TrPP-based catalysts can be classified into three groups according to their catalytic activity (based on the

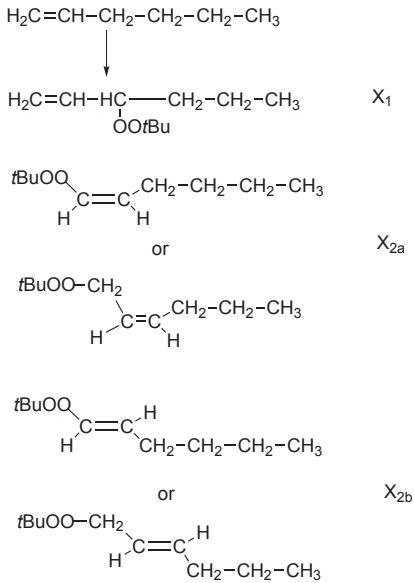


FIG. 4
Conversion scheme indicating the tentative structures of the major primary products

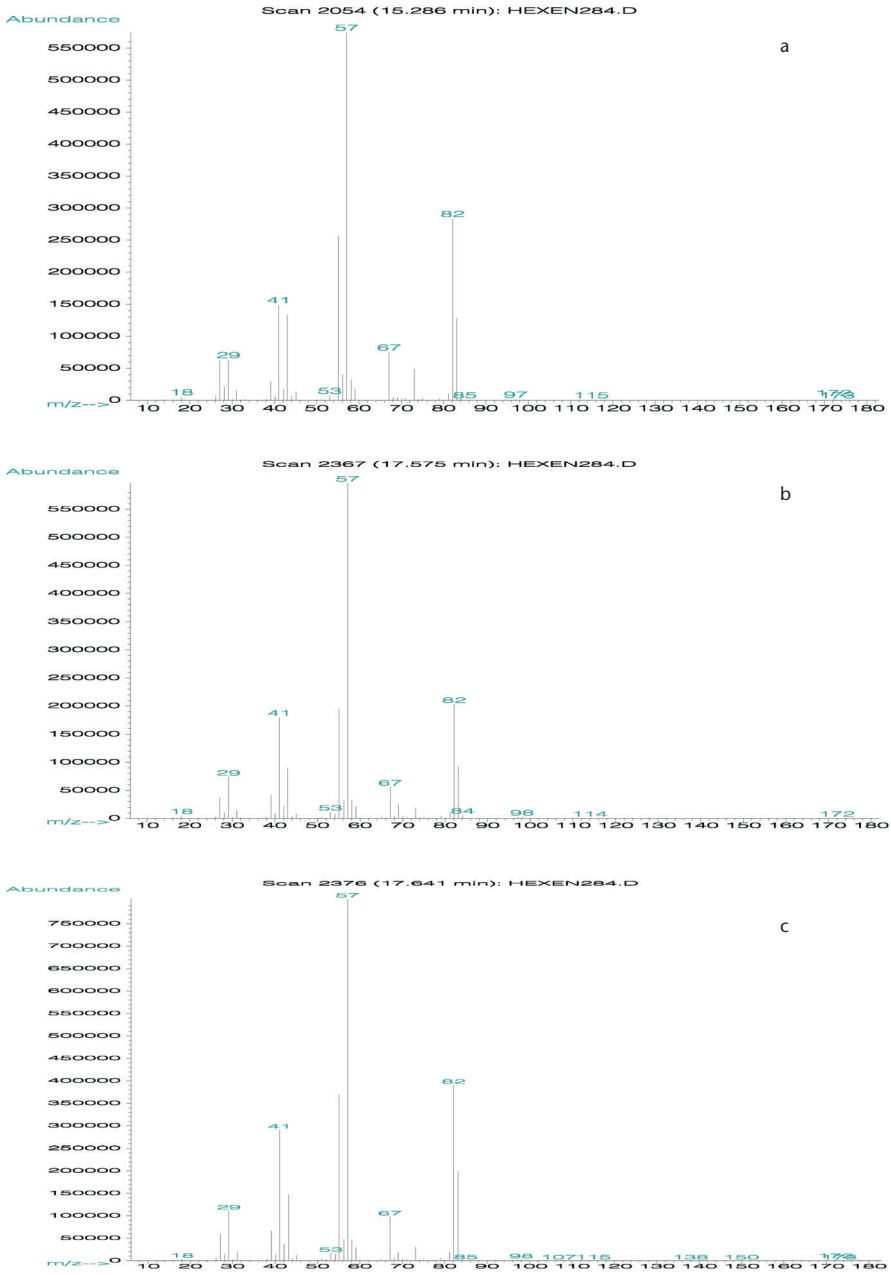


FIG. 5
Measured mass spectra of compounds X_1 (a), X_{2a} (b) and X_{2b} (c); M = 172

results obtained after 7 days of the reaction): (i) active catalysts (5, 6; the sum of the primary product peak areas is above 200% of the internal standard peak area), (ii) medium active catalysts (1, 3, 8–10; the sum of the primary product peak areas is 50–120% of the internal standard peak area), and (iii) inactive catalysts (2, 4, 7; the sum of the primary product peak areas is less than 50% of the internal standard peak area, which is roughly equivalent to the result obtained in blank experiment with no catalyst present in the system).

Generally, the catalysts with Fe^{2+} as a central ion are usually slightly more active than the catalysts with Mn^{2+} or Co^{2+} as a central ion with one exception for the Mn^{2+}-TrPP-Y1-aminoPS catalyst (5), which demonstrated the highest activity followed by the Fe^{2+}-TrPP-Y2-aminoPS catalyst (6).

Cobalt as a central atom provided relatively low efficiencies independently on the structure of the catalyst (Table I).

Next experiments with selected catalysts were focused on the reaction kinetics, i.e. the rate of X_1, X_2 and 2-hexenal formation and the rate of the decrease of *t*-BuOOH concentration in 1, 5, 9, and 12 days of the catalysis at the laboratory temperature (Table II).

Table II shows that after 12 days even in the blank experiment with no catalyst present in the system some decrease in *t*-BuOOH takes place and a low amount of the products is formed.

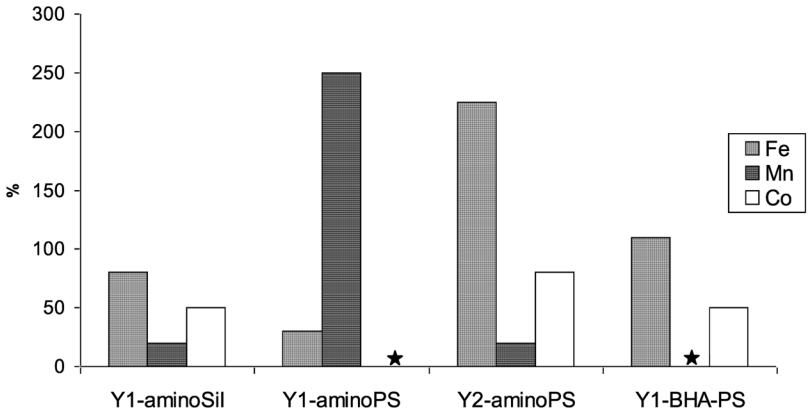


FIG. 6

The efficiency of the studied M-TrPP-based catalysts immobilized on aminopropyl-silica gel (aminoSil), aminopolystyrene (aminoPS), and benzhydrylamine polystyrene (BHA-PS) used for 1-hexene oxygenation with *t*-BuOOH expressed as a peak areas of compounds X_1+X_2 in % of the peak area of the internal standard (pyridine) obtained after 7 days of the reaction, ★data not measured

The decrease in *t*-BuOOH amount is partially caused by its spontaneous decomposition to 1-butanol, which was proven in the reaction mixture after long standing. Limited and slow decrease in the oxidant amount and at the same time the creation of low amount of the products was observed in experiments with the catalysts Mn^{2+}-TrPP-Y1-aminoSil (2) and Fe^{2+}-TrPP-Y1-aminoPS (4), the inactive catalysts. Slightly better results were obtained for the catalysts Fe^{2+}-TrPP-Y1-BHA-PS (9) and Co^{2+}-TrPP-Y3-aminoSil (12), where a moderate decrease of *t*-BuOOH and the production of certain amount of the primary products X_1 and X_2 were reached immediately in the first day of the experiment. The best results were obtained with the catalysts Mn^{2+}-TrPP-Y1-aminoPS (5) and Fe^{2+}-TrPP-Y2-aminoPS (6), which proved their activity in previous experiments (vide supra). In the case of the catalysts (5) and (6) a fast and pronounced decrease of the oxidant concentration in the first day of the reaction was registered. Correspondingly, the primary products X_1 and X_2 were quickly formed in the first day of the reaction and then their concentration decreased in the following time, which was in contrast to the secondary product 2-hexenal, which amount was slowly and continuously increasing during 12 days.

Finally, the influence of the amount of the oxidant *t*-BuOOH on the progress of the reaction for two catalysts, one active (Mn^{2+}-TrPP-Y1-aminoPS (5)) and the other inactive (Fe^{2+}-TrPP-Y1-aminoPS (4)), differing by the central atom only was studied (Table III).

As can be seen from Table III in the case of the inactive catalyst Fe^{2+}-TrPP-Y1-aminoPS (4) the increasing amount of the oxidant in the reaction mixture had practically no influence on the amount of the formed products X_1 and X_2 even for a long reaction time. A significant amount of the oxidant remained in the system unreaceted even after 11 days. On the other hand, for the active catalyst Mn^{2+}-TrPP-Y1-aminoPS (5) the situation was quite different. The amount of the primary reaction products X_1 and X_2 was strongly dependent on the amount of the oxidant in the system.

TABLE I
Efficiencies[a] of the Co^{2+}-metalloporphyrin catalysts

TrPP-Y1-aminoSil (catalyst No. 3)	TrPP-Y2-aminoPS (catalyst No. 8)	TrPP-Y1-BHA-PS (catalyst No. 10)	TrTolP-Y1-aminoSil (catalyst No. 11)	TrPP-Y2-aminoSil (catalyst No. 12)
50	80	50	80	80

[a] The efficiency of the catalysts is expressed as a sum of peak areas of X_1+X_2 in % of peak area of the internal standard (pyridine). Data obtained in the first day of the experiment. The measurements were carried out after 7 days of the reaction.

TABLE II
Time dependence for X_1, X_2 and 2-hexenal formation and decrease of *t*-BuOOH amount using selected catalysts[a]

Catalyst No.	Compound															
	t-BuOOH				X_1				X_2				2-hexenal			
	Time, day				Time, day				Time, day				Time, day			
	1[b]	5	9	12	1[b]	5	9	12	1[b]	5	9	12	1[b]	5	9	12
5	30	0	0	0	110	110	95	–[c]	150	140	120	–[c]	3	8	17	–[c]
6	10	0	0	0	105	100	95	–[c]	125	120	120	–[c]	5	11	19	–[c]
9	250	200	–[c]	140	40	50	–[c]	50	70	70	–[c]	60	3	6	–[c]	9
12	270	220	–[c]	110	20	30	–[c]	60	30	50	–[c]	80	1	2	–[c]	9
2	400	350	350	200	10	10	10	15	10	20	20	20	1	2	5	5
4	400	350	250	200	10	20	30	40	20	30	40	60	1	2	5	5
blank	–[c]	–[c]	–[c]	180	–[c]	–[c]	–[c]	20	–[c]	–[c]	–[c]	25	–[c]	–[c]	–[c]	5

[a] The amount of the product is expressed as relative % of the peak area of the internal standard (pyridine). [b] These measurements were carried out within 2–3 h after the mixing of the catalyst with the substrate and they represent the starting points of the experiment. [c] – Values not measured.

TABLE III
The influence of the amount of *t*-BuOOH on the oxygenation reaction[a]

Catalyst No.	Compound								
	t-BuOOH			X_1			X_2		
	Time, day			Time, day			Time, day		
	1[c]	11	17	1[c]	11	17	1[c]	11	17
5 – 1×[b]	90	0	–[d]	50	80	–[d]	70	100	–[d]
5 – 2×[b]	110	0	–[d]	90	150	–[d]	150	250	–[d]
5 – 5×[b]	250	70	–[d]	120	250	420	210	380	570
5 – 10×[b]	1300	900	900	340	460	440	460	670	530
4 – 1×[b]	400	–[d]	–[d]	10	–[d]	–[d]	20	–[d]	–[d]
4 – 2×[b]	600	600	–[d]	20	60	–[d]	30	80	–[d]
4 – 5×[b]	900	900	–[d]	10	40	–[d]	10	80	–[d]

[a] The amount of the product is expressed as relative % of the peak area of the internal standard (pyridine). [b] 1×–10× corresponds with the corresponding multiple of the original amount of *t*-BuOOH. [c] These measurements were carried out within 2–3 h after the mixing of the catalyst with the substrate and they represent the starting points of the experiment. [d] – Values not measured.

Doubling the amount of *t*-BuOOH provided about twice as much of the oxidation products X_1 and X_2. However, a further increase in *t*-BuOOH concentration led to a moderate increase of the oxidation products only and this trend was independent on the reaction time. This finding was in a good agreement with a fast decrease in *t*-BuOOH concentration in the first day in the case that its amount in the system was doubled, and on the contrary with its only moderate concentration decrease when its amount in the system was ten times higher.

CONCLUSIONS

The oxygenation of α-olefins using *t*-BuOOH was studied with the set of twelve porphyrin-based catalysts with various central metal ions, porphyrin ring substitution, type of linkage to the support, and the support itself, all of which mimicking cytochrome P-450. It was demonstrated that efficiency of the individual catalysts depended on a combination of all the above mentioned parameters and differed significantly even for the catalysts of the similar structures. The most active catalysts were Mn^{2+}-TrPP-Y1-aminoPS (5) and Fe^{2+}-TrPP-Y2-aminoPS (6).

The primary reaction products were identified as the compounds containing peroxidic –OO*t*-Bu group, which slowly converted to common oxygenation products i.e., epoxides, alcohols, aldehydes and ketones.

The conversion of the primary to the secondary products was slow at the laboratory temperature. An increase of the reaction temperature would accelerate the conversion process but then polymerization reactions could take place as it was demonstrated when the GC injection temperature was set above 150 °C.

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REFERENCES

1. de Sousa A. N., de Carvalho M. E. M. D., Idemori Y. M.: *J. Mol. Catal. A: Chem.* **2001**, *169*, 1.
2. Nam W., Kim I., Lim M. H., Choi H. J., Lee J. S., Jang H. G.: *Chem. Eur. J.* **2002**, *8*, 2067.
3. Rose E., Andrioletti B., Zrig S., Quelquejeu-Etheve M.: *Chem. Soc. Rev.* **2005**, 34, 573.
4. Liu Y., Zhang H.-J., Lu Y., Cai Y.-Q., Liu X.-L.: *Green Chem.* **2007**, *9*, 1114.
5. Stephenson N. A., Bell A. T.: *J. Mol. Catal. A: Chem.* **2007**, *275*, 54.
6. Atkinson S. T., Brady S. P., James J. P., Nolan K. B.: *Pure Appl. Chem.* **1995**, *67*, 1009.
7. Yang J., Weinberg R., Breslow R.: *Chem. Commmun.* **2000**, 531.

8. Breslow R., Gabriele B., Yang J.: *Tetrahedron Lett.* **1998**, *39*, 2887.
9. Cai Y., Liu Y., Lu Y., Gao G., He M.: *Catal. Lett.* **2008**, *124*, 334.
10. Yang J., Gabriele B., Belvedere S., Huang Y., Breslow R.: *J. Org. Chem.* **2002**, *67*, 5057.
11. Yu X. Q., Huang J. S., Yu W. Y., Che C. M.: *J. Am. Chem. Soc.* **2000**, *122*, 5337.
12. Nestler O., Severin K.: *Org. Lett.* **2001**, *3*, 3907.
13. Pulkkinen J., Zachar P., Borek V. A., Laatikainen R., Král V.: *J. Mol. Catal. A: Chem.* **2004**, *219*, 21.
14. Pulkkinen J. T., Laatikainen R., Ahlgren M. J., Perakyla M., Vepsalainen J. J.: *J. Chem. Soc., Perkin Trans. 2* **2000**, 777.
15. Mammana A., Asakawa T., Bitsch-Jensen K., Wolfe A., Chaturantabut S., Otani Y., Li X., Li Z., Nakanishi K., Balaz M., Ellestad G., Berova N.: *Bioorg. Med. Chem.* **2008**, *16*, 6544.
16. Kubát P., Lang K., Anzenbacher P., Jursíková K., Král V., Ehrenberg B.: *J. Chem. Soc., Perkin Trans. 1* **2000**, 933.
17. Griesbeck A. G., Bartoschek A.: *Chem. Commun.* **2002**, 1594.