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Isolation of Aroclor1254 degrading bacteria in contaminated soil by transformer oil

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ABSTRACT- Aerobic biodegradation is an environmental-friendly biological method that allows micro organisms to remove persistent organic pollutants. Aroclor1254 is a mixed compound containing polychlorinated biphenyls (PCBs) along with persistent organic materials. There is no estimate of Aroclor1254 usage and its release into the environment in Iran. A transformer manufacturing plant in Shiraz (located in southwest of Iran) uses transformer oils containing Aroclor1254. Soil samples contaminated by transformer oil were taken from the manufacturing plant's site and were tested for the presence of certain bacteria, level of degradation and pH. In all samples, 13 bacterial strains were isolated on C-media, and their growth was measured using a spectrophotometer. Of the isolated strains, eight could degrade Aroclor1254. Of these eight strains, three showed weak growth (OD between 0.1 to 0.195), three showed low growth (OD between 0.2 to 0.3) and two showed good growth (OD between 0.3 to 0.5). Some of the strains grew well during the first week, but their growth decreased and eventually stopped in the second week. For all the growth media on which the bacteria from the soil was separated and cultured, pH decreased as the bacterial growth increased. For some media, in which bacterial growth had stopped, pH increased after 14 or 21 days but it was always below 7.0. This indicated that environmental pH or increasing carbon dioxide levels were not suitable for the growth of these strains. However, this trend was not observed in bacteria belonging to *Pseudomonas*, *Curtobacterium* and *Burkholderia*. These bacteria could grow and degrade Aroclor1254 even at pH of 4.5. In fact, their degradation efficiency increased at a constant pH of about 5.5.

INTRODUCTION

Synthetic organic materials that are widely used in industries cause serious environmental pollution even when they are released into the biosphere in small quantities. Polychlorinated biphenyls (PCBs) are synthetic organic materials that are not only a pollutant but also very toxic and persistent organic compounds (POCS). PCBs contain a biphenyl nucleus carrying 1–10 chlorines. Approximately 209 types of PCBs are available that differ with respect to the number and position of chlorines. Because PCBs cause environmental pollution, their production and use have been prohibited worldwide since 1976. However, because of their non-flammability, chemical stability, high boiling point and electrical insulating properties, they are still used in electrical equipment (e.g. heat transfer and hydraulic equipment) as plasticizers in paint sand plastics and rubber products, and in pigments, dyes and carbonless copy paper. PCBs serve as coolants and insulating fluids (especially in capacitors), components of early fluorescent light, fitting, sand locomotive electrical transformers, as stabilising additives in flexible PVC, as a coating of electrical wiring and electronic components, as pesticide extenders and in cutting oils, reactive flames and hydraulic fluids (Rudel et al., 2008).

PCBs have very low solubility in water but high in organic solvents. The boiling point of PCBs (such as monochlorobiphenyl to decachlorobiphenyl) ranges from 25°C to 306°C, their vapour pressure at 25° Cranges from 1.1 to1.4×10^{-6} Pa and their water solubility at 25°C ranges from 4.0 to 7.6×10^{-4} g/m^3. PCBs used for industrial purposes have complex compounds called Aroclors that contain approximately 130 PCB congeners. PCBs have carcinogenic, mutagenic and teratogenic properties (NwinyiObinna, 2011).

The amount of PCBs released in Iran is unknown because of the unknown amounts used in various industries. Because of their hydrophobic nature and specific gravity of greater than 1, PCBs tend to accumulate in sediment and soil and thus probably in the food chain. PCBs are soluble in organic solvents such as lipids. Their solubility in organic solvents increases their transportation and persistence in the environment. The solubility of PCBs in water decreases with an increase in the degree of chlorination. The

solubility of a compound plays an essential role in its degradation. Compounds that are soluble in water can be easily degraded by microorganisms compared with those that have low solubility in water (Imamoglu et al., 2002). Two methods — physicochemical and biological — have been used to treat PCBs. The use of physical and chemical methods for degrading PCBs is associated with some problems. First, these methods are expensive; second, they are associated with larger problems during solvent extraction and subsequent thermal destruction. PCBs derivatives like polychlorinated dibenzo-p-dioxins (PCDDs) and dibenzofurans(PCDFs), which are derived by the incineration of PCBs, have much higher toxicity than PCBs. PCBs can be treated using microorganisms without the production of other pollutants; moreover, this treatment is associated with minimal cost and can be performed in situ. However, the application of this method is difficult. It is difficult to treat high concentrations of PCBs because of their own toxicities and because of the availability of other carbon sources in the environment that the microorganisms commonly use. A limited number of microorganisms can treat a few congeners. For this reason, fundamental studies, including searching for useful microorganisms and developing genetically engineered ones have been performed to overcome this difficulty. This has resulted in remarkable advances in the field of microbiology and genetic engineering over the last 20 years.

Monsanto's PCBs were manufactured under the trade name Aroclor. Aroclor mixtures are labelled with a four-digit number, of which the first two digits indicate its molecular structure while the last two digits indicate its chlorine content by percentage weight. The prefix 12 is used to classify PCBs. Other major PCB brands include Clophen by Bayer, West Germany; Phenoclor by Caffaro, Italy; Kanechlor by Kanegafuchi, Japan; Pyralene by Prodelec, France and Sovol by a company in the former USSR.

The first step of in situ microbial degradation is the availability of useful microorganisms that can live in an aqueous medium or soil.

The objective of this study was to degrade Aroclor1254 by using suitable microorganisms under standard laboratory conditions and pH tolerance. Aroclor1254 was selected for this research because it is widely used in industries and has a wide range of PCBs congeners (31 of 209 congeners).

MATERIALS AND METHODS

Sample Collection

Five soil samples (5 kg each) were collected from an area that was used to dump transformer oil at a transformer manufacturing plant site in southeast of Shiraz, I.R. of Iran. Soil samples were collected from 15-cm depth of dumped transformer oil area, from 5-cm depth of soil near the root of trees, from 5-cm depth in furrow of tree's row that was the pollutant water ways and finally from the 15-cm depth in furrow of tree's row that was the pollutant water ways. These samples were stored at lower than 4°C before transferring them to nutrient agar media. The suspensions of soil samples were transferred to nutrient agar media and incubated for 2 days at 30ºC.

C-medium was used to screen and isolate bacterial strains (KyungSu, Na et al., 1998). This media is clear and does not contain any carbon source for microorganisms. Therefore, if microbes in the media degrade Aroclor1254, the turbidity of the media will increase, indicating the growth of these microorganisms. The C-medium included (per a litre of the medium) 5g $(NH_4)_2SO_4$, 2.93g KH_2PO_4, 5.87g K_2HPO_4, 0.3g $MgSO_4 \cdot 7H_2O$, 2g NaCl, 0.03g $CaCl_2$, 0.01g $FeSO_4 \cdot 7H_2O$, 0.6mg $NiSO_4 \cdot 7H_2O$, 0.2g yeast extract and 2ml trace element solution (pH= 7.0). The trace element solution included 4mg MoO_3, 28mg $ZnSO_4 \cdot 5H_2O$, 2mg $CuSO_4 \cdot 5H_2O$, 4mg H_3BO_3, 4mg $MnSO_4 \cdot 5H_2O$ and 4mg $CoCl_2 \cdot 6H_2O$. Aroclor1254 used in this study was obtained from Sigma-Aldrich (Cat. No. 48586). Aroclor at a concentration of 0.01% was added to the C-medium as a carbon source (KyungSu, et al., 1998).

Identification of Isolates

Morphological and biochemical tests such as gram staining, oxidase and catalase tests and O/F were performed, as described by Schaad et al., (2001) (Table 1).

Determination of Growth

Three steps were used for determining Aroclor1254 biodegradation: (1) identification of strains that used Aroclor1254 as the carbon source; (2) Indication of growth with measured turbidity and (3) alteration of C-medium's pH during the test.

The strains were cultivated on C-medium with 0.1% of Aroclor1254 and inoculating 1% of the strain then incubated at 35°C for 7 days. Bacterial growth in the C-medium was checked using spectrophotometer by measuring optical density (OD) at 600nm (Kyung-Su Na, et al., 1998).Thirteen cavities of spectrophotometer were selected without any bacteria to control and 13 with two replications were cultivated as described.

RESULTS AND DISCUSSION

The morphological and physiochemical characteristics of the 8 strains that could degrade Aroclor1254 are summarised in Table 1.

Based on the physiological and biochemical characteristics, the strains were identified as *Curtobacterium* (C_1), *Burkholderia* (B_{41a} and C_3), *Clavibacter* (B_{32a}, C_4 and B_{21}), *Bacillus* (B_{43}) and *Pseudomonas* (B_{32}).

Table 1. Characteristic of the isolated bacteria

Colony	c1	b32a	Colony	c4	b21	b41a	c3	b32
Gram reaction	+	+	+	+	+	-	-	-
Growth on YDC	-	-	-	-	-	-	+	-
Growth at 40	-	-	V	V	-	-	-	-
Fluorescent on KB	-	-	-	-	-	-	-	+
Fermentation	-	-	+	-	-	-	-	-
Oxidation	+	+	+	+	+	+	+	+
Oxidas	-	+	-	-	-	+	+	+
Catalase	+	+	+	+	+	+	-	+
Arginine dihydrolase	-	+	+	-	-	+	+	+
Ureas	-	+	nd	+	+	+	+	-
Utilization of Rhamnose	+	-	-	-	-	+	-	+
Glucose	+	-	-	-	-	+	+	-
Inositol	+	-	-	-	-	+	-	-
Sorbitol	+	-	-	-	-	+	-	-
Melibiose	+	-	-	-	-	+	+	-
Saccharose	+	-	-	-	-	-	-	-
Mannitol	+	-	-	-	-	+	+	-
Production of H2s	-	+	-	-	+	-	-	-

nd: Not define, V :between 21%-79% of strain positive ⁺ :Positive reaction ,⁻ :Negative reaction

Determination of Bacterial Growth by Using Aroclor1254 as the Sole Carbon Source

Some strains grew in the presence of Aroclor1254 and used it as the sole carbon source. Three strains C_1, C3 and C_4 were isolated from soil polluted with transformer oil, and 10 strains B_{22}, B_{43a}, B_{31}, B_{32a}, B_{23}, B_{43}, B_{41a}, B_{21}, B_{32} and B_{41b} were isolated from soil near the roots of trees that came in contact with the runoff from the area polluted with transformer oil.

Results of utilisation of Aroclor1254 as the carbon source by bacterial strains are shown in Table 2 . Table 2 (case1 indicates first replication and case 2 indicates second replication) shows turbidity and growth. OD<0.1 indicated no growth (-), from 0.1 to 0.195 indicated low growth (†), from 0.2 to 0.3 growth (††) and from 0.3 to 0.5 good growth (†††). StrainsB_{22}, B_{43a}, B_{31}, B_{23} and B_{41b} could not use Aroclor1254 as the sole carbon source (OD<0.1). Figs. 1,3, 5, 6 and 7 show that after 21 days, these isolates could not decrease pH below 6.9. Table 2 shows that none of these strains could use Aroclor 1254 as the carbon source. Strains B_{41a} and B_{32}showed good growth and good degradability (OD, 0.3–0.5), indicating that these strains may be applicable for the in situ bioremediation of contaminated soil. Table 3 and Figs. 8 and 13 showed that both these strains could decrease the pH to <3.4. It must be known that these strains were grown in a medium supplemented with a complicated compound containing 31 PCB congeners. Strains B_{32a}, B_{21} and C_1 showed growth, and hence may be applicable for the bioremediation of contaminated soil (OD, 0.2–0.3). The pH for these strains was between 4 and 5 (Figs. 2, 9 and 10) and decreased with an increase in the growth of these strains. Strains B_{43}, C_3 and C_4 showed low growth and hence weak degradability; therefore, they cannot degrade Aroclor1254 in nature (OD, 0.1–0.2).

Thus, the isolated strains can degrade some PCB congeners. If a mixed culture of the isolated strains is developed, biological treatment efficiency against recalcitrant organic compounds in industrial wastewater can highly be improved. There are the combinations of 75 of 129 PCB's congeners in Aroclor 1254. In this case, the conditions of degradation for bacteria are very difficult. This congeners are from low to high chlorinated that may be in our environment.

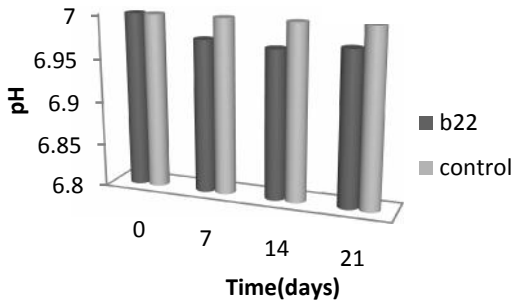

Fig. 1. pH alteration during 21 days by B22 isolated from soil near trees

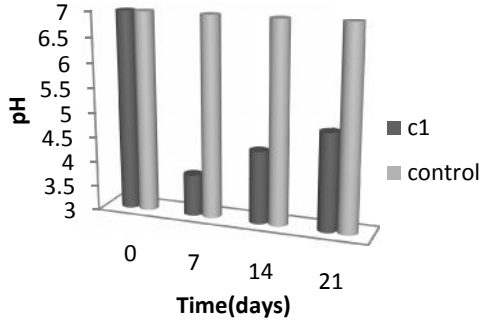

Fig. 2. pH alteration during 21 days by C1 isolated from sand pollute with transformer oil

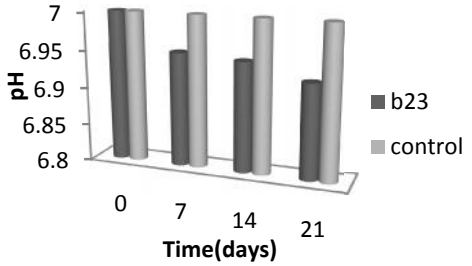

Fig. 3. pH alteration during 21 days by B23 isolated from soil near trees

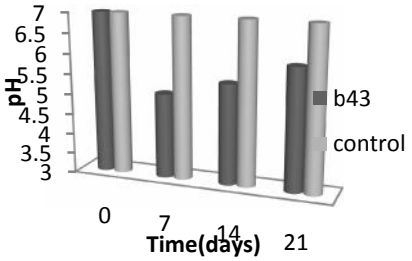

Fig. 4. pH alteration during 21 days by B43 isolated from soil near trees

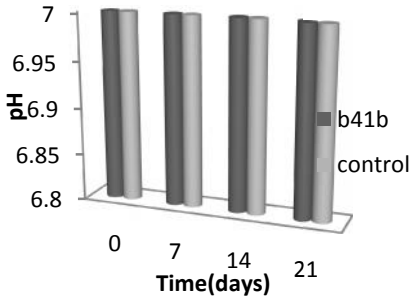

Fig. 5. pH alteration for three weeks by B41b isolated from soil near trees

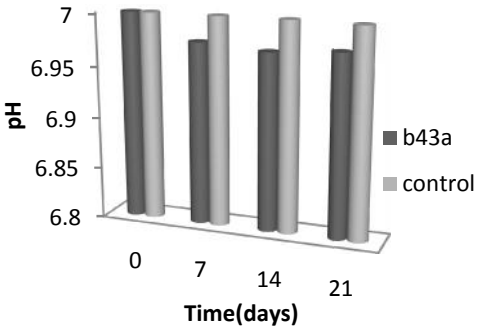

Fig.6. pH alteration during 21 days by B43a isolated from soil near trees

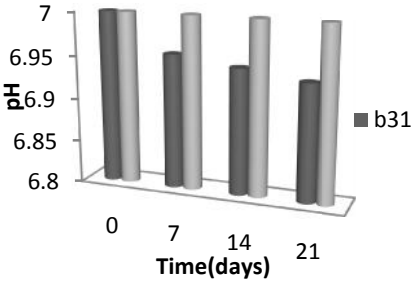

Fig. 7. pH alteration during 21 days by B31 isolated from soil near trees

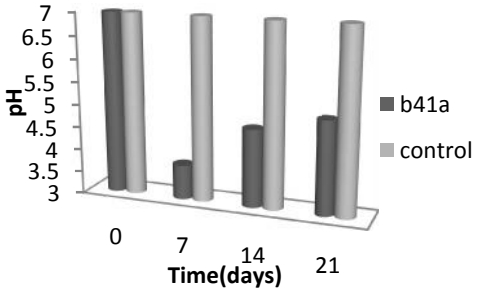

Fig. 8. pH alteration during 21 days by B41a isolated from soil near trees

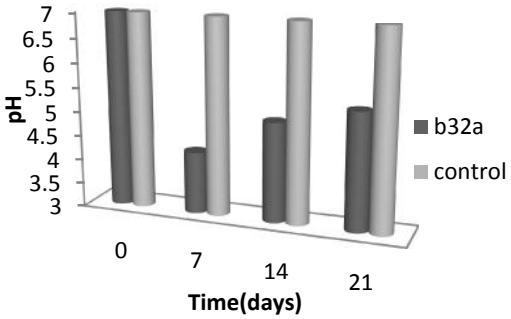

Fig. 9. pH alteration for three weeks by B32a isolated from soil near trees

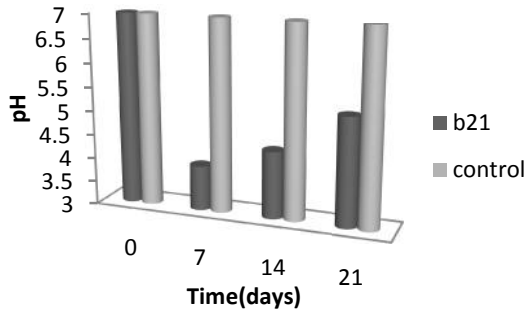

Fig.10. pH alteration during 21 days by B21 isolated from soil near trees

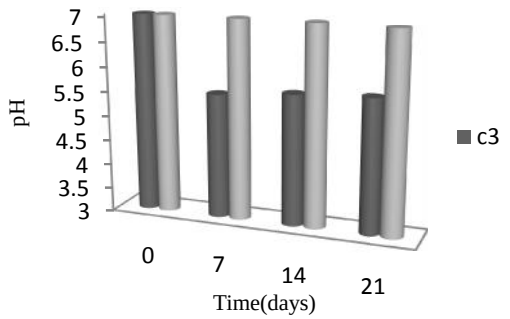

Fig. 11. pH alteration during 21 days by C3 isolated from sand pollute with transformer oil

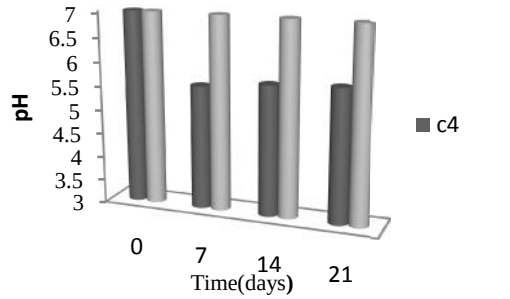

Fig. 12. pH alteration during 21 days by C4 isolated from sand pollute with transformer oil

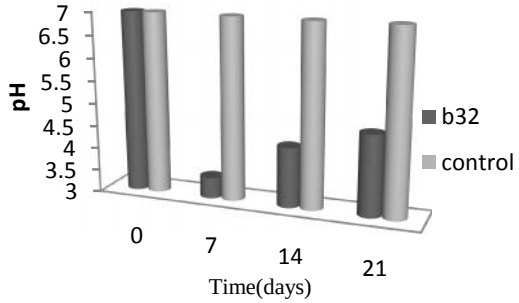

Fig. 13. pH alteration during 21 days by B32 isolated from soil near trees

Alteration of Media's pH by Bacterial Growth

An alternative method can be used to determine the strains degrade Aroclor 1254. In the beginning of cultivation, all cultures had a pH of 7.0. However, after one, two or three weeks of growth, the pH decreased in some cultures while remained constant in others. According to data presented in Table 3, pH of <7.0 is an indicator of Aroclor 1254 degradation.

The results showed that if OD was higher, then the decrease in pH to <7.0 was more ($B_{32}$with OD= 0.351 has pH=3.46).

In some strains like B_{32}, pH increased and growth stopped after two or three weeks, indicating that pH must be considered during biological treatment even in nature.

Curtobacterium and *Burkholderia* can grow and live in lower pH up to 4.5. Therefore, they can efficiently be used for biological treatment even in difficult conditions. They can degrade some of Aroclor's congeners and can survive. For other bacterial strains like B_{43}, the pH should be approximately 6.0.

The results of this research showed that pH lower than 7.0 is a good indicator of Aroclor 1254 degradation. The pH decrease occurred in aerobic bacteria, but it may or may not occur in anaerobic bacteria. Therefore, we can recognize the Aroclor 1254 degradation according to pH instead of other difficult methods. This method may be suggested for degradation of other persistent organic pollutants.

CONCLUSIONS

Seedlings were successfully infected by arbuscular mycorrhizal fungus but root colonization decreased with increased levels of water stress. Water stress significantly decreased shoot dry weight in two citrus rootstocks. Mycorrhizal rough lemon rootstocks had significantly higher shoot dry weight than the control at all levels of water stress. By increasing water stress, shoot N, P, Mn, Cu, and Fe uptake of two citrus rootstocks decreased, but shoot Zn uptake increased. Shoot N and P uptake were significantly higher in mycorrhizal seedlings than NM seedlings of two citrus rootstocks, whereas there was no significant difference in shoot Mn, Cu, Zn and Fe uptake of two citrus root stocks at all levels of water stress. As water stress increased, SOD, CAT, G-POD and APX activities of two citrus rootstocks leaves increased. With inoculation of seedlings by *G. mosseae* compared with the control, antioxidant enzymes activities increased. The results suggested that the AM inoculation may play an important role in water stress tolerance.

Table 2. Result of O.D. spectrophotometer after one week growth

	b22	b43a	b31	b32a	b23	b43	b41a	b21	c1	c4	c3	b32	b41b	O.D		Results
Replication1	—	—	—	††	—	†	†††	††	††	†	†	†††	—	<0.1	No growth	—
eplication2	—	—	—	††	—	†	††	††	††	†	†	†††	—	0.1-0.195	Low growth	†
Control	—	—	—	—	—	—	—	—	—	—	—	—	—	0.2-0.3	Growth	††
Result	—	—	—	††	—	†	†††	††	††	†	†	†††	—	0.3-0.5	Good growth	†††

— : No growth, †: Low growth, ††: Growth, †††: Good growth Replication1: First iteration Replication2: Second iteration

Table 3. Media's pH alteration after the growth of bacterial strains

		b22	b43a	b31	b32a	b23	b43	b41a	b21	c1	c4	c3	b32	b41b
pH after 7 days	Replication 1	6.98	6.97	6.96	4.12	6.96	5	3.52	3.98	3.65	5.58	5.51	3.4	7
	Replication 2	6.97	6.98	6.95	4.4	6.94	5.19	4	3.89	4	5.53	5.49	3.52	7.01
	Mean	6.975	6.975	6.955	4.26	6.95	5.095	3.76	3.935	3.825	5.555	5.5	3.46	7.005
	Control	6.99	7	7	7	7	7	7	7	7	7	7	7	7
pH after 14 days	Replication 1	6.96	6.97	6.95	4.9	6.95	5.35	4.56	4.45	4.4	5.7	5.65	4.3	7
	Replication 2	6.96	6.97	6.94	5.15	6.94	5.6	4.86	4.36	4.5	5.68	5.6	4.35	7
	Mean	6.96	6.97	6.945	5.025	6.945	5.475	4.71	4.405	4.45	5.69	5.625	4.325	7
	Control	7	7	7	7	7	7	7	7	7	7	7	7	7
pH after 21 days	Replication 1	6.95	6.98	6.94	5.3	6.93	5.9	5	5.15	5	5.75	5.69	4.75	7.01
	Replication 2	6.96	6.97	6.93	5.45	6.92	6.1	5.1	5.34	4.9	5.76	5.69	4.8	6.99
	Mean	6.955	6.975	6.935	5.375	6.925	6	5.05	5.245	4.95	5.755	5.69	4.775	7
	Control	7	7	7	7	7	7	7	7	7	7	7	7	7

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REFERENCES

Adebusoye, S.A., Picardial, F.W., Ilori, M.O., Amund, O.O., & Fuqua, C. (2008). Characterization of multiple novel aerobic polychlorinated biphenyl (PCB) utilizing bacterial strains indigenous to contaminated tropical frican soils. *Biodegradation*, 19, 145-159.

Adriaens, P., Kohler, H.P.E., KohlerStaub, D., & Focht, D.D. (1989). Bacterial Dehalogenation of Chlorobenzoates and Coculture Biodegradation of 4,4'-Dichlorobiphenyl. Applied and Environmental. *Microbiology*, 55(4), 887-892.

Bopp, L.H., (1986). Degradation of highly chlorinated PCBs by *Pseudomonas* strain Lb400. *Journal of Industrial Microbiology*, 1, 23-29.

Furukawa, K. (1994). Molecular genetics and evolutionary relationship of PCB-degrading bacteria. *Biodegradation*, 5, 289-300.

Furukawa, K., Simon, J., & Chakrabatry, A.M. (1983). Common induction and regulation of biphenyl, xylene/toluene, and salicylate catabolism in Pseudomonas *Paucimobilis*. *Journal of. Bacteriology*, 154(3), 1356-1362.

Furukawa, K., Tonomura, K., & Kamibayashi, A. (1979). Effect of chlorine substitution on the bacterial metabolism of various of polychlorinated biphenyls. Applied. and Environmental. *Microbiology*, 38(2), 301-310.

Imamoglu, M.G., Rayne, S., & Addison, R.F. (2002). Exponential increase of brominated flame retardants, polybrominated diphenyl ethers. Environmental *Science and Technology*, 36,1886-92.

Kiyohara, H., Nagao, K., & Yano, K. (1982). Rapid screen for bacteria degrading water-insoluble, solid hydrocarbons on agar plates. Applied and. Environmental *Microbiology*, 42, 454-457.

KyungSu, N., YongWoon, L., JinSook, JaeSuk, L.L., Motoki, K., & SeonYong C. (1998). Isolation and Characterization of Polychlorinated Biphenyls (PCBs) Degrading Bacteria from a Municipal Sewage Treatment Plant. *Environmental Engineering Research*, 3(2), 67-78.

Leigh, M.B., Prouzova, P., Mackova, M., Macek, T., Nagle, D.P., & Fletcher, J.S. (2006). Polychlorinated Biphenyl (PCB)-Degrading Bacteria Associated with Trees in a PCB-Contaminated. *Applied and Environmental Micribiology*,, 72(4), 2331-2342.

NwinyiObinna, C. (2011). Enrichment and Identification of Askarel Oil (PCB blend) Degrading Enriched from Landfill Sites in Edo State, Nigeria.

Nwinyi, O.C., Nwodo, C.S., & Amund, O.O. (2008). Biodegradation potential of two Rhodoccocus strain capable of utilizing aniline as Carbon Source in tropical ecosystem *Research Journal of Microbiology*, 3(2), 99-104.

Prescott, M., Harley, J.P., & Klein, D.A. (2001). Industry and Biotechnology in general Microbiology. 5th ed. McGraw-Hill.

Robinson, G.K., & Lenn, M.J. (1994). The bioremediation of polychlorinated biphenyls (PCBs): Problems and perspectives. *Biotechnology and Genetic Engineering. Reviews*, 12,139-188.

Rudel, R.A, Seryak, L.M., & Brody, J.G. (2008). PCB-containing Wood Floor Finish is a Likely Source of Elevated PCBs in Residents' blood, Household air and Dust: A Case Study of Exposure. *Environmental Health*, 7(2),1-15.

Schaad, N.W, Jones, J.B., & Chun,W. (2001). Laboratory Guide for Identification of Plant Pathogenic Bacteria. 2nd ed. St. Paul, MN, USA. Amer PHytopathol Soc Press.

Sierra, I., Valera, J.L., Marina, M.L., & Laborda, F. (2003). Study of the biodegradation process of polychlorinated biphenyls in liquid medium and soil by a new isolated aerobic bacterium (Janibactersp). *Chemosphere*, 53, 609-618.

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جداسازی باکتری های تجزیه کننده Aroclor1254 ازخاک آلوده به روغن ترانسفورماتور

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واژه های کلیدی:
Aroclor1254
تجزیه بیولوژیکی
Burkholderia
Curtobacterium
محیط کشت-C
واکنش خاک(pH)
Pseudomonas

چکیده- تصفیه آلاینده ها به صورت هوازی توسط میکروارگانیسم ها فرایندی است سازگار با محیط زیست که در آن آلاینده ها توسط فرایند بیولوژیک پاکسازی می گردند. Aroclor1254 از مشتقات متنوع (PCBs) تشکیل شده است که این ترکیبات خود یک آلاینده پایدار به شمار می آید. میزان مصرف یا رهاسازی دقیقی از این ماده در محیط زیست ایران وجود ندارد. یک کارخانه تولید ترانسفورماتور در جنوب شرق شیراز از روغن ترانسفورماتور که شامل Aroclor1254 می باشد برای ساخت استفاده می نماید. نمونه هایی از خاک آلوده به این روغن در داخل کارخانه برای بررسی وجود باکتریهای مناسب، میزان تجزیه و تغییرات pH، برداشت گردید. از باکتری های موجود ۱۳ سویه جداسازی و در محیط کشت C- گذاشته شد و میزان رشد آنها در کنار Aroclor1254 توسط دستگاه اسپکتروفتومتر بررسی گردید. از این میزان باکتری کشت داده شده، ۸ سویه آنها موفق به تجزیه Aroclor گردیدند. از این ۸ سویه باکتری، ۳ سویه تجزیه ضعیف، ۳ سویه مقدار کمی تجزیه و ۲ عدد آنها تجزیه خوبی داشتند. بعضی از این باکتریها در هفته اول رشد خوبی داشتند ولی در هفته دوم رشد آنها کاهش و یا حتی متوقف گردید. در تمام باکتری های کشت شده میزان pH محیط با افزایش رشد، کاهش یافته است. در نمونه هایی که رشد بعد از ۱۴ یا ۲۱ روز متوقف شده بود میزان pH شروع به افزایش نمود ولی همیشه کمتر از ۷ بود. این نتایج نشان داد که pH محیط رشد یا میزان دی اکسید کربن تولید شده برای رشد این باکتری ها مناسب نمی باشد. البته این روند افزایش pH و کاهش رشد در باکتری های *Pseudomonas*، *Curtobacterium* و *Burkholderia* مشاهده نگردید. به عبارت دیگر این باکتری ها می توانند حتی تا pH=4.5 رشد داشته باشند. در واقع این باکتری ها عمل تجزیه خود را می توانند با راندمان بالایی در pH حدود ۵/۵ انجام دهند.