Hydrodechlorination of Aroclor 1260 in Aqueous Two-phase Mixture Catalyzed by Biogenerated Bimetallic Catalysts

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Original Research Article

ABSTRACT

The PCBs are known recalcitrant and toxic pollutants and significant values of contamination could be found in water. PCBs can be hydrodechlorinated using Pd-based catalysts and hydrogen, but the research to identify more efficient heterogeneous catalysts, able to work in an aqueous phase, less sensible to deactivation and easily removable at the end of the treatment, remains a

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considerable interesting goal. A strain of *Klebsiella oxytoca*, DSM 29614, known to produce a specific exopolysaccharide (EPS), was grown in different media with sodium citrate or with sodium citrate plus ferric citrate, as sole energy and carbon sources under anaerobic conditions. The cultures were amended with 50 mg of palladium as $Pd(NO_3)_2$ to generate Pd-EPS (Pd content 13%) or FePd-EPS (Pd content 8.4%; Fe 7.4%) species which were secreted from the cells, isolated by treatment with a cold ethanol solution (70%) and dried under vacuum as powders. The catalytic ability of these mono- and bi-metallic species was tested in the hydrodechlorination reaction of the Aroclor 1260 PCBs mixture under aqueous biphasic conditions. Here we demonstrate that the degree of PCBs hydrodechlorination is dependent by the nature of catalyst and of base used to neutralize HCl produced, the bimetallic species being more active and an organic base resulting more effective. Working with a substrate/catalyst 8/1 molar ratio, at 3 MPa H_2 and 60°C in 20 h a significant removal of highly chlorinated PCBs was obtained under the best conditions. The result seems promising for remediation of groundwater contaminated with PCBs.

1. INTRODUCTION

Polychlorinated biphenyls (PCBs) are a class of 209 organic pollutants produced in complex mixtures with a different mean chlorination degree [1]. Due to their hydrophobic nature and high stability, PCBs are characterized by strong persistence in the environment, and significant values of contamination could be found in water [2]. In combination with their toxicity this stimulates researches for reducing their concentration in the ecosystem. PCBs can be hydrodechlorinated using Pd-based catalysts that show a very high potential for the destruction of such contaminants in water [3,4]. This process may be highly efficient and selective in clean water even under mild conditions. However, palladium is very sensitive to deactivation, especially in waste-waters, due to its sequestration and precipitation with suspended organic matter. In addition, loss of Pd(0) reactivity is mostly due to hydrogen sulfide (H_2S and HS^-), which is ubiquitous in anoxic ecosystem [5,6]. For these reasons, Pd-based catalysts are rarely used for applications in water treatment. Until now only a few field-scale studies have investigated the use of Pd-based catalysts for hydrodechlorination (HDC) in groundwater, but the interest for its application remains very high [3,7-9]. Conventional Pd catalysts have been flanked in HDC processes also by biological dechlorination and reductive dechlorination of PCBs by anaerobic microorganisms has been demonstrated to occur [10-13]. However, biological reductive dechlorination usually is a slow reaction and it is often difficult to avoid conflicts when a mixture of indigenous and external microorganisms is applied [14].

Over the last decades, metal nanoparticles with different composition, size, shape and dispersity have been produced through the use of microorganisms. Several years ago some approaches were identified to obtain Pd(0) nanoparticles which were catalytically very active and more resistant to deactivation. Nanoscale metal particles (NPs) are usually characterized by high surface area to volume ratios, high levels of stepped surfaces and high surface energies showing so improved reactivity, even at high loads of compounds to be degraded, including PCBs, by dechlorination [15-22].

In this study, encouraged by our previous results obtained in the hydrodechlorination of mono and trichloro benzenes using purified biogenerated Pd-binding polysaccharide [23,24] we decided to verify the possibility to work with more complex polychlorinated biphenyls, such as Aroclor 1260.

2. EXPERIMENTAL DETAILS

2.1 Materials and Instrumentation

$NaHCO_3$, NH_4Cl, $MgSO_4$, NaH_2PO_4, KCl, Na-citrate, Fe(III)-citrate, $Pd(NO_3)_2$, Na_2CO_3, ammonium acetate, Aroclor 1260, 95% Ethanol, Methanol, were Sigma-Aldrich products. n-Hexane (pesticide grade) was a Romil product. Nutrient broth was a Difco product. Non-labelled PCBs were AccuStandard products. All reagents were used as received. PCB determinations were performed in full scan mode on a fused silica capillary column (HP5-MS 30 m, 0.25 mm x 0.25 μm; Agilent Technologies) installed in a ThermoFinnigan (Trace GC 2000) coupled to a quadrupole mass spectrometer (ThermoFinnigan Trace MS). The GC-MS parameters were as

follows: solvent delay 4.5 min; initial temperature 150°C for 5 min; heating ramp 10°C/min; final temperature 300°C for 30 min; injector temperature (split) 280°C; temperature (interface) 300°C; temperature (source) 200°C; detector voltage 350 V; gas carrier flow (He) 0.8 mL/min; injection volume 1 µL; mass range (amu) 35-500.

We searched for 126 different congeners, corresponding, in our conditions, to 83 chromatographic peaks, including some typical Aroclor PCBs and dioxin-like PCBs.

2.2 Preparation of Catalysts

The Pd-EPS (Pd content 13%) and FePd-EPS (Pd content 8.4%; Fe 7.4%) catalysts were biogenerated by *Klebsiella oxytoca,* DSM 29614 (ex BAS-10), according to the procedure recently published [24], by adding to the culture broth, maintained under anaerobic conditions, only 50 mg of $Pd(NO_3)_2$ or 50 mg of $Pd(NO_3)_2$ plus 50 mg of Fe(III)-citrate, respectively. All cultures were first centrifuged to eliminate bacterial cells. The supernatants were treated with 800 ml of cooled ethanol (95%) to precipitate the metal polysaccharides. The purifications were repeated twice. Each colloidal material was finally dried out under vacuum and grinded to powder to obtain both catalysts.

2.3 Element Determination in Poly-saccharides

Samples (1 mg) of dry pulverized Pd-EPS and FePd-EPS were digested with 2 ml of aqua regia, heating the mixture at 333 °K until a solution was obtained. The total amounts of elements were determined versus their relative standards solutions by inductively coupled plasma atomic emission spectrometry (ICP-AES) (Optima 3100, Perkin Elmer).

2.4 Hydrodechlorination of Aroclor 1260. General Procedure

If not otherwise indicated (see footnotes of Table 1), 2 mg of Pd-EPS or 3 mg of FePd-EPS were stirred in a Schlenk tube under nitrogen in 2 ml of distilled water for about 10 min. A solution of 7 mg of Aroclor 1260 (~0.0195 mmol assuming MW 358 as medium value) in 1 ml of methanol was then added to the aqueous phase to obtain a substrate/Pd = 8/1 mol ratio and as well as 1.7 mg (0.021 mmol) of ammonium acetate. The Schlenk tube was then transferred into a 150 ml stainless steel autoclave under nitrogen, pressurized with 3 MPa of H_2 and stirred at 333° K for 20 h. The reactor was then cooled to room temperature and the residual gas released. Then the sample was extracted three times via ultrasonic bath using aliquots of 10 mL of n-hexane (pesticide grade, Romil). The organic phases were collected and dried on Na_2SO_4, concentrated to 100 µL using a nitrogen flow (Turbovap 2, Caliper Science) and analyzed by GC-MS.

3. RESULTS AND DISCUSSION

In Table 1 results of different runs obtained by changing some reaction parameters (catalyst, amount and type of base) are reported. A% of the individual congener is calculated by integrating all considered peaks (83) and dividing the area of the individual congener for the total area. For each homologous class (same number of chlorine atoms) the areas of the components of the class are added together and similarly related to the total area. To rationalize the identification of defined PCB components in Table 2, the method, developed by K. Ballschmiter and M. Zell [25] and based on the IUPAC rules of substituent characterization in biphenyls, is followed. The number of position according to this arrangement is taken as an abbreviation for the PCB structure. The distribution of congeners before and after the reactions is better visualized in Fig. 1. In Fig. 2 is reported the chromatogram of used Aroclor 1260 and in Fig. 3 the chromatogram of the final mixture in run 5. In absence of any palladium catalyst, as expected, no significant variation of congeners distribution of Aroclor 1260 occurs (run 1). On the basis of our recent good results in the hydrodechlorination of 1,2,4-trichlorobenzene [24], initially we worked in absence of a base (runs 2 and 3), so avoiding to neutralize the HCl formed. The distribution displacement of PCBs concentrations from the heaviest (n°Cl 6-10) to the lightest (n°Cl 0-5) compounds resulted increased, suggesting an encouraging improvement of the hydrodechlorination process. Surprisingly the mono metallic catalyst worked slightly better than the bimetallic catalyst while in the hydrodechlorination of 1,2,4-trichlorobenzene an opposite result was found [24]. In the presence of an inorganic base a relevant loss of activity of the catalyst was observed (run 4) ; on the contrary a synergic positive effect was found using ammonium acetate as base with the most relevant decrease in the concentrations of PCBs with higher number of chlorine atoms (n°Cl 6-10;

-59%). In particular it is to underline the significant A% decrease , between -49% and -85%, of some dioxin–like PCBs congeners: BZ 118, 156, 157, 167, 170, 180, 189 (see Table 2, where in the column Aroclor 1260 the A% of the congeners present in the original PCBs mixture are reported, while in the columns Run 2, Run 3 and Run 5 are reported the A% of the single congeners found after the hydrodechlorination experiments carried out at different reaction conditions described in Table 1). Finally a preliminary attempt to recycle the catalyst embedded in the aqueous phase was, in our opinion, positive (run 6); the catalyst was still enough active, even if a loss of activity was observed. However we believe that the observed decreased activity is mostly due to unwanted and not quantifiable losses of catalyst suspended in the aqueous phase, during the work-up and recycle.

Table 1. Results in the hydrodechlorination experiments starting from Aroclor 1260

n°Cl	Aroclor 1260[a]	Run1[b]	Run 2[c]	Run 3[d]	Run 4[e]	Run 5[f]	Run 6[g]
	A%[h]	A%[h]	A%[h]	A%[h]	A%[h]	A%[h]	A%[h]
0	0.02	0.03	0.21	2.57	0.05	3.37	0.17
1	0.06	0.06	0.27	1.21	0.40	2.04	0.38
2	0.25	0.17	1.28	4.28	0.67	8.41	2.48
3	0.52	0.43	4.97	8.59	1.82	15.40	5.30
4	0.61	0.45	8.88	10.62	2.88	14.42	5.21
5	10.6	10.3	25.5	21.4	14.5	20.18	14.8
6	46.1	46.9	37.6	29.7	43.0	21.22	38.9
7	34.0	33.3	18.0	17.8	29.9	12.24	26.9
8	7.00	7.46	3.07	3.50	6.26	2.48	5.22
9	0.80	0.84	0.28	0.31	0.49	0.23	0.59
10	0.04	0.01	0.01	0.00	0.01	0.00	0.03

[a] starting material; [b] general procedure but absence of any Pd catalyst (blank test); [c] general procedure in the presence of FePd-EPS but in absence of base; [d] general procedure in the presence of Pd-EPS but in absence of base; [e] general procedure in the presence of FePd-EPS but using sodium carbonate instead of ammonium acetate; [f] general procedure in the presence of FePd-EPS; [g] reaction carried out using the aqueous phase containing the catalyst of previous run and adding further amount of Aroclor 1260; [h] sum of congeners
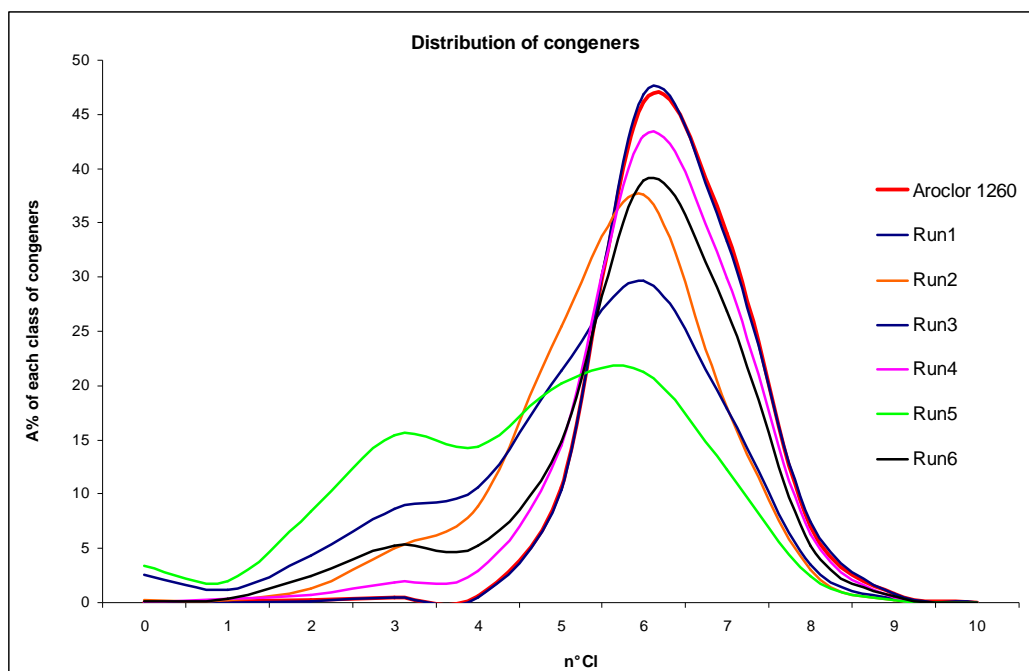

Fig. 1. Distribution and area % of congeners in Aroclor 1260 and in the different experiments
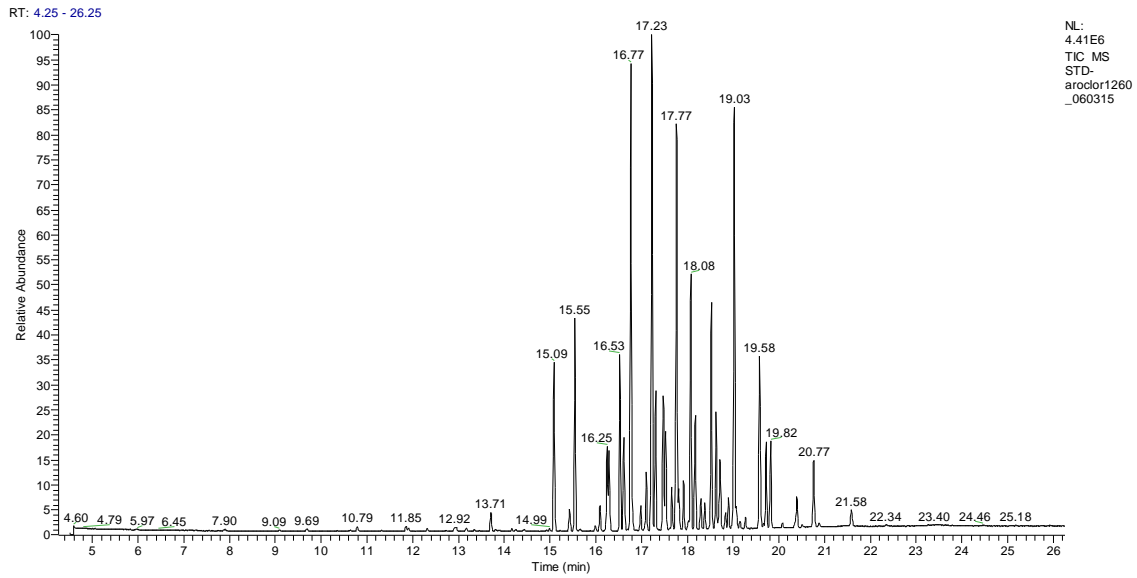
Fig. 2. Chromatogram of used Aroclor 1260
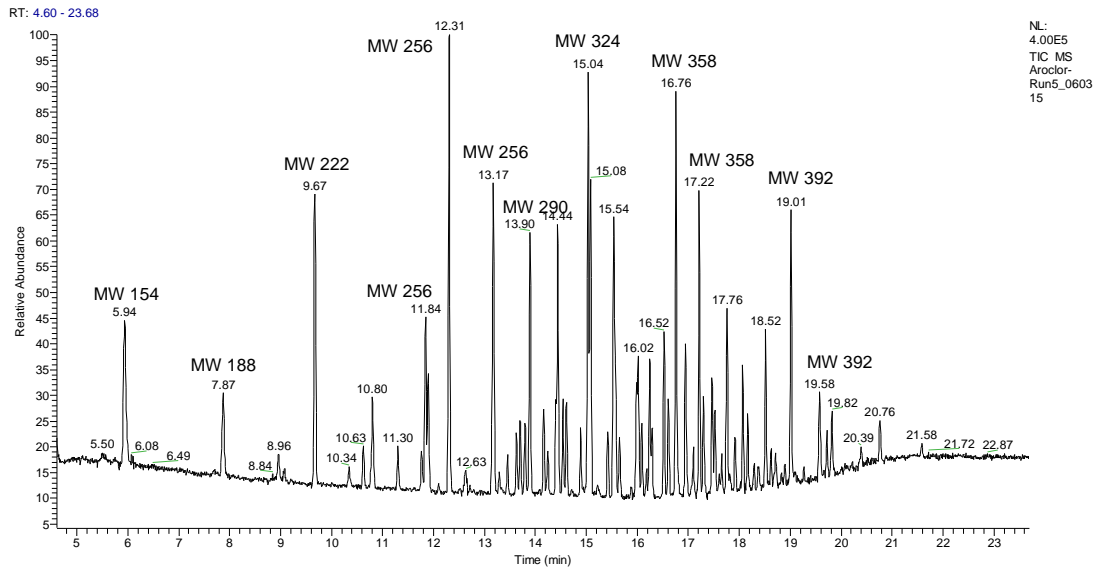
Fig. 3. Chromatogram of product mixture (run 5)

Table 2. PCB concentrations (A%) of analyzed Aroclor 1260 and of three hydrogenated mixtures

#	n°Cl	MW	BZ [1]	ID[2]	Aroclor 1260 A%	Run 2 A%	Run 3 A%	Run 5 A%
1	Biphenyl	154		STD	0.01	0.21	2.57	3.37
2	1	188		no ID	0.04	0.21	1.02	1.49
3	1	188		no ID		0.04	0.16	0.38
4	1	188		no ID	0.01	0.03	0.02	0.17
5	**2**	**222**	**4**	**STD**	**0.06**	**0.51**	**2.81**	**4.66**
6	2	222		no ID	0.01	0.03	0.11	0.32

5

#	n°Cl	MW	BZ[1]	ID[2]	Aroclor 1260 A%	Run 2 A%	Run 3 A%	Run 5 A%
7	2	222		no ID	0.02	0.15	0.45	0.64
8	**2**	**222**	**5**	**STD**	**0.11**	**0.30**	**0.49**	**1.54**
9	3	256		no ID	0.01	0.19	0.49	0.65
10	2	222		no ID		0.07	0.20	0.54
11	**3**	**256**	**18**	**STD**	**0.11**	**0.82**	**1.45**	**2.02**
12	2 + 3[3]			no ID	0.06	0.43	0.46	1.43
13	3	256		no ID		0.08	0.12	0.11
14	3	256		no ID	0.07	0.92	2.79	6.12
15	3	256		no ID	0.01	0.05	0.05	0.09
16	3	256		no ID		0.03	0.01	0.04
17	**3**	**256**	**31**	**STD**	**0.09**	**0.07**	**0.03**	**0.04**
18	**3**	**256**	**28**	**STD**	**0.07**	**0.06**	**0.01**	**0.02**
19	3	256		no ID	0.08	2.17	3.17	5.10
20	3	256		no ID	0.03	0.06	0.05	0.27
21	4	290		no ID		0.28	0.43	0.45
22	4	290		no ID		0.35	0.74	0.82
23	**4**	**290**	**52**	**STD**	**0.39**	**1.00**	**0.85**	**1.03**
24	4	290	49	cfr	0.05	0.79	0.56	0.93
25	4[3]	290	47+48	cfr		1.77	2.37	3.44
26	**4**	**290**	**44**	**STD**	**0.06**	**0.83**	**1.11**	**1.06**
27	3 + 4[3]			no ID	0.03	0.61	0.38	0.50
28	4	290		no ID	0.03	2.39	3.05	4.42
29	5	324	96	cfr		0.43	0.65	1.11
30	4	290	74	cfr		0.40	0.83	1.22
31	4	290	70	cfr	0.02	0.61	0.45	0.78
32	4	290	66	cfr	0.06	0.17	0.04	0.03
33	5	324		no ID		3.44	3.15	4.47
34	5	324		no ID	3.51	5.04	3.99	3.63
35	5	324		no ID	0.48	1.09	0.91	0.80
36	**5**	**324**	**101**	**STD**	**4.37**	**6.03**	**4.17**	**3.67**
37	5	324	99	cfr		0.83	1.74	1.03
38	5	324	119	cfr	0.04	1.32	0.65	0.65
39	5	324	83	cfr	0.10	1.51	1.76	1.08
40	5	324	97	cfr		1.03	1.69	1.66
41	5[3]	324	87 + 115	cfr	0.52	1.81	1.04	0.87
42	5	324	85	cfr		0.87	0.31	0.32
43	6	358	136	cfr	1.52	2.10	1.65	1.50
44	5	324	110	cfr	1.66	2.11	1.34	0.89
45	6	358	151	cfr	3.64	3.57	2.78	1.92
46	6	358	135	cfr	2.12	2.20	1.85	1.18
47	**5 + 6[3]**		**118 + 149**	**STD**	**10.18**	**9.12**	**7.34**	**5.16**
48	6	358		no ID		1.58	1.60	2.02
49	6	358		no ID	0.50	0.29	0.16	0.06
50	6	358		no ID	1.25	1.16	0.83	0.42
51	**6**	**358**	**153**	**STD**	**10.30**	**6.06**	**5.10**	**3.38**
52	6	358	132	cfr	2.79	2.96	2.15	1.21
53	6	358	141	cfr	2.75	2.06	2.28	1.36
54	7	392	179	cfr	2.03	1.60	1.30	1.00
55	6	358	137	cfr		0.46	0.21	0.08
56	7	392	176	cfr	0.85	0.78	0.54	0.44
57	**6**	**358**	**138**	**STD**	**9.28**	**4.81**	**3.18**	**2.47**

#	n°Cl	MW	BZ[1]	ID[2]	Aroclor 1260 A%	Run 2 A%	Run 3 A%	Run 5 A%
58	6	358		no ID	0.59	0.36	0.17	0.18
59	7	392	178	cfr	0.99	1.08	1.53	0.72
60	7	392	187	cfr	0.16	0.14	0.06	0.02
61	7	392	183	cfr	5.13	2.45	2.44	1.48
62	7	392	185	cfr	2.33	1.26	1.20	0.84
63	6	358	167	STD	0.72	0.84	0.37	0.29
64	7	392	174	cfr	0.54	0.35	0.51	0.25
65	7	392	177	cfr	4.58	2.99	3.00	1.92
66	7	392	171	cfr	2.43	0.82	0.53	0.46
67	6 + 7[3]		156 + 173	cfr + STD	1.84	0.80	0.44	0.39
68	6 + 8[3]		157+ 197	cfr + STD	0.31	0.24	0.24	0.07
69	7	392		no ID	0.65	0.35	0.30	0.25
70	**7**	**392**	**180**	**STD**	**8.88**	**3.99**	**4.55**	**3.27**
71	7	392	193	cfr	0.32	0.06	0.04	0.06
72	7	392	191	cfr	0.14	0.05	0.01	0.01
73	**8**	**426**	**199**	**STD**	**0.23**	**0.18**	**0.07**	**0.15**
74	7[3]		170 + 190	cfr	4.25	1.60	1.60	1.33
75	8	426	203	cfr	1.69	0.58	0.71	0.63
76	8	426	196	cfr	1.93	0.98	1.17	0.79
77	**7**	**392**	**189**	**STD**	**0.13**	**0.04**	**0.02**	**0.02**
78	8 + 9[3]		195 + 208	cfr	0.66	0.23	0.29	0.10
79	9	460	207	cfr	0.07	0.01	0.01	0.01
80	**8**	**426**	**194**	**STD**	**1.57**	**0.56**	**0.94**	**0.58**
81	8	426	205	cfr	0.08	0.01	0.01	0.02
82	9	460	206	cfr	0.42	0.16	0.15	0.16
83	**10**	**494**	**209**	**STD**	**0.05**	**0.01**	**0.00**	**0.00**

[1] BZ = Congener number according to ref. [25]; [2] STD = Identification by standard injection; cfr = Tentatively identified by retention time and elution order according to ref. [26]; no ID when a defined congener was not identified but only the number of chlorine atoms was determined; [3] Determined by mass analysis

4. CONCLUSION

In conclusion, the development of new bio-generated metal catalysts, prepared by a greener synthetic approach, is a highly promising area. Noteworthy, these new types of Pd catalysts were purified by alcoholic extraction of exopolysaccharide binding metals in order to eliminate cells as much as possible from culture broth: this procedure distinguishes our catalysts from other biogenerated ones containing whole cells [27]. In our opinion, they present less drawbacks and risks, therefore they are more reliable for use and reuse in comparative experiments with respect to other conventional heterogeneous Pd catalysts. The use of these catalysts, embedded in the aqueous phase and active only in the presence of water, may improve the reductive dehalogenation of pollutants present in aqueous environment, in particular those containing a higher number of chlorine atoms that are more recalcitrant for a microbial dehalogenation. At the present, FePd-EPS, combined with ammonium acetate, results the more efficient catalyst in this reaction that might be improved using a continuous flow reactor as reported in the literature for other dehalogenation reactions [6]; furthermore it is prepared easier than the monometallic catalyst and contains a lower amount of expensive precious metal.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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