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| **Ionic chromatography analysis of the bioremediation medium in contact with Gellan cryogels with and without bacteria**In order to estimate the composition of the MSM in presence of Gellan cryogel an adsorption of ions was performed using AAS spectroscopy. AAS results indicate that gellan gum does not significantly affect metal ion concentration, therefore it won’t affect the viability of bacteria in the MSM over the bioremediation experiment. Initial concentration of chloride ions was 7497 ppm. It seems that in presence of free Rhodococcus and Pseudomonas in the solution containing 4CP in MSM, HPLC is needed to confirm some decline of chloride(excel file). The decrease of the chloride ions content in the solution with unknown bacteria most probably is related to the growth of the biomass. The control cryogels based on GellanG revealed some sodium adsorption from the MSM solution Monitoring of 4CP concentration over one month reveal gradual decrease of the concentration down to 60% of initial concentration. For comparison we have previously showed that these strains can degrade phenol at concentration 50mg/L within 5-7 days. Therefore another 4CP degrading bacteria should be used. Poor degradation activity of 4CP by cryogels based on gellan gum compare to synthetic polymers (PEIal and PVAal) is most probably related to consumption of gellan gum as a source of carbon. Another reason of low degradation activity may be poor diffusion of 4CP inside of the cryogel, due to Gellan Gum cryogels characterized by comparatively small pores and thick walls(low flow through properties) compare to conventional cryogel

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| **Wet weight g of bacteria in the cryogel**

|  |  |  |
| --- | --- | --- |
| 0.091419 | 0.09433 | 0.099151 |

 **Number of viable cells in each cryogel**

|  |  |  |
| --- | --- | --- |
| **1.46E+08** | **1.51E+08** | **1.59E+08** |

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**Bioremediation of chlorophenol, m-cresol and phenol**

4CP resistant Psedomonas and **Rhodococcus koreenisis** bacteria were isolated after 3 weeks in carbonate buffer containing 50 mg/L of 4-chlorophenol. 4CP resistant, Psedomonas and **Rhodococcus koreenisis** free bacteria and cryogels showed moderate degradation of 4CP over 30 days. Concurrently to 4CP concentration measurement the number of alive bacteria was estimated using OD. It was typical for that number of bacteria decline over the bioremediation process. Number of cells declined over the duration of the experiment, therefore in following calculations the initial Number of cells was excluded. More or less the same initial number **5E+07** of cells were used in each experiment. Poor degradation activity of 4CP by cryogels based on gellan gum compare to synthetic polymers (PEIal and PVAal) is most probably related to consumption of gellan gum as a source of carbon. Another reason of low degradation activity may be poor diffusion of 4CP inside of the cryogel, due to Gellan Gum cryogels characterized by comparatively small pores and thick walls (low flow through properties) compare to conventional cryogel. It was illustrated that Psedomonas and **Rhodococcus koreenisis can degradate 4CP only in presence of MSM, these strains did not show significant bioremediation activity in the carbonate buffer.**

PVA-al and combinations of PVA-al &Pei-al are more suitable for processes performed at pH7.2 or alkaline, since Schiff’s base group is not stable even at slightly acidic conditions, and it can lead to lose of mechanical strength of the material in long term use. To overcome this disadvantage physically interrupt bacteria cryogels can be used.

Cryogels based on Gellan is more suitable for immobilization of bacteria which will be utilized in slightly acidic or acidic environment, due to according to preliminary experiments showed good stability over time. The main drawback may be that bacteria should not consume the gellan gum as a source of carbon.