Contamination Issues in Asian Developing Countries

Tatsuya Kunisue and Shinsuke Tanabe

Abstract This chapter focuses on the contamination, bioaccumulation, and toxicological effects of dioxins and related compounds (DRCs), such as polychlorinated dibenzo-*p*-dioxins (PCDDs), polychlorinated dibenzofurans (PCDFs), and dioxin-like polychlorinated biphenyls (DL-PCBs), in Asian developing countries, with a particular emphasis on open dumping sites of municipal waste. A comprehensive investigation of soils has suggested clearly that dumping sites (DS) are potential sources of DRCs, whereas the concentrations of DRCs in soils from urban and agricultural areas in Asian developing countries were comparable to or lower than those in general background soils from developed nations. In India, notably higher concentrations of DRCs were detected in human milk from women residing around DS, compared with those from reference sites (RS) and other Asian developing countries, indicating that the residents around DS ingest greater amounts of DRCs, possibly via the intake of contaminated bovine milk and fish. Elevated concentrations of DRCs were also detected in wild animals inhabiting the Indian DS area, such as crow and pig, and the accumulated DRC profiles suggested direct transfer of these contaminants from contaminated soil. Toxic equivalents (TEQs) of DRCs and the liver to adipose concentration ratios of PCDD/Fs in pigs had statistically significant positive correlations with the levels of hepatic cytochrome P450 (CYP) 1A-like protein, suggesting the induction of CYP1A by DRCs and CYP1A-dependent hepatic sequestration of PCDD/Fs. In addition, decreases in plasma-free thyroxine and immunoglobulin G were observed in pigs from the DS. Thus, DS in developing countries are one of the main challenges for further research due to the long-term effects on environmental quality and human/animal health. The continuous formation of DRCs in DS and their elevated residues

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detected in breast milk from residents living around such DS warrant effect studies of these contaminants on their offspring. Comprehensive and long-term monitoring programs are urgently needed with proper capacity building in Asian developing countries, to mitigate DRC emission and their risk on ecosystems and human health.

Keywords Asian developing countries, Bioaccumulation, Cytochrome P450, Dioxins and related compounds, Human exposure, Open dumping sites

Contents

1 Introduction

It is a well-known fact that dioxins and related compounds (DRCs) including polychlorinated dibenzo-*p*-dioxins (PCDDs), polychlorinated dibenzofurans (PCDFs), and dioxin-like polychlorinated biphenyls (DL-PCBs) are persistent organic pollutants (POPs). DRCs have been detected in various environmental media and animals including humans because of their persistence in the environment and highly bioaccumulative nature. Especially, higher trophic animals accumulate elevated levels of these contaminants, and consequently their toxic effects have become a social concern [1].

DRCs are unintentionally formed during various combustion processes and are impurities of chlorinated chemicals that were used in large quantities as herbicides and wood preservatives. Combustion is believed to be the major source of PCDDs and PCDFs (PCDD/Fs) to the environment [2]. It has been found that DL-PCBs are also formed in municipal waste incineration [3], while these contaminants are

contained in commercial PCB mixtures [4]. During the past few decades, numerous monitoring surveys on DRC pollution have been conducted mainly in developed nations. In general, emission and exposure levels of DRCs into the environment and for humans have decreased since the 1980s [5]. Despite the fact that DRC contamination has been extensively studied in developed nations, little is known about their behavior, fate, ultimate sources, temporal trends, and animal exposure in developing countries where investigations on DRCs were recently undertaken.

In recent years, public media have voiced concern about open dumping sites (DS) in Asian developing countries where large amounts of municipal solid waste have been dumped. Unfortunately, in most Asian developing countries, open DS areas are located near human habitats; therefore, exposure to various toxic chemicals originating from DS is of serious concern because of the potential effects on human health, wildlife, and environmental quality [1]. Uncontrolled burning of solid waste by waste pickers, generation of methane gas, lack of advanced waste incineration technology, and natural low-temperature burning are major problems in DS at present. These are favorable factors for the formation of DRCs. However, studies on contamination status, animal exposure, and biochemical effects of DRCs are limited in DS.

Our research group has conducted comprehensive investigations of POPs in Asian regions and suggested the presence of DRC sources in Asian developing countries such as India, Cambodia, Vietnam, and the Philippines [6], which have large open dumping sites of municipal waste in the suburbs of major cities. Typically, in DS, a variety of municipal waste are dumped continuously and burnt under low temperature by spontaneous combustion or intentional incineration (Fig. 1). DRCs are formed by this low-temperature combustion, in addition to leaching out of DL-PCBs from dumped electric appliances. Consequently, the surrounding environment may be polluted by these contaminants.

The present chapter discusses the contamination issues and toxicological effects of DRCs in Asian developing countries, with a particular emphasis on open DS from the outcome of comprehensive investigations conducted previously. To date, it is believed that environmental pollution and potential health effects by dioxins are major issues in developed nations. No dioxin problems were known in developing countries other than sporadic incidents such as herbicide agent orange in Vietnam. Here we provide scientific data that DS can be a significant emission source of DRCs that lead to adverse effects on humans and wildlife in developing countries.

Fig. 1 Photos of open dumping sites in Asian developing countries. (**a**) Perungudi dumping site, Chennai city, India. (**b**) Stoeung Meanchey dumping site, Phnom Penh, Cambodia

2 Contamination Status in the Environment: Soil Research

2.1 Residue Levels

To understand the contamination status of DRCs in Asian developing countries, our research group has conducted firstly a comprehensive research of soils [7]. Soil samples were collected from urban and agricultural areas and DS in the Philippines, Vietnam, Cambodia, and India. The DS in each country were located in the suburbs of major cities, Manila (Philippines), Hanoi and Ho Chi Minh (Vietnam),

Phnom Penh (Cambodia), and Chennai (India), and the urban and agricultural areas were more than 30 km away from the DS.

Elevated levels of DRCs were detected in soils from the Asian DS, with the highest concentrations of 200,000 pg/g (dry weight basis) in soils from the Cambodian DS, suggesting the formation and emission of these contaminants in the DS environment. Interestingly, the magnitude of DRC contamination in DS soils was significantly greater than that of urban and agricultural areas. When comparing DRC concentrations in various soil types globally, DS in Asian developing countries showed higher concentrations than general background soils reported in other countries [8, 9] and comparable levels to the DRC-contaminated sites in developed nations [10–12]. On the other hand, DRC concentrations observed in soils from urban and agricultural areas in the Philippines, Vietnam, Cambodia, and India were comparable to or lower than those in general soils from other countries. Furthermore, toxic equivalents (TEQs) of DRCs in some soil samples collected from DS in Asian developing countries, which were estimated based on human/mammal toxic equivalency factors (TEFs) proposed by WHO [13], exceeded the environmental quality standard of 1,000 pg/g TEQs set forth by the Japanese government and US Department of Health [7]. These results suggest clearly that DS are a major source of DRCs in Asian developing countries, while the magnitude of DRC contamination derived from the impurities of agrochemicals and from urban activities was relatively small by comparison. Though reaction mechanisms for DRC formation are believed to be complex, the combustion of chlorinated waste is the major source of PCDD/Fs to the global environment [14]. A previous study evaluated the contribution to dioxin formation from combustion of some polymer materials such as polyethylene (PE), polystyrene (PS), and polyvinyl chloride (PVC) and showed that PVC contributed significantly to the formation of PCDD/Fs and DL-PCBs [15]. Common applications of plastics in daily use products and industries together with the lack of proper management of waste materials in developing countries have led to significant disposal of chlorinated waste-containing products such as PVC, chloromethane, and chlorophenols in open DS every day. Accordingly, we suggest the possibility of the considerable formation of DRCs in these DS.

2.2 Homologue Profiles

To further understand the role of DS as a source of DRCs, homologue profiles of PCDD/Fs were examined in DS soils. Their PCDD/F homologue profiles were then compared with typical profiles of samples representing environmental sources, which were the emission from a typical municipal waste incinerator in the United States [14] and an average of 12 different combustion sources [16] (Fig. 2). In general, the homologue profiles of samples representing environmental sources are characterized by the predominance of lower chlorinated dibenzofurans and an increasing proportion from tetra- to hexa-chlorinated dibenzo-*p*-dioxins (T_4-H_6CDDs). Interestingly, homologue profiles of the DS soils from the Philippines,

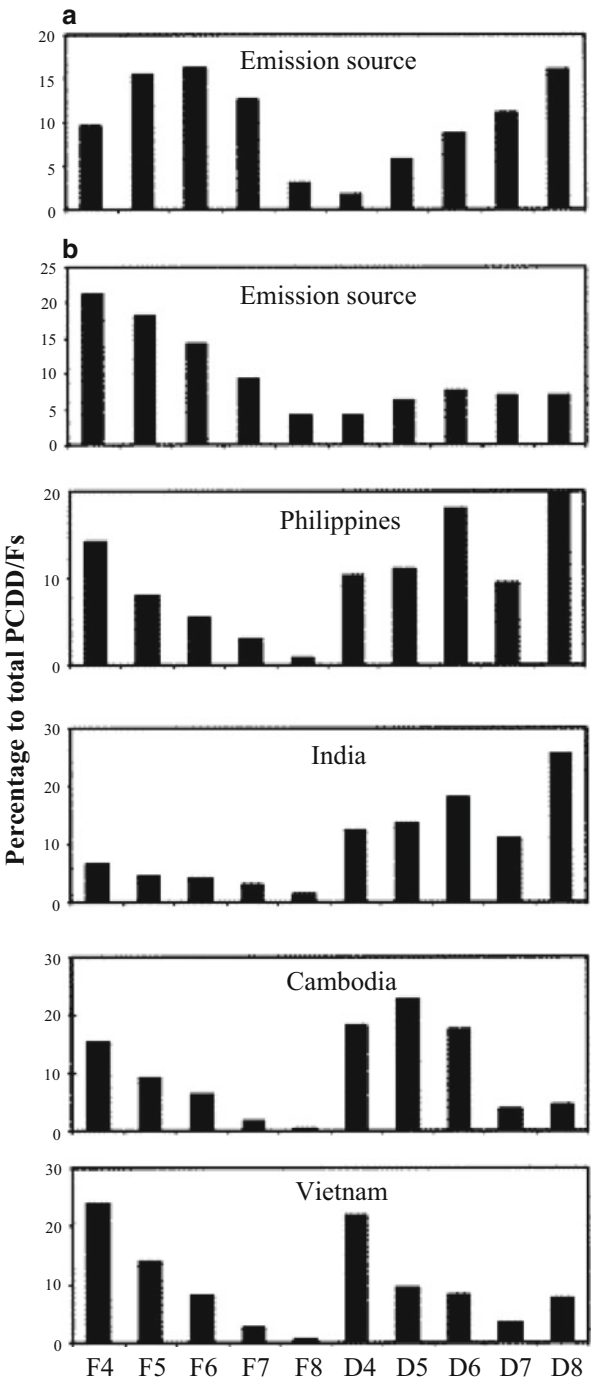

Fig. 2 Homologue profiles of PCDD/Fs in soils from dumping sites in Asian developing countries in comparison with the profile of samples representing emission sources (municipal waste incinerators). Vertical bars represent the percentage of each homologue to total PCDD/F concentrations. F and D refer to dibenzofurans and dibenzo-*p*-dioxins, respectively, and numbers indicate the degree of chlorination. Data for emission source samples were cited from Brzuzy and Hites [14], (**a**) a typical emission of a municipal waste incinerator, and Baker and Hites [16], (**b**) an average of 12 different combustion sources

Vietnam, Cambodia, and India reflected a pattern of emission sources (Fig. 2). PCDD/F profiles of DS soils from the Philippines and Cambodia were similar to those of emission sources, implying recent formation of these contaminants in each DS. On the other hand, the typical pattern of environmental sink samples, which were soils collected from various locations over the world, contains octachlorinated dibenzo-*p*-dioxin (O_8CDD) as a predominant congener [16]. PCDD/F profiles observed in the urban and agricultural soils from Asian developing countries were similar to those of typical environmental sinks [7].

As for DL-PCB congener patterns, non-*ortho* congener CB-126 contributed predominantly to total TEQs in most of the soil samples surveyed in Asian developing countries. The formation of DL-PCBs has been hypothesized through three alternative processes including the release from commercial PCB mixtures, emission from combustion, and, to a lesser extent, photolysis of higher chlorinated PCBs [17]. A study in the United Kingdom reported that TEQ input of DL-PCBs from Aroclor formulations into the environment was mainly contributed by CB-77, CB-105, CB-118, CB-156, and to a lesser extent CB-126 [18]. Combustion source emissions were dominated by non-*ortho* DL-PCBs, in which CB-126 contributed predominantly to total TEQs [17]. In addition, it should be noted that CB-126 can be formed during the domestic burning process [19]. Our result suggests that uncontrolled burning of solid wastes in Asian DS could be a source of DL-PCBs.

2.3 Flux and Load of DRCs to DS

Soil is a useful environmental matrix to estimate the deposition of PCDD/Fs on a global scale [9, 14, 20]. Using the same approach that was reported in the previous studies [9, 14, 20], we estimated the flux of PCDD/Fs to the soils and their load to the DS areas in Asian developing countries. Flux to soils can be calculated by the following equation:

$$F = CM/(St),$$

where F is depositional flux to soils (ng m^{-2} $year^{-1}$), C is the concentration in soils, M is the mass of soils collected (g), S is the surface area of soil sample (m^2), and t is the accumulation time of PCDD/Fs in the soil compartment (year). For soils in open DS, t values were calculated on the basis of the time when DS began to be used and the time when soil samples were collected [7]. Accordingly, we set t values for soils in DS in the Philippines; Cambodia; India; Hanoi, Vietnam; and Ho chi minh, Vietnam, at 7, 21, 15, 3, and 11 years, respectively. The loading rate (R) of PCDD/Fs to a DS (considered as the annual amount of PCDD/Fs received by surface area of the DS, mg TEQs/year) can be calculated by multiplying flux value to surface area (A) of the DS:

Table 1 Estimated flux of PCDD/Fs to dumping site soils and estimated loadings of PCDD/Fs to dumping site areas in Asian countries

Country	Flux (ng m^{-2} $year^{-1}$)		Dumping site area (m^2)	Load	
	Mean	Range		mg/year	mg TEQ/year
Philippines	17,000	13000–21,000	230,000	3,900	35
Cambodia	2,900	31–19,000	30,000	87	1.1
India	990	290–4,500	1,400,000	1,400	8.8
Vietnam–Hanoi	4,100	83–34,000	50,000	210	3.2
Vietnam–Ho Chi Minh	67	3.8–160	300,000	20	0.12

$$R = FA.$$

Estimated fluxes of PCDD/Fs to soils in DS in the Philippines, Cambodia, India, and Vietnam are given in Table 1. It is interesting to note that fluxes to DS soils from the Philippines and Cambodia were greater than those from other locations in the world reported previously [9]. This result indicates that DS are potential sources of PCDD/Fs; the elevated fluxes observed in these DS could be attributed to uncontrolled combustion processes. The load of PCDD/Fs to the DS indicates that DS in the Philippines and India with a large area of approximately 23 and 140 ha could receive the highest annual amount of 3,900 and 1,400 mg/year PCDD/Fs (35 and 8.8 mg TEQs/year), respectively (Table 1). The DS in Ho chi minh, Vietnam, had the lowest loading rate due to the less contamination of PCDD/Fs in soils. For comparison, total annual fluxes to the Kanto region in Japan, one of the polluted areas in the world, were estimated to range from 50 to 900 g TEQ with a total area of 32,000 km^2 (approximately 3 million ha) [21]. The area of DS in India is 140 ha, which is 21,000 times smaller than that of the Kanto region, and this area was estimated to receive 8.8 mg TEQs/year. These estimates suggest that DS in India and the Philippines may be significant reservoirs for PCDD/Fs [7].

3 Human Exposure: Human Milk Research

3.1 Contamination Status

To evaluate the status of human exposure to DRCs, we have analyzed human breast milk from Asian countries [22]. Mean lipid-normalized concentrations of TEQs of DRCs, which were estimated based on human/mammal TEFs proposed by WHO [13], in human breast milk collected from general public in India, Cambodia, Vietnam, the Philippines, Malaysia, China, and Japan [23–26] are illustrated in Fig. 3. Relatively high concentrations of TEQs were found in human milk from Japan where a large amount of DRCs have been released into the environment in the

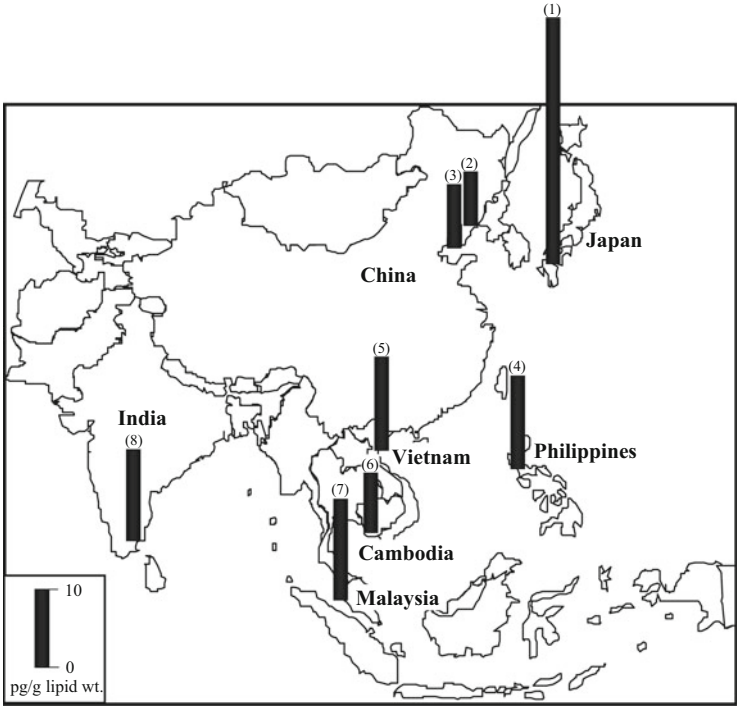

Fig. 3 TEQs of DRCs (PCDDs, PCDFs, and DL-PCBs) in human breast milk collected from general public in Asian countries. (*1*) Fukuoka, Japan (Kunisue et al. [23]), (*2*) Shenyang, China (Kunisue et al. [24]), (*3*) Dalian, China (Kunisue et al. [24]), (*4*) Quezon, Philippines (Kunisue et al. [25]), (*5*) Hanoi, Vietnam (Kunisue et al. [25]), (*6*) Phnom Penh, Cambodia (Kunisue et al. [25]), (*7*) Penang, Malaysia (Sudaryanto et al. [26]), (*8*) Palaverkadu, India (Kunisue et al. [25])

past. Our studies also demonstrated elevated DRC concentrations in wildlife inhabiting Japan [27, 28]. From the outcome of our soil survey described above, we presumed that the residents living around DS may be exposed to DRCs, because most of them earn their livelihood by doing DS-dependent labor. It is expected that in utero and lactational exposure to DRCs may adversely affect the brain development and immune systems of infants and children [29–32]. So, we attempted to elucidate the contamination status of DRCs in human breast milk collected from the residents around DS in India, Cambodia, and Vietnam and compared with those in general public from reference sites (RS) [25].

In India, the concentrations of DRCs in human breast milk from the DS were significantly higher than those from the RS and the other two countries, while levels of these contaminants in human breast milk from Cambodia and Vietnam were not significantly different between the DS and RS (Fig. 4). This result indicates that significant pollution sources of DRCs are present in/around the DS of India, and the surrounding residents may be exposed to relatively high levels of these

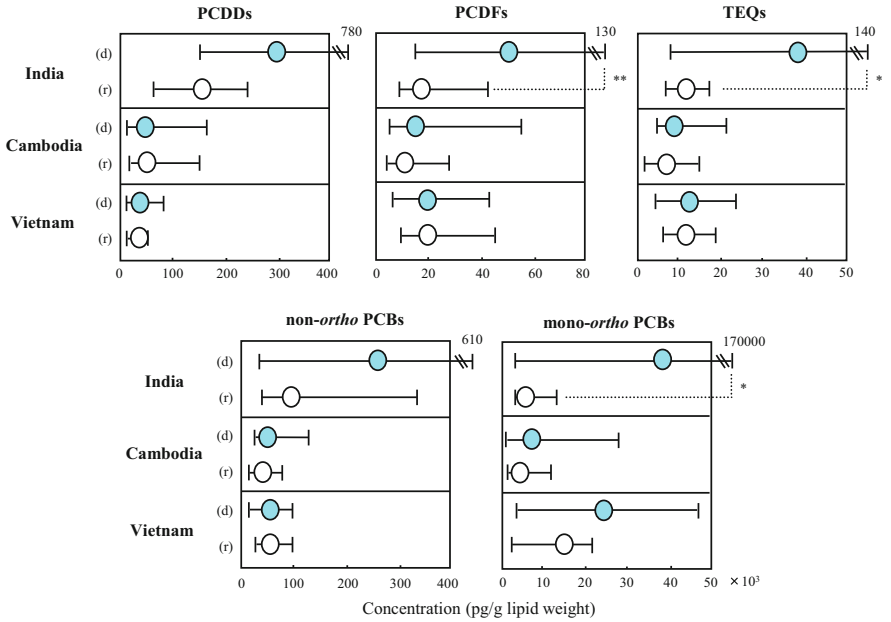

Fig. 4 Comparison of DRC concentrations in human breast milk from dumping (d) and reference (r) sites. The circles and bars represent mean and range values, respectively. $*p < 0.05$, $**p < 0.01$. Data were cited from Kunisue et al. [25]

contaminants. To understand the magnitude of contamination in human breast milk from the Indian DS, TEQ levels were compared with the values for human breast milk from general public of other countries since 1990. The TEQ levels in human breast milk from the Indian DS were comparable to or higher than those from developed countries [23, 33–38], suggesting that the DS residents have been exposed to comparable levels of DRCs with the general public in developed countries. On the other hand, the TEQs in human breast milk from Cambodia and Vietnam were lower than those from developed countries and comparable to those from other developing countries [24, 26, 37, 39]. In this international comparison, however, there are some uncertainties such as age and parity of the mother, sampling period, sample number, and accuracy of the analytical techniques involved. In addition, very little data are available on mono-*ortho* DL-PCBs in the literature. Because of such uncertainties, it was difficult to draw any firm conclusion from the above comparison. However, the observation that TEQ levels in human breast milk from the DS of India were comparable to or higher than TEQ values, which were estimated from PCDD/F and DL-PCB concentrations, from some developed countries including Japan is noteworthy. In developed countries, concentrations of DRCs in human breast milk have recently decreased [40], because of the installation of highly efficient incinerators and strict regulations on the production and usage of various chemicals. On the other hand, in Asian developing countries, it can be anticipated that the residue levels of DRCs in

human breast milk may increase in the future, because the release of these contaminants is poorly controlled currently.

3.2 Variation Associated with Parity and Age

Concentrations of DRCs in human breast milk vary by various factors such as the age, parity, and breast-feeding period of the mother [40, 41]. In the case of primiparae, it is observed that DRC concentrations in human breast milk were positively correlated with the age of mothers [23]. However, our study on Asian developing countries showed no significant correlations between DRC concentrations and primiparae age [25]. Although it cannot be clearly explained why no significant correlation was observed in Asian developing countries, a narrow range of age and recent exposure of DRCs may be possible reasons. Most women in Asian developing countries have many children in their life with the first infant often born by the mother at a young age.

In developing countries, it can be anticipated that the parity of mother is one of the focal factors influencing concentrations of DRCs in human breast milk. Therefore, we examined the relationship between the number of deliveries by the mothers and TEQs in human breast milk from women in Asian developing countries. TEQ levels in human breast milk from the DS of India tended to decrease with increase in the number of deliveries. One of the primipara donors had an exceptionally high TEQ level (140 pg/g lipid wt.). These results suggest that mothers who have been exposed to relatively high levels of DRCs may transfer higher amounts of these contaminants to the first infant than to the infants born afterward through breast-feeding, and hence the firstborn children might be at higher risk by DRCs. In developed countries, DRC concentrations in human breast milk from primiparae were also higher than those from multiparae [23, 41].

3.3 Risk Assessment for Infant

The presence of DRCs in human breast milk is of great concern, because these lipophilic chemicals are readily transferred and absorbed to infants. It is reported that one- to three-month-old infants absorb above 90% of most DRC congeners containing in their mothers' milk [42–44]. To understand the magnitude of exposure to DRCs by infants, we estimated daily intake (DI) from the concentrations of these contaminants in human breast milk observed in Asian developing countries, based on the assumption that an infant ingests 700 ml milk per day and the weight of an infant is 5 kg, and compared to the guideline standard proposed by the WHO [45]. As expected, relatively higher DIs of TEQs were observed in infants residing around DS in India compared with those from other countries, and DIs in all cases exceeded 1–4 pg TEQs/kg/day, the tolerable daily intake (TDI) (Fig. 5). DRCs

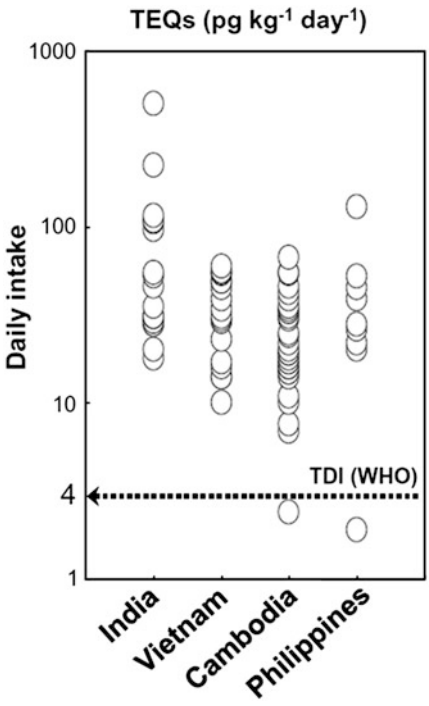

Fig. 5 Estimated daily intake (DI) of DRCs (TEQs) by infants in Asian developing countries. DI was estimated using TEQ data in human breast milk reported by Kunisue et al. [25]. TDI: Tolerable Daily Intake [45]

induce various toxic effects, e.g., cancer, in animal bodies [46]. These observations imply that abundance of DRCs in human breast milk may adversely affect development and reproductive systems of Asian children. However, it is difficult to draw any firm conclusions from Fig. 5 whether or not adverse effects by DRCs have already occurred in Asian infants, because TDIs used here are estimated on the basis of life-span exposure. Not only TDIs from life span but also TDIs of DRCs estimated from breast-feeding period are needed.

4 Potential Sources for Dumping Site Residents

4.1 Bovine Milk

Although greater contamination of DRCs was observed in DS soils compared to urban and agricultural soils in Asian developing countries [7], the DRC concentrations in human breast milk collected from the DS residents in Cambodia and Vietnam were not significantly higher than those from RS. As described earlier, however, residue levels of DRCs in Indian samples from the DS were notably higher (Fig. 4). These observations imply that the residents around the DS in Cambodia and Vietnam have not been greatly exposed to DRCs originating from

the DS. For humans, food intake, especially meat and dairy products, accounts for 98.8% of exposure to DRCs, and consumption of water, ingestion of soil, and inhalation of air are not major sources [47]. In addition, residue levels and composition of DRCs in human tissues generally reflect those in foods ingested [48–51]. In India, buffalo and cows reared near the DS feed mainly on dumped leftovers (Fig. 6). The residents around the DS constantly drink the milk collected from these bovines (Fig. 6). On the other hand, in Cambodia and Vietnam, livestock such as buffalo and cows are not reared around the DS. To elucidate whether or not bovine milk is a potential source of DRCs for the residents around the DS in India, residue levels of these contaminants in buffalo' and cows' milk collected were investigated and compared with those in bovine milk collected from RS [25].

DRCs were detected in all of the bovine milk samples analyzed, revealing that bovines in India have been exposed to these contaminants. Concentrations of DRCs in bovine milk collected from the DS were significantly higher than those from the RS (Fig. 7). This result indicates that buffalo and cows feeding in the Indian DS consume greater amounts of DRCs through contaminated soils and/or garbage and that daily intake of these bovine milk by the residents around the DS is one of the possible reasons why elevated TEQ levels were observed in human breast milk collected from the DS. Interestingly, compositions of PCDD/F congeners in bovine milk showed different patterns depending on the area of collection. In bovine milk collected from the DS, lower chlorinated congeners such as 2,3,7,8-T_4CDD, 1,2,3,7,8-P_5CDD, and 2,3,4,7,8-P_5CDF predominated, while the residue levels of 1,2,3,4,6,7,8-H_7CDD and O_8CDD were relatively high in those from the RS (Fig. 8). As described in our soil study, concentrations of T_4-, P_5-, and H_6-CDD/Fs in soils from the DS in India were higher than those from urban and agricultural areas [7]. These observations indicate that T_4-, P_5-, and H_6-CDD/Fs are formed via combustion of municipal waste and that buffalo and cows feeding in and around the Indian DS accumulate greater amounts of these compounds through contaminated soils, leftovers, and/or pastures. In soils collected from Indian DS, however, 1,2,3,4,6,7,8-H_7CDD and O_8CDD were predominant among all the 2,3,7,8-substituted congeners [7]. A previous study reported that the average percent contribution of high-chlorinated DD/Fs in pastures effected through soil particle adhesion was higher than low-chlorinated DD/Fs, and during summer, the period of high atmospheric temperature, the uptake of PCDD/Fs by pasture from vapor phase increased with the increasing degree of chlorination (increasing K_{OA}) [52]. Additionally, another study showed that PCDD/F contamination in cow milk reflected not only the intake from pastures but also ingestion through contaminated soils [53]. These findings suggest that the intake of high-chlorinated DD/DFs such as 1,2,3,4,6,7,8-H_7CDD and O_8CDD by buffalo and cows in and around the Indian DS is greater than that of low-chlorinated DD/DFs. In bovine milk from the DS, however, higher concentrations of low-chlorinated DD/DFs such as T_4-, P_5-, and H_6-CDD/Fs were observed, indicating that buffalo and cows in and around the Indian DS preferentially transfer more amounts of low-chlorinated DD/Fs to their milk. Fries et al. [54, 55] investigated a mass balance of PCDD/Fs in cows following administration of pentachlorophenol-treated wood, and they reported

Fig. 6 Photos of buffalo and cows rearing in/around Perungudi dumping site, Chennai city, India

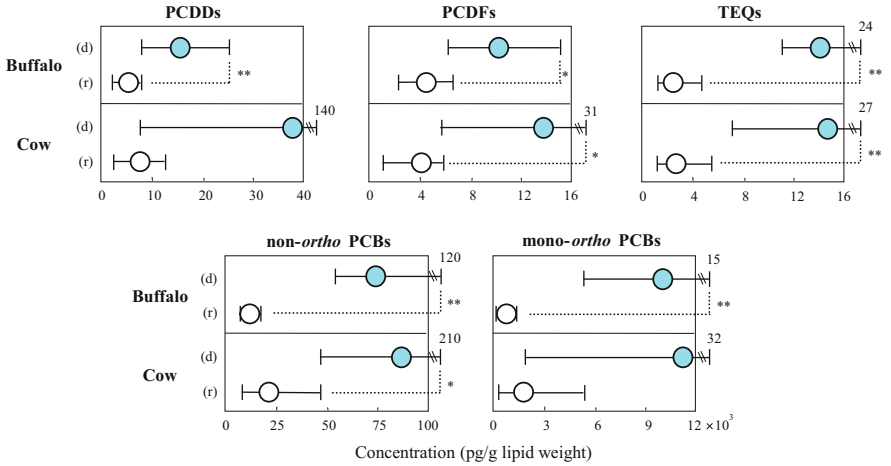

Fig. 7 Comparison of DRC concentrations in bovine milk from dumping (d) and reference (r) sites in India. The circles and bars represent mean and range values, respectively. $*p < 0.05$, $**p < 0.01$. Data were cited from Kunisue et al. [25]

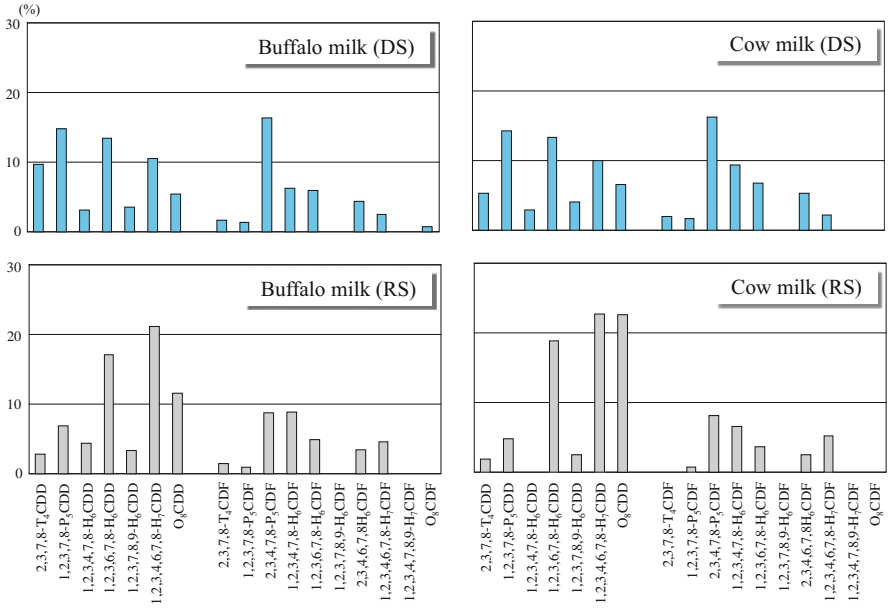

Fig. 8 Compositions of PCDD/Fs in bovine milk collected from the dumping and reference sites in India. DS and RS in the parentheses represent dumping site and reference site, respectively. Data were cited from Kunisue et al. [25]

that transfer to milk and storage in body fat increased with decreasing degree of chlorination, while excretion in feces increased with increasing degree of chlorination. Thus, bovines from the Indian DS transfer considerable amounts of low-chlorinated DD/Fs to their milk. Residents who constantly drink bovine milk are at high risk because of their high TEF values. In India, dietary consumption of dairy products is generally higher than other countries, and average consumption of milk in India by a person per day rose from 135 g in 1980 to 176 g in 1990 [56]. Assuming that an adult weighing 60 kg drinks 176 g of the buffalo or cow milk investigated in India per day, the estimated daily intake of TEQs from bovine milk from the DS was above 1 pg TEQs/kg/day, and only one buffalo milk sample had a value that exceeded the TDI proposed by the WHO [45] (Fig. 9). Even though the values are within TDI, the residents around the DS in India are exposed to considerably high levels of DRCs and hence may be at greater risk of exposure to these contaminants via bovine milk.

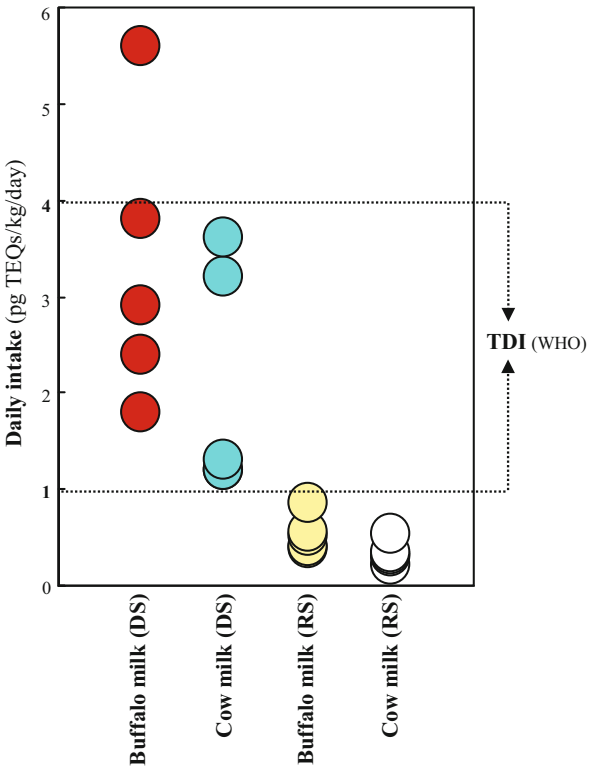

Fig. 9 Estimated daily intake of TEQs by adults through bovine milk collected from the dumping and reference sites in India. DS and RS in the parentheses represent dumping site and reference site, respectively. Daily intake was estimated using TEQ data in bovine milk reported by Kunisue et al. [25] and based on the assumption that an adult (60 kg) ingests 176 g of bovine milk per day (John et al. [56])

4.2 Fish

Recently, we have detected elevated concentrations of DRCs, especially DL-PCBs, in human breast milk collected from residents around a DS in Kolkata, India, which is located in the northeastern region and is the second largest city in India [57]. The TEQ levels were higher than those in human breast milk from the DS in Chennai described earlier [25] and recent levels in Japanese milk [23] (Fig. 10). These observations indicate that the magnitude of pollution by DRCs in Indian DS could be different domestically, and the residents around such DS ingest greater amounts of DRCs compared with general public in developed countries. Unlike the DS in Chennai, livestock animals such as buffalo and cows were not reared around the DS in Kolkata. However, there is a pond adjacent to the DS in Kolkata, and the interview with the DS residents showed that they consume fish collected from the pond. When the relationships between DRC concentrations in human breast milk and frequency of food consumption by the DS residents were examined, the DRC concentrations significantly increased with the frequency of fish consumption, but not with those of meat and dairy products [57]. Extremely high concentrations of DRCs (mean: 500 pg TEQs/g lipid wt.) were detected in fish collected from the pond adjacent to the DS, compared with those (31 pg TEQs/g lipid wt.) in fish collected from a RS pond. These results clearly suggest that fish consumption is a major source of DRCs for the DS residents in Kolkata. Furthermore, assuming that an adult weighing 60 kg eats 30 g of fish investigated in Kolkata per day, estimated daily intake of TEQs (4.6–16 pg TEQs/kg/day) from DRC concentrations in fish samples collected from the pond adjacent to the DS exceeded the WHO-TDI [45].

Considering the above observations, it is likely that not only residents are affected by DRCs, but also many other animals inhabiting the DS areas may be exposed to considerably high levels of DRCs derived from the DS and may suffer adverse effects.

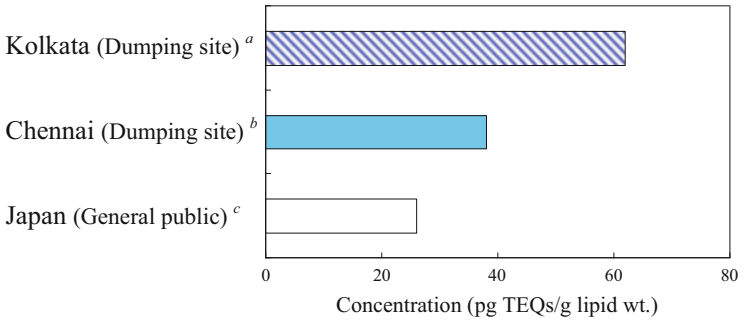

Fig. 10 Comparison of TEQ levels in human breast milk collected from the residents around dumping sites in Kolkata and Chennai and from the general public in Japan. [a]Someya et al. [57], [b]Kunisue et al. [25], [c]Kunisue et al. [23]

5 Animal Exposure and Toxicological Impacts

In the DS of Chennai in India, wild crows (house crow [*Corvus splendens*] and jungle crow [*Corvus macrorhynchos*]) and pigs (*Sus scrofa*) feed on the raw garbage with contaminated soils (Fig. 11). Because biomagnification of DRCs through the contaminated garbage and further adverse effects on these animals are speculated, our group clarified the contamination levels and accumulation features of DRCs and assessed the biochemical effects in crows and pigs inhabiting the DS [58, 59].

Fig. 11 Photos of crows and pigs in Perungudi dumping site, Chennai city, India

5.1 Crow

5.1.1 Accumulation Patterns

DRCs in pectoral muscle samples of crows collected from the DS and a RS in Chennai were analyzed. As with humans and bovine described earlier, the concentrations of DRCs detected in the DS crows were significantly higher than those from the RS [58]. Another study showed that DRC concentrations in general population of various animals from India were lower than those reported in Japan [60]. However, the magnitude of DRC contamination in crows from the Indian DS was significantly greater than those from Japan [58], suggesting that crows in the DS have been exposed to these contaminants derived from burning wastes by their DS feeding activity. When the compositions of DRCs were examined, the profiles observed in DS crows were similar to those in soils collected from the DS [7], but not for those in crows from the RS. This result indicates that crows inhabiting the DS ingest contaminated soil together with raw garbage. A further scale-dependent analysis, principal component analysis (PCA), supported that the DRC profiles in crows from the DS were influenced by DRC congeners present in the DS soils [58].

5.1.2 Bioconcentration

To verify whether or not the DRC congeners in crows from the DS were directly affected by on-site contamination, bioconcentration factors (BCFs) of PCDD/F congeners were estimated from concentrations in crows and soils from the DS, and compared with the theoretical BCF values, which were calculated from water-particle and lipid-water partitioning coefficients. BCFs of individual congeners in crows from the DS were calculated as the ratios of concentrations in crow muscles (C_{lipid} on lipid-weight basis) to the concentration in soils (C_{particle} on dry weight basis); $\text{BCF}_{\text{measured}} = C_{\text{muscle}}/C_{\text{soil}}$. In addition to BCFs based on the congener concentrations in muscle and soil, theoretical BCFs ($\text{BCF}_{\text{theoretical}}$) were calculated assuming that transfer of congeners contributed to the body of crow from the DS was dependent upon their partitioning between soil particle and lipid in tissue. The partition of individual congeners between particle and lipid was estimated using the following formula:

$$\text{BCF}_{\text{theoretical}} = \frac{C_{\text{muscle}}}{C_{\text{soil}}} = \frac{C_{\text{lipid}}}{C_{\text{particle}}} = \frac{C_{\text{water}}}{C_{\text{particle}}} \times \frac{C_{\text{lipid}}}{C_{\text{water}}} = \frac{1}{K_{\text{pw}}} \times K_{\text{bw}},$$

where particle-water partition coefficient (K_{pw}) and biotic lipid-water partition coefficient (K_{bw}) were referred from Govers and Krop [61]. Regression analyses revealed that the averages of log $\text{BCF}_{\text{measured}}$ calculated in the DS crows were positively correlated with log $\text{BCF}_{\text{theoretical}}$ (Fig. 12). This result elucidated that

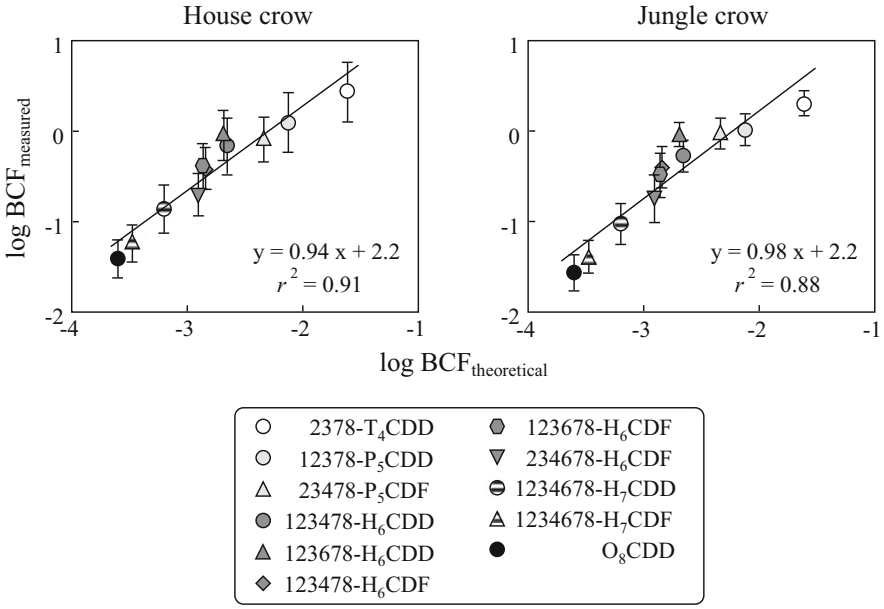

Fig. 12 Relationships between log $BCF_{measured}$ and log $BCF_{theoretical}$ for PCDD/Fs in crows from the Indian dumping site. Each plot and bar means average and standard deviation, respectively. Data were cited from Watanabe et al. [58]

PCDD/F congener profiles in the muscle of crows from the DS were mostly determined by the soil particle-lipid partitioning, based on the physicochemical characteristics of each congener. However, the $BCF_{measured}$ was approximately 10^2-fold higher than the $BCF_{theoretical}$. Some factors including intake pathway and diet composition might have influenced the distribution of PCDD/Fs in crows. The K_{bw} used for estimating $BCF_{theoretical}$ was calculated from the experimental data on fish exposed to PCDD/Fs dissolved in water [61]. In fish, the major entry route of xenobiotics dissolved in water is a direct pathway across the blood-water interface at the gills, and uptake of xenobiotics in the diet can be ignored when estimating internal concentration of xenobiotics [62]. Conversely, oral intake of contaminated soil is the main route for crows. Hack and Selenka [63] suggested that the action of enzymes on alimentary lipids and the potential of bile to form mixed micelles with fatty acids and monoglycerides enhance xenobiotic mobilization to a high degree in a gastrointestinal model. Such gastrointestinal absorption could contribute to higher $BCF_{measured}$ than $BCF_{theoretical}$. Higher organic content in particles can result in strong binding of 2,3,7,8-T_4CDD onto the particle, lowering its bioavailability [64]. When K_{pw} was calculated, sediment was substituted for the particle phase [61]. The sediment might contain higher organic content than soil from the DS. Higher K_{pw} leads to lower $BCF_{theoretical}$. Furthermore, Stephens et al. [65] reported that PCDD/Fs in soils ingested by chicken are readily absorbed and are bioaccumulated in the tissues. Their estimated BCFs ($BCF_{chicken}$) of PCDD/Fs from

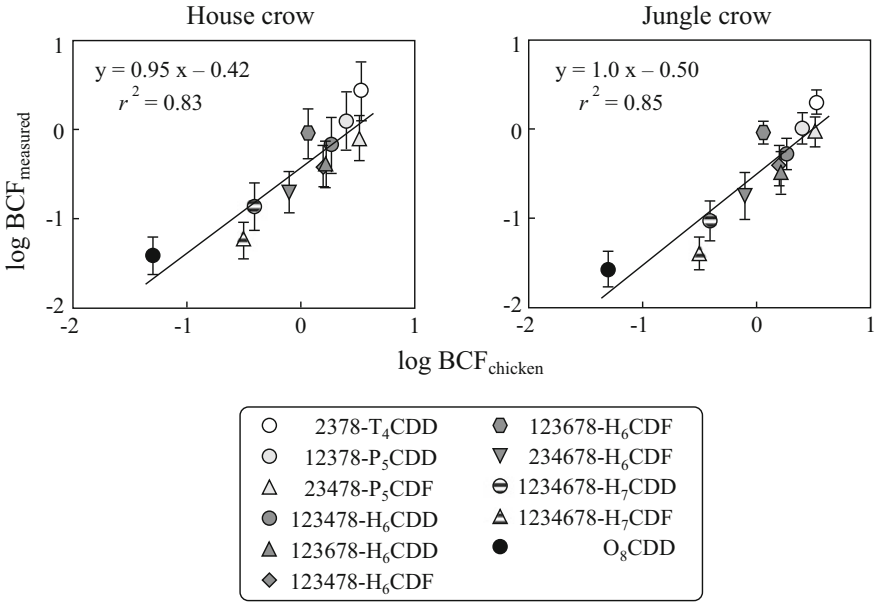

Fig. 13 Relationships between log $BCF_{measured}$ and log $BCF_{chicken}$ for PCDD/Fs. Each plot and bar means average and standard deviation, respectively. Data of $BCF_{measured}$ and $BCF_{chicken}$ were cited from Watanabe et al. [58] and Stephens et al. [65], respectively

soils to thigh muscle after feeding a diet mixed with 10% of highly contaminated soil for 80 days were generally consistent with our data (Fig. 13), supporting that crows in the DS consume soil with raw garbage.

Lower BCFs for congeners with larger molecules such as H_7- and O_8-CDDs/Fs were shown in Figs. 12 and 13. This may be due to the lower uptake efficiency of these congeners through the gastrointestinal tract. A previous study for wild tufted ducks reported that biomagnification factors (BMFs) of PCDD/F congeners tended to decrease with their K_{ow} [66]. In humans, the net absorption of PCDD/F congeners is likely to be diminished with the degree of chlorination [67]. Mean log BCF values in the DS crows had significant negative correlations with log K_{ow} and molecular weight of PCDD/F congeners (Fig. 14). Opperhuizen and Sijm [68] pointed out a lack of membrane permeation for hydrophobic chemicals with widths over 0.95 nm. In wild common cormorants, congeners with large molecules such as H_7CDD/F and O_8CDD showed no life-stage-dependent accumulation probably because of gastrointestinal barrier, whereas T_4- to H_6-CDDs and P_5- and H_6-CDFs showed significant increase with growth [27]. The results shown in Fig. 14 clearly demonstrate that molecular configuration may limit the dietary uptake of PCDD/F congeners in crows.

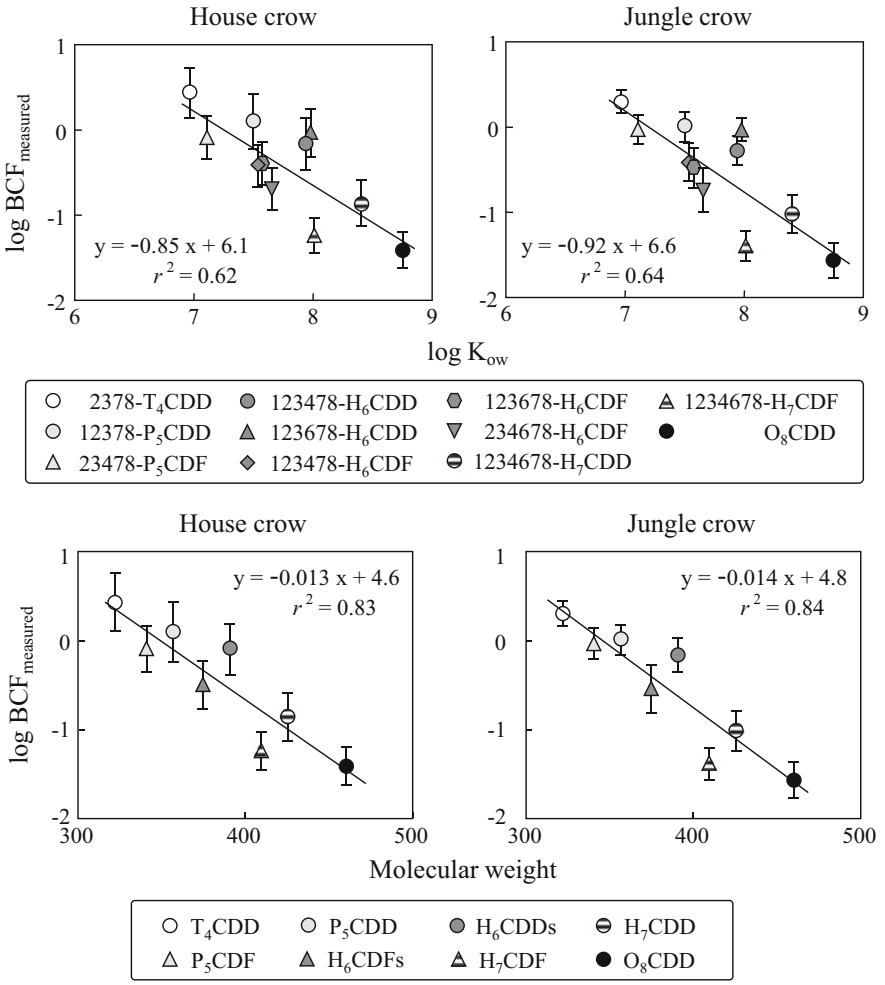

Fig. 14 Relationships between log $BCF_{measured}$ for PCDD/Fs in crows from the DS and log K_{ow} or molecular weight of each congener. Each plot and bar means average and standard deviation, respectively. Data were cited from Watanabe et al. [58]

5.2 Pig

5.2.1 Accumulation Features

As in the case of crows described above, pigs inhabiting the DS in Chennai also feed on raw garbage with contaminated soils (Fig. 11), and hence considerable exposure to DRCs and adverse effects on their health are expected. Our group recently examined exposure levels, accumulation features, and toxicological effects of DRCs by analyzing samples of liver, abdominal fat, muscle, and plasma

collected from pigs in the DS and a RS in Chennai [59]. Concentrations of DRCs in tissue samples from the DS pigs were significantly higher than those from the RS. In addition, similar DRC congener profiles between pig tissue and soil from the DS were shown. These observations support that wild animals, such as crows and pigs, in the DS are highly exposed to these contaminants through ingestion of on-site garbage contaminated with soil.

5.2.2 Relationships with Hepatic Cytochrome P450 Enzymes

Availability of fresh liver from the pigs enabled the measurement of cytochrome P450 (CYP) 1A1, CYP2B1, and CYP4A1 by immunoblotting assays. A single band of protein cross-reacted with each antibody around the corresponding rat CYP standard was detected in pig hepatic microsomes (Fig. 15). When the relationships between TEQs and CYP1A-, CYP2B-, or CYP4A-like protein levels were examined in the pig liver, hepatic TEQs (wet weight basis) were positively correlated with the levels of CYP1A-like protein ($p < 0.05$, Fig. 16). Induction of CYP1A enzymes through the aryl hydrocarbon receptor (AhR) has been used extensively as a sensitive indicator of exposure and effects of DRCs. Schmitz et al. [69] and Zeiger et al. [70] reported no-observed-effect level (NOEL) and half-maximum induction (EC_{50}) values for TCDD-induced EROD in HepG2 cells, H4IIE cells, and Wistar rat primary hepatocytes; the EC_{50} values were 220, 16, and 6.4 pg/ml, and NOEL values were 11, 0.064, and 0.013 pg/ml, respectively. TEQs (mean $\pm$ SD: 3.9 ± 3.2 pg/g wet weight) in the liver of all pigs from India were higher than the NOELs for EROD in H4IIE and rat primary hepatocytes, but were lower than that in HepG2. Silkworth et al. [71] reported that human hepatocytes are about 10–1,000 times less sensitive for CYP1A induction by certain DRC congeners than rat and monkey cells. These observations suggest that pigs may be more sensitive to

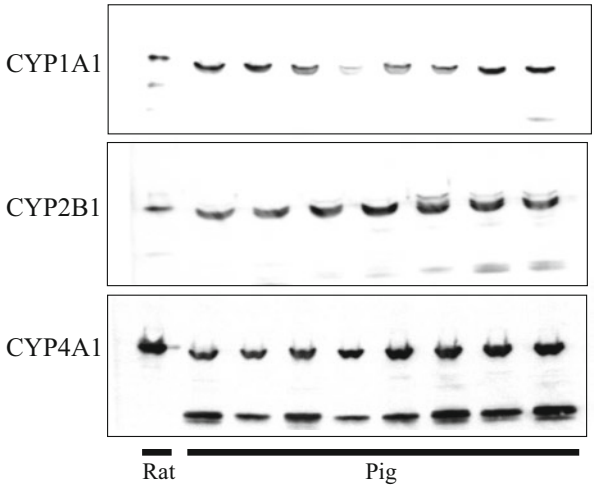

Fig. 15 Results of immunoblot analyses of pig hepatic microsomes using anti-rat CYP1A1, CYP2B1, and CYP4A1 polyclonal antibodies. Data were cited from Watanabe et al. [59]

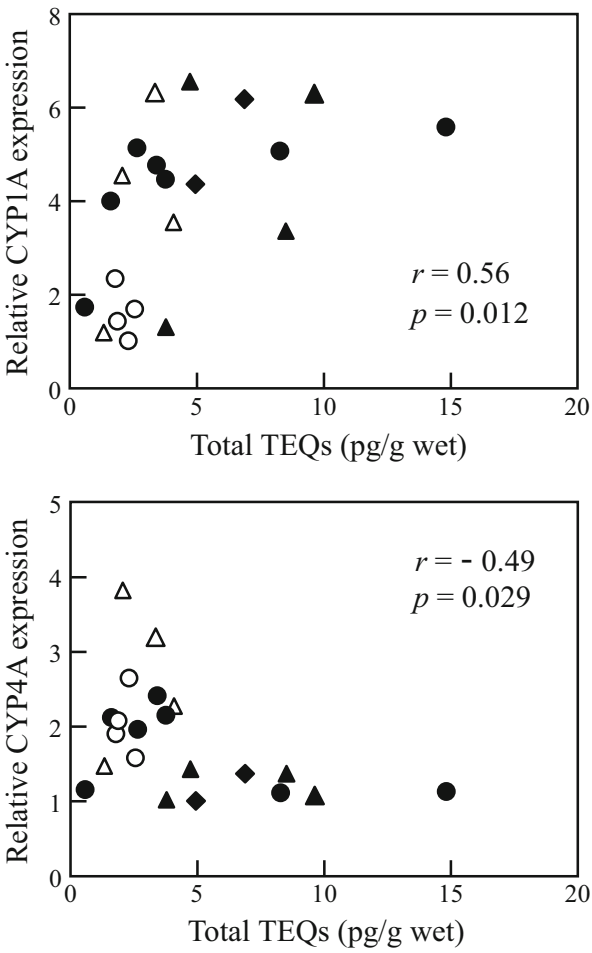

Fig. 16 Relationships between hepatic TEQs and expression levels of CYP1A- or CYP4A-like protein in the liver microsomes of pigs from India. *Filled circle* = female from the dumping site; *filled triangle* = male from the dumping site; *open circle* = female from the reference site; *open triangle* = male from the reference site; *filled diamond* = piglets from the dumping site. Data were cited from Watanabe et al. [59]

CYP1A induction by DRCs than humans. On the other hand, CYP4A-like protein content was negatively correlated with TEQ levels in the pig liver ($p < 0.05$, Fig. 16), whereas CYP2B-like protein revealed no correlation with hepatic TEQs. The negative correlation between hepatic CYP4A-like protein and TEQ levels is consistent with a previous study which reported the suppression of the CYP4A protein in rat treated with AhR ligand [72]. Koga et al. [73] demonstrated decreased hepatic CYP4A1 expression in rat treated with CB-77. Given that CYP4A1 is induced through peroxisome proliferator-activated receptor α (PPARα), it is likely that DRCs have a potential to affect PPARα-signaling pathways in pigs. Disruption of PPARα-signaling pathway may pose health hazards, as PPARα is involved in development, physiology, and inflammatory response [74].

5.2.3 Hepatic Sequestration

On the lipid-weight basis, the concentrations of PCDD/F and non-*ortho* DL-PCB congeners in the liver were higher than those in the adipose and muscle tissues of pigs, while the mono-*ortho* DL-PCB congener levels were almost similar among these different tissues. This result suggests the lipid-dependent accumulation of mono-*ortho* DL-PCB congeners and the specific binding of PCDD/F and non-*ortho* DL-PCB congeners to proteins. Table 2 shows liver/adipose concentration ratios (L/A ratios on lipid-weight basis) and their relationship with hepatic CYP1A-like protein content. L/A ratios of most PCDD/F congeners were significantly positively correlated with CYP1A-like protein content ($p < 0.05$), indicating that CYP1A is involved in the hepatic sequestration of these congeners. The ratios for all mono-*ortho* DL-PCB congeners were near 1.0, which means no hepatic sequestration of these congeners. As for PCDD congeners, the L/A ratios increased with an increasing number of chlorine substitutions. Similar trends for PCDD/F congeners were reported in our earlier investigations on wild animals [27, 28, 75–77]. A possible mechanism to explain dose-dependent hepatic sequestration is the induction of hepatic microsomal protein, CYP1A2, and the subsequent binding of PCDD/F congeners to this protein. Comparisons between DRC-dosed CYP1A2 knockout and parental strains of mice provided direct evidence that CYP1A2 was the target protein for the binding of 2,3,7,8-T_4CDD, 1,2,3,7,8-P_5CDD, and 2,3,4,7,8-P_5CDF in the liver, but not for CB-153 [78]. Interspecies comparison of the L/A (or liver/muscle) ratios showed that the capacity of hepatic sequestration of DRCs in pigs was comparable to that in raccoon dog [28], but higher than those in Baikal seal [75], common cormorant [27], and jungle crow [77].

5.2.4 Maternal Transfer

To understand the maternal transfer of DRC congeners in pigs, the hepatic concentrations of DRCs in a dam-piglets pair (two piglets and their dam) from the DS were measured and compared. Concentration ratios of DRCs between piglets and their dam (piglets/dam) exceeded 1.0, and especially the congeners with a molecular weight between 360 and 400 were detected at higher concentrations in piglets than in their dam (Fig. 17); this shows maternal transfer of DRCs. Such transfers of DRCs from dams to neonates via milk have been considered to be more significant than placental transport [79]. Iwata et al. [75] showed significant declines in 2,3,7,8-T_4CDD, 1,2,3,7,8-P_5CDD, 2,3,4,7,8-P_5CDF, CB-126, CB-169, and CB-157 concentrations with age in the liver of wild female Baikal seals, suggesting that these congeners are easily eliminated through lactation. Low molecular weight congeners with a molecular weight less than 360, however, might be metabolized by the piglets' hepatic CYP1A, whose expressions were as high as in the adults (Fig. 16). This would explain lower concentration ratios (piglets/dam) for lower molecular weight congeners than for those with 360 to 400 molecular weights.

Table 2 Liver to adipose concentration ratios (L/A, lipid basis) of DRCs and their relationships with hepatic CYP1A-like protein contents in pigs from India

Congener	n[a]	L/A concentration ratio		Relationship between CYP1A and L/A	
		Mean	Range	S[b]	r^2
PCDDs					
2,3,7,8-T_4CDD	3	5.1	2.2–7.8	na	na
1,2,3,7,8-P_5CDD	7	14	0.43–63	10	0.61*
1,2,3,4,7,8-H_6CDD	9	21	2.9–55	8.2	0.62*
1,2,3,6,7,8-H_6CDD	12	19	0.47–83	8.5	0.39*
1,2,3,7,8,9-H_6CDD	4	18	4.4–31	na	na
1,2,3,4,6,7,8-H_7CDD	12	55	2.8–210	21	0.35*
O_8CDD	12	91	4.2–210	23	0.44*
PCDFs					
2,3,7,8-T_4CDF	4	4.2	2.2–7.0	na	na
1,2,3,7,8-P_5CDF	0	na	na	na	na
2,3,4,7,8-P_5CDF	11	130	3.9–400	62	0.66**
1,2,3,4,7,8-H_6CDF	11	73	1.7–270	37	0.61**
1,2,3,6,7,8-H_6CDF	9	93	2.4–320	42	0.57*
1,2,3,7,8,9-H_6CDF	0	na	na	na	na
2,3,4,6,7,8-H_6CDF	5	85	5.2–190	39	0.45
1,2,3,4,6,7,8-H_7CDF	12	120	4.3–360	44	0.54**
1,2,3,4,7,8,9-H_7CDF	1	55	55–55	na	na
O_8CDF	7	360	13–1,000	140	0.35
Non-ortho DL-PCBs					
PCB-77	12	7.5	0.35–77	1.2	0.008
PCB-81	10	8.7	2.3–25	2.3	0.16
PCB-126	12	24	2.4–110	9.9	0.32
PCB-169	12	2.7	0.73–10	0.77	0.24
Mono-ortho DL-PCBs					
PCB-105	12	1.7	0.24–10	−0.17	0.010
PCB-114	12	0.82	0.26–2.9	0.0017	<0.001
PCB-118	12	1.0	0.24–4.2	0.0061	<0.001
PCB-123	9	1.0	0.35–3.0	−0.12	0.026
PCB-156	12	0.73	0.36–1.6	−0.0031	0.013
PCB-157	12	0.85	0.39–1.9	0.028	0.009
PCB-167	12	0.89	0.21–1.9	0.19	0.38*
PCB-189	12	0.81	0.14–1.9	0.077	0.047

[a]The number of specimens in which the congener was detected both in the liver and adipose tissue
[b]S = the slope of the following equation; Concentration ratio (liver/adipose) = (CYP1A-like protein content) × S + intercept. This calculation was done when n was 5 or more
*$p < 0.05$, **$p < 0.01$; *na* no data available

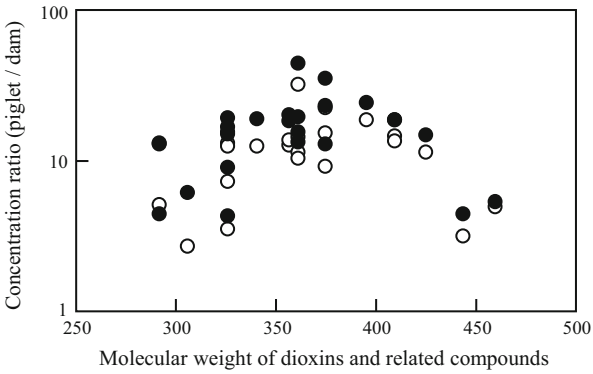

Fig. 17 Relationship between molecular weight of dioxins and related compounds (DRCs) and concentration ratios (piglets/dam) of DRCs in the liver of pigs from the dumping site. *Solid and open circles* represent different piglets whose mother is the same individual. Data were cited from Watanabe et al. [59]

Iwata et al. [75] also reported poor elimination of H_7CDD to O_8CDD congeners in aged mothers, suggesting less excretion of such highly chlorinated congeners through lactation. Data from Van den Berg et al. [80] also support the results, showing decreased excretion rates via dam milk with increasing chlorine content.

5.2.5 Biochemical Effects

To assess the biochemical effects in pigs inhabiting the DS, concentrations of plasma hormones, immunoglobulins, and vitamin A were measured and compared to those in the RS pigs. Interestingly, plasma immunoglobulin G (IgG) levels were significantly lower in male pigs from the DS than those from the RS ($p < 0.05$, Fig. 18). The immune system is one of the most sensitive targets of 2,3,7,8-T_4CDD [81], and plasma IgG is reported to be suppressed by 2,3,7,8-T_4CDD exposure through the suppression of antigen-responding B-cell proliferation during germinal center formation in mice [82]. In humans from Seveso, Italy, plasma 2,3,7,8-T_4CDD concentrations (3.5–90 pg/g lipid) were negatively associated with plasma IgG concentrations [83]. TEQs in pigs (170 ± 87 pg/g lipid in males and 110 ± 110 pg/g lipid in females) from the DS were similar to those reported for the Seveso population, indicating that DRCs may affect the immune system in the DS pigs. A significant difference between the DS and RS was also observed for plasma-free thyroxine (FT_4) levels in females (Fig. 18). When all specimens were analyzed together (plasma levels of FT_4 had no significant difference between gender), a decrease in plasma FT_4 in the pigs from the DS was detected ($p = 0.039$), compared with those from the RS. The competitive binding of DL-PCBs and T_4 to transthyretin and glucuronidation of T_4 by dioxin-inducible UDP-glucuronyl transferase may account for the decrease of FT_4 in the DS pigs. Correlation analyses between hepatic TEQs and plasma hormone levels showed no specific patterns.

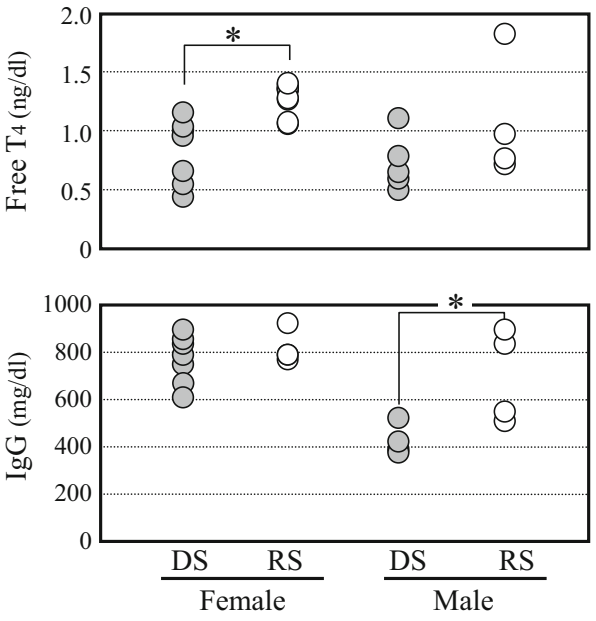

Fig. 18 Comparison of plasma-free thyroxine (T_4) and immunoglobulin G (IgG) levels between sampling sites in the pigs from India. DS and RS represent dumping and reference sites, respectively. $^*p < 0.05$

5.2.6 Hydroxylated Metabolites

We have recently analyzed hydroxylated metabolites of PCBs (OH-PCBs) in the blood of pigs from India and found higher concentrations of OH-PCBs in the DS pigs, especially piglets, than in the RS pigs [84]. In addition, OH-PCB concentrations in the blood were positively correlated with hepatic CYP1A-like protein content ($p < 0.01$), indicating the CYP1A-dependent formation of OH-PCBs in the liver and the subsequent retention of these metabolites in the blood of pigs. Considering that hepatic levels of DRCs and CYP1A expression were higher in the DS pigs, as described earlier, OH-PCBs in the DS pigs could be preferentially formed from PCBs through CYP1A-mediated metabolism induced by DRC exposure. Thus, DRCs in the liver of DS pigs pose effects on hepatic CYP1A and CYP4A expression and are probably sequestered by the induced CYP1A protein, and subsequently hydroxylated metabolites of xenobiotic chemicals including OH-PCBs are formed. Plasma IgG and T_4 levels may also be affected by DRCs accumulated in the DS pigs. Swine is considered as a prospective model animal to predict bioavailability and biotransformation of environmental chemicals in humans, due to the similarities of gastrointestinal tract function [85], nutritional requirements [86], and CYP activities [87]. The similar phenomena observed in the pigs from the Indian DS may arise in the residents living around the DS, and hence more attention should be paid for the human risk from not only DRCs but also hydroxylated metabolites, which are formed by DRC-induced CYP1A protein. Management of the DS is crucial to protect the health of the inhabiting wild animals and humans.

6 Conclusions and Future Consideration (E-Wastes)

Recent studies have demonstrated that DS in Asian developing countries is a potential source of DRCs. Levels, profiles, and estimated fluxes of DRCs observed for soils in DS suggested that these contaminants are formed by uncontrolled burning of solid waste, and DS in India and the Philippines may be a significant reservoir for DRCs. In India, the concentrations of DRCs in human breast milk from two DS in Chennai and Kolkata were significantly higher than those from RS and other Asian developing countries, and elevated levels of these contaminants were observed in bovine milk collected from buffalo and cows feeding in the Chennai DS and in fish collected from a pond adjacent to the Kolkata DS. These results indicate that residents around these DS have been exposed to considerably high levels of DRCs, probably through the intake of contaminated bovine milk and fish. Wild crows and pigs inhabiting the DS in Chennai were also contaminated by DRCs, and direct transfer of these contaminants from contaminated soil was suggested. In addition, the study on pigs suggested that DRCs pose effects on hepatic CYP1A and CYP4A expression and are probably sequestered by the induced CYP1A protein. Plasma IgG and T_4 levels may also be affected by DRCs in pigs from the DS. Because there is no control and measure of DRC release in DS, it can be anticipated that pollution by DRCs will become exacerbated further. In view of these observations, we suggest that further investigations on the pollution sources and animal exposure of DRCs in Asian developing countries, especially DS, are needed to elucidate future pollution trends and to assess the health risk to humans and wildlife.

In recent years, increasing activities of electrical and electronic waste (e-waste) recycling in developing countries have received international attention, because of the emission of toxic chemicals resulting from the uncontrolled recycling processes of e-waste facilities. In Asia, environmental fate and human exposure of hazardous substances released from e-waste recycling sites (EWRS) have been extensively investigated in China, where e-waste recycling plays an important economic role, since 2000. EWRS have been identified as hotspots of not only polybrominated diphenyl ethers (PBDEs) which are contained in e-wastes as flame retardants but also DRCs such as PCDD/Fs and polybrominated dibenzo-*p*-dioxins/furans (PBDD/Fs) [88]. Despite the presence of EWRS in developing countries, researches on DRCs in EWRS are exceedingly limited in Asian countries other than China. Our group has recently found significantly higher concentrations of PCDD/Fs, PBDD/Fs, and DL-PCBs in dust from two EWRS in North Vietnam compared with those from an urban site (Hanoi) and suggested a substantial release of these DRCs by recycling activities [89]. Furthermore, dioxin-like activities in the extract of EWRS dust, estimated using the dioxin-responsive chemically activated luciferase gene expression (DR-CALUX) assay, were also greater than those in the urban dust, and higher percentage of unknown dioxin-like activities was observed in the dust extract, indicating large contribution from unidentified DRCs (other than PCDD/Fs, PBDD/Fs, and DL-PCBs) [88]. Given the above results, the role of "e-wastes" as a significant source of DRCs to the environment should be urgently elucidated in Asian developing countries.

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