



AIKU AMBIENTE SRL

ADVANCED REMEDIATION TECHNOLOGY



HIP-Petrohemija a.d. Pancevo (Serbia)

TESTING OF GROUNDWATER CONTAMINATED WITH CHLORINATED SOLVENTS

06.09.2021 - 21.10.2021

FINAL REPORT

Milan, 03/02/2022

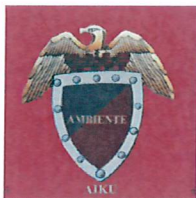
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INTRODUCTION

The MPCD® protocol for the degradation of chlorinated solvents has been applied. This activity began in September 6, 2021, and was carried out at the HIP industrial site in Pancevo (Serbia), with analytical results up to 45 days of treatment. The test results and related considerations and evaluations are reported below.

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MATERIALS AND METHODS

The test was conducted on portions of groundwater contaminated with chlorinated solvents, in particular vinyl chloride (VCM) and 1, 2, dichloroethane (ECD). The waters were taken from piezometer RB-12.

The procedure involved the following steps:

- take 1m³ of water from piezometer RB-12 and pour it into a 1.000 Lt. aerated tank (sample A).
- take 800 Lt. of water from piezometer RB-12 and pour it into a 1.000 Lt. aerated tank (sample B).
- take 800 Lt. of water from piezometer RB-12 and pour it into a 1.000 Lt. aerated tank (sample C).

The 3 portions of groundwater formed were treated according to the following scheme:

1. Sample A: nothing has been added to this portion, it is the “white” control sample.
2. Sample B: A volume of 1,8 Lt. of MPCD® super-concentrated, equivalent to 200 Lt. of MPCD Petrol 97, was added to this portion. The solution was shaken to mix well.
3. Sample C: A volume of 1,8 Lt. of super concentrated MPCD® was added to this portion, equivalent to 200 Lt. of MPCD Petrol 97. **After 10 days, 2 Lt. of probiotics** supplied by us (probiotics specially selected for the degradation of chlorinated solvents) were added and the solution was shaken to mix well.

All tanks containing the portions to be treated were placed under cover from the weather, and the tanks were provided with ventilation of the contents by means of openings in the upper part of each tank.

The solution of each portion was also mixed once a week.

Protocol application activities were carried out by HIP staff according to the procedure in the following table 1.

Only for the day of 6 September 2020 (T=0), AIKU Environment staff was also present on site, ones who started the activities by providing MPCD® and Probiotics.



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**TABLE1 - SUMMARY OF THE
PROCEDURE**

| Portion Name | Composition | Treatment | Monitoring |
|--------------|---|--|-------------|
| SAMPLE A | 1,000 litres of groundwater contaminated with chlorinated solvents | Guaranteed aeration, mixed once x week | See table 2 |
| SAMPLE B | 800 litres of groundwater contaminated with chlorinated solvents + litres 200 of MPCD Petrol 97 | Guaranteed aeration, mixed once x week | See table 2 |
| SAMPLE C | 800 litres of groundwater contaminated with chlorinated solvents + 200 litres of MPCD Petrol 97 + 2 litres of Probiotics(added after 10 days from start of activity). | Guaranteed aeration, stirred once a week | See table 2 |

The monitorings carried out over time, according to the schedule in TABLE 2 below, were performed by HIP staff.

TABLE 2 - MONITORING PLAN

| Timing | SAMPLE A | SAMPLE B | SAMPLE C |
|--|---|---|---|
| Start of activity, after portion formation. T=0 | Took a sample. Performed analysis of chlorinated solvents, BTEX, Total Dissolved Hydrocarbons, counting of | Took a sample. Performed analysis of chlorinated solvents, BTEX, Total Dissolved Hydrocarbons, counting of | Took a sample. Performed analysis of chlorinated solvents, BTEX, Total Dissolved Hydrocarbons, Counting of |

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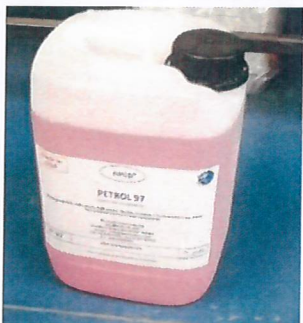



| | | | |
|--|---|---|---|
| | total bacteria. | total bacteria. | total bacteria. |
| After 15 days from the start of activities | Taken a sample. Executed | Taken a sample. Executed | Taken a sample. Executed |
| Timing | SAMPLE A | SAMPLE B | SAMPLE C |
| T=15 | analysis of chlorinated solvents, BTEX, total dissolved hydrocarbons, counting total bacteria. | analysis of chlorinated solvents, BTEX, total dissolved hydrocarbons, counting total bacteria. | analysis of chlorinated solvents, BTEX, total dissolved hydrocarbons, counting total bacteria. |
| 30 Days after start of activity T=30 | Took a sample. Performed analysis of chlorinated solvents, BTEX, Total Dissolved Hydrocarbons, counting total bacteria. | Took a sample. Performed analysis of chlorinated solvents, BTEX, Total Dissolved Hydrocarbons, counting total bacteria. | Took a sample. Performed analysis of chlorinated solvents, BTEX, Total Dissolved Hydrocarbons, counting total bacteria. |
| 45 Days after start of activity T=45 | Took a sample. Performed analysis of chlorinated solvents, BTEX, Total Dissolved Hydrocarbons, counting total bacteria. | Took a sample. Performed analysis of chlorinated solvents, BTEX, Total Dissolved Hydrocarbons, counting total bacteria. | Took a sample. Performed analysis of chlorinated solvents, BTEX, Total Dissolved Hydrocarbons, counting total bacteria. |
| Total samples analysed | 4 | 4 | 4 |

The analytical determinations were carried out in Petrohemija's own laboratory.



Materials & Equipment

| Description | |
|---|--|
| n. 3 1000 Lt tanks - (supplied by Petrohemija). To allow adequate ventilation, the upper part of the tank was removed. |  |
| Description | |
| Sampling material (PVC-PET bottles, glass, sterile, etc.) - provided by Petrohemija. |  |
| MPCD Petrol 97- 400 Lt. (supplied by AIKU Ambiente srl) |  |
| Selected probiotics to degrade chlorinated solvents (supplied by AIKU Ambiente srl) |  |



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Photo 1 shows the three 1,000 Lt. tanks forming the three samples A, B and C.



Photo:1

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ANALYTICAL OUTCOMES

The table in Annex 1 shows the analytical results for Samples A, B and C, according to the monitoring program as per Table 2 above. Analyses performed by Petrohemija's trusted laboratory.

RESULTS

Sample A (control sample)

The control sample had no addition of MPCD® or probiotics. The analysis of the control sample are needed to understand the decontamination trend according to a "natural" trend. Therefore, from the monitoring data over time, it is expected to have a constant analytical picture (perhaps with a slight tendency to degradation) in the expression of the measured values.

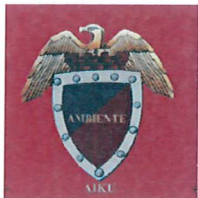
A somewhat confusing analytical picture emerges from viewing the data.

First of all, it should be noticed that the concentration value of the EDC compound, measured in the HIP laboratory and compared with that measured by the external laboratory, for the sample T=0, differs by two orders of magnitude. That is, a concentration of 366 mg/l against a concentration of 6,8 mg/l.

The values measured at time T=0 for the parameter "total chlorinated solvents", on samples A, B and C (when neither MPCD® nor probiotics had been added yet), do not overlap. There are similar values for samples B and C but a lower order of magnitude for sample A.

While the values are superimposable for the "1,2 EDC" parameter.

The trend in EDC concentration over time (over the 45 days of monitoring) is rather strange, as it shows a sudden decrease in the compound from 6,8 to 0,0767 mg/l well below the legal limit, a concentration value that drops further over the 45 days to <0,01 mg/l.



This decreasing trend suggests that the system, even without the addition of any formulated (such as MPCD® or others) or probiotic, is able to reclaim itself in 30 days!

The same oddity in the expression of the data applies to the parameter "Hydrocarbons C10 - C40." From a concentration of a few ppm at time T=0 (24 ppm of dissolved hydrocarbons), it rose to around ppm 8.000 after 15 days (which could be explained if MPCD® was present - see next paragraph) and in-fact the hydrocarbons disappeared at times T=30 and T=45, reaching the clean-up objectives. Again, it seems quite natural!!

On the other hand, the bacterial count is stable over time, showing a bacterial population on the order of 10^3 - 10^4 .

The uncertainty of the analytical framework of Sample A, with this wide variability, does not allow us to take in consideration the control Sample A, as a comparison for other existing tests and in the evaluation considerations regarding the purpose of the test.

However, the possibility that the system will naturally reclaim itself in just 30 days cannot be ruled out and is totally excluded!

The reason for this variability remains to be investigated. It could be in the way of sampling, interference in instrumental analyses, or something else. It is however an aspect that will have to be corrected in the next tests.

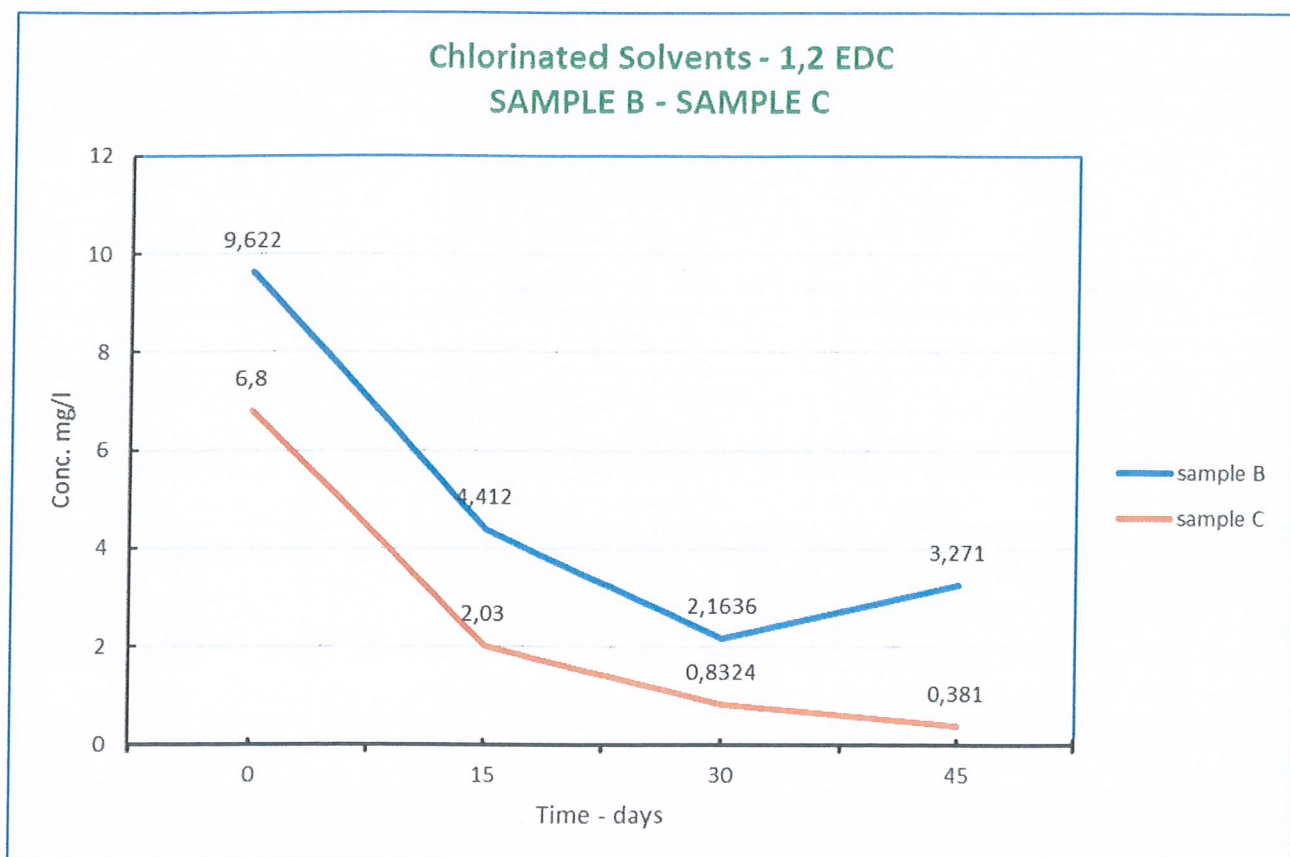
Sample B (MPCD®) and Sample C (MPCD® and Probiotics)

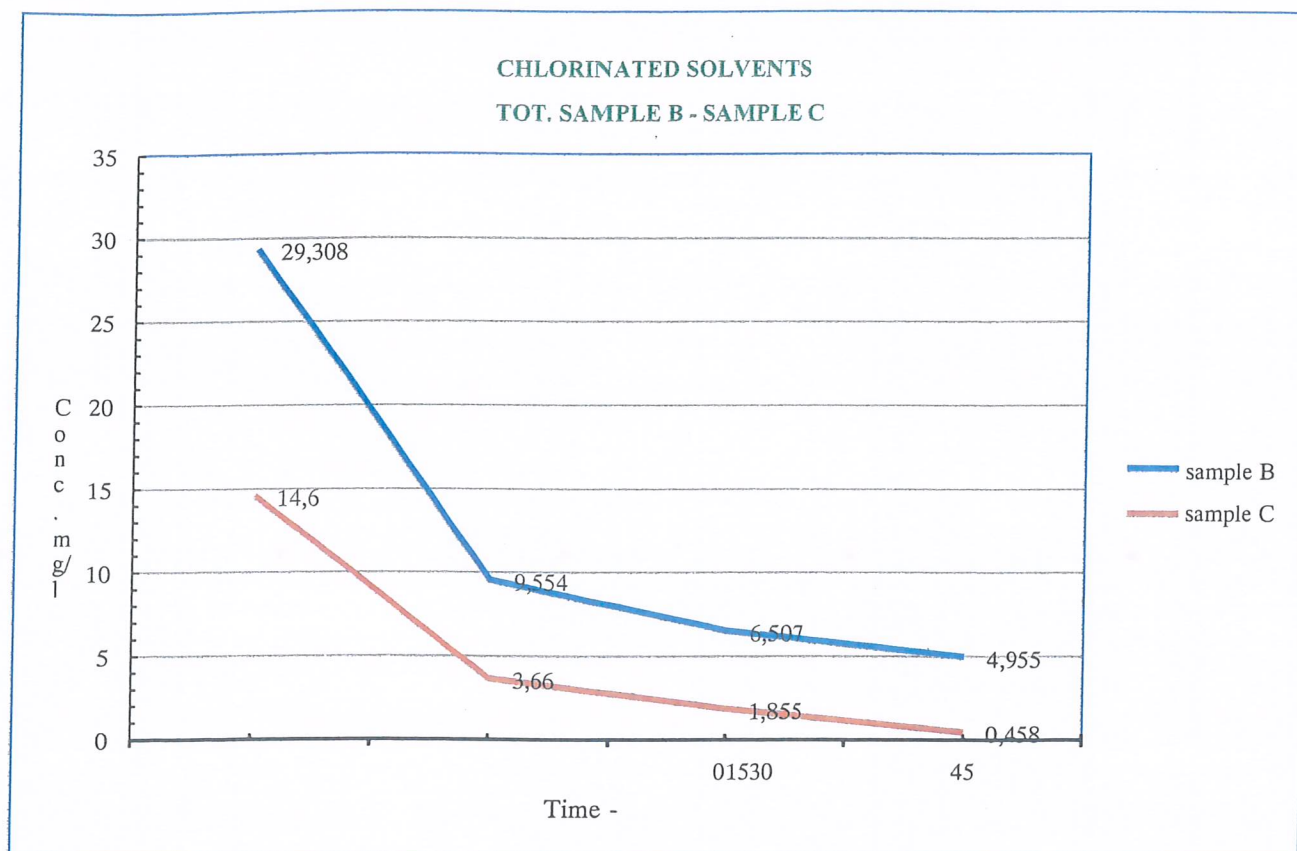
These two samples, show a degradation trend, comparable to that which was found in our laboratories and illustrated to you in our report of 21 January 2020, sent on the date 31.01.2020, that is attached to this document again (Annex 2).

Chlorinated compounds (both as total compounds and as EDCs) have a constant decreasing trend, with a higher degradation rate in Sample C in the presence of probiotics (see graphs below), with values reaching remediation limits around day 45.



The graphs below visually illustrate the decontamination progress made by the MPCD® protocol.





The temporary rise shown by C10 - C40 hydrocarbons is also identical to that found in our laboratories. Increase due to dechlorination of chlorinated compounds. Subsequently (time T=30 and T=45), the concentration of hydrocarbons is decreasing due to the breakdown activity of MPCD® and degradation by probiotics.

The subsequent felling of the hydrocarbons has a slower rate on Sample C, due to the fact that from day 30 onwards the bacterial load in Tank C was abruptly reduced from 10^2 to <10 CFU, slowing down the final hydrocarbon degradation step.

On the other hand, in Sample B, the bacterial population remained constant over time (with some variation in the positive direction), ensuring a greater rate of degradation of the dissolved hydrocarbons.



This analytical framework, with the final data at T=45, available to date, confirms that the application of the MPCD® + probiotics protocol shows its effectiveness in the degradation of chlorinated compounds and in this case the EDC 1,2 compound.

The disruptive action of MPCD® combined with the degrading action of probiotics is able to break down both chlorine and hydrocarbon contaminants.

FINAL CONSIDERATIONS

The application of the MPCD® Protocol, consisting in the addition of the MPCD® formulation and subsequently of AIKU Ambiente probiotics, allows the establishment of a disintegration/degradation system that **is able** to proceed to the dechlorination of chlorinated compounds in an aerobic environment, highlighting a totally innovative mechanism. In fact, the degradation of chlorinated solvents usually takes place in an anaerobic/reducing environment, an environment that is "created" by injecting substances into the water table that break down dissolved oxygen, making the water table anoxic. Technologies that, after having achieved the objectives of remediation in the long term (remember that anaerobic phenomena are much slower than aerobic ones, in the order of years!), in fact leave a water table extremely poor in oxygen and altered in its essential chemical and physical parameters, and therefore unable to 'purify' itself from other possible contaminations (even if of modest entity) according to a natural process (natural attenuation).

As demonstrated by the test data, the use of the MPCD® Protocol ensures better quality of the water table post-remediation.

For any subsequent test phases (on-site test field on small areas), the deficiencies highlighted in this test must be taken in consideration and the analytical panel must be completed by inserting other chemical-physical parameters that are useful and necessary for obtaining the operational information required for full-scale remediation.

ANNEXES

1. Summary table of analytical results
2. AIKU Ambiente Report of 21.01.202