Available online at www.sciencedirect.com
SCIENCE DIRECT®
Talanta 60 (2003) 515–522
Talanta
www.elsevier.com/locate/talanta

A demonstration model for a selective and recyclable uptake of metals from water: Fe(III) ions complexation and release by a supported natural fluorescent chelator

M. Mureseanu [1], G. Renard [a,*], A. Galarneau [a], D.A. Lerner [a]

[a] *UMR 5618 ENSCM/CNRS (Institut Gerhardt), 8 Rue Ecole Normale, 34295 Montpellier, Cedex, France*

Received 17 July 2002; received in revised form 4 December 2002; accepted 20 December 2002

Abstract

We describe here the preliminary stage of development of a process aiming at the selective uptake and release of metal ions from water. The process envisioned involves the encapsulation of highly selective natural chelates secreted by bacteria or other living species in mesoporous solids that could be used as usual resins. To demonstrate the feasibility of the concept, we use a model system involving pyoverdin, a natural Fe(III) ions chelator from a *Pseudomonas fluorescens* strain, encapsulated in a mesoporous templated silica. For this model study, the native fluorescence of the chelator allows a simpler follow-up and quantification of the uptake and release processes.

Keywords: Metal ion uptake; Fluorescence; Pyoverdin; Mesoporous silica

1. Introduction

In the course of evolution, micro-organisms learned to deal with metal ions present in their environment. They can protect themselves by binding toxic metal ions which entered the cell cytosol using phytochelatins [1] or they can absorb metal ions they need for their growth from the surrounding medium. To reach that goal, they excrete natural ligands which complex metal ions reversibly and with a high selectivity. These complexes are then taken up by micro-organisms.

Various uses may be envisioned for such selective ligands. In the field of metal ions recovery from water, one could imagine encapsulate a set of natural complexants, each one on a porous mineral support with good chemical and mechanical properties, and packing each of these hybrid materials in some sort of cartridge. In the next step, these cartridges, one for each metal to remove, would be arranged one on top of another in a column and the water sample introduced at one end. Water percolating through the column would loose one type of metal after another following its uptake by the ad hoc complexant.

* Corresponding author. Tel.: +33-4-67-1443-76; fax: +33-4-67-1443-49.
E-mail address: renard@cit.enscm.fr (G. Renard).
[1] On leave from the Faculty of Chemistry, Department of Analytical chemistry, University of Craiova, A. I. Cuza 13, Romania.

0039-9140/03/$ - see front matter
doi:10.1016/S0039-9140(03)00103-6

After saturation of the column, a regeneration step, such as washing with a small volume of an acid, could recover separately the pure metal ions from each type of cartridges. A similar technology could be used to reduce the complexity of the analysis of complex metal ions mixtures, especially when interferences are expected or to effect a preconcentration step. Information gathered from such systems would also be of interest for developers of metal ion sensors.

The work presented here constitutes the first step of such a project. For safety reason and convenience in the monitoring of the complexation and decomplexation step, pyoverdin, a natural siderophore was used. Bacteria, like all living species require iron for their growth, but the insoluble nature of iron in neutral and oxidising environments, limits its concentration to levels well below that required for growth. A response to this limitation is the extra-cellular release of soluble, low molecular weight siderophores [2]. These organic molecules exhibit a high affinity for Fe(III) ($K_f = 10^{20}$–10^{40}) [3] and are even able to dissolve common soil Fe minerals (e.g. hematite, goethite) [4]. The fluorescent pseudomonas are a major group of gram-negative bacteria [5]. As a group, these bacteria are characterised by the production of water soluble yellow–green pigments, called pseudobactins or pyoverdins, which are siderophores [6,7]. Pyoverdins are chromopeptides consisting in a 2,3-diamino-6,7-dihyroxyquinoline fluorescent chromophore linked to a short peptide [8]. The length and composition of the oligopeptide varies among different pyoverdins. Binding of iron is mediated by the catecholate group of the chromophore and by two-hydroxamate groups substituting the peptide backbone [2]. The resulting neutral octahedral complex has a 1:1 stoichiometry and a stability constant of approximately 10^{24} M^{-1} at neutral pH [9,10].

Furthermore, methods of physical or chemical immobilisation of biomolecules on solid surfaces are well known [11,12]. Some require the covalent attachment to the solid, while others will spontaneously be strongly adsorbed to the supporting surface [13]. Various supports are used such as semi permeable membrane, mesoporous silicas [14,15] or sol–gels for entrapment, or polymers microcapsules, for which comprehensive reviews are available [16,17]. For instance, pyoverdin was immobilised on controlled pore glass in order to obtain a biosensor for ferric ion [18] and total inorganic iron determination [19].

Micelles templated silica (MTSs) of the MCM-41 family discovered in 1992 [20,21] are materials that possess a hexagonally packed array of mesopores with a very narrow pore size distribution. In the following years, new families of templated silicas were obtained in which the pore size can be systematically varied from 2 to 10 nm or more [22–24]. Such mesoporous materials with large pores may encapsulate small biomolecules within the channels which can be several hundred nanometers long [13,25–27].

The present work describes the encapsulation of a pyoverdin in the mesopores of a MTS-type silica support, its use for the selective uptake of the Fe(III) from a multimetallic solution and for the controlled release of these ions.

2. Experimental

2.1. Apparatus

Electronic absorption spectra were recorded on a UVIKON XL UV–Visible spectrometer from Bio-Tek Instruments and diffuse reflectance in the UV–visible range were obtained on a Lambda 14 spectrometer (Perkin–Elmer Inc., Shelton, USA) with an integrating sphere (Labsphere, North Sutton, USA) for solid samples. The latter were held in 0.05 mm thick cuvettes (100 QS, Hellma, Mülheim, Germany). Atomic adsorption spectroscopy (AAS) measurements were performed on a SpectrAA-220 Varian spectrometer.

X-ray powder diffraction patterns were collected on a CGR θ 60 diffractometer using Cu Kα monochromatic radiation. Nitrogen adsorption–desorption isotherms were recorded on a Micromeritics ASAP 2000 (Collecting pH data was made on a G810 pH-meter (Schott)).

Steady-state fluorescence spectra were recorded on a fluorimeter built around two Jobin Yvon M25 monochromators, each fitted with continuously variable slits and a grating with 1200 line-

s mm^{-1} (linear dispersion of 3 nm mm^{-1}). The detector is a low noise R928 photomultiplier (Hamamatsu). The spectral bandwidths were 3 nm for the excitation monochromator, and 1.5 nm for the emission monochromator. All fluorescence spectra were uncorrected.

2.2. Reagents and materials

Pseudomonas fluorescens strains (IP–69-13T, Institute Pasteur, Paris,) were a gift from Prof. J. Guiraud, Montpellier or were obtained from NRRL (B-2641, PF-US, United States).

Analytical reagent-grade chemicals were used to prepare the solutions required for the biosynthesis and purification of pyoverdin from *Pseudomonas fluorescens*.

Freshly prepared MilliQ (Millipore) ultra pure water was used in all experiments.

A 1000 mg l^{-1} Fe(III) stock solution was prepared by dissolving $FeCl_3$ anhydrous (Prolabo) in 1% HCl. The multimetallic solution was prepared by dissolving $K_2Cr_2O_7$, $CuCl_2$, $NiCl_2 \cdot 2H_2O$, $Pb(NO_3)_2$, $ZnCl_2 \cdot 6H_2O$ in 1% HCl. The Fe, Cr, Cu, Ni, Pb and Zn standard aqueous solutions were prepared following the recommended preparation methods of standard solutions for AAS.

Tetraethyl orthosilicate (TEOS, Aldrich), dodecylamine (Aldrich), 3-sn-phosphatidylcholine (Fluka) and β-D-lactose (Aldrich) were employed for the pyoverdin encapsulation.

The optimum pH for the formation of the Fe(III)–pyoverdin complex was obtained with a 0.01 M solution of phthalic acid/biphthalate (Prolabo) at pH 4.5.

2.3. Procedures

2.3.1. Production, isolation and characterisation of the pyoverdin

The stock culture medium was prepared from 3 g yeast extract, 3 g malt extract, 5 g yeast peptone and 10 g glucose l^{-1}, adjusted to pH 6.2 before sterilisation.

For pyoverdin production, the nutrient medium was made up of 6 g K_2HPO_4, 3 g KH_2PO_4, 1 g $(NH_4)_2SO_4$, 0.2 g $MgSO_4$ and 4 g succinic acid l^{-1}, adjusted to pH 7.0 before sterilisation [9].

The pseudomonas strains were grown in 2-l flasks at 28 °C for 48 h with orbital shaking (130 rpm) using a temperature controlled 3031 shaker/incubator (GFL).

The pigment was extracted and purified using the technique proposed by Meyer and Abdallah [9]: complexation with Fe(III), centrifugation, saturation with NaCl, extraction with chloroform–phenol (1/1;v/p), another extraction with diethyl-ether–water (10/1; v/v), exclusion chromatography (Sephadex G25), decomplexation with a 5% 8-hydroxyquinoleine, solution, exclusion chromatography (Sephadex G25).

Absorption spectra were obtained both for the purified pyoverdin and for its ferric complex. The extinction coefficient of ferric pyoverdin at its absorption maximum was computed from the absorbance of an aqueous solution in which the concentration of iron was measured by atomic absorption spectroscopy. The extinction coefficient of pyoverdin was deduced following its titration with a 10^{-4} M ferric chloride solution [28].

2.3.2. Encapsulation of pyoverdin in an MTS mesoporous material by direct synthesis

Three millilitre of 8.73×10^{-4} M pyoverdin solution (pH 7.0) was mixed with 0.15 g lactose and added to a mixture of 0.5 g lecithin and 0.1 g dodecylamine in 5.2 g ethanol. The TEOS (2.7 g) was added under stirring for 15 min and the mixture was left to stand for 24 h. The yellow–white powder was washed 5 times with a mixture of ethanol and water (1/1) and dried for 24 h at 50 °C. The same procedure was applied for the reference material (mesoporous support without pyoverdin). The washing step softly removes most of the lecithin from the channels.

Aliquots of the rinsing waters were analysed by spectrofluorimetry at 460 nm to quantify the amount of siderophore that was not encapsulated.

In order to release pyoverdin from the solid support we proceeded to a basic hydrolysis of the hybrid material (pH 9.0) followed by elution through a G25 Sephadex column. The fractions obtained were analysed by spectrofluorimetry at 433 nm to quantify the amount of siderophore

formerly encapsulated in the MTS mesoporous material.

2.3.3. Spectrofluorimetric quantification of Fe(III)

The response curve of the 2.9×10^{-4} M pyoverdin solution to the presence of Fe(III) was obtained by successive additions of aliquots of a 4×10^{-4} M Fe(III) solution.

The relative fluorescence intensity was recorded in the 410–600 nm range (excitation at 400 nm). The calibration graph was obtained by plotting the decrease of the fluorescence intensity at its maximum versus Fe(III) concentration.

2.3.4. Quantification of Fe(III) uptake

AAS calibration graphs for Fe and for the other metallic ions were obtained by plotting the increase of the absorption intensity versus metallic ion concentration.

Reference MTS (0.5 g) was mixed with 5 ml of a 4 mg l^{-1} multimetallic solution (pH 4.5) containing Fe(III) ions at increasing concentrations, left to equilibrate, centrifuged for 15 min at 3000 rpm. Then Fe(III) was quantified in the supernatant by spectrofluorimetry. A volume of 0.1 ml of the supernatant was added to 3 ml of the 2.9×10^{-4} M pyoverdin solution (pH 7.0) and the decrease of the fluorescence intensity was recorded. The procedure was repeated until the concentration of Fe(III) in the supernatant became constant, indicating the saturation of the solid. The amounts of Fe and other metals present in all the samples were quantified by FAAS. The same procedure was applied for the MTS with the encapsulated pyoverdin. The amount of ferric ions complexed by the encapsulated pyoverdin was quantified by difference.

3. Results and discussion

3.1. Properties of pyoverdin and its Fe(III) chelate in solution

The pyoverdin from the *Pseudomonas fluorescens* strain selected and cultivated in an iron-poor medium shows after extraction and purification, an electronic absorption spectrum characteristic

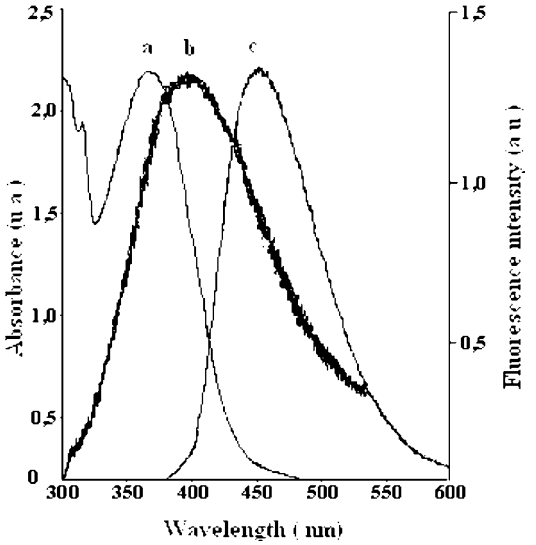

Fig. 1. Electronic absorption (a) and fluorescence emission (c) spectra of pyoverdin (2.9×10^{-4} M in water at pH 7, excitation wavelength 400 nm)—emission of pyoverdin encapsulated in the MTS (b)(excitation wavelength 290 nm). For (b) and (c), the bandwidth is 3 nm in excitation and 1.5 nm in emission.

for pyoverdins (Fig. 1a). Its absorption maximum is observed at $\lambda_{max} = 380$ nm ($\varepsilon = 24\,534$ $M^{-1}\,cm^{-1}$) at pH 5.0. The chromopeptide emits a strong fluorescence with a maximum at $\lambda_{em} =$ 452 nm for an excitation at 400 nm (Fig. 1c). The spectrum is independent of the exciting wavelength.

A study of the fluorescence dependence on pH for pyoverdin shows that the maximum in emission occurs at pH 7.0. It also reveals the disappearance of the fluorescence when the pH becomes lower than 3.0 (Fig. 2). This loss of fluorescence has its origin in the protonation of the catechol groups. As said above, these phenols groups are involved in the complexation of Fe(III) so that it is expected that at the same pH pyoverdin should release the metal ion [3].

The natural fluorescent pigment used forms a 1:1 complex with Fe(III) [3]. After control, this complex was shown to be non-fluorescent either free in solution or adsorbed on mesoporous silica, so that the amount of iron ions in a solution can be quantified by the decrease of the fluorescent signal of the pigment. The absorption spectrum of the iron complex of pyoverdin has a maximum at

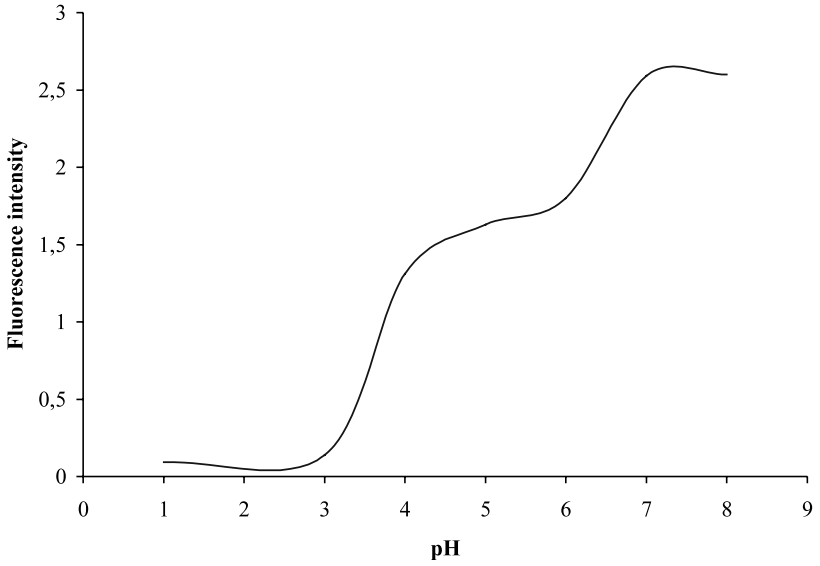

Fig. 2. pH dependence of the fluorescence intensity of pyoverdin (2.9×10^{-4} M), excited at 400 nm and monitored at 450 nm.

$\lambda_{max} = 400$ nm ($\varepsilon = 24\,804$ $M^{-1}\,cm^{-1}$) and two shoulders at 480 nm ($\varepsilon = 6615$ $M^{-1}\,cm^{-1}$) and 550 nm ($\varepsilon = 2949$ $M^{-1}\,cm^{-1}$) well into the visible [29], which gives it a reddish colour (Fig. 3). This spectrum strongly overlaps that of pyoverdin.

Due to the very high affinity of Fe(III) for pyoverdin, the collisional quenching of free pyoverdin by Fe(III) ions, if it existed, would be difficult to prove. In absolute such a quenching is expected since these ions are paramagnetic. We carried several tests, for instance with dansyl-labelled bovine serum albumin, and did not observe a quenching effect in the presence of different concentration of Fe(III) in the range

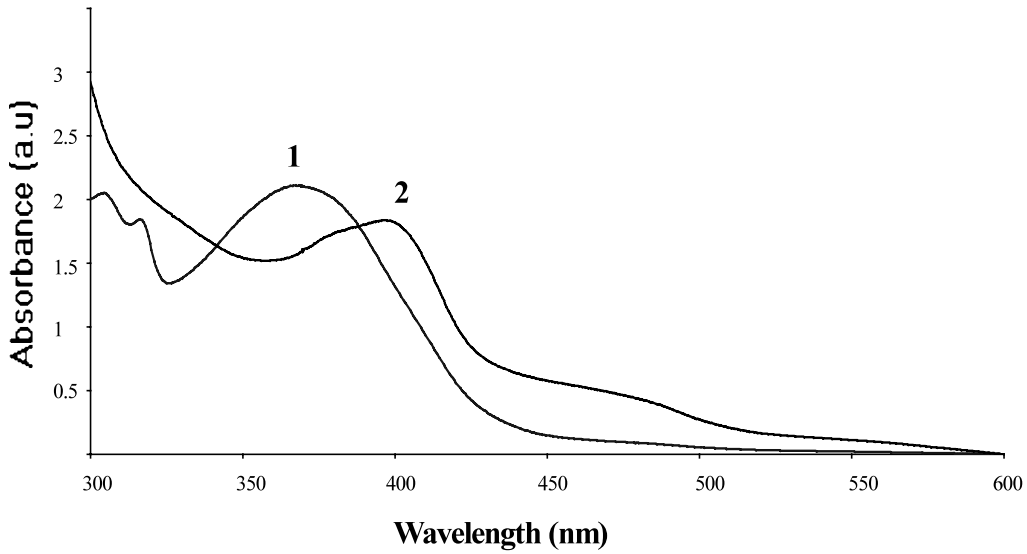

Fig. 3. Electronic absorption spectra of the Fe(III) complex of pyoverdin 2 and pyoverdin 1 (in water at pH 7) revealing their strong overlap.

used in this work. No significant decrease of the fluorescence intensity was observed. Palanché et al. arrived also to the same conclusions, using isobutylamino-NBD [29].

The parameters affecting the determination of Fe(III) with pyoverdin in solution and the effect of the presence of interfering elements were then investigated.

For a given concentration of pyoverdin, the data expressed as the maximum relative fluorescence intensity versus iron concentration define a portion of a straight line before showing some upwards curvature at higher concentrations. For instance with 2.9×10^{-4} M pyoverdin loaded on the solid, fluorescence allows the determination of 3–43 μg Fe(III) (1–14.3 mg l^{-1}) and the data fit a linear regression given by $y = -0.0297x + 1.274$ ($r^2 = 0.969$). This confirms that pyoverdins in general and related fluorescent siderophore ligands can be used for sensing iron at quite low levels [18,29].

3.2. Selectivity

In view of our objective, the selectivity of the complexation was tested. To this effect, different ions were added, separately or together, at varying concentrations to evaluate their effect on the Fe(III) quantification. None of these ions tested interfere as they have no affinity or an extremely reduced affinity for the ligand compared to iron. We could then quantify Fe(III) ions in the same concentration range as above (1–14.3 mg l^{-1}) in the presence of concentrations of up to 15 mg l^{-1} for Cr, Cu, Ni, Zn and Pb ions. It is interesting to note that for a different siderophore, an analogous study showed that Cu(II) could compete albeit with a much lower efficiency [29], so that the choice of the chelators will be important for future developments.

3.3. Synthesis and characterisation of the mesoporous MCM containing pyoverdin

Encapsulation of the pyoverdin in the mesopores of a MTS was achieved by direct synthesis in soft conditions that do not affect the structure and activity of the biomolecule. The direct synthesis results in a mesoporous solid in which the pores are entirely filled by surfactant. This material is known as an 'as synthetised' solid. In our case, the surfactant is replaced by lecithins already used to prepare mesoporous aluminosilicates [26].

After the washing steps with ethanol which should remove the lecithin from the pores, the textural features of the hybrid mesoporous mineral obtained were determined by means of X-ray diffraction and nitrogen volumetry. The X-ray diffraction spectrum is similar to that of a MTS with an ordered hexagonal mesoporous texture. The lattice parameter of the hexagonal array determined from d_{100} is given in Table 1 together with other textural properties. The isotherm (type IV, not shown) presents an almost vertical step for relative pressure p/p_o, in the vicinity of 0.3–0.4, without hysteresis. It is typical of monodisperse pores. However, the surface area S resulting from analysis of these isotherms by the BET equation in the p/p_o range 0.05–0.2 (Table 1) as well as the mesoporous volume V_m, measured at the top of the pore-filling step indicate that the pores are only partly filled. Effectively these materials usually have larger microporous volumes. This may be due to the fact that some lecithin is left after the washing, since the reference solid prepared without pyoverdin is yellowish. The pore diameter is close to 3 nm. In pores of that size diffusion is unhindered for metal ions.

Almost identical properties and values were found for the reference solid obtained in the same conditions but for the presence of pyoverdin.

3.4. Uptake and release of Fe(III) ions

To establish a mass balance for the solid loaded with pyoverdin, the fluorescence spectra were obtained for the starting pyoverdin solution and for the supernatant solution after synthesis and washing of the solid material. Fluorescence spectra

Table 1
Textural properties of the MTS

a (Å)	S ($m^2\ g^{-1}$)	V_m (ml g^{-1})
35	514	0.3

were also recorded for the supernatant solution after hydrolysis of the hybrid solid and chromatography on Sephadex G25 of the solution obtained. The difference in solution concentrations calculated from the maximum fluorescence emission intensity at 460 and 435 nm, respectively yielded the amount of pyoverdin loading of the support. The result is that 44% of the initial quantity used for immobilisation was adsorbed in the channels and resisted the ethanol extraction.

We have then used this encapsulated pyoverdin in the MTS mesoporous material to uptake Fe(III) ions from the multimetallic solution. The optimum pH for the formation of the Fe(III)–pyoverdin complex was obtained with a 0.01 N phthalic acid/biphthalate solution (pH 4.5) [19]. With 1 g of mesoporous material containing immobilised pyoverdin, 80 μg Fe(III) were adsorbed from the solution. When blanks were run with the reference material, 11 μg Fe(III) only were adsorbed. These tests are reproducible. All of the trapped iron could be removed from the solid by treatment with a dilute HCl solution or with a sodium sulfite solution. In the latter case, Fe(III) is reduced to Fe(II) for which pyoverdin has no affinity.

Compared to an aqueous solution where the emission maximum of pyoverdin is at 450 nm for an excitation at 400 nm, the emission maximum is shifted to 390 nm after immobilisation (Fig. 1b). This shift is due to a change in the luminophore environment following its adsorption on silica. The UV–Vis reflectance spectra of the immobilised pyoverdin recorded versus the reference mesoporous reveals also the presence of the chromophore.

These results mean that the coupling of several techniques, especially spectroscopic techniques, will help ascertain the presence of the chelators in the channels of the porous solid.

4. Conclusion

We have shown in that preliminary study that a mesoporous solid encapsulating a selective chelator (pyoverdin for ferric ions) could be used to remove selectively and then release these ions under control with an acid or a reducing solution. The results were monitored by FAAS and by spectrofluorimetry and are in good agreement. The amount of metal complexed by the chelator is small, but as this quantification was based on the fluorescence properties of pyoverdin, we restricted the study to the concentration range giving a linear response. The loading of such a solid could be made much higher. These hybrid metal sponges could be used for preconcentration purpose. However much work remains to perfect the system along different lines such as increasing the number of cycles that the solid may resist, compare spontaneous adsorption with covalent grafting for the chelator, screen existing natural chelators for other metals, control the size and shape of the particles. Recently published results give strong hope to solve these problems [30–32] and we actively pursue our work.

Acknowledgements

Support from the ‘Ministère de la Recherche’ is acknowledged for an ‘ACI Jeune Chercheur’ grant N°4100 for M. M.

References

[1] I. Leopold, D. Günther, Fresenius J. Anal. Chem. 359 (1997) 364.
[2] J.M. Meyer, F. Halle, D. Hohnandel, P. Lemanceau‘, H. Ratefiarivelo, VCH Verlagsgesellchaft, Weinheim, 1987, pp. 188.
[3] A.-M. Albrecht-Gary, S. Blanc, N. Rochel, A.Z. Ocaktan, M.A. Abdallah, Inorg. Chem. 33 (1994) 6391.
[4] T.A. Kendall, J.M.F. Hochella, in: Eleventh Annual V.M. Goldschmidt Conference, 2001.
[5] D. De Vos, A.J. Lim, J.P. Pimay, R.L. Duinslaege, H. Revets, A. Vanderkelen, R. Hamers, P. Cornelis, Burns 23 (1997) 379.
[6] H. Budzikewicz, Microbiol. Rev. 104 (1993) 209.
[7] M. Julich, K. Budzikiewicz, V. Geoffory, J.M. Meyer, L. Gardon, J. Biosciences 56 (2001) 687.
[8] H. Georgias, K. Taraz, H. Budzikiewicz, V. Geoffroy, J.M. Meyer, Z. Naturforsch. 54c (1999) 301.
[9] J.M. Meyer, M.A. Abdallah, J. Gen. Microbiol. 107 (1978) 319.
[10] S. Wandenbaum, P. Demange, A. Dell, J.M. Meyer, M.A. Abdallah, Tetrahedron Lett. 24 (1983) 4877.

[11] O. Zaborsky, Immobilized Enzymes, CRC Press, Cleveland, 1973.
[12] W. Hartmeier, Immobilized Biocatalysts—An Introduction, Springer, Berlin, 1988.
[13] J.F. Diaz, K.J. Balkus, Jr., J. Mol. Catal. B. Enzymatic 2 (1996) 115.
[14] M. Reetz, Adv. Mater. 9 (1997) 943.
[15] T. Asefa, M.J. MacLachan, N. Coombs, G.A. Ozin, Nature 402 (1999) 867.
[16] D. Avnir, S. Braun, O. Lev, M. Ottolenghi, Chem. Mater. 6 (1994) 1605.
[17] D. Brunel, in: D.C.S. a. A.P. Kybett (Ed.), Supported catalysts and their applications RSC, Cambridge, 2001.
[18] J.M. Barrero, M.C. Moreno-Boudi, M.C. Pérez-Condé, C. Camara, Talanta 40 (1993) 1613.
[19] P. Pulido Tofino, J.M. Barrero-Moreno, M.C.M.C. Pérez-Conde, Talanta 51 (2000) 537.
[20] J.S. Beck, J.C. Vartuli, W.J. Roth, M.E. Leonovicz, C.T. Kresge, K.D. Schmitt, C.T.W. Chu, D.H. Olson, E.W. Sheppard, J. Am. Chem. Soc. 114 (1992) 10834.
[21] C.T. Kresge, M.E. Leonowicz, W.J. Roth, J.C. Vartuli, J.S. Beck, Nature 359 (1992) 710.
[22] Q. Huo, D.I. Margolese, U. Ciesla, P. Feng, T.E. Gier, P. Sieger, R. Leon, P.M. Petroff, F. Schüth, G.D. Stucky, Nature 386 (1994) 317.
[23] A. Imhof, D.J. Pine, Nature 389 (1997) 948.
[24] A. Galarneau, D.A. Lerner, M.F. Ottaviani, F.D. Renzo, F. Fajula, Stud. Surf. Sci. Catal. 405 (1998).
[25] C.-Y. Chen, S.-Q. Xi, M.E. Davis, Microporous Mater. 4 (1995) 1.
[26] Y.G. Goltsov, L.A. Matkovskaya, Z.V. Smelaya, V.G. lI'in, Mendeleev Commun. (1999) 241.
[27] C. Tourné-Péteilh, D.A. Lerner, C. Charnay, L. Nicole, S. Bégu, J.-M. Devoisselle, Chem. Phys. Chem, in press.
[28] P. Demange, A. Bateman, A. Dell, M.A. Abdallah, Biochemistry 27 (1988) 2745.
[29] T. Palanché, F. Marmolle, M.A. Abdallah, A. Shanzer, A.-M. AIbrecht Gary, JBIC 4 (1999) 188.
[30] S. Kawi, M.W. Lai, Chem. Commun. (1998) 1407.
[31] I. Kiricsi, A. Fudala, Z. Konya, K. Hernadi, P. Lentz, J.B. Nagy, Appl. Catal. A: Gen. 203 (2000) L1.
[32] C. Boissière, A. Larbot, A. van der Lee, P.J. Kooyman, E. Prouzet, Chem. Mater. 12 (2000) 2902.