Chapter 8

The Role of Bacteria on the Breakdown of Recalcitrant Polychlorinated Biphenyls (PCBs) Compounds in Wastewater

Spar Mathews and Patricia Sithebe

Additional information is available at the end of the chapter

http://dx.doi.org/10.5772/intechopen.75400

Abstract

Pseudomonas aeruginosa was used to assess their potentials to degrade PCBs at concentrations of 1.0 μg/mL. An aliquot of 1.0 μL of the bacterial suspension with an optical density of 1.0 at 600 nm was used as an inoculum of the assay. Isolates were analysed for their ability to degrade PCB (Aroclor 1260) by measuring a shift in the wavemax using Cary 300 UV-visible spectrophotometer for a period of 96 hours. The presence /absence of the compounds was checked using high performance liquid chromatography (HPLC) UFLC Shimadzu using florescence detector pump RF-20A and system gold column C18 (CTO-20A) after 96 h. PCBs were extracted from wastewater samples from both Gaborone and Mafikeng using the Quick, Easy, Cheap, Effective, Rugged, and Safe (QuEChERS) extraction kit, and analysis was performed using the gas chromatography mass spectrometer (GC-MS). The bacteria were able to degrade these compounds under different pH values of 5.0, 7.0, 8.0, and 9.0 and temperatures of 20, 27, 30, and 35°C. Degradation occurred at the most at 35°C and the least at 20°C for PCB samples that were used in the study. The bacteria strain was able to completely degrade Aroclor 1260 that was incoperated into the wastewater samples within 96 h. This was shown by a shift in the wavelength from 224 to 270 nm, which indicated that Aroclor 1260 was degraded and therefore forming a chlorobenzoate derivative. From this finding, it can be concluded that the sewage water samples did not possess PCB (Aroclor 1260) after treatment with bacteria and can be safely recycled.

Keywords: *Pseudomonas aeruginosa*, sewage water, PCBs, recycled, breakdown

1. Background

One of the key areas in sustainable development entails the promotion of environmental management and introduction of new technologies to treat large quantities of waste. This includes treatment of wastewater for recycling purposes [1]. The adverse effects of global warming have mostly been experienced by countries in Africa, resulting in scarcity of water as a natural resource. This has prompted a great global concern to recycle and conserve water, especially in sub-Saharan Africa where the problem of water scarcity has affected most countries [2]. South Africa is faced with freshwater scarcity, which is exacerbated by its increasing demand, pollution, unsustainable use, and climate change [3].

The presence of chemicals in the environment calls for quantification of such so as to come up with a risk analysis posed by these chemicals [4, 5]. According to Guillen et al., substances such as pharmaceuticals, perfluorinated acids, perfluorosulfonates, PAHs, PCBs, pesticides, and surfactants are mostly found in wastewater [4]. Ying et al. also noted that the presence of pharmaceutically active compounds in wastewater is a major concern [6]. Several methods may be used to determine quantitatively, these substances from wastewater, which is mainly from sewage treatment plants [6]. According to Ying et al., although much research has been done regarding the removal of these substances, it was mainly on activated sludge and no work has been done on wastewater [6].

Some strains of organism *Acinetobacter* have the ability to degrade pollutants such as biphenyls from wastewater [7]. *Enterobacter cloacae* secretes an emulsifier that increases the hydrophobicity of the bacterial cell surface and also neutralizes the surface charge of cells [8]. This as a result increases the ability of the bacteria to degrade PCBs [8, 9]. Biosurfactants are also effective at extremes of temperature, pH, and salinity [9, 10], a property that is essential in the biodegradation of PCBs as they are hydrophobic organic compounds [11]. This property causes these recalcitrant compounds to be removed through physico-chemical means or treatments, limited bioavailability to microorganisms, and limited availability to oxidative and reductive chemicals when applied in treatments [8].

This study is very important in contributing toward addressing sustainable development goal 6. With the global emphasis of conservation of natural resources and the three Rs, that is, reduce, reuse, and recycle, this research is very important. The research is my original proposal which was stimulated by quite a number of issues, such as the scarcity of water although there is a lot of water that is being let to waste. Also the high prevalence of cancer cases with no direct link to water but with a view to eliminate the possibility of such cancer causing chemicals with direct attention on PCBs, from sewage which in most cases the effluent is released into the environment.

2. Brief literature

2.1. Physio-chemical properties of polychlorinated biphenyls (PCBs)

Polychlorinated biphenyls (PCBs) consist of two benzene rings with a carbon-to-carbon bond between carbon 1 on one ring and carbon 1 on the other ring [12]. PCBs have varying number of chlorines in their structure [12–14], as shown in **Figure 1**.

http://dx.doi.org/10.5772/intechopen.75400

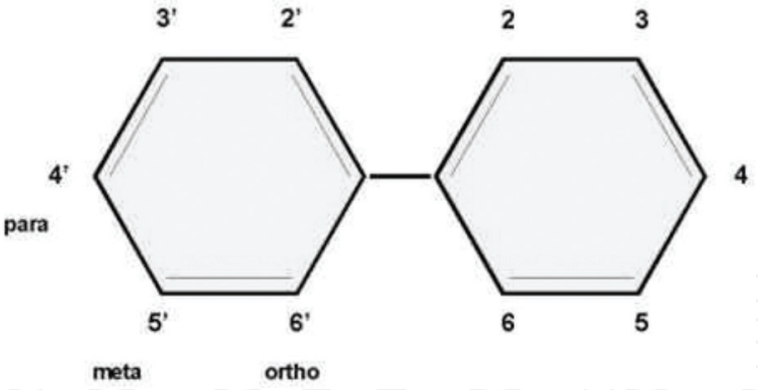

Figure 1. Structure of PCBs (*source*: [14]).

Toxicity of PCBs is dependent upon the number of chlorines present on the biphenyl structure and their position, that is, the co-planar congeners [13, 14]. The PCB congeners that have been deemed to be highly toxic were those that had chlorine atoms attached to the 3,4-ortho positions, followed by those with 5–10 chlorine atoms in the para and meta positions [13].

PCBs toxicity has been largely associated with their structure. This has resulted in PCBs being placed into categories, namely coplanar or non-*ortho*-substituted "*arene*" substitution patterns or noncoplanar or *ortho*-substituted congeners [15]. The coplanar group members are characterized by a fairly rigid structure, with the biphenyl rings in the same plane giving them a molecule structure similar to polychlorinated dibenzo-p-dioxins (PCDDs) and polychlorinated dibenzofurans. Based on this structure, this group of PCBs act in the same way as these molecules as an agonist of the aryl hydrocarbon receptor (AhR) in organisms [16]. This group of PCBs is considered contributors to overall dioxin toxicity [16].

On the other hand, the other group of PCBs, noncoplanar PCBs, has chlorine atoms at the *ortho* positions. According to Ross [17], they have not been found to activate the AhR and are not considered part of the dioxin group; however, they have been implicated in having some neurotoxic and immunotoxic effects, although at levels much higher than normally associated with dioxins, and thus of much less concern to regulatory bodies [18, 19].

According to Rudel et al. PCBs are very stable compounds and do not decompose readily [18]. Their chemical inability to oxidize and reduce in the natural environment gives them this characteristic; they have a long half-life (8–15 years) and are insoluble in water, thus the recalcitrant property [18]. The biodegradability (and solubility in water) of PCBs is also dependent upon the number of chlorine molecules it has [12, 13]. The more chlorine molecules contained in a compound renders that compound less biodegradable [12]. PCBs are mostly hydrophobic; some are less hydrophilic [20, 21]. These properties result in bioaccumulation of these compounds as they do no dissolve in water, and thus, they render them difficult to be biodegraded [12, 14, 20, 22].

2.2. Elimination of PCBs

Although the Stockholm Convention on Persistent Organic Pollutants (POPs) (of which PCBs are part of) signed in 2001 was aimed at eliminating and/or restricting the production and use of POPs [23], more of these ubiquitous substances are still being introduced into the

environment through various human activities [24]. Water has become a widely used environmental matrix for monitoring POPs [20, 25, 26], although most studies on PCBs have been carried out on contaminated soils than water [27].

The destruction of PCBs by chemical, thermal, and biochemical processes is extremely difficult and presents the risk of generating extremely toxic dibenzodioxins and dibenzofurans through partial oxidation [12, 16, 27].

2.3. Effects of polychlorinated biphenyls (PCBs) on human health

PCB mixtures have been associated with cancer incidents in animals from long time back [17, 28, 29]. PCBs were found to induce liver tumors, thyroid adenomas, intestinal metaplasia, and adenocarcinomas in rats and mice [29]. Exposure to some environmental chemicals such as DDT and PCBs has been associated with a drop in sperm count, breast cancer, testicular cancer, and hypospadias, which are all associated with endocrine disruption caused by these chemicals [30]. This comes as a result of some PCB congeners being able to occupy thyroid receptors, thus interrupting their action [17, 30].

PCBs accumulate in the fats of organisms and get passed on from one organism to the other in food chains [31, 32], thus causing bioaccumulation. They get entry into the human body and animals through the skin, lungs, and gastrointestinal tract [13]. PCBs then get distributed to various parts of the body via blood and accumulate in different tissues [31, 33]. The effects of PCBs on humans depend on age, sex, and part of the body affected by chemicals [13]. The liver, as the major organ for removal of toxins in the body, is usually highly affected by PCBs [13, 29]. Humans become exposed to PCBs through consumption of contaminated fish, meat, and dairy products [28] and also through grains grown in PCB contaminated soils [13, 28]. PCBs have been isolated from human milk and serum [31, 34] and have been found to have effects on breastfed children leading to low IQ and endocrine-related ailments [28, 31, 34]. Some studies have shown an increase in cancer mortality in workers exposed to PCBs [13].

2.4. Biodegradation

Biodegradation is the metabolic ability of microorganisms to transform or mineralize organic contaminants into less harmful, non-hazardous substances, which are integrated into natural biochemical cycles [27, 35]. Specific bacteria having bio-degradative potential for various chemical substances in wastewater as well as raw water may be used to treat water [35] for purposes of safe recycling. Bacteria, unlike other organisms, have the ability to interact better with man-made and naturally occurring compounds, which result in such compounds being changed structurally and eventually degraded [35]. This is in a way a better cleanup strategy that can be used in the cleanup of wastewater as it is environment friendly [35]. Use of mixed population of microbes is usually recommended as it has been seen to yield faster results as the two different microbes attack different parts through different mechanisms resulting in effective breakdown of the toxic compound [21, 33]. This activity also creates a condition of co-metabolism [33].

PCBs may not be readily biodegradable, but studies have shown that some bacteria species such as *Vibrio cholera*, *Acinetobacter 1woffnii*, *Aeromonas hydrophila*, *Pseudomonas aeruginosa*, *Pseudomonas putida*, *Rhodococcus* sp., *Bacillus* sp., and *Burkholderia* sp. have the ability to breakdown these compounds, although it is through a very long route [11, 13, 21, 36, 37, 51]. This may be achieved through co-metabolism and mineralization [8, 35]. They use of a metabolic pathway similar in all these bacteria, which comprise four steps catalyzed by enzymes BphA, BphB, BphC, and BphD [37]. The pathway, according to Petric et al., is initiated by insertion of two oxygen atoms at the carbon positions 2, 3 of one aromatic ring [37]. This is followed by dehydrogenation meta-cleavage and hydrolysis forming a 5-carbon compound [37]. The process follows a biphenyl catabolic pathway [37].

2.5. Biodegradation of xenobiotic compounds

According to Heider and Rabus, xenobiotic compound due to its recalcitrant nature is hard to break down [38]. The recalcitrant nature of these compounds is a result of the complexity of its chemical composition [8]. Breakdown of these compounds occurs when enzymes act on certain groups present in the compound [38]. The halocarbons, for example, the halogen group, are targeted, with enzymes such as oxygenases playing a major role in their breakdown [8]. The enzymes target the bonds such as ester-, amide-, or ether bonds present in the compounds leading to break down of these compounds [39, 40]. The enzymes may target the aliphatic chains and in aromatic compounds, the aromatic components may be targeted [40]. The mode of attack as well as the site depends primarily on the action of enzyme, its concentration, and favorable conditions [40]. According to Abor-Amer [40], the xenobiotics do not act as a source of energy to microbes and as a result, they are not degraded while the presence of a suitable substrate induces its breakdown [39]. These substrates are known as co-metabolites, and the process of degradation is known asco-metabolism [39]. Gratuitous metabolism is another process in which xenobiotics serve as substrates and are acted upon to release energy [8].

The processes described cannot be achieved through the use of *Moringa oleifera* in treating wastewater to remove PCBs. It is evident from literature that the removal of these compounds using plant protein has not been fully studied [52]. Plant protein has been found to be slightly efficient with the reduction of fecal coliforms and other bacteria [41, 42], which has made Moringa treatment to be applicable. The use of *Moringa oleifera* seed powder in water treatment plants has been found to target mainly microorganisms, thus reducing turbidity [43]. Although this mode of water treatment has been used, especially in rural areas of the developing countries, synthetic polymers, aluminum sulfate, ferric chloride, and poly aluminum chlorides used together with this powder have been reported to be unsafe [41, 43, 44]. The action of *Moringa oleifera* seed powder has been reported to be based on the ability of the protein contained in the seeds to be able to form coagulants, which reduce water turbidity by acting on coliforms [45]. The bacteria found to be mainly involved in biodegradation of POPs and PCBs have been found mostly not to be coliforms [11, 13]. After treating water with Moringa seed powder, 10^1–10^5 of bacteria is left [45]. Taking into cognizance that Moringa is a tree, sustainability of tree growth and productivity, which relies on environmental conditions, may not be viable. This will therefore affect production and maintenance of the Moringa tree

species, given the global warming and related environmental problems. Growing of bacteria indoors is quite sustainable, when compared to growth of plants although Lea argues that propagation is affordable [44].

3. Materials and method

3.1. Sample collection

Water released from the wastewater treatment plant (effluent) was obtained from Notwane Sewage Treatment Plant situated in Gaborone, Botswana. It was collected from the sampling site in sterile 250 ml Duran bottles and immediately placed on ice in a cooler box with ice. The samples were taken to the Department of Biological Sciences, North-West University, Mafikeng for analysis. Samples were analyzed within 24 h of sampling. The treatment plant treats 40,000 m^3 per day of sewage.

3.2. Biodegradation of PCBs in wastewater by isolate *Pseudomonas aeruginosa*

In the study carried out by the author, out of the many bacteria stated in literature, only *Pseudomonas aeruginosa* isolated from the wastewater sampled during the study was used. The water samples were divided into two parts, one part was sterilized by autoclaving at 121°C for 15 min and the other half was left unsterilized. The wastewater samples were treated with Aroclors of polychlorinated biphenyls (PCBs) obtained from SUPELCO Solutions Within™, USA, through Lehlabile Scientific, South Africa. The PCBs were supplied as Aroclors. Aroclor 1242 (Lot No. LB8851), 1248 (Lot No. LB88969), and 1260 (Lot No. LB92109) in 1 ml ampoules at concentration 1000 μg/ml dissolved in isooctane were used in this study. The purity for each Aroclor was not stated.

To each 100 ml wastewater sample in a 250 ml flask, 10 μl of polychlorinated biphenyls Aroclors mixture, herein referred to as PCBs, was added. The sterilized wastewater samples were inoculated with a colony of the 18 h old culture of the test organism, which was identified as *Pseudomonas aeruginosa* (with accession number from the gene bank of CP 006832 in a study carried out in 2014). Non-sterilized wastewater without bacterial inoculation (Control 1) and sterilized wastewater without inoculation (Control 2) were both treated with PCBs and were the controls. The flasks were wrapped with aluminum foil to exclude light and were incubated at 30°C in the dark in a rotary shaker at 150 rpm [46]. A 5 ml was aseptically taken at 24 h intervals from each setup/flask for detection of PCBs using HPLC and spectral changes were checked at 200–800 nm using Cary 300 UV-visible spectrophotometer, for a period of 96 h.

Analysis for PCB using HPLC was carried out as described by Roy et al. with some modifications [46]. A 1 ml was sampled from each setup to check for residual PCB at 24 h interval. The compounds were extracted by adding 10 ml each of dichloromethane and acetone. The mixture was incubated in a rotary shaker for 24 h at 30°C. After incubation, the mixture was centrifuged for 10 min at 12,000 rpm at 4°C using a Hermle Z326k high speed microcentrifuge, Labortechnik GmbH (LASEC, South Africa). The extra water was pipetted and 4 g

http://dx.doi.org/10.5772/intechopen.75400

of anhydrous sodium sulfate mixed with a PCB-containing solvent to remove residual water. The extract was concentrated to 1.5 ml using a rotary evaporator Stuart RE300DB, LASEC, South Africa and filtered with 0.45 μm PTFE syringe filters. Extracts were analyzed by high performance liquid chromatography (HPLC) UFLC Shimadzu using a florescence detector pump RF-20A and system gold column C18 (CTO-20A). The excitation level was set at 254 nm, emission level at 390 nm. The mobile phase used was a mixture of acetonitrile and water (80:20) as described by Roy et al. [46]. Data analysis was computed using real-time analysis. All chemicals used were of HPLC grade supplied by Sigma Aldrich through Lehlabile Scientific, South Africa.

4. Results

4.1. Degradation of PCBs by *Pseudomonas aeruginosa*

Samples of wastewater from the Notwane Sewage Treatment Plant were used in this study to find out the degree of biodegradation of PCBs in wastewater un-inoculated and inoculated with the test organism. Spectral changes (a shift in wavelength (λ_{max}) in nm), detected using the UV-visible spectrophotometer, were used as an indication that the compounds were broken down into new products. The results of the wavemax (λ_{max}) nm obtained are presented in **Figure 2**.

The results shown by chromatogram indicated that there was a shift in λ_{max} from 224 to 270 nm in 0 h of incubation to 96 h of incubation at 30°C on a rotary shaker in the dark. These results were obtained using a Cary 300 UV-visible spectrophotometer at a wavelength range of 200–800 nm. The results were an indication that isolates *Pseudomonas aeruginosa* was able to degrade Aroclor 1260 into chlorobenzoates and derivatives, which have wavelength ranging from 244 to 270 nm, hence the shift in wavelength.

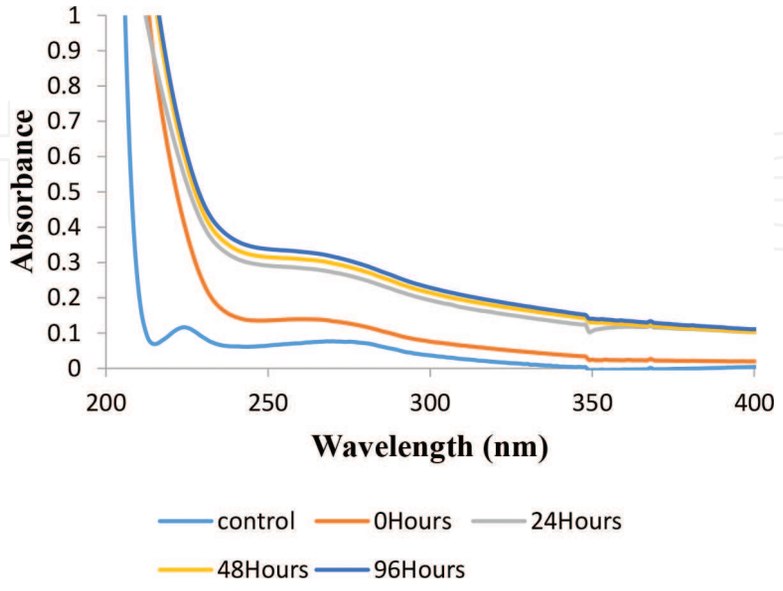

Figure 2. Spectral changes of PCB degradation in water inoculated with isolate *Pseudomonas aeruginosa*.

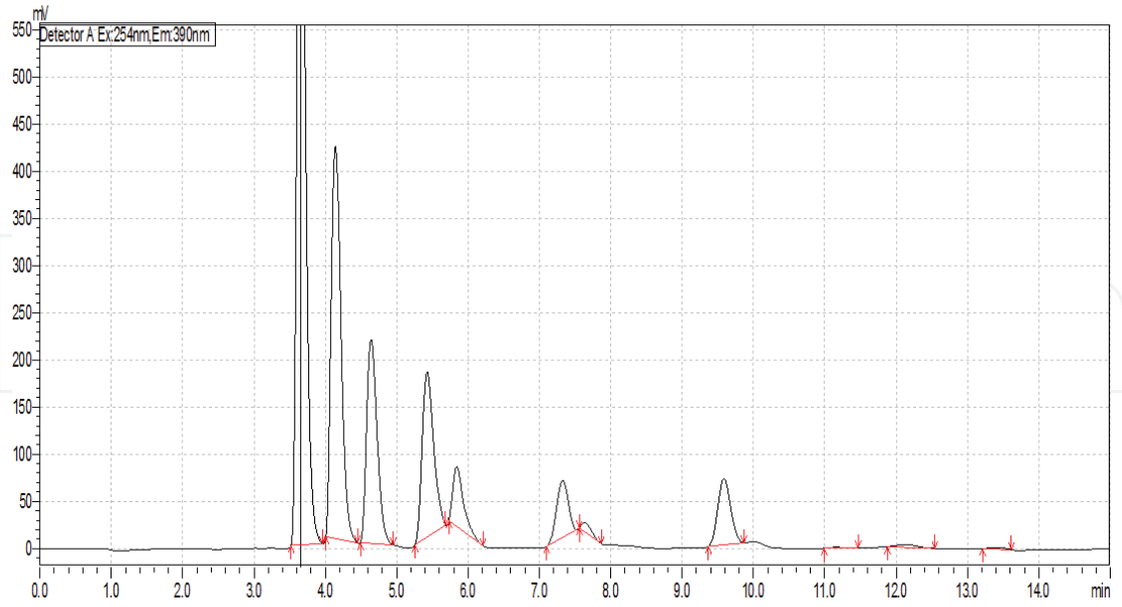

Figure 3. HPLC chromatogram for PCBs standard using a florescence detector method at 10 µl injection volume.

4.2. Results from high performance liquid chromatography (HPLC)

HPLC chromatogram depicted differences in picks obtained for the experiments, the controls, and the standards. Although concentrations were not determined, no PCBs were detected by HPLC after 96 h of incubation. This was an indication that the bacteria degraded the compounds, hence the chromatogram shown in **Figure 4**. The chromatogram obtained for the experiments was compared with the chromatogram for the PCB standard using the retention times for the compounds in the standard, which was represented in **Figure 3**. The results for

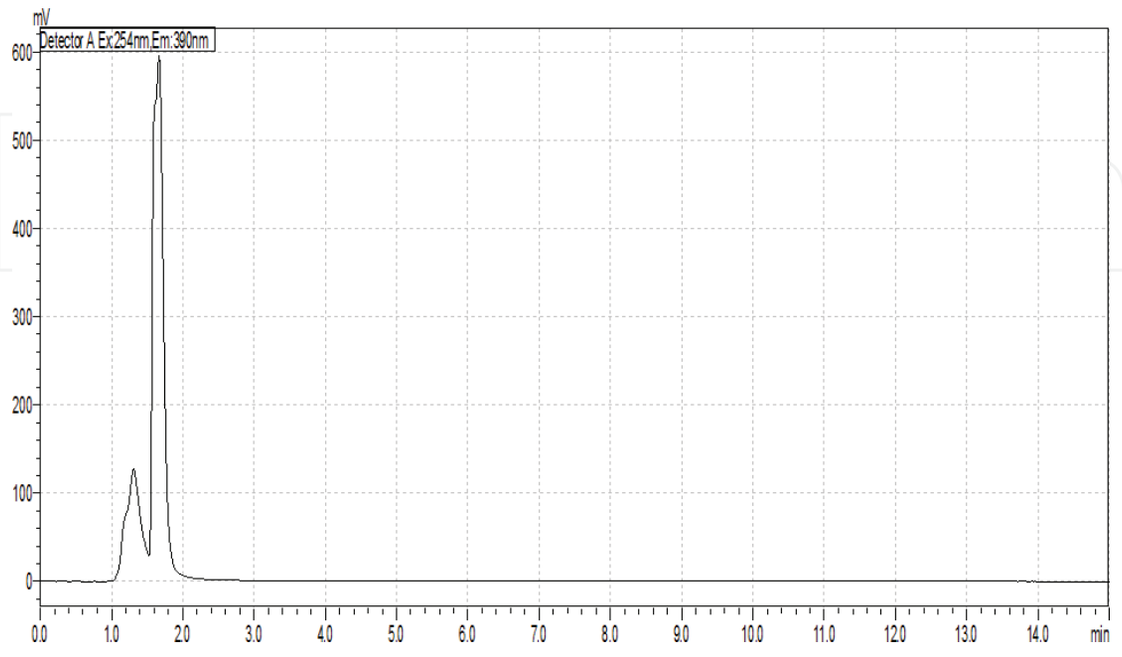

Figure 4. Chromatogram for PCB biodegradation experiment after 96 h of incubation.

PCB standard presented in **Figure 3** showed that there were several compounds in the Aroclor; thus, the many picks, which are not found when the same treatment was extended to the experiment, resulted in **Figure 4**.

The chromatogram shows that the components of the standard had retention times ranging from 3.647 to 13.119 min. The components picked by the instrument are indicated by the red line marking the beginning and ending of the peak. The instrument could not detect the actual names of the compounds though.

The wastewater was sterilized and thereafter inoculated with the test organism and PCBs mixture added. **Figure 4** shows that no peaks were picked by the HPLC. This is an indication that the polychlorinated biphenyls added to the wastewater were completely broken down by isolate *Pseudomonas aeruginosa* resulting in complete elimination.

Representation of **Figure 4** is a clear indication that there were no detectable amounts of compounds after Aroclor was subjected to bacterial treatment. The compounds were broken down in 5 days. There are no peaks shown as compared to the chromatogram shown in **Figure 3**.

5. Discussion

The wavelength maximum (λ_{max}) observed after 96 h ranged from 264 to 269 nm on average for PCBs (**Figure 2**). These results indicated that the PCBs were broken down forming new products with different wavelength and thus the change. This implies that the bacteria were able to use them as their sole source of carbon; thus, the biodegradation of the PCBs added to wastewater. The shifts are an indication of the presence of initial ring oxidation metabolites and ring fission metabolites [19, 47]. PCBs first get degraded into chlorobenzoates [47] that have been found to have λ_{max} ranging from 210 to 214 nm in the B-band and 244 to 270 nm in the C-band when dissolved in water [48], a range that was observed in the results obtained after 24 h of culturing the organism used in this study in PCBs Aroclor mixture, results of which are shown in **Figure 2**. The results depicted that the bacteria was able to breakdown the PCBs, which were similar to results in a similar studies by Vrchotova et al. and Seeger et al. [47, 49]. In their studies [47, 49], the product chlorobenzoate was biodegraded into benzoate and eventually pyruvate and acetyladehyde, which are essential in the tricarboxylic cycle (TCA) [47, 49].

The HPLC run confirmed that the compound was biodegraded by the bacteria isolate *Pseudomonas aeruginosa* as presented in **Figures 3** and **4**, which was also proved by Heider and Rabus, Roy et al. and Raja et al. [38, 46, 50] in their studies. No PCB compound was detected after 96 h of exposure to the bacteria in wastewater.

6. Conclusion

Pseudomonas aeruginosa, isolated from wastewater in the Notwane Sewage Treatment Plant was successfully used in biodegradation of recalcitrant polychlorinated biphenyls (PCBs). This having been successfully employed at the micro level, and further tests can be carried out to

validate the results obtained in this study. With this recommendation in place, it is ideal to say that employing bacteria in the biodegradation processes of recalcitrant PCBs will be highly cost effective as it is a biotechnological process. The process will enable developing countries to employ effective but easy to maintain at a cost effective mode means of wastewater treatment. This wholly will also enable these countries to address the problem of water shortage at the same time practicing water conservation strategies. This is in a way contributing toward addressing the sustainable development goals (SDGs). With the findings from this study, a recommendation for further experimentation on a larger scale is made so as to safely recycle the sewage water for purposes of redirecting to Gaborone dam. This in a way will aid in curbing the problem of water shortage, of course, taking into consideration other factors, such as total coliforms, *Escherichia coli,* and other pathogenic organisms and chemicals. These have to be within the expected standards according to Botswana Bureau of Standards (BOBS) limits as well as international World Health Organization (WHO) standards.

Acknowledgements

My gratitude goes to the Department of Biological Sciences at North-West University, Mafikeng Campus for financial support while carrying out the study. I extend my gratitude to Professor C.N. Ateba, Professor N.P. Sithebe, and Dr. K. Sichilongo for all their support and guidance. I would like to thank the Ministry of Basic Education of Botswana for providing me time to carry out the research and financial support in one way or the other.

Conflict of interest

The author declares that there is no conflict of interests regarding the publication of this paper.

Other declarations

This chapter was extracted from the thesis (unpublished) of my research for PhD, which was undertaken in 2014 with University of North West, Mafikeng Campus in South Africa.

Author details

Spar Mathews* and Patricia Sithebe

*Address all correspondence to: sparmat@gmail.com

North West University, Mafikeng Campus, South Africa

References

[1] Khadhraoui M, Belaid C. Wastewater treatment for a possible water reuse in semi-arid climate zone. Journal of Arid Land Studies. 2012;**22**(1):333-336

[2] Ateba CN, Maribeng MD. Detection of enterococcus species in groundwater from some rural communities in the Mmabatho area, South Africa: A risk analysis. African Journal of Microbiology Research. 2011;**5**(23):3930-3935

[3] Department of Water Affairs (DWA). The Annual National State of Water Resources Report: October 2011 to September 2012. Water Resource Information Programmes. South Africa: Department of Water Affairs; 2012

[4] Guillen D, Ginebreda A, Farre M, Darbra RM, Petrovic M, Gros M, Barcelo D. Prioritization of chemicals in the aquatic environment based on risk assessment: Analytical, modeling and regulatory perspective. Science of the Total Environment. 2012;**440**:236-252. DOI: 10.1016/j.scitotenv.2012.06.064

[5] Urbaniak M. Biodegradation of PCDDs/PCDFs and PCBs. Biodegradation—Engineering and Technology. 2013. pp. 73-100

[6] Ying G, Kookana RS, Kolpin DW. Occurrence and removal of pharmaceutically active compounds in sewage treatment plants with different technologies. Journal of Environmental Monitoring. 2009;**11**:1498-1505

[7] Gumaa NHH, Potrus WM, Mohammed SI. The effect of cultural and environmental conditions on biodegradation and biosurfatant production by Serratiamarcescens utilizing weathered diesel oil. Journal of Al-Nahrain University. 2010;**13**(1):112-120

[8] Pacwa-Plociniczak M, Plaza GA, Piotrowska-Seget Z, Cameotra SS. Environmental applications of biosurfactants: Recent advances. International Journal of Molecular Science. 2011;**12**:633-654

[9] Md F. Biosurfactant: Production and application. Journal of Petroleum and Environmental Biotechnology. 2012;**3**(4):124-129

[10] Kapadia SG, Yagnik BN. Current trend and potential for microbial biosurfactants. Asian Journal of Experimental Biological Sciences. 2013;**4**(1):1-8

[11] Blokesch M. Chitin colonization, chitin degradation and chitin-induced natural competence of vibrio cholera are subject to catabolite repression. Journal of Environmental Microbiology. 2012;**14**(8):1898-1912

[12] Environmental Protection Agency. Health Effects of Polychlorinated Biphenyls (PCBs). Washington, DC: United States of America EPA; 2013

[13] Anyasi RO, Atagana HI. Biological remediation of polychlorinated biphenyls (PCB) in the soil and sediments by microorganisms and plants. African Journal of Plant Science. 2011; **5**(7):373-389

[14] Barbalace RC. The Chemistry of Polychlorinated Biphenyls.Environmental Chemistry. com. 2003. [Accessed 21/09/2013]

[15] Jensen WB. The origins of the ortho-, meta-, and para- prefixes in chemical nomenclature. Journal of Chemical Education. 2006;**83**(3):356

[16] Arsalan J, Stuart H, Sadegh H, Stuart H. Concentrations and chiral signatures of polychlorinated biphenyls in outdoor and indoor air and soil in major UK conurbation. Environmental Science and Technology. 2007;**41**(7):2153-2158

[17] Ross G. The public health implications of polychlorinated biphenyls (PCBs) in the environment. Ecotoxicology and Environmental Safety. 2004;**59**(2004):275-291

[18] Rudel RA, Seryak LM, Brody JG. PCB containing wood floor finish is a likely source of elevated PCBs in residents blood, household air, and dust: A case study of exposure. Journal of Environmental Health. 2008;**7**(21):1-8

[19] Wethington DM, Hornbuckle KC. Milwaukee WI as a source of artmospheric PCBs to LAKE Michigan. Environmental Science and Technology. 2005;**39**(1):57-63

[20] Muir D, Lohmann R. Water as a new matrix for global assessment of hydrophilic POPs. Trends in Analytical Chemistry. 2013;**46**(2013):163-172

[21] Nwinyi OC. Enrichment and identification of Askarel oil (PCB blend) degrading bacteria enriched from landfill sites in Edo state, Nigeria. Agriculture and Biology Journal of North America. 2011;**2**(1):89-100

[22] Schafer KS, Kegley SE. Persistent toxic chemicals in the US food supply. Journal of Epidemiology Community Health. 2002;**56**:813-817

[23] Andrews W. Manual of Food Control.4 Rev. 1. Rome: Microbiological Analysis Food and Agriculture Organisation of the United Nations (FAO); 1992

[24] Teran T, Lamon L, Marcomini A. Climate change effects on POPs environmental behaviour: A scientific perspective for future regulatory actions. Atmospheric Pollution Research. 2012;**3**(2012):466-476

[25] Van Leeuwen SPJ, Van Bavel B, DeBoer J. First worldwide UNEP interlaboratory study on persistent pollutants (POPs), with data on polychlorinated biphenyls and organochlorine pesticides. Trends in Analytical Chemistry. 2013;**46**(2013):110-117

[26] Van Leeuwen SPJ, Van Bavel B, Abad E, Leslie HA, Fiedler H, DeBoer J. POPs analysis reveals issues in bringing laboratories in developing countries to a higher quality level. Trends in Analytical Chemistry. 2013;**46**(2013):198-206

[27] Leigh MB, Prouzova P, Mackova M, Macek T, Nagle DP, Fletcher JS. Polychlorinated biphenyl (PCB) – degrading bacteria associted with trees in a PCB-contaminated site. Applied and Environmental Microbiology. 2006;**72**(4):2331-2342

[28] Lynch CD, Jackson LW, Kostyniak PJ, BM MG, GMB L. The effect of prenatal and postnatal exposure to polychlorinated biphenyls and child neurodevelopment at age twenty four months. Reproductive Toxicology. 2012;**34**:451-456

[29] Pavuk M, Cerhan JR, Lynch CF, Schecter A, Petrik J, Chovancova J, Kocan A. Environmental exposure to PCBs and cancer incidence in Eastern Slovakia. Chemosphere. 2004; **54**(2004):1509-1520

[30] Rogan WJ, Ragan NB. Some evidence of effects of environmental chemicals on the endocrine system in children. Journal of Hygiene and Environmental Health. 2007;**210**(2007):659-667

[31] Man YB, Lopez BN, Wang HS, Leung AOW, Chow KL. Cancer risk assessment of polybrominateddiphenyl ethers (PBDEs) and polychlorinated biphenyls (PCBs) in former agricultural soils of Hong Kong. Journal of Hazardous Materials. 2011;**195**(2011): 92-99

[32] Zhao G, Wang Z, Zhou H, Zhao Q. Burdens of PBBs, PBDEs, and PCBs in tissues of the cancer patients in the e-waste. Science of the Total Environment. 2009;**407**(2009):4831-4837

[33] Martins LF, Peixoto RS. Biodegradation of petroleum hydrocarbons in hypersaline environments. Brazillian Journal of Microbiology. 2012;**2012**:865-872

[34] Linderholm L, Biague A, Mansson F, Norrgren H, Bergman A, Jakobsson K. Human exposure to persistent organic pollutants in West Africa—A temporal trend study from Guinea-Bissau. Environmental International. 2010;**36**:675-682

[35] Dhall P, Kumar R, Kumar A. Biodegradation of sewage water using autochthonous bacteria. The Scientific World Journal. 2012;**861903**:1-8

[36] Hamzah A, Rabu A, Azmy RFHR, Yussoff NA. Isolation and characterisation of bacteria degrading Sumandak and South Angsi oils. Sains Malaysiana. 2010;**39**(2):161-168

[37] Petric I, Hrsak D, Fingler S, Voncina E, Cetcovik H, Kolar AB, Kolic NU. Enrichment and characterization of PCB-degrading bacteria as potential seed cultures for bioremediation of contaminated soils. Food Technology and Biotechnology. 2007;**45**(1):11-20

[38] Heider J, Rabus R. Genomic Insights in the Anaerobic Biodegradation of Organic Pollutants. Microbial Biodegradation: Genomics and Molecular Biology. Caister: Academic Press; 2008. http://www.link.springer.com/referenceworkentry [Accessed: November 04, 2014]

[39] Cyon M, Zmijowska A, Wojcik M, Piotrowska-Seget Z. Biodegradation and bioremediation potential of diazinon-degrading Serratiamarcescens to remove other organophosphorus pesticides from soils. Journal of Environmental Management. 2013; **117**(2013):7-16

[40] Abor-Amer AE. Biodegradation of diazinon by Serratiamarcescens DI101 and its use in bioremediation of contaminated environment. Journal of Microbiology and Biotechnology. 2011;**21**(1):71-80

[41] Dalen MB, Pam JS, Izang A, Ekele R. Synergy between Moringaoleifera seed powder and alum in the purification of domestic water. Science World Journal. 2009;**4**(4):6-11

[42] Kawo AH, Daneji IA. Bacteriological and physcio-chemical evaluation of water treated with seed powder of Moringaoleifera LAM. Bayero. Journal of Pure and Applied Sciences. 2009;**4**(2):208-212

[43] Mumuni A, Oloruntoba EO, Sridhar MKC. Use of Moringaoleifera (LAM) seed powder as a coagulant for purification of water from unprotected sources in Nigeria. European Scientific Journal. 2013;**9**(24):214-229

[44] Lea M. Bioremediation of Turbid surface water using seed extract from *Moringa oleifera* (LAM.) (Drumstick) Tree. Current Protocols in Microbiology. 2014;**1G.2.1-1G.2.8**

[45] Mangale SM, Chonde SG, Jadhav AS, Raut PD. Study of *Moringa oleifera* (drumstick) seed as natural absorbent and antimicrobial agent for river water treatment. Journal of National Production and Plant Resources. 2012;**2**(1):89-100

[46] Roy M, Khara P, Basu S, Dutta TK. Catabolic versatility of sphingobiumspstrain PNB capable of degrading structurally diverse aromatic compounds. Journal of Bioremediation and Biodegradation. 2013;**4**(1):1-6. http://www.academia.edu/ [Accessed: November 04, 2014]

[47] Seeger M, Hernandez M, Mendez V, Ponce B, Cordova M, Gonzalez M. Bacterial degradation and bioremediation of chlorinated herbicides and phenyls. Journal of Soil Science & Plant Nutrition. 2010;**10**(3):320-332

[48] Forbes WF. Light absorption studies: The ultraviolet absorption spectra of chlorobenzenes. Canadian Journal of Chemistry. 1960;**38**(7):1104-1112

[49] Vrchotova B, Mackova M, Macek T, Demnerova K. Bioremediation of chlorobenzoic Acids. Agricultural and Biological Sciences. "Applied Bioremediation—Active and Passive Approaches". 2013. http://www.intechopen.com/books/applied-bioremediation [Accessed: November 04, 2014]

[50] Raja CE, Selvam GS, Omine K. Isolation, Identification and Characterisation of heavy metals resistant bacteria from sewage. In: Fukuoka JS, editor. International Joint Symposium on Geo-disaster Prevention and Geo-environment in Asia. 2009. pp. 205-211. http://www7.civil.kyushu-u.ac.jp/jeotech [Accessed: November 04, 2014]

[51] Martinkova L, Uhnakova B, Patek M, Nesvera J, Kren V. Biodegradation potential of the genus Rhodococcus. Environment International. 2009;**35**:162-177

[52] Farhadian M, Duchez D, Vachelard C, Larroche C. Monoaromatics removal from polluted water through bioreactors—A review. Journal of Water Research. 2008;**42**:1325-1341