

Geochemical evaluation of Tang and Pirgal mud volcanoes in order to estimate Sulfate-reducing bacteria growth

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Abstract

Mud volcanoes are important complications morphological that made of clay. In this study, geochemistry factors of Tang & Pirgal mud volcanoes on growth of Sulfate-Reducing Bacteria (SRB) is investigated based on chemical analysis required factors for growth this group of bacteria (temperature, EC, pH, Na, Ca, Mg, Fe, K and total sulfur) and growth in their specific medium. Temperature, EC, and pH evaluated by their specific devices, sodium and potassium elements evaluated by flame photometer method and calcium, magnesium and iron evaluated with atomic absorption method. The growth of SRB assessed in their specific medium (Postgate's B Medium). The results were as follows that 11 samples showed growth in their medium and results of chemical analysis included that T:24/46°C, pH:8/76, EC:18/29ms/cm, Na:38ppm, K:37ppm, Mg:1/499ppm, Ca:0/264ppm, Fe:0/181ppm and TS:886/91ppm for samples of Tang and T:28/56°C, pH:9/03, EC:15/86ms/cm, Na:32ppm, K:52ppm, Mg:0/707ppm, Ca:0/141ppm, Fe:0/021ppm and TS:724/90ppm for samples of Pirgal. All archived factors are effective for growth of SRB.

INTRODUCTION

Soil pollution is one of the major problems of the environment. There are lots of ways to solve this problem, such as Bioremediation which is the use of either naturally occurring or deliberately introduced microorganisms to consume and break down environmental pollutants in order to clean polluted site. Microorganisms, especially bacteria, have a key role in Bioremediation process. One of the oldest group of this kind of bacteria is Sulfate-Reducing Bacteria (SRB) which can get their energy from oxidation organic components or molecular hydrogen via reduction sulfate to hydrogen sulfide. They are obligatory anaerobic.

"SRB can cause a serious problem for industries, such as the offshore oil industry, because of the production of sulfide, which is highly reactive, corrosive and toxic. However, these organisms can also be beneficial by removing sulfate and heavy metals from waste streams. Although SRB has been studied for more than a century, it is only with the recent emergence of new molecular biological and genomic techniques that we have begun to obtain detailed information on their way of life" [1].

Sulfite reductase and sulfite oxidase are essential enzymes for the growth of SRB bacteria, these enzymes need iron for their activities. These bacteria can remove some hydrocarbons from soil, for instance, benzene, toluene, ethylbenzene and xylene in order to bioremediation of soil polluted. This kind of bacteria can isolate from anaerobic habitats. Mud volcanoes are one of the places to find these bacteria [2,3].

A mud volcano is a mass of materials, clay or stones that come out with together gas and accumulates on the surface of the earth and conical shapes are created. Exterior mud is so sticky and can smell odors of methane and sulfur gasses [4]. In Iran, mud volcanoes are frequently being in beaches of northern and southern. Tang and Pirgal mud volcanoes are the most important place in Chabahar port and southern side of Iran. Tang is the greatest hydrocarbonic mud volcano of Iran. Based on compounds of mud volcanoes, many species of microorganisms can growth and presence in there.

Based on the important role of SRB in bioremediation of polluted soil with hydrocarbons components, therefore, in the present study strived to the evaluated connection between the amount of sulfur and other geochemical factors of mud volcanoes.

MATERIAL & METHODS

In summer of 2011, Sampling was done from craters of Tang and Pirgal mud volcanoes (10 samples from each) in aseptic conditions by used plastic strilled pipes(in sizes of 5, 10, 15 &30 cm). In time of sampling, thermometer stilled in craters of mud volcanoes for 30 minutes. Following analysis have been conducted as suggested by Gibson.M [5]:

1. pH & EC :10 gr of sample shaken in 50cc of distilled water and stilled for 24h then pH and EC were evaluated from surface liquid with 3310JENWAY, UK, and EC meter, respectively
2. Measured Na & K:For evaluation, these elements applied Flame photometric method
3. Measured Fe, Mg & Ca:These elements estimated with atomic absorption method
4. Measured Total Sulfur(TS):In order to this purpose, total sulfur method had used

5. Screening SRB: 1 gr of sample add to 9cc of Ringer's solution, then, shook well. 1 cc of each sample solutions adds to 9 cc strilled Postgate's B medium. All inoculated media were incubated at 30° C for at least 5 months. Appearance black color in Postgate's B confirmed presence of SRB and reduction of sulfate to hydrogen sulfide.

Hydrogen sulfide reacts with iron of medium and produces sulfide ferro and then black color [6].

RESULTS

Temperatures of mud volcanoes have been measured and the result is shown in table 1. Result of pH & EC is shown in Table 2. Result of Na & K is shown in Table 3.

Table 1: Results of temperature

Place	Temperature (°C)
Tang	24/46°C
Pirgal	28/56°C

Table 2: Results of pH and EC

Place	pH	EC(ms/cm)
Tang	8.76	18/29
Pirgal	9.03	15/86

Table 3: Results of Na and K

Place	Na(ppm)	K(ppm)
Tang	38(5 times dilution)	37
Pirgal	32(25 times dilution)	52

Result of Fe, Mg & Ca is shown in Table 4. Total sulfur (TS) is presented in table 5. Finally, for Screening SRB, among 20 samples, 11 of them showed positive result as

shown in Figure 6. "T" in the table presents Tang Mud-Volcano and "P" presents Pirgal mud volcano.

Table 4: Results of Fe, Mg and Ca

Place	Fe(ppm)	Mg(ppm)	Ca(ppm)
Tang	0.181(10 times dilution)	1.499	0.264 (25 times dilution)
Pirgal	0.021(25 times dilution)	0.707	0.141 (25 times dilution)

Table 5: Results of TS

Place	TS (ppm)
Tang	886.91
Pirgal	724.70

Table 6: Period of time for positive results of Postgat's B media

Samples	Time for became positive
T2	2 months and 11 days
T3	5 months
T4	2 months and 23 days
T5	2 months and 2 days
T6	5 months and 17 days
P1	23 days
P2	2 months and 2 days
P3	23 days
P4	1 month and 23 days
P5	1 month and 20 days
P6	23 days

CONCLUSION

Today, Bioremediation is one of important action to save our world from pollutions, therefore, evaluated biodiversity of different ecosystems in order to find powerful microorganisms for this goal is so noticeable. Sulfate Reducing Bacteria are one of these kinds of bacteria.

The presence of these bacteria was confirmed in some mud volcanoes, such as, Pecele Mici in Carpathian mountains of Romania, Salsedi Nirano in Italy and etc. [6,7].

There are lots of studies in the field of chemical analysis of effective factors on the growth of SRB. It is showed that Mg^{2+} has the critical role in beginning of life cycle of SRB [2]. If the concentration of this ion, as same as Ca^{2+} , increase at first step of the life cycle, reduction of sulfate will increase. He mentioned that NaCl is essential

for the survival of these bacteria and can increase it. Then he proved that, Iron essential for activities of sulfite reductase and sulfite oxidase that have important roles in sulfur cycle. Even he showed that Total Sulfur is the main substrate for growth of SRB [2]. In the present study amounts of these Ca^{2+} , Mg^{2+} and Fe were so high, therefore can prepare suitable conditions for growth of SRB. High amounts of Na^+ , K^+ , and EC in both mud volcanoes affect on the survival of these bacteria and increase that. The high amount of TS for both mud volcanoes and smell of sulfide hydrogen and presence of yellow sulfur compounds are another reasons to prove that both mud volcanoes are good habitats for SRB.

REFERENCES

- [1] Garcia.C.et.al, 2001, "Bioremediation of an industrial acid mine water by metal-tolerant sulfate-reducing bacteria", Minerals engineering, Vol:14, No:9, pp:997-1008.
- [2] Gerard Muyzer¹ & Alfons J. M. Stams, 2008, The ecology and biotechnology of sulphate-reducing bacteria, Nature Reviews Microbiology, Vol 6, pp:441-454 .
- [3] Gibson.M, 1987, "Sulfate mineral from a mud volcano in the Katakolo area, western Peloponnesus, Greece", American Mineralogist. Vol:72, pp:839-841.
- [4] Heller .C.et.al, 2010, "Microbial ecology of terrestrial methane-emitting mud volcanoes in Italy", Goldschmidt Conference Abstracts, Vol:38, pp:78-124 .
- [5] Jun-ya.C.et.al, 2008, "Influence of Mg^{2+} on the growth and activity of sulfate reducing bacteria", Hydrometallurgy journal, Vol:95, pp:127-134.
- [6] Lazar.C.S.et.al, 2011, "Archaeal populations in hypersaline sediments underlying orange microbial mats in the Napoli mud volcano", Environmental Microbiology, Vol: 77, N:9, pp:241-256.
- [7] Treude .T.et.al, 2006, "Microbiological investigation of methane- and hydrocarbon-discharging mud volcanoes in the Carpathian Mountains Romania", Environmental Microbiology, Vol: 8, N:4, pp: 574-590.

