Biological treatment of a brominated micropollutant α-hexabromocyclododecane (α–HBCDD) from a raw hospital wastewater

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Abstract

The brominated micropollutants were not removed with conventional biological treatment processes. In this study it was aimed to treat the α–HBCDD which is a hydrophobic organic substance using a sequential treatment process consisting from an upflow anaerobic batch reactor (UASB) and a completely stirred tank reactor (CSTR) since α–HBCDD is a hydrophobic brominated micropollutant and its removals was very low under long retention times in anaerobic sediments (in monitored natural attenuation environments) and under aerobic stimulated environments (in conditions adding limiting nutrient and electron acceptors for support the microorganisms growth), separately, a sequential anaerobic/aerobic reactor system proses was choosen. The anaerobic and aerobic removals of α–HBCDD were 7% and 12%, respectively, at a SRT of 55 days. The effects of SRTs on α–HBCDD and its metabolite removals was investigated. The yields increased as the SRT was increased from 5 days up to 55 days. The total α–HBCDD yields in the whole sequential biological system was recorded as 18.3%. The metabolite of α–HBCDD is α2-Bromocyclododecan-1-ol (2–BCD). This metabolite was produced under anaerobic conditions and it was removed in aerobic reactor with a yield of 17% at 55 days SRT. The maximum 2–BCD removal efficiency was 63.6% in the sequential reactor process.

Introduction

Hexabromocyclododecane (HBCDD or HBCD) is used in large quantities as a flame retardant in building materials made of expanded- and extruded polystyrene (EPS/XPS) [1]. In addition, HBCDD is increasingly used as a flame retardant in electronics as a substitute for the banned flame retardants: Polybrominated diphenyl ethers (PBDEs). HBCDD is a hydrophobic and persistent organic pollutant which has been detected in air, water, soil and sediments worldwide, and HBCDD is found in indoor air and dusts and contaminates building waste [2]. HBCDD is an endocrine disruptor which accumulates in natural organisms and magnifies through the food chain, leading to progressively increasing background levels in wildlife and in human tissues, including in human milk. Human exposures are mainly from food and indoor dusts.

In May 2015 HBCDD became listed in Annex A of the Stockholm Convention [2]. No isomerization of α– to β– or γ–HBCDD forms occurred, while OH–HBCDD was identified as a product of α–HBCDD metabolism. Hexabromocyclododecane (HBCDD) is a brominated aliphatic cyclic hydrocarbon mainly used as a flame retardant additive in thermal insulation materials, especially in extruded (XPS) and expanded (EPS) polystyrene, which used to represent 80% of uses in Europe [3]. Due to its bioaccumulative, persistent and toxic characteristics, HBCDD has been listed in Annex A of the Stockholm Convention on Persistent Organic Pollutants in a rapid degradation of β– and γ–HBCDD compared to α–HBCDD, through debromination and hydroxylation [4]. Moreover, the bio-isomerization of γ–HBCDD into β– and mostly α–HBCDD, as well as the recalcitrance of α–HBCDD to bio-isomerization has been demonstrated in mammals [1].
The recent α-HBCDD degradability studies performed under separate anaerobic and aerobic environments exhibited low yields: For example, in anaerobic aquatic sediments 5650 ng/g HBCDD concentration showed 6.5% reduction in 113 days without any extraneous substance [5]. Natural attenuation and biostimulation sets within the first 8 days exhibited 13% α-HBCDD yields, then the decrease slowed down, in the natural attenuation set, there was 11.5% decrease of total-HBCDD [6]. In a microbial community, gram-positive bacteria population decreased the α-HBCDD removals (6.5%) and gram positive bacteria like Brassica rizosphere, Sphingomonas sp. increased the aerobic degradation of α-HBCDD (13%) [7]. Besides, 13 bacterial strains were isolated from a contaminated soil sample and tested for the degradation of γ-HBCDD in aerobic environment. It was found that Pseudomonas sp. (HB01 strain), degraded more than 10% of γ-HBCDD within 5 days [8]. 8% α-HBCDD anaerobic degradation was observed in microcosms with soil, aquatic sediments and digester sludge [9]. Also, in a study, anaerobic degradation was examined in the digested sewage sludge by adding primers and nutrients, which are starch and yeast. As a result of the study, it was found 9% HBCDD degradation by approximately more than 35 days half-life in the heat sterilized set, and in the biostimulation set (with primers and nutrients) HBCDD degradation half-life was found as 0.66 day [10]. The anaerobic degradation of α-HBCDD after 4 months incubation exhibited 23% degradation [11]. α-HBCDD degradation under 35 days half-life in the heat sterilized environments and in the biostimulation environments (with electron donors and nutrients) were low (10% ana 13%) [10]. The biodegradation of HBCDD was examined in the aerobic microcosms with soil and aquatic sediments [9]. The aerobic degradation of the α-HBCDD in the soil was high (23%) due to the microbial biotransformation. In viable soil (i.e. natural attenuation) 15% of the α-HBCDD was degraded during 119 days [12].

Micropollutants are detected in hydrosphere (surface waters, groundwaters, drinking waters), geosphere and biosphere owing to the developing measurement techniques. Hospitals are one of the main sources of micropollutant emissions because of large quantities of consumption such as: medical activities performed inside, laboratory researches and drugs eliminated from the human body via excretion with urine and feces. Usually, hospital wastewaters are not treated before sending to wastewater treatment plants (WWTPs) [13]. The micropollutants in the hospital wastewaters are directly discharged into the sewage system in Turkey without treatment since the conventional WWTPs can only treat macropollutants discharged into the sewage system in Turkey without treatment or washed from waste products, then released into municipal wastewater treatment plants or was washed from municipal wastewater treatment plants or was washed from waste products and infiltrated various ecosystems. α-HBCDD was reported in water, soil, sediment, plants, animals, humans, and even indoor dust [16]. Due to its highly hydrophobic nature (log KOW = 7.74), α-HBCDD released to aquatic environments tends to partition to and be accumulated in soil, sediment or sludge [17]. α-HBCDD is persistent, bioaccumulative and known to undergo long-range transport. α-HBCDD is an enzyme inducer, endocrine disruptor, and developmental neurotoxicant. Specifically, α-HBCDD exposure leads to changes in thyroid hormone systems, neurodevelopmental effects in children or mice, alters rat hepatic gene expression profiles for cholesterol biosynthesis and lipid metabolism in a sex specific manner and induces genetic recombination [18]. There are limited data for the treatability of α-HBCDD in the literature. Davis, Gonsior, Markham, & Marti [19] studied aerobic treatability of α-HBCDD and reported that it was not significant and did not follow further degradation to CO₂ and H₂O because of its physicochemical properties like low solubility (2.089×10⁻⁵ g L⁻¹) and high hydrophobicity (log KOW = 7.74) [20]. Peng, et al. [21] reported that α-HBCDD can be biodegraded by the anaerobic bacteria at the end of 300 days for 500 µg L⁻¹ initial concentration at pH 7.00 at 30°C together with the carbon and accumulates in time because of low treatment efficiencies. Also, the sludge cake obtained from the sludge dewatering process of the WWTPs containing the micropollutants can be applied onto soils as fertilizer. Entrained micropollutants in fertilizers leaks into the groundwaters with the help of rain waters. In this way, a hydrologic cycle occurs for the untreated micropollutants and human take the micropollutants into his body repeatedly .Therefore, treating the hospital wastewaters having a wide range of micropollutants is a very important issue in wastewater treatment. α-HBCDD is commonly added to polystyrene foams, upholstery textiles, and electronic devices as it exhibits excellent properties for flame protection purposes in these commercial products [15].

HBCD is a solid, white powder that is used as a flame retardant additive for thermoplastic polymers. Its principal use is in expanded polystyrene foams and other styrene resins. It may also be used in latex binders, unsaturated polyesters, and polyvinyl chloride wire, cable, and textile coatings. When used in textiles, it is applied as a back coating to the fabric, encapsulated in a polymer matrix. Textile applications include residential and commercial furniture, up-holstery seating in transportation, draperies, and wall coverings (FRA 1998). HBCD is usually applied with antimony trioxide as a back coating in a mass ratio of 2:1 (i.e., about 6–15% HBCD and 4–10% antimony oxide by weight).

HBCD was reported to be rapidly metabolized and eliminated in the feces and urine following absorption, with 70% of the administered radioactivity eliminated in the feces and another 16% eliminated in the urine 72 hr after dosing. HBCD is rapidly absorbed from the gastrointestinal tract, distributed primarily to the body fat, and eliminated rapidly, primarily in the feces.

α-HBCDD can be discharged during the industrial process or washed from waste products, then released into municipal wastewater treatment plants or was washed from waste products and infiltrated various ecosystems. α-HBCDD was reported in water, soil, sediment, plants, animals, humans, and even indoor dust [16]. Due to its highly hydrophobic nature (log KOW = 7.74), α-HBCDD released to aquatic environments tends to partition to and be accumulated in soil, sediment or sludge [17]. α-HBCDD is persistent, bioaccumulative and known to undergo long-range transport. α-HBCDD is an enzyme inducer, endocrine disruptor, and developmental neurotoxicant. Specifically, α-HBCDD exposure leads to changes in thyroid hormone systems, neurodevelopmental effects in children or mice, alters rat hepatic gene expression profiles for cholesterol biosynthesis and lipid metabolism in a sex specific manner and induces genetic recombination [18]. There are limited data for the treatability of α-HBCDD in the literature. Davis, Gonsior, Markham, & Marti [19] studied aerobic treatability of α-HBCDD and reported that it was not significant and did not follow further degradation to CO₂ and H₂O because of its physicochemical properties like low solubility (2.089×10⁻⁵ g L⁻¹) and high hydrophobicity (log KOW = 7.74) [20]. Peng, et al. [21] reported that α-HBCDD can be biodegraded by the anaerobic bacteria at the end of 300 days for 500 µg L⁻¹ initial concentration at pH 7.00 at 30°C together with the carbon
source (glucose) for a degradation rate of 92.4% in a synthetic wastewater [21].

In Turkey, there are not any limitations for the discharge of raw hospital wastewater into sewage channels. Moreover, there are no WWTPs to treat both macro and micropolllutants simultaneously in wastewater for hospitals in Turkey. The Ministry of Environment and Urbanization should take some precautions and impose some restraints about treating or pre-treating raw hospital wastewaters in Turkey.

Materials and methods

Reactor configurations

The quartz aerobic reactor used in this study has a volume of 1.6 liter and consists of a continuous flow stirred tank reactor without sludge return. The quartz upflow anaerobic sludge reactor has a volume of 1 liter and has no sludge return. The 37 °C and 22 °C temperatures in anaerobic and aerobic reactors were provided with a digital heater.

1, for the bacterial growth and the mixing liquor. Turbulence produced from the air pump in suspended liquid provided homogenous mixed liquor in the aerobic reactor. The sedimentation of the microorganisms was provided with the stability of the mixed liquor by the hesitation of the air pump before the sampling.

Operational conditions

The anaerobic reactor was fed with raw hospital wastewater for 20 days and the effects of SRTs (5 days, 30, 45 and 55 days) on the α-HBCDD removals was studied in anaerobic, aerobic and sequential reactor systems. At the beginning, the MLVSS values were 3400 mg L−1, 27000 mg L−1 and 29000 mg L−1 in the aerobic, anaerobic and sequential reactor systems, respectively. The anaerobic/aerobic sequential reactor system reached steady-state conditions after 5 days of continuous operation which was defined as having the same effluent COD and the α-HBCDD concentrations for 2 consecutive weeks. The pH values in anaerobic, aerobic reactors were 8.25 ± 0.50 and 7.3 ± 0.20 while the organic carbon loadings in the anaerobic and aerobic reactors were 0.19-0.29 and 0.10-0.16 g C/L.

Results

Total α-HBCDD removals in anaerobic biological reactor

α-HBCDD removal efficiency of the anaerobic reactor was obtained as 1.7% with an α-HBCDD effluent of 0.0113 μg L−1 at an α-HBCDD influent of 0.0115 μg L−1 during the 5 days of SRT operation (Table 1). α-HBCDD removal efficiency was measured as 2.6% with an α-HBCDD effluent of 0.0112 μg L−1 for an α-HBCDD influent of 0.0115 μg L−1 at 30 days SRT (Table 1). For 45 days SRT operation, α-HBCDD removal efficiency was found as 5.2% with an α-HBCDD effluent of 0.0109 μg L−1 for an α-HBCDD influent of 0.0115 μg L−1 (Table 1). For 55 days SRT operation, α-HBCDD removal efficiency was calculated as 7.0% with an α-HBCDD effluent of 0.0107 μg L−1 for an α-HBCDD influent of 0.0115 μg L−1 (Table 1).

The results showed that as the SRT was increased from 5 days to 55 days α-HBCDD removal efficiency increased slightly from 1.7% to 7.0% in the anaerobic reactor (Table 1). However, the ANOVA (one-way) statistical analysis results showed that it was not observed a significant regression between α-HBCDD...
removal and SRT increase (P = 0.052, α = 0.05, F = 5.86). The α-HBCDD is slightly removed in the anaerobic step.

In this study, the presence of 2-Bromocyclododecan-1-ol (2-BCD) which is the main metabolite of α-HBCDD was monitored in the effluents of the anaerobic reactor at all SRTs. 2-BCD was a less hydrophobic (log KOW = 4.90) micropollutant than its parent compound of α-HBCDD (log KOW = 7.74) based on its log KOW value. The effluent concentrations of 2-BCD were determined as 0.0015, 0.0015, 0.0013 and 0.0011 μg L⁻¹ at 5, 30, 45 and 55 days SRTs, respectively in the anaerobic reactor (Table 1). Based on Davis, et al. [20], dehalogenation is caused by breaking off the bromines from α-HBCDD and as a result, 2-BCD formed in the anaerobic conditions [20].

**Total α-HBCDD removals in aerobic biological reactor**

During 5 days of SRT operation, α-HBCDD removal efficiency of the aerobic biological reactor was found as 3.5% with an α-HBCDD effluent of 0.0109 μg L⁻¹ at an α-HBCDD influent of 0.0113 μg L⁻¹ (Table 1). α-HBCDD removal efficiency reached to 6.3% with an α-HBCDD effluent of 0.0105 μg L⁻¹ for an α-HBCDD influent of 0.0112 μg L⁻¹ at 30 days SRT (Table 1). For 45 days SRT operation, α-HBCDD removal efficiency was obtained as 8.3% with an α-HBCDD effluent of 0.0100 μg L⁻¹ for an α-HBCDD influent of 0.0109 μg L⁻¹ (Table 1). When the SRT was reached to the highest value of 55 days, α-HBCDD removal efficiency was determined as 12.1% with an α-HBCDD effluent of 0.0094 μg L⁻¹ for an α-HBCDD influent of 0.0107 μg L⁻¹ (Table 1).

Total removal of α-HBCDD was higher in the aerobic reactor (12.1%) than the anaerobic reactor (7.0%) at 55 days of SRT (Table 1). The low removals of α-HBCDD in both reactors showed that this micropollutant is partially resistant to biodegradation due to its physicochemical properties (low water solubility = 2.09×10⁻⁵ g L⁻¹ and high octanol/water partition coefficient = 7.74) in both reactors.

α-HBCDD removal efficiency in the aerobic reactor was calculated between 3.5% and 12.1% as the SRT was increased from 5 days to 55 days, respectively (Table 1). According to the ANOVA (one-way) statistical test results α-HBCDD could not treated effectively biologically in the aerobic reactor at all SRTs (P = 0.055, α = 0.05, F = 5.65).

In the presence of 2-BCD (0.0015, 0.0015, 0.0013 and 0.0011 μg L⁻¹ at 5, 30, 45 and 55 days SRTs, respectively) produced from the reductive dehalogenation in the anaerobic reactor, the 2-BCD was analysed in the samples taken from the effluent of the aerobic reactor at all SRTs (Table 1). The effluent concentrations of 2-BCD were found as 0.0012, 0.0010, 0.0008 and 0.0004 μg L⁻¹ at 5, 30, 45 and 55 days SRTs, respectively in the aerobic reactor effluents (Table 1). Aerobic microorganisms could degrade 20.0% of 2-BCD with an effluent concentration of 0.0012 μg L⁻¹ at an influent 2-BCD concentration of 0.0015 μg L⁻¹ at 5 days SRT in the aerobic reactor (Table 1). When the SRT was increased to 50 days, the removal efficiency of 2-BCD was obtained as 33.3% with an effluent concentration of 0.0010 μg L⁻¹ at an influent 2-BCD concentration of 0.0015 μg L⁻¹ in the aerobic reactor (Table 1). The 2-BCD removal efficiency was found as 38.5% with an effluent concentration of 0.0008 μg L⁻¹ at an influent 2-BCD concentration of 0.0013 μg L⁻¹ at 45 days SRT in the aerobic reactor (Table 1). Finally, 2-BCD removal efficiency increased to 63.6% with an effluent concentration of 0.0004 μg L⁻¹ at an influent 2-BCD concentration of 0.0011 μg L⁻¹ at the highest SRT of 55 days in the aerobic reactor (Table 1). As the SRT was increased from 5 days to 55 days, 2-BCD yields increased from 20.0% to 63.6%, respectively in the aerobic reactor (Table 1). The ANOVA (one-way) test statistics results showed that there was a linear correlation between SRTs and the removals of 2-BCD remaining from the anaerobic reactor and this regression was significant (R² = 0.93, P = 0.007, α = 0.05, F = 15.78).

**Total α-HBCDD removals in anaerobic/aerobic sequential biological reactor system**

Firstly, α-HBCDD removal efficiency of the anaerobic/aerobic sequential biological reactor system was measured as 5.2% with an α-HBCDD effluent of 0.0109 μg L⁻¹ at an α-HBCDD influent of 0.0115 μg L⁻¹ at 5 days SRT (Table 1). After 30 days SRT, α-HBCDD removal efficiency was determined as 8.7% with an α-HBCDD effluent of 0.0105 μg L⁻¹ for an α-HBCDD influent of 0.0115 μg L⁻¹ at 30 days SRT (Table 1). When the SRT was increased to 45 days, α-HBCDD removal efficiency reached to 13.0% with an α-HBCDD effluent of 0.0100 μg L⁻¹ for an α-HBCDD influent of 0.0115 μg L⁻¹ (Table 1). At the highest SRT of 55 days, α-HBCDD removal efficiency was calculated as 18.3% with an α-HBCDD effluent of 0.0094 μg L⁻¹ for an α-HBCDD influent of 0.0115 μg L⁻¹ at 55 days SRT (Table 1). In this study, total α-HBCDD removal efficiency (18.3%) calculated in the anaerobic/aerobic sequential biological reactor system was higher than the results obtained by other researchers. Davis, et al. [20], also studied the aerobic treatability of α-HBCDD and reported that this micropollutant was not removed (0.00% removal efficiency) in the aerobic activated sludge process for an initial concentration of 3.6 mg L⁻¹ at 56 days HRT and at 22±3 °C [20].

The influent concentrations of 2-BCD were determined as 0.0015, 0.0015, 0.0013 and 0.0004 μg L⁻¹ at 5, 30, 45 and 55 days SRTs, respectively in the anaerobic/aerobic sequential biological reactor (Table 1). The effluent concentrations of 2-BCD were detected as 0.0012, 0.0010, 0.0008 and 0.0004 μg L⁻¹ at 5, 30, 45 and 55 days SRTs, respectively (Table 1). 2-BCD removal efficiency increased with the increasing SRT and the maximum 2-BCD yield was obtained as 63.6% in the anaerobic/aerobic sequential biological reactor system at the highest SRT of 55 days (Table 1). The maximum 2-BCD removal efficiency (63.6%) found in the anaerobic/aerobic sequential biological reactor system was higher than the study performed by Kassotaki, Buttiglieri, Ferrando-Climent, Rodriguez-Roda & Pijuan [24]. They found 6.5% of 2-BCD removal efficiency in a sequencing batch reactor having a volume of 8 L treating synthetic wastewater at 24 h HRT and at 30 °C for 100 μg L⁻¹ initial α-HBCDD concentration [25-28].
Aerobic and anaerobic/aerobic sequential biological reactors at 5, 30, 45 and 55 days SRTs (Aerobic reactor.

Table 1: Anaerobic reactor

| SRT (days) | α-HBCDD inf. (μg/L) | α-HBCDD eff. (μg/L) | α-HBCDD % removal | α-HBCDD removed mass (μg/L) | 2- BCD inf. (μg/L) | 2- BCD eff. (μg/L) | 2- BCD % removal |
|------------|---------------------|---------------------|-------------------|---------------------------|-------------------|-------------------|------------------|
| 5          | 0,0115              | 0,0113              | 1,7               | 0,0002                    | 0,001             | 0,0015            | 0                |
| 30         | 0,0115              | 0,0112              | 2,6               | 0,0003                    | 0,001             | 0,0015            | 0                |
| 45         | 0,0115              | 0,0109              | 5,2               | 0,0006                    | 0,001             | 0,0013            | 0                |
| 55         | 0,0115              | 0,0107              | 7                 | 0,0008                    | 0,001             | 0,0011            | 0                |

Anaerobic/Aerobic Sequential Reactor System.

| SRT (days) | α-HBCDD inf. (μg/L) | α-HBCDD eff. (μg/L) | α-HBCDD % removal | α-HBCDD removed mass (μg/L) | 2- BCD inf. (μg/L) | 2- BCD eff. (μg/L) | 2- BCD % removal |
|------------|---------------------|---------------------|-------------------|---------------------------|-------------------|-------------------|------------------|
| 5          | 0,0113              | 0,0109              | 3,5               | 0,0004                    | 0,0015            | 0,0012            | 20               |
| 30         | 0,0115              | 0,0105              | 6,3               | 0,0007                    | 0,0015            | 0,001             | 33,3             |
| 45         | 0,0109              | 0,01              | 8,3               | 0,0009                    | 0,0013            | 0,0008            | 38,5             |
| 55         | 0,0107              | 0,0094              | 12,1              | 0,0013                    | 0,0011            | 0,0004            | 63,6             |

Conclusions

A hydrophobic brominated micropollutant α-HBCDD was removed with a total yields of 18% under biological sequential anaerobic/aerobic conditions at a SRT of 55 days. The metabolite of α-HBCDD is 2-BCD is produced in anaerobic reactor and it was removed with a yield of 18,3% at aerobic reactor. The maximum total 2-BCD removal efficiency was 63,6% in the whole biological reactor system.

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