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Distribution of polychlorinated biphenyls in effluent from a large municipal wastewater treatment plant: Potential for bioremediation?

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ABSTRACT

This study involved an evaluation of the potential for bioremediation of polychlorinated biphenyls (PCBs) in the effluent from a large municipal wastewater treatment plant. It was focused on the presence of PCBs in two types of effluents: the continuous effluent present during dry weather conditions and the intermittently present effluent that was present during wet weather due to incoming stormwater. The annual discharge of PCBs for both types of effluent was calculated based on a five-year dataset (2011–2015). In addition, the toxicity and bioremediation potential of the PCBs in the effluent were also assessed. It was found that the continuous effluent was responsible for the majority of the discharged PCB into the receiving river (1821 g for five years), while the intermittent effluent contributed 260 g over the five years. The average number of chlorine per biphenyl for the detected PCB congeners showed a 19% difference between the two types of effluent, which indicated a potential for organohalide respiration of PCBs during the continuous treatment. This was further supported by a high level of tri-, tetra- and penta-chlorinated congeners accounting for 75% of the anaerobically respired PCBs. Potential for aerobic degradation and thus biomineralization of PCBs was identified for both effluents. Furthermore, toxicity of 12 dioxin-like PCBs showed that normal operation of the wastewater reduced the toxicity throughout the wastewater treatment plant.

Introduction

Polychlorinated biphenyls (PCBs) are a group of persistent organic pollutants (POPs) with 10 homologs and 209 congeners (Focant et al., 2004). They have globally been used for industrial purposes since the 1920s (Abbas et al., 2014). Many studies have over time shown that PCBs are highly toxic with undesirable effects on the environment and humans (Cheng and Hu, 2010; Lallas, 2001). Studies have documented the PCB contaminations in the surface water and groundwater (Rodenburg et al., 2010; Wu et al., 2015). PCBs can potentially enter a wastewater treatment plant (WWTP) *via* the sewer system and/or stormwater (in areas with combined sewer and stormwater systems) and be discharged *via* wastewater effluent. Balasubramani et al. (2014) determined the total PCB (Σ209) concentration from 16 different effluent outfalls which collected from municipal and industrial

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wastewater treatment plants in the Houston area. The PCB congeners with low-chlorine contents had the highest concentrations in the dissolved phase (*i.e.*, wastewater) (1.01 to 8.12 ng/L). The highly chlorinated PCB congeners in the suspended phase (*i.e.*, suspended solids) were the most abundant and ranged from 2.03 to 31.2 ng/L. Therefore, PCBs from a wastewater effluent with a large volume discharging into an aquatic environment can result in bioaccumulation through the food chain and thus expose aquatic organisms to PCBs with toxicity as a result (Kulkarni et al., 2008; North, 2004).

Biodegradation of PCBs can take place under aerobic conditions that can lead to biomineralization or during anaerobic conditions, where organohalide respiration can take place (Laroe et al., 2014; Zanaroli et al., 2015). The total amount of PCBs discharged in the effluents and their toxicity can potentially be controlled by aerobic and anaerobic processes that transform the PCBs. The toxicity of the highly chlorinated PCB congeners can be reduced by anaerobic organohalide respiring bacteria. Some dioxin-like PCBs (*e.g.*, PCB-114, PCB-156, and PCB-157) can be reduced to lower chlorinated congeners by anaerobic organohalide respiring bacteria such as *Dehalobium chlorocoercia DF1, Desulfitobacterium dehalogenans, Desulfomonile tiedjei, and Dehalococcoides mccartyi* (Kranzioch et al., 2013; Payne et al., 2017). Subsequently, these products containing less than four chlorines per molecule can be degraded by aerobic bacteria (*e.g.*, *Burkholderia xenovorans* strain LB400, *Alcaligenes xylosoxidans, Pseudomonas stutzeri*, *Ochrobactrum anthropi*, and *Pseudomonas veronii*) (Correa et al., 2010; Murínová et al., 2014; Passatore et al., 2014). As a result, their toxicity were eliminated due to biodegradation into chlorobenzoic acid as well as other degradation products and eventually into carbon dioxide (Chang et al., 2013; Pieper, 2005). One way to assess PCB toxicity is the Toxic Equivalency (TEQ) that can be calculated by applying Toxic Equivalency Factors (TEFs) that have been defined in comparison to the most toxic dioxin, 2,3,7,8-tetrachlorodibenzodioxin (2,3,7,8-TCDD) (Van den Berg et al., 2006). However, the calculation of TEQs is limited to the toxicity of the 12 dioxin-like PCB congeners, since TEFs do not exist for the remaining 197 PCB congeners (Baars et al., 2004). To data, numerous studies have documented the presence of organohalide respiration with PCBs in the natural environment (Ahmed and Focht, 1973; Su et al., 2015). However, limited studies and information are available on PCB bioremediation in conventional wastewater treatment systems and sewage sludge (Dong et al., 2015; Patureau and Trably, 2006). Most of the PCB organohalide respiration studies were conducted by using mesocosm experiments in soil or sediments with the presence of PCBs. It is currently uncertain whether WWTP processes have the ability to perform organohalide respiration with PCBs or aerobically mineralize PCBs during treatment (Deblonde et al., 2011; Kiedrzyńska et al., 2017).

This paper presented an assessment of the annual PCB amount discharged from effluents originating from an intermittent (wet weather) discharge and a continuous effluent from a large municipal WWTP. In addition, the toxicity and bioremediation potentials of PCBs were discussed based on an evaluation of the proportion of PCBs that can be anaerobically or aerobically transformed. The number of chlorines on the biphenyl rings as well as the positions affects the toxicity of the PCBs thus the discharge of PCBs influence the potential health effects downstream of the discharge area.

1. Materials and methods

1.1. Wastewater treatment processes

The WWTP in this study has a daily treatment capacity of above 200 million gallons of wastewater. The sewer collection system in the service area is a combined sewer system that covers one-third of the area, while separate sanitary sewer systems cover the remaining area. Under normal operational conditions when the influent flow is below the treatment capacity of the plant, two separately primary influents drain directly into the WWTP, where large particles from untreated wastewater are initially removed using screens and gravity settling basins. The organic matter present in the effluent is then consumed through biological treatment (secondary treatment) with aeration. After this step, the microorganisms convert ammonia into nitrate and nitrite through a nitrification process. These compounds are subsequently converted to nitrogen gas. As a final step, the wastewater passes through a filtration unit made by sand and anthracite. Before discharge to the river the effluent is disinfected by sodium hypochlorite-based chlorination and the residual chlorine is removed before the treated wastewater is discharged into the river. During periods of rain, the combined sewer systems in the area will also transport stormwater thus increasing the required volume for treatment above the capacity of the WWTP. In this case, the volume that cannot be treated as described above will pass through the primary processes, but the effluent will be discharged into the river without any biological treatment. Disinfection takes place to comply with the requirements for discharge (Gray, 2004).

1.2. Wastewater sampling and PCBs congener analysis

In this study, 10 PCB homologs and 209 PCB congeners were evaluated in effluent discharge during normal treatment and in case of stormwater overflow during a five-year study period (2011–2015). The principal wastewater treatment flowchart is shown in Fig. 1. The samples evaluated in this study were collected from two wastewater discharge points: One was a bypass effluent (intermittent effluent) after the primary treatment process, while the other was from regular treatment effluent (continuous effluent). The samples from the bypass effluent were collected during rain events as grab samples. The sample was pumped from the dechlorination tank to the sample bottle after the pump had been running for at least 15 min prior to sampling to flush the stale volume out of the pipe. At this point the effluent was flushed out for another 5 min, then the flow was reduced to allow for sampling. The influent flow was required to be approximately 40% higher than the daily capacity for sampler initiation from this effluent discharge. Composite samples were collected from the normally treated effluent during planned dry weather events twice per quarter and during wet weather events twice per quarter using a Sigma 900Max #006 composite sampler (HACH Company, Colorado, USA). Teflon-lined Tygon (American Durafilm Co. Inc., Holliston, USA) tubing with a strainer attached to the

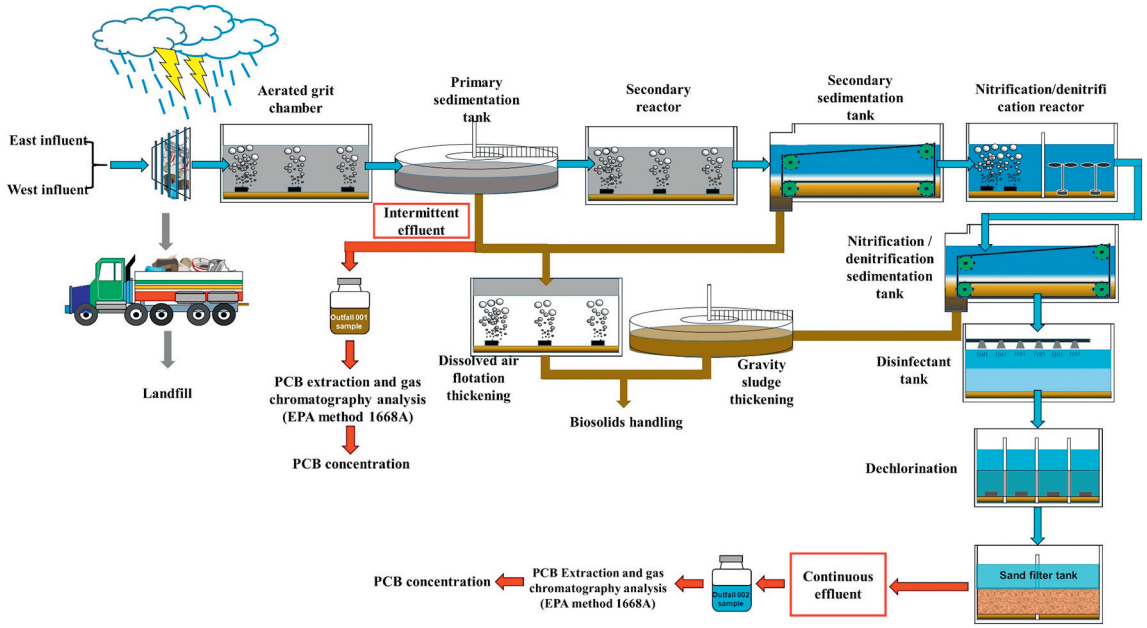

Fig. 1 – Principal flow diagram showing the major wastewater treatment processes at the waste water treatment plant that was analyzed in this study.

pump tubing with a stainless steel connector and two hose clamps at each end of the connector. The sampling bottles were placed into a double-bagged drum liner bag to minimize contamination from outside sources and the tubing from the sampler head was carefully placed into the sample bottle. Trace rainfall amounts were not reported 72 hr prior to the dry weather sampling events. Deionized water was applied as field blank samples for each sampling batch (EPA, 2008). All sampling operations were performed with gloves and the samples were handled in a biological safety cabinet to prevent exposure to humans and contamination of the samples.

Extraction of PCBs from the samples was performed using solid-phase extraction (SPE) or a continuous liquid/liquid extraction (CLLE) (EPA, 2008) by SGS AXYS Analytical Services Ltd., British Columbia, Canada. After extraction, a cleanup standard was spiked into the extracts and additional clean up using back-extraction with sulfuric acid and Florisil chromatography was performed. The PCB extracts were concentrated to 20 μL, and internal standards were injected into each extract prior to analysis by gas chromatography (GC) and high-resolution mass spectrometer (MS). All 209 PCB congeners were analyzed at a certified laboratory by using EPA method 1668B as discussed above (Rushneck et al., 2004). The analytical results were corrected by the a rinsate blank (Equipment Blank) and a method blank prior to any future calculation and data analysis (Johnson et al., 2008; Keith, 1996).

1.3. Calculation of annual PCB discharge

The collection of effluent samples from continuous treatment occurred approximately 11 times per year, while collection of grab samples from the stormwater overflow took place 6–7 times per year over the five-year study period. Missing data points between the sampling events were estimated based on the existing sampling data. The calculation of the total PCB mass distribution and contribution from PCB homologs was based on the concentration from the analyzed samples and converted into mass by multiplying with the effluent flowrate (Appendix A Eqs. (S1) and (S2)).

1.4. Analysis of toxicity equivalent (TEQ) for 12 dioxin-like PCBs

The TEQ concentration for each data point was calculated by multiplying the concentration of each of the 12 dioxin-like PCB congener with the corresponding toxicity equivalent factor (TEF) thus obtaining a total TEQ for each sample (Van den Berg et al., 2006). In this study, TEFs from the World Health Organization (WHO) (Van den Berg et al., 2006) were applied. The calculated total TEQ concentrations represent the equivalent amount of toxicity that 2,3,7,8-TCDD would have impacted the environment with.

1.5. Analysis of chlorine/biphenyl for PCBs

The number of chlorines per biphenyl is a measurement for the weathering of PCBs throughout the sewer and wastewater processes. This was calculated based on the total mass (g) of each PCB homolog, the number of chlorines and the molecular weight (g/mol) for each PCB homolog. The number of chlorine per biphenyl ring for each day then was calculated by using the calculated mole of chlorine number for 10 PCB homologs divided by the their total moles. Finally, the number of chlorine per biphenyl ring for each day was calculated by adding the average number of chlorine/biphenyl for each year divided by the WWTP operation days (365 days) (Appendix A Eqs. (S3) and (S4)).

1.6. Estimated aerobic and anaerobic biodegradation potential

Two biological processes can be responsible for transformation of PCBs: aerobic oxidation and anaerobic organohalide respiration

(Abramowicz, 1995; Nyholm et al., 2010). The total mass (g) of PCB homologs with four or less chlorine atoms (*i.e.*, mono-, di-, tri- and tetra-chlorobiphenyls) were firstly calculated by their mass concentration (g/L) from both effluents for 365 days and their corresponding flowrates (L/day). Aerobic biodegradation potential of PCBs was calculated by total mass (g) of PCB homologs divided by the annual total mass (g) of all PCB homologs. The calculation of the anaerobic biodegradation potential was calculated by adding the mass of the homologs with five to ten chlorine atoms and subtracting the mass (g) of PCB congeners, where chlorine atoms were solely placed in *ortho* positions and divide by the total mass of (g) of PCBs. The congeners with *ortho* chlorinated positions were subtracted since multiple studies have shown that organohalide respiring bacteria cannot utilize these chlorines as electron acceptors (Wu et al., 2002) (Appendix A Eqs. (S5) and (S6)).

1.7. DNA extraction and molecular analysis

Deoxyribonucleic acid (DNA) was extracted from 10 locations at the WWTP: primary sedimentation tank (PST), nitri/denitrification reactor (NDR), primary effluent (PE), anaerobic digestion reactor (Digested biosolids: DB), final product biosolids (FPB), nitri/denitrification sedimentation (NDS), secondary reactor (SR), secondary sedimentation tank (SST), centrifuge pre-dewatering (liquid and biosolid: LB), east primary influent (EPI). DNA was extracted using the MoBIO PowerSoil DNA Isolation Kits (Qiagen Inc., Germantown, USA) (Krzmarzick et al., 2012). The extracted DNA from the nine samples was analyzed on the nano-drop (Thermo Fisher Scientific, Waltham, USA) to measure the concentration of the DNA and the purity. Detectable concentrations of DNA were found in six of 10 samples (*i.e.*, PST, PE, NDS, SR, SS and EPI). After that, polymerase chain reaction (PCR) was performed by using DreamTaq Green PCR Master Mix (Thermo Fisher Scientific, Waltham, USA) and primers set 348F/884R specific for the 16S rRNA gene targeting putative PCB-dechlorinating *Chloroflexi* (Chun et al., 2013; Fagervold et al., 2005; Kjellerup et al., 2012). Each PCR reaction was conducted by using 12.5 μL of DreamTaq Green PCR Master Mix, 8.5 μL nuclease-free water, 1ul forward and reverse primers, and 2 μL of sample DNA. Thermocycler conditions were as follows: (1) 1 cycle of 2 min at 95°C; (2) 45 sec at 95°C; (3) annealing for 45 sec at 58°C, (4) a last cycle of 60 sec at 72°C, (5) 40 cycles of step 2–4; and (5) a final extension of 30 sec at 72°C. The PCR products were evaluated *via* gel electrophoresis and bands were compared to the amplicon length for the positive control (536 bp) and a molecular ladder (Genprice Inc., San Jose, USA) (Appendix A Fig. S1). Four PCR-amplified 16S rRNA gene products of these 10 samples showed the expected size (536 bp) (*i.e.*, Appendix A Fig. S1. NDR, DB, NDS and LB) from 10 locations of the WWTP when the genomic DNA was targeted with primers 348F and 884R.

2. Results

2.1. Annual PCB discharge

After the production of PCBs was banned in the 1970s in USA, the amount of PCBs present in the environment was expected to decrease, since existing PCB sources would be eliminated (Zhang et al., 2004). The results from this study showed that current sources still exist and are discharged to a WWTP in the wastewater influent and subsequently discharged in the effluent from a WWTP. The continuous effluent contributed the major PCB discharge (80.7%–94.7%) of total PCB discharge from both intermittent effluent and continuous effluent over the five-year period (Fig. 2). A significantly ($p < 0.01$) larger contribution (1821.4 g) came from the continuous effluent discharge compared to the stormwater overflow (260.3 g) thus showing that the sources of PCBs in the effluent originate from the wastewater and not from the stormwater contribution in the combined sewer system (Table 1). A similar result of total PCBs discharge (1.65 g PCB/day) during dry weather was observed from a WWTP located in Philadelphia North East discharging to the Delaware River (Hansler et al., 1998). This PCBs discharge was estimated to 602.3 g/year, which was comparable to the results from this study.

The distribution of PCB homologs for the two types of effluent showed that tri-, tetra-, penta-, hexa-, and hepta-chlorinated PCBs were the most abundant congeners in the stormwater overflow with penta- and hexa-chlorinated congeners having the highest abundance (Fig. 3). The contributions from these five homologs ranged on average for 49.5%–58.1% of the total annual PCBs (Table 2). For the continuous effluent, di-, tri-, tetra-, penta-, and hexa-chlorinated congeners were the most abundant (Fig. 3). Here, tetra- and penta-chlorinated congeners accounted for annual mass discharge ranging from 47.8%–51.1% of the total PCBs over the five-year period.

2.2. Toxicity equivalent (TEQ) for 12 dioxin-like PCBs

The toxicity of PCBs depends on the number of chlorines atoms, but also the positions of the chlorines on the biphenyl rings (Barbalace, 2003). Non-*ortho* PCB are generally considered to be "dioxin-like", and they are more toxic than the rest of the congeners, since the chlorine atoms will line up in a single plane as coplanar, when there is one or no chlorines in the *ortho* position (Van den Berg et al., 2006). The arithmetic mean of calculated TEQs over five-year periods for four non-*ortho* and eight mono-*ortho* dioxin-like PCBs in the two effluents were summarized in Table 3. The maximum contaminant level in water for TCDD established by US EPA is 0.03 ng/L (Rodriguez et al., 2008). None of the mean value for the 12 dioxin-like PCBs observed in the discharged effluent over the five-year period exceeded the health standard of 30 pg TCDD/L. In addition, the TEQ concentration of the total 12 dioxin-like PCB congeners were 1.65 pg TEQ/L(intermittent) and 0.261 pg TEQ/L (continuous), respectively.

2.3. PCB organohalide respiration patterns for discharged effluents

The average number of chlorine/biphenyl for 209 PCB congeners from the continuous effluent was 3.83–4.03 over a five-year study period (Table 4), which was 19% lower than that of the intermittent effluent (4.80 to 4.93). This difference of an average number of chlorine/biphenyl indicated that a PCB organohalide respiration process might occur during normal wastewater treatment. This was mainly attributed by the mass of tetra- and

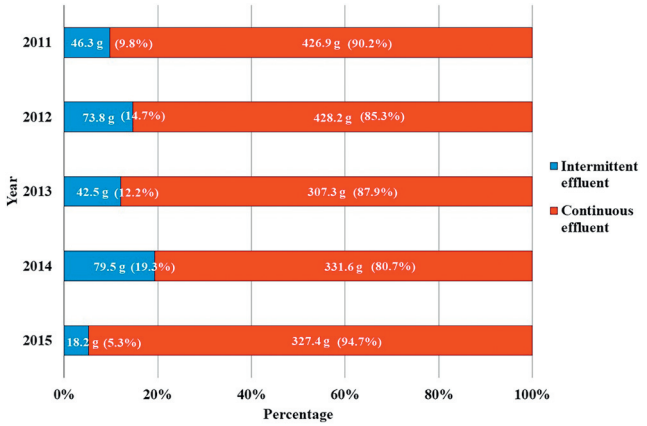

Fig. 2 – Comparison of estimated annual PCBs discharged from the intermittent and continuous effluents during 2011 to 2015.

penta-chlorinated congeners that accounted for 47.8%–57.8% of the total annual PCBs over five years for the continuous effluent (Table 2). In contrast, penta- and hexa-chlorinated congeners had an increased abundance in the intermittent effluent with 49.5%–58.2% of total annual PCBs from 2011 to 2015. Higher chlorinated PCB congeners can often be biotransformed to lower chlorinated PCBs by anaerobic organohalide respiration processes thus explaining the difference in chlorine per biphenyl values.

2.4. Aerobic and anaerobic biodegradation potential

Aerobic degradation of PCBs can occur for congeners with four or less chlorines per biphenyl. More specifically, of the total annual PCB mass discharging to the river from the intermittent effluent only 27.8% can theoretically be aerobically biodegraded (Fig. 4a), where tri- (4.60 g) and tetra- (7.85 g) chlorinated congeners had the highest abundance. In contrast, the potential for aerobic biodegradation for the continuous effluent was more pronounced and 58.3% (Fig. 4b) could potentially be mineralized compared to the intermittent effluent. Similarly, tri- (73.7 g) and tetra- (97.2 g) chlorinated congeners contributed the most toward the total mass. On the other hand, intermittent effluent had a high aerobically biodegraded PCBs accounting for 72.2% of the total annual PCB discharges over the five-year period. For the intermittent effluent, penta- (13.1 g), hexa- (14.5 g), and hepta- (7.9 g) chlorinated congeners contributed the most with a total of 94.1% of the total PCB mass toward the anaerobic biodegradation potential. In comparison, the continuous effluent was dominated by penta- (89.8 g), hexa- (43.9 g) and hepta- (14.6 g) chlorinated congeners that altogether made up 97.5% of the anaerobic biodegradation potential. More specifically, both the intermittent and continuous effluents had high anaerobic PCB degradation potentials ($R_{anaerobic}$ = 98.7% for intermittent effluent, $R_{anaerobic}$ = 93.7% for continuous effluent) considering all the potential PCB congeners that would be anaerobically degraded to *ortho*-PCB congeners (Appendix A Fig. S2).

Table 1 – Summary of total PCB discharges calculated based on the annual PCB loading model.

	Intermittent effluent	Continuous effluent
Year	Total mass (g)	Total mass (g)
2011	46.3	426.9
2012	73.8	428.2
2013	42.5	307.3
2014	79.5	331.6
2015	18.2	327.4
Average	52.1 (±25.0)	364.3 (±58.3)
Total	260.3	1821.4

2.5. DNA extraction and PCR amplification

The results from the PCR analysis showed that putative PCB-dechlorinating *Chloroflexi* such as *Dehalococcoides* species might exist in the wastewater samples and play a role in dechlorination of PCBs. To identify the present bacterial phylotypes, it would be necessary to sequence the PCR amplicons to compare the sequence similarity with known PCB dechlorinating bacteria. This is beyond the scope of this study. In support of this finding, a study published by Smith et al. (2015) showed the presence of putative PCB-dechlorinating *Chloroflexi* in wastewater treatment plants. This study reported that *Dehalococcoides mccartyi* was found in eight WWTP anaerobic digesters in the central and northeastern Oklahoma region and averaged 3.3×10^7 gene copies per gram biosolids. Despite the abundance of *Dehalococcoides mccartyi* were relatively small that only represents 0.02% of the total bacterial community. The discovery of *Dehalococcoides mccartyi* existing in the anaerobic digesters is important that indicated the organohalide respiration of PCBs might occur in WWTPs. The differences of their populations among different digesters were less than one order of magnitude revealing a similar capability of PCB organohalide respiration in anaerobic digesters in the WWTP at central and northeastern Oklahoma region.

3. Discussion

The evaluation of the PCB abundance and homolog analysis in the two effluents from the large municipal WWTP showed the presence of high levels of PCBs. In the continuous effluent, the average number of chlorines per biphenyl over the five-year period was 3.94 ± 0.28 chlorines per biphenyl compared to 4.87 ± 0.30 chlorines per biphenyl for intermittent effluent. Furthermore, the number of chlorines per biphenyl for west and east primary influents were also calculated based on two sampling events at November 10, 2010 and June 14, 2017 respectively (Appendix A Table S1). The average number of chlorines per biphenyl was 4.42 and 5.03 for the west primary influent and 4.41 and 4.4 for the east primary influent. These two types of primary influents were directly discharged into the WWTP without any physical and biological treatment resulting similar characteristics of wastewater to the intermittent effluent that discharged through the stormwater overflow during wet weathers after the primary treatment process. Therefore, this difference indicated that stormwater can quickly travel through the sewers thereby showing no PCB organohalide respiration.

The anaerobic zones in the tanks such as denitrification reactors are not completely mixed. Therefore, it might have enough time for organohalide respiration of PCBs during the

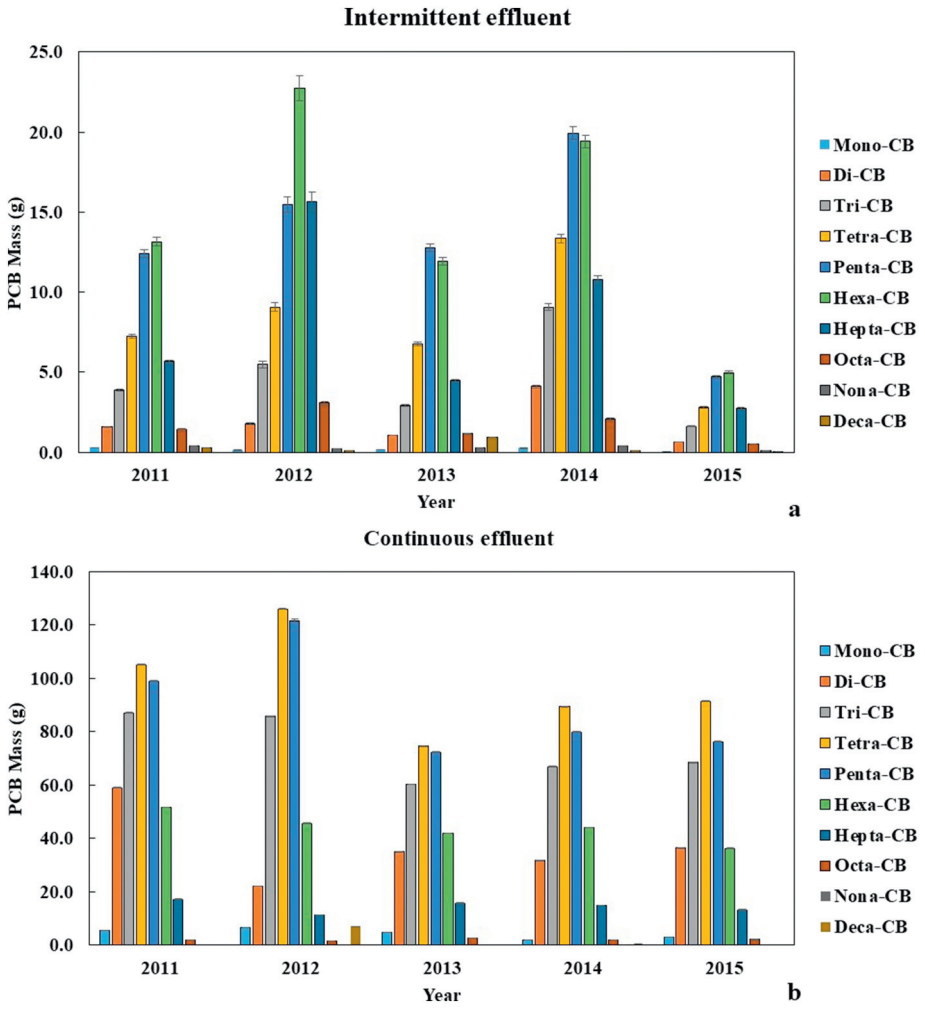

Fig. 3 – PCB homolog distribution from (a) intermittent effluent and (b) continuous effluent.

continuous treatment. In addition, the rate and extent of PCB respiration depended on the number and positions of chlorines assuming the reductive conditions are present in the anaerobic environment (Grimm et al., 2015). Moreover, a relatively high Cl/biphenyl number of primary influents and intermittent effluent (4.42, 4.41, 5.03, 4.40 and 4.87) indicated that the pattern of the chlorine atoms distribution were comparable to Aroclor 1248 (Cl/biphenyl = 3.97) and Aroclor 1254 (Cl/biphenyl = 5.15).

In the continuous effluent, the mol% distribution of PCB homologs showed that di-, tri-, tetra-, and penta-chlorinated PCBs were the most abundant homologs. The contributions from these four homologs accounted for 13.22%, 22.90%, 26.64%, and 9.74% of the total annual PCBs (Appendix A Table S2). Similar results were reported by Balasubramani et al. (2014), who showed that the concentrations of PCB homologs measured at the16 different effluents from municipal and industrial wastewater treatment plants in the Houston area, TX, were dominated by lower chlorinated congeners (*i.e.*, mon-, di-, tri-, tetra-, penta-, and hexa-chlorinated PCBs). On the contrary, tri- (11.24%), tetra- (16.91%), penta- (25.18%), and hexa-chlorinated PCBs (25.20%) were the most abundant in the intermittent effluent sharing a similar mol% distribution of PCB homologs for the west and east primary influents (Appendix A Table S2). These PCB homologs distribution are also comparable to Aroclor 1254 which is a commonly manmade PCB mixture with relatively high chlorine content (54% of chlorine by weight). However, the PCB homologs distribution of the continuous effluent was similar to Aroclor 1242 that contain 42% of chlorine by weight (Appendix A Table S3). This anaerobic degradation is rarely reported at such low concentrations of PCBs (1.07×10^{-5} to 5.19×10^{-5} mg/L for intermittent effluent; 7.90×10^{-7} to 1.14×10^{-6} mg/L for continuous effluent) without a specific inoculation of organohalide respiring bacteria. Nevertheless, this study reported the first observation of PCB organohalide respiration at low PCB level in a wastewater system. This biotransformation from highly chlorinated PCB homologs to low-chlorine content PCBs was consistent with findings from other studies (Kaya et al., 2018; Kuipers et al., 2003). Kuipers et al. (2003) investigated the anaerobic dechlorination of weathered Aroclor 1260 in soil collected from Resolution Island, Canada. hexa- and heptachlorobiphenyls were found to be the major homolog groups. PCB-47 and PCB-51 were formed as the dechlorination products after inoculation with anaerobic sediments. The average number of chlorine/biphenyl was reduced from 6.6 to 5.1. Kaya et al. (2018) determined the dechlorination of weathered Aroclor 1254 from three different sites (Grasse River, Fox River, and Baltimore Harbor) which

mainly contained penta-, hexa- and hepta-chlorinated PCB congeners. The results of the mesocosms indicated that the major dechlorination products of these high-chlorine content PCB congeners were tetra- and tri-chlorinated congeners.

More specifically, the occurrence of PCB organohalide respiration could be further supported by comparison of the level (mol %) of *ortho*-PCBs (*i.e.*, PCB-1, PCB-4, PCB-10, PCB-19, PCB-54) in continuous and intermittent effluents (Table 5). The PCB-dechlorinating bacteria in the environment predominantly dechlorinate chlorines located at *meta*- and *para*-positions (Fagervold et al., 2005), and only few studies have reported the reductive dechlorination of *ortho*-chlorines (Bedard et al., 2007; May et al., 2006; Sowers and May, 2013). As a result, this dechlorination pathway resulted in the formation of the *ortho* dechlorination PCBs. A significantly ($p < 0.001$, Appendix A Table S4) higher mol% of *ortho*-PCBs came from the continuous effluent which is ranging from 6.03% to 9.89% over five years. However, the mol% of *ortho*-PCBs in intermittent effluent was only 1.19% to 2.04%. This is mainly the fact that the anaerobic organohalide respiring bacteria can attack highly chlorinated PCB congeners through removal of *meta*- and *para*-chlorines resulting a higher mol% of *ortho*-PCBs in the continuous effluent (Lombard et al., 2014). In addition, a relatively high percentage of *ortho*-PCBs in the continuous effluent could imply that the PCB organohalide respiration has been occurred in the sewage collection system before transferring to the WWTP (Rodenburg et al., 2010). Sewers can serve as a "pretreatment" zone that contain *Dehalococcoides* to support dechlorination of PCBs (Fagervold et al., 2007; Rodenburg et al., 2010). These differences indicated that a PCB organohalide respiration could potentially occur during the sewage collection system and wastewater treatment process (continuous effluent) thereby resulting a change of PCB homologs distribution. More importantly, Appendix A Table S5 indicates PCB-4 and PCB-19 were the major contribution that could be potentially organohalide respiring products among these 5 *ortho*-PCBs for both continuous and intermittent effluents. The mol% of PCB-4 and PCB-19 in continuous effluent were ranging from 3.61% to 6.08% and 0.70% to 1.51% respectively over five years. On the other hand, the mol% of PCB-4 and PCB-19 in intermittent effluent only counted for 0.70% to 1.51% and 0.04% to 0.15% respectively over five years. As a result, highly chlorinated PCB congeners can be transformed to five different PCB congeners solely having *ortho* chlorine atoms (*i.e.*, 2-Monochlorobiphenyl, 2,6-Dichlorobiphenyl, 2,2′-Dichlorobiphenyl, 2,2′,6′-Trichlorobiphenyl, 2,2′,6,6′-tetrachlorobiphenyl). More interestingly, PCB-11 (3,3′-dichlorobiphenyl) and their possible precursors were also detected from both effluents in this study. The PCB-11 is inadvertently produced during the manufacture of diarylide yellow pigments (Herbst and Hunger, 2006). Use of these yellow pigments in consumer goods such as newspapers, and cardboard boxes can result in PCB-11 entering WWTPs and combined sewer overflows (Rodenburg et al., 2010). Three non-*ortho* PCB congeners: PCB-77, PCB-126, and PCB-169 (*i.e.*, 3,3′,4,4′-tetrachlorobiphenyl, 3,3′,4,4′,5-pentachlorobiphenyl, and 3,3′,4,4′,5,5′-hexachlorobiphenyl) could be the precursors of PCB-11, because removal of *ortho* chlorines from a PCB molecule is unusual in a natural environment (Bedard, 2004). A higher level of PCB-11 ($p < 0.001$) was observed from the continuous effluent (ranging from 1.8%–5.8% over five years) compared to the intermittent effluent (1.0%–1.8%). A low level of

Table 2 – Five-year summary of PCB homolog discharges from intermittent and continuous effluents.

Year	Mono-CB (g)	Di-CB (g)	Tri-CB (g)	Tetra-CB (g)	Penta-CB (g)	Hexa-CB (g)	Hepta-CB (g)	Octa-CB (g)	Nona-CB (g)	Deca-CB (g)
Intermittent effluent										
2011	0.28 (± 0.01)	1.58 (± 0.03)	3.89 (± 0.08)	7.26 (± 0.14)	12.44 (± 0.24)	13.14 (± 0.28)	5.66 (± 0.11)	1.43 (± 0.03)	0.39 (± 0.01)	0.27 (± 0.01)
2012	0.13 (± 0.005)	1.78 (± 0.06)	5.48 (± 0.19)	9.07 (± 0.27)	15.48 (± 0.46)	22.74 (± 0.76)	15.67 (± 0.56)	3.08 (± 0.11)	0.24 (± 0.01)	0.10 (± 0.002)
2013	0.18 (± 0.003)	1.05 (± 0.02)	2.94 (± 0.06)	6.78 (± 0.15)	12.77 (± 0.30)	11.94 (± 0.27)	4.46 (± 0.09)	1.16 (± 0.02)	0.26 (± 0.01)	0.94 (± 0.03)
2014	0.26 (± 0.01)	4.11 (± 0.2)	9.07 (± 0.22)	13.35 (± 0.27)	19.93 (± 0.39)	19.43 (± 0.38)	10.79 (± 0.21)	2.09 (± 0.04)	0.38 (± 0.01)	0.09 (± 0.002)
2015	0.06 (± 0.001)	0.66 (± 0.01)	1.62 (± 0.03)	2.78 (± 0.05)	4.70 (± 0.09)	5.00 (± 0.10)	2.75 (± 0.06)	0.53 (± 0.01)	0.09 (± 0.002)	0.03 (± 0.001)
Continuous effluent										
2011	5.68 (± 0.02)	58.99 (± 0.13)	87.09 (± 0.14)	105.06 (± 0.16)	98.99 (± 0.10)	51.62 (± 0.036)	17.22 (± 0.005)	2.01 (± 0.001)	0.10 (± 0.001)	0.09 (± 0.001)
2012	6.71 (± 0.01)	22.10 (± 0.04)	85.82 (± 0.13)	125.93 (± 0.19)	121.67 (± 0.29)	45.70 (± 0.09)	11.43 (± 0.03)	1.64 (± 0.01)	0.18 (± 0.001)	7.01 (± 0.09)
2013	4.82 (± 0.01)	34.97 (± 0.02)	60.31 (± 0.04)	74.52 (± 0.05)	72.22 (± 0.08)	41.90 (± 0.06)	15.79 (± 0.03)	2.53 (± 0.01)	0.04 (± 0.003)	0.20 (± 0.001)
2014	2.07 (± 0.01)	31.81 (± 0.02)	66.85 (± 0.04)	89.30 (± 0.08)	79.86 (± 0.09)	44.16 (± 0.06)	15.04 (± 0.02)	2.08 (± 0.01)	0.08 (± 0.001)	0.35 (± 0.001)
2015	2.95 (± 0.004)	36.50 (± 0.03)	68.51 (± 0.06)	91.30 (± 0.09)	76.21 (± 0.07)	36.32 (± 0.04)	13.28 (± 0.02)	2.15 (± 0.005)	0.06 (± 0.0004)	0.17 (± 0.001)

Table 3 – Average TEQs of 12 dioxin-like PCBs for the intermittent effluent (33 observations) and for the continuous effluent (56 observations) over the five-year study period.

	Intermittent effluent										
	TEQ PCB-77 (pg-TEQ/l)	TEQ PCB-81 (pg-TEQ/l)	TEQ PCB-105 (pg-TEQ/l)	TEQ PCB-114 (pg-TEQ/l)	TEQ PCB-118 (pg-TEQ/l)	TEQ PCB-123 (pg-TEQ/l)	TEQ PCB-126 (pg-TEQ/l)	TEQ PCB-156 (pg-TEQ/l)	TEQ PCB-157 (pg-TEQ/l)	TEQ PCB-167 (pg-TEQ/l)	TEQ PCB-169 (pg-TEQ/l)	TEQ PCB-189 (pg-TEQ/l)	Total TEQs (pg-TEQ/l)
Mean	4.96×10^{-3}	9.67×10^{-3}	2.02×10^{-2}	5.10×10^{-4}	1.39×10^{-2}	2.72×10^{-3}	1.09	2.87×10^{-3}	4.18×10^{-4}	1.40×10^{-3}	5.07×10^{-1}	7.54×10^{-4}	1.65
STD	1.68×10^{-2}	4.30×10^{-2}	2.12×10^{-2}	1.17×10^{-3}	1.37×10^{-2}	9.88×10^{-3}	4.06	3.39×10^{-3}	1.88×10^{-3}	2.33×10^{-3}	1.87	1.81×10^{-3}	4.61
Max	9.63×10^{-2}	2.41×10^{-1}	8.57×10^{-2}	6.63×10^{-3}	5.70×10^{-2}	4.74×10^{-2}	23.1	1.45×10^{-2}	1.07×10^{-2}	1.22×10^{-2}	10.2	8.67×10^{-3}	23.5
Min	1.93×10^{-4}	3.11×10^{-4}	3.40×10^{-4}	2.84×10^{-5}	4.86×10^{-5}	1.92×10^{-5}	1.50×10^{-1}	4.22×10^{-5}	4.22×10^{-5}	2.88×10^{-5}	6.50×10^{-2}	3.78×10^{-5}	2.28×10^{-1}
	Continuous effluent												
	TEQ PCB-77 (pg-TEQ/l)	TEQ PCB-81 (pg-TEQ/l)	TEQ PCB-105 (pg-TEQ/l)	TEQ PCB-114 (pg-TEQ/l)	TEQ PCB-118 (pg-TEQ/l)	TEQ PCB-123 (pg-TEQ/l)	TEQ PCB-126 (pg-TEQ/l)	TEQ PCB-156 (pg-TEQ/l)	TEQ PCB-157 (pg-TEQ/l)	TEQ PCB-167 (pg-TEQ/l)	TEQ PCB-169 (pg-TEQ/l)	TEQ PCB-189 (pg-TEQ/l)	Total TEQs (pg-TEQ/l)
Mean	2.03×10^{-4}	3.19×10^{-4}	1.19×10^{-3}	3.92×10^{-5}	8.23×10^{-4}	2.34×10^{-5}	1.93×10^{-1}	1.29×10^{-4}	4.53×10^{-5}	4.42×10^{-5}	6.57×10^{-2}	4.13×10^{-5}	2.61×10^{-1}
STD	2.18×10^{-4}	6.05×10^{-5}	2.49×10^{-3}	5.10×10^{-5}	1.73×10^{-3}	3.06×10^{-5}	2.24×10^{-16}	3.20×10^{-4}	2.22×10^{-5}	9.47×10^{-5}	4.40×10^{-3}	2.86×10^{-5}	6.42×10^{-3}
Max	1.77×10^{-3}	7.59×10^{-4}	1.88×10^{-2}	3.21×10^{-4}	1.31×10^{-2}	2.46×10^{-4}	1.93×10^{-1}	2.29×10^{-3}	2.06×10^{-4}	7.23×10^{-4}	9.63×10^{-2}	2.49×10^{-4}	2.95×10^{-1}
Min	6.22×10^{-5}	3.11×10^{-4}	3.75×10^{-4}	1.38×10^{-5}	1.43×10^{-4}	1.81×10^{-5}	1.93×10^{-1}	3.13×10^{-5}	3.00×10^{-5}	8.04×10^{-6}	6.50×10^{-2}	2.27×10^{-5}	2.59×10^{-1}

the precursors of PCB-11 (PCB-77, 126, 169) was found in the intermittent effluent ranging from 0.06%–1.16%. In addition, the level of precursors of PCB-11 in the continuous effluent ranged from 0.03%–0.11% thus the levels of PCB-11 in the continuous and intermittent effluents combined were higher than that of the precursors to PCB-11. These levels of precursors were not sufficient to generate high the observed levels of PCB-11, which could imply that PCB-11 originated from other sources in the wastewater such as dyes and pigments and not entirely from organohalide respiration.

Moreover, another strong evidence indicating this PCB anaerobic organohalide respiration is that similar results of mol% of *ortho*-PCBs in west and east primary influents (0.90% and 1.98%) as compared to that of intermittent effluent were also observed as shown in Appendix A Table S4. This could indicate that specific types of bacteria such as putative PCB-dechlorinating *Chloroflexi* could be enriched, when the microorganisms are present in an anoxic/anaerobic zone thus anaerobically dechlorinate PCBs in these parts of the plant (Bedard, 2008). Therefore, it is important to identify the proportion of PCBs that can be anaerobically and aerobically biodegraded based on the annual PCB discharge. However, it should be noted that the mol% of *ortho*-PCBs from two influence was calculated based on single sampling event instead of randomly taking samples over an entire year. Accumulation of *ortho*-PCBs indicating PCB dechlorination activity has also been confirmed by other studies although most of the studies of PCBs organohalide respiration to date have been conducted in microcosms (Mattes et al., 2017; Fagervold et al., 2011; Kjellerup et al., 2014; Payne et al., 2011; Wang and He, 2013). In this study, an increase in *ortho*-PCBs (*i.e.*, PCB-1, PCB-4, PCB-10, PCB-19, PCB-54) appears in both continuous and intermittent effluents which are correspond with a low mol% of *non-ortho*-PCBs. Moreover, a higher mol% of ortho-PCBs (6.03% to 9.89%) from the continuous effluent indicated a potential organohalide respiration of PCBs. Similar results were observed in a study of PCB dechlorination hotspots in sediments from a contaminated wastewater lagoon (Mattes et al., 2017). The results indicated that mol% of *ortho*-chlorinated was 0.9% to 2.2% in 14 out of 27 sediment samples and the remaining 13 samples had mol% values ranging from 3% to 31%. In addition, some bioaugmentation studies of indigenous PCB dechlorinating activity in sediment microcosms that shown accumulation of *ortho*-PCBs were also reported (Fagervold et al., 2011; Kjellerup et al., 2014; Payne et al., 2011; Wang and He, 2013).

The results from this study indicated that there was a potential for aerobic and anaerobic biodegradation of PCBs present in the effluent. In the intermittent effluent from stormwater overflow, 27.8% of total PCBs was estimated to be susceptible to aerobic biodegradation, while 58.3% could undergo aerobic degradation in the continuous effluent. This difference could be explained by the intermittent effluent can be quickly discharged through the sewers thereby indicating no potentials for organohalide respiration of PCBs unlike the continuous effluent with a longer residence time. The presence of organohalide respiration bacteria, therefore, can respire with the high chlorinated PCBs entering the WWTP (>4 chlorines/biphenyl) (De et al., 2006; Field and Sierra-Alvarez, 2008) thereby transforming the higher chlorinated PCBs to lower chlorinated PCBs. As a result, the PCB distribution pattern in the continuous effluent had

Table 4 – Average number of chlorine/biphenyl for total PCB discharges from intermittent and continuous effluents.

	Intermittent effluent	Continuous effluent
Year	Cl/Biphenyl	Cl/Biphenyl
2011	4.88 (± 0.14)	3.83 (± 0.30)
2012	4.88 (±0.54)	4.02 (±0.39)
2013	4.93 (±0.18)	3.94 (±0.25)
2014	4.87 (±0.22)	4.03 (±0.15)
2015	4.80 (±0.37)	3.91 (±0.15)
Average	4.87 (±0.30)	3.94 (±0.28)

increased potential for aerobic PCB degradation as compared to that in the intermittent effluent, since this effluent by-passed all the biological processes in the WWTP. The result of aerobic biodegradation of the lower chlorinated congeners was complete mineralization to carbon dioxide *via* the intermediate products *cis*-dyhydriol and 2,3-dihydroxy intermediate with low toxicity

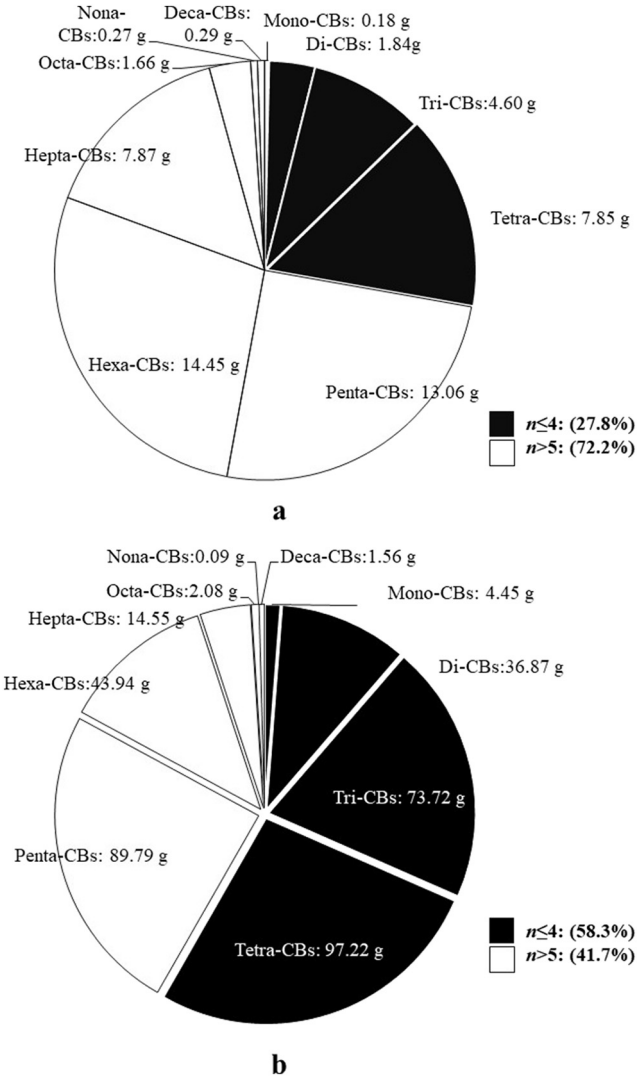

Fig. 4 – The potential for anaerobic organohalide respiration ($n > 5$) and aerobic degradation ($n \leq 4$) for PCBs (a) intermittent effluent and (b) continuous effluent.

Table 5 – Mol% of the *ortho*-PCBs for continuous effluent and intermittent effluent.

Year	Continuous effluent	Intermittent effluent
2011	7.44%	1.87%
2012	6.03%	1.19%
2013	9.89%	1.81%
2014	7.80%	0.51%
2015	8.72%	2.04%

(Tu et al., 2011). This process was performed by aerobic bacteria that possess the genes that encode for biphenyl-2,3-dioxygenase (bphA gene) and a dehydrogenase (bphB) (Tu et al., 2011).

On the other hand, both effluents had high PCB anaerobic degradation potential (organohalide respiration) if all potential PCB congeners that would be anaerobically degraded to the *ortho*-PCB congeners are considered. This was mainly attributed that tetra- through hexa-chlorinated congeners that partially dechlorinated during the wastewater treatment processes that occur under normal flow conditions. Therefore the mass of tri- though penta-chlorinated congeners were higher in continuous effluent. Furthermore, the TEQ concentrations of the dioxin-like PCBs from this effluent were reduced by 1–2 order of magnitude compared to the intermittent effluent. This change in toxicity indicated a reduction of dioxin-like PCBs in the WWTP with stabilized anaerobic processes such as denitrification. Some studies have also shown that PCBs organohalide respiration occurred in urban wastewater treatment systems (Rodenburg et al., 2012; Rosińska and Karwowska, 2017). Rosińska and Karwowska (2017) suggested that PCB 169 was dominant in the wastewater sludge with concentrations ranging from 8.2 to 23.4 μg/kg. However, a significant degradation (77.8% to 80.5%) was observed for this dioxin-like PCB congener after the organohalide respiration process during anaerobic digestion. The PCB organohalide respiration signal from the continuous effluent was also supported by results from molecular analysis of nine sampling locations from the metropolitan WWTP (Appendix A Table S6). PCBs have been considered as legacy pollutants thus potential sources could enter stormwater and sewage system (in case of combined sanitary and stormwater systems) thereby continuously release PCBs into the sewers, the WWTP and the receiving river. Therefore, a long-term plan for upstream monitoring of PCBs in the sewer system and thus PCB source control would be a potential solution to overcome the reduction in PCB discharge required. Such a long-term plan could also include alternatives to reduce the possible PCB sources in the WWTP influent such as *in-situ* bioremediation at historical PCB contaminated sites and promotion of organohalide respiration of PCBs in the sewer pipe system/network (Rodenburg et al., 2012).

4. Conclusions

Annual PCB discharges were determined from intermittent and continuous effluent in a WWTP by using five years of data from 2011 to 2015. The continuous effluent contributed a significantly larger mass of PCBs with a total mass (1821.4 g) for five years, which showed that this effluent contributed the majority of PCBs into the nearby river. A 19% difference in the

average number of chlorine/biphenyl for the 209 PCB congeners for intermittent *versus* continuous effluent indicated a potential for organohalide respiration of PCBs during normal wastewater treatment. Also, a high mass distribution of tri-, tetra- and pentachlorinated PCBs accounted for approximately 75% of the anaerobically biodegraded PCBs further indicated the possibility of PCB organohalide respiration occurrence for the continuous effluent. Enhanced biodegradation of PCBs *via* bioaugmentation could be an alternative for removal PCBs. A combination of anaerobic organohalide respiration followed by aerobic biodegradation would be the best solution to achieve complete removal of PCBs by final biomineralization. This study is contributing to a continued evaluation of the PCB annual discharge and biodegradation potential in the wastewater effluents thus more recommendations will be gained from subsequent studies.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jes.2018.06.007.

REFERENCES

Abbas, S.H., Ismail, I.M., Mostafa, T.M., Sulaymon, A.H., 2014. Biosorption of heavy metals: a review. J. Chem. Sci. Technol. 3, 74–102.

Abramowicz, D.A., 1995. Aerobic and anaerobic PCB biodegradation in the environment. Environ. Health Perspect. 103, 97.

Ahmed, M., Focht, D., 1973. Degradation of polychlorinated biphenyls by two species of *Achromobacter*. Can. J. Microbiol. 19, 47–52.

Baars, A., Bakker, M., Baumann, R., Boon, P., Freijer, J., Hoogenboom, L., et al., 2004. Dioxins, dioxin-like PCBs and non-dioxin-like PCBs in foodstuffs: occurrence and dietary intake in the Netherlands. Toxicol. Lett. 151, 51–61.

Balasubramani, A., Howell, N.L., Rifai, H.S., 2014. Polychlorinated biphenyls (PCBs) in industrial and municipal effluents: concentrations, congener profiles, and partitioning onto particulates and organic carbon. Sci. Total Environ. 473, 702–713.

Barbalace, R.C., 2003. The chemistry of polychlorinated biphenyls. https://environmentalchemistry.com.

Bedard, D.L., 2004. Polychlorinated Biphenyls in Aquatic Sediments: Environmental Fate and Outlook for Biological Treatment, Dehalogenation. Springer, pp. 443–465.

Bedard, D.L., 2008. A case study for microbial biodegradation: anaerobic bacterial reductive dechlorination of polychlorinated biphenyls—from sediment to defined medium. Annu. Rev. Microbiol. 62, 253–270.

Bedard, D.L., Ritalahti, K.M., Löffler, F.E., 2007. The *Dehalococcoides* population in sediment-free mixed cultures metabolically dechlorinates the commercial polychlorinated biphenyl mixture Aroclor 1260. Appl. Environ. Microbiol. 73, 2513–2521.

Chang, Y.-C., Takada, K., Choi, D., Toyama, T., Sawada, K., Kikuchi, S., 2013. Isolation of biphenyl and polychlorinated biphenyl-degrading bacteria and their degradation pathway. Appl. Biochem. Biotechnol. 170, 381–398.

Cheng, H., Hu, Y., 2010. Curbing dioxin emissions from municipal solid waste incineration in China: re-thinking about management policies and practices. Environ. Pollut. 158, 2809–2814.

Chun, C.L., Payne, R.B., Sowers, K.R., May, H.D., 2013. Electrical stimulation of microbial PCB degradation in sediment. Water Res. 47, 141–152.

Correa, P.A., Lin, L., Just, C.L., Hu, D., Hornbuckle, K.C., Schnoor, J.L., et al., 2010. The effects of individual PCB congeners on the soil bacterial community structure and the abundance of biphenyl dioxygenase genes. Environ. Int. 36, 901–906.

De, J., Ramaiah, N., Sarkar, A., 2006. Aerobic degradation of highly chlorinated polychlorobiphenyls by a marine bacterium, *Pseudomonas* CH07. World J. Microbiol. Biotechnol. 22, 1321–1327.

Deblonde, T., Cossu-Leguille, C., Hartemann, P., 2011. Emerging pollutants in wastewater: A review of the literature. Int. J. Hyg. Environ. Health 214, 442–448.

Dong, B., Chen, H.Y., Yang, Y., He, Q.B., Dai, X.H., 2015. Biodegradation of polychlorinated biphenyls using a moving-bed biofilm reactor. Clean Soil Air Water 43, 1078–1083.

EPA, 2008. Method 1668B. chlorinated biphenyl congeners in water, soil, sediment, biosolids, and tissue by HRGC/HRMS. Test Methods for Evaluating Solid Waste (Washington, DC).

Fagervold, S.K., May, H.D., Sowers, K.R., 2007. Microbial reductive dechlorination of Aroclor 1260 in Baltimore Harbor sediment microcosms is catalyzed by three phylotypes within the phylum *Chloroflexi*. Appl. Environ. Microbiol. 73, 3009–3018.

Fagervold, S.K., Watts, J.E., May, H.D., Sowers, K.R., 2005. Sequential reductive dechlorination of meta-chlorinated polychlorinated biphenyl congeners in sediment microcosms by two different *Chloroflexi* phylotypes. Appl. Environ. Microbiol. 71, 8085–8090.

Fagervold, S.K., Watts, J.E., May, H.D., Sowers, K.R., 2011. Effects of bioaugmentation on indigenous PCB dechlorinating activity in sediment microcosms. Water Res. 45, 3899–3907.

Field, J.A., Sierra-Alvarez, R., 2008. Microbial transformation and degradation of polychlorinated biphenyls. Environ. Pollut. 155, 1–12.

Focant, J.-F., Sjödin, A., Patterson, D.G., 2004. Improved separation of the 209 polychlorinated biphenyl congeners using comprehensive two-dimensional gas chromatography–time-of-flight mass spectrometry. J. Chromatogr. A 1040, 227–238.

Gray, N.F., 2004. Biology of Wastewater Treatment. World Scientific.

Grimm, F.A., Hu, D., Kania-Korwel, I., Lehmler, H.-J., Ludewig, G., Hornbuckle, K.C., et al., 2015. Metabolism and metabolites of polychlorinated biphenyls. Crit. Rev. Toxicol. 45, 245–272.

Hansler, G.M., Fikslin, T.J., Greene, F.W., Greene, R.W., 1998. Study of the Loadings of Polychlorinated Biphenyls From Tributaries and Point Sources Discharging to the Tidal Delaware River.

Herbst, W., Hunger, K., 2006. Industrial Organic Pigments: Production, Properties, Applications. John Wiley & Sons.

Johnson, P.D., Girinathannair, P., Ohlinger, K.N., Ritchie, S., Teuber, L., Kirby, J., 2008. Enhanced removal of heavy metals in primary treatment using coagulation and flocculation. Water Environ. Res. 80, 472–479.

Kaya, D., Imamoglu, I., Sanin, F.D., Sowers, K.R., 2018. A comparative evaluation of anaerobic dechlorination of PCB-118 and Aroclor 1254 in sediment microcosms from three PCB-impacted environments. J. Hazard. Mater. 341, 328–335.

Keith, L.H., 1996. Compilation of EPA's Sampling and Analysis Methods. CRC Press.

Kiedrzyńska, E., Urbaniak, M., Kiedrzyński, M., Jóźwik, A., Bednarek, A., Gągała, I., et al., 2017. The use of a hybrid sequential biofiltration system for the improvement of nutrient removal and PCB control in municipal wastewater. Sci. Rep. 7.

Kjellerup, B., Naff, C., Edwards, S., Ghosh, U., Baker, J., Sowers, K., 2014. Effects of activated carbon on reductive dechlorination of PCBs by organohalide respiring bacteria indigenous to sediments. Water Res. 52, 1–10.

Kjellerup, B.V., Paul, P., Ghosh, U., May, H.D., Sowers, K.R., 2012. Spatial distribution of PCB dechlorinating bacteria and activities in contaminated soil. Appl. Environ. Soil Sci. 2012.

Kranzioch, I., Stoll, C., Holbach, A., Chen, H., Wang, L., Zheng, B., et al., 2013. Dechlorination and organohalide-respiring bacteria dynamics in sediment samples of the Yangtze Three Gorges Reservoir. Environ. Sci. Pollut. Res. 20, 7046–7056.

Krzmarzick, M.J., Crary, B.B., Harding, J.J., Oyerinde, O.O., Leri, A.C., Myneni, S.C., et al., 2012. Natural niche for organohalide-respiring *Chloroflexi*. Appl. Environ. Microbiol. 78, 393–401.

Kuipers, B., Cullen, W.R., Mohn, W.W., 2003. Reductive dechlorination of weathered Aroclor 1260 during anaerobic biotreatment of Arctic soils. Can. J. Microbiol. 49, 9–14.

Kulkarni, P.S., Crespo, J.G., Afonso, C.A., 2008. Dioxins sources and current remediation technologies—a review. Environ. Int. 34, 139–153.

Lallas, P.L., 2001. The Stockholm Convention on persistent organic pollutants. AJIL 95, 692–708.

Laroe, S.L., Fricker, A.D., Bedard, D.L., 2014. *Dehalococcoides mccartyi* strain JNA in pure culture extensively dechlorinates Aroclor 1260 according to polychlorinated biphenyl (PCB) dechlorination process N. Environ. Sci. Technol. 48, 9187–9196.

Lombard, N.J., Ghosh, U., Kjellerup, B.V., Sowers, K.R., 2014. Kinetics and threshold level of 2, 3, 4, 5-tetrachlorobiphenyl dechlorination by an organohalide respiring bacterium. Environ. Sci. Technol. 48, 4353–4360.

Mattes, T.E., Ewald, J.M., Liang, Y., Martinez, A., Awad, A., et al., 2017. PCB dechlorination hotspots and reductive dehalogenase genes in sediments from a contaminated wastewater lagoon. Environ. Sci. Pollut. Res. 1–13.

May, H.D., Cutter, L.A., Miller, G.S., Milliken, C.E., Watts, J.E., Sowers, K.R., 2006. Stimulatory and inhibitory effects of organohalides on the dehalogenating activities of PCB-dechlorinating *bacterium o-17*. Environ. Sci. Technol. 40, 5704–5709.

Murínová, S., Dercová, K., Dudášová, H., 2014. Degradation of polychlorinated biphenyls (PCBs) by four bacterial isolates obtained from the PCB-contaminated soil and PCB-contaminated sediment. Int. Biodeterior. Biodegrad. 91, 52–59.

North, K.D., 2004. Tracking polybrominated diphenyl ether releases in a wastewater treatment plant effluent, Palo Alto, California. Environ. Sci. Technol. 38, 4484–4488.

Nyholm, J.R., Lundberg, C., Andersson, P.L., 2010. Biodegradation kinetics of selected brominated flame retardants in aerobic and anaerobic soil. Environ. Pollut. 158, 2235–2240.

Passatore, L., Rossetti, S., Juwarkar, A.A., Massacci, A., 2014. Phytoremediation and bioremediation of polychlorinated biphenyls (PCBs): State of knowledge and research perspectives. J. Hazard. Mater. 278, 189–202.

Patureau, D., Trably, E., 2006. Impact of anaerobic and aerobic processes on polychlorobiphenyl removal in contaminated sewage sludge. Biodegradation 17, 9–17.

Payne, R.B., Ghosh, U., May, H.D., Marshall, C.W., Sowers, K.R., 2017. Mesocosm studies on the efficacy of bioamended activated carbon for treating PCB-impacted sediment. Environ. Sci. Technol. 51, 10691–10699.

Payne, R.B., May, H.D., Sowers, K.R., 2011. Enhanced reductive dechlorination of polychlorinated biphenyl impacted sediment by bioaugmentation with a dehalorespiring bacterium. Environ. Sci. Technol. 45, 8772–8779.

Pieper, D.H., 2005. Aerobic degradation of polychlorinated biphenyls. Appl. Microbiol. Biotechnol. 67, 170–191.

Rodenburg, L.A., Du, S., Fennell, D.E., Cavallo, G.J., 2010. Evidence for widespread dechlorination of polychlorinated biphenyls in groundwater, landfills, and wastewater collection systems. Environ. Sci. Technol. 44, 7534–7540.

Rodenburg, L.A., Du, S., Lui, H., Guo, J., Oseagulu, N., Fennell, D.E., 2012. Evidence for dechlorination of polychlorinated biphenyls and polychlorinated dibenzo-*p*-dioxins and-furans in wastewater collection systems in the New York metropolitan area. Environ. Sci. Technol. 46, 6612–6620.

Rodriguez, C., Cook, A., Devine, B., Van Buynder, P., Lugg, R., Linge, K., et al., 2008. Dioxins, furans and PCBs in recycled water for indirect potable reuse. Int. J. Environ. Res. Public Health 5, 356–367.

Rosińska, A., Karwowska, B., 2017. Dynamics of changes in coplanar and indicator PCB in sewage sludge during mesophilic methane digestion. J. Hazard. Mater. 323, 341–349.

Rushneck, D.R., Beliveau, A., Fowler, B., Hamilton, C., Hoover, D., Kaye, K., et al., 2004. Concentrations of dioxin-like PCB congeners in unweathered Aroclors by HRGC/HRMS using EPA method 1668A. Chemosphere 54, 79–87.

Smith, B.J., Boothe, M.A., Fiddler, B.A., Lozano, T.M., Rahi, R.K., Krzmarzick, M.J., 2015. Enumeration of organohalide respirers in municipal wastewater anaerobic digesters. Microbiol. Insights 8 (Suppl. 2), 9–14.

Sowers, K.R., May, H.D., 2013. In situ treatment of PCBs by anaerobic microbial dechlorination in aquatic sediment: Are we there yet? Curr. Opin. Biotechnol. 24, 482–488.

Su, X., Zhang, Q., Hu, J., Hashmi, M.Z., Ding, L., Shen, C., 2015. Enhanced degradation of biphenyl from PCB-contaminated sediments: The impact of extracellular organic matter from *Micrococcus luteus*. Appl. Microbiol. Biotechnol. 99, 1989–2000.

Tu, C., Teng, Y., Luo, Y., Li, X., Sun, X., Li, Z., et al., 2011. Potential for biodegradation of polychlorinated biphenyls (PCBs) by *Sinorhizobium meliloti*. J. Hazard. Mater. 186, 1438–1444.

Van den Berg, M., Birnbaum, L.S., Denison, M., De Vito, M., Farland, W., Feeley, M., et al., 2006. The 2005 World Health Organization reevaluation of human and mammalian toxic equivalency factors for dioxins and dioxin-like compounds. Toxicol. Sci. 93, 223–241.

Wang, S., He, J., 2013. Phylogenetically distinct bacteria involve extensive dechlorination of Aroclor 1260 in sediment-free cultures. PLoS One 8, e59178.

Wu, Q., Watts, J.E., Sowers, K.R., May, H.D., 2002. Identification of a bacterium that specifically catalyzes the reductive dechlorination of polychlorinated biphenyls with doubly flanked chlorines. Appl. Environ. Microbiol. 68, 807–812.

Wu, C., Zhu, H., Luo, Y., Teng, Y., Song, J., Chen, M., 2015. Levels and potential health hazards of PCBs in shallow groundwater of an e-waste recycling area, China. Environ. Earth Sci. 74, 4431–4438.

Zanaroli, G., Negroni, A., Häggblom, M.M., Fava, F., 2015. Microbial dehalogenation of organohalides in marine and estuarine environments. Curr. Opin. Biotechnol. 33, 287–295.

Zhang, Z., Huang, J., Yu, G., Hong, H., 2004. Occurrence of PAHs, PCBs and organochlorine pesticides in the Tonghui River of Beijing, China. Environ. Pollut. 130, 249–261.