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POPs in Free-Range Chicken Eggs from Indonesia

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1 Introduction

Eggs have been found to be sensitive indicators of persistent organic pollutants (POPs) contamination in soils and are an important exposure pathway from soil pollution to humans. Eggs from contaminated areas can readily lead to exposures which exceed thresholds for the protection of human health[1-5]. Chickens and eggs might therefore be considered ideal "active samplers" and indicator species for POPs contaminated sites. In this study, we sought to broaden the available data on POPs contamination at selected sites in Indonesia. It is based on larger reports released in 2020-2024, where more information about the sites can be found[6-10]. The analyses focused on POPs listed[11-13] and/or nominated[14] to be listed under the Stockholm Convention. Technical chemicals intentionally used in industrial processes and consumer products, very often as additives to plastic or paper packaging[9,15,16] as well as unintentionally generated POPs as by-products in industrial processes and during combustion of waste[9] were chosen to be analyzed in eggs for this study.

Many of the POPs selected for analyses in this study are endocrine disruptors[17,18] or carcinogens[19]. All of them accumulate in animals, mostly in fatty tissues[20], with exemption of per- and polyfluoroalkyl substances (PFASs) which bind to proteins[21]. Their presence in food of animal origin such as eggs can cause severe health problems. This study highlights the level of POPs contamination and food safety in Indonesia.

2 Materials and Methods

Nineteen pooled samples of free-range chicken eggs were collected from seven localities in Java, with an additional sample obtained from Morowali in Central Sulawesi. Two reference samples were purchased from a supermarket in Jakarta and a convenience store in Karawang, respectively. In this study, the number of individual eggs in each pooled sample ranged from 2 to 9.

All samples were analyzed for their content of polychlorinated dibenzo-p-dioxin and dibenzofuran (PCDD/F) and dioxin-like polychlorinated biphenyl (dl PCB) congeners and six samples also for polybrominated dibenzo-p-dioxins and dibenzofurans (PBDD/Fs) using GC/HRMS in an ISO 17025 accredited laboratory[1], with a resolution >10,000 using ^{13}C isotope labeled standards. The analysis of PCDD/Fs and dl PCBs in eggs followed the methods prescribed for controlling levels of these substances in foodstuffs according to EU regulations[22]. The results are presented in pg WHO-TEQ/g fat. Toxic equivalency factors (TEFs) defined in 2005[23] were used to evaluate dioxin toxicity in all samples.

Analyses of polybrominated diphenyl ethers (PBDEs), 3 isomers of hexabromocyclododecane (HBCD), tetrabromobisphenol A (TBBPA), six novel brominated flame retardants (nBFRs)[2], 16 PFASs[3], short and medium chain paraffins (SCCPs, MCCPs), 13 polychlorinated naphthalene (PCN) congeners, two stereoisomers of dechlorane plus (DP), DDT and its metabolites, 3 stereoisomers of hexachlorocyclohexane (HCH), pentachlorobenzene (PeCB), hexachlorobenzene (HCB), hexachlorobutadiene (HCBD), and seven non dioxin-like PCB (ndl PCBs) congeners. Analyses for other POPs than PCDD/Fs and dl PCBs were conducted in a Czech certified laboratory at the Department of Food Chemistry and Analysis of the University of Chemistry and Technology in Prague. Some of the analyzed chemicals were not included in this study (see Tables 1 and 2), but they are included in previous reports, their levels were rather low or below LOQ.[6-10]

3 Results

Results of chemical analyses for pooled samples of free-range chicken eggs are summarized in Tables 1 and 2. The results of POPs analyzed in chicken egg samples were also compared with EU and Indonesian standards set for eggs as foodstuff.[24-26]

Table 1: Results of the POPs analyses in pooled egg samples from various locations in Java and from Morowali, Sulawesi – first set of chemicals (unintentionally produced POPs and chlorinated paraffins). Levels exceeding limit values set for eggs as food in EU are marked in yellow.

[1] State Veterinary Institute in Prague and Muenster Analytical Solutions (MAS)
[2] 1,2-bis(2,4,6-tribromophenoxy) ethane (BTBPE), decabromodiphenyl ethane (DBDPE),hexabromobenzene (HBB), octabromo-1,3,3-trimethylpheny-1-indane (OBIND),2,3,4,5,6-pentabromoethylbenzene (PBEB), and pentabromotoluene (PBT)
[3] PFBA, PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnDA, PFDoDA, PFTrDA, PFTeDA, PFBS, PFHxS, PFOS, PFDS, PFOSA

Locality	Sample ID (eggs)	N	Fat	PCDD/Fs	dl PCBs	PCDD/F/dl PCBs	PeCB	HCB	SCCPs	MCCPs
-	Units		%	pg TEQ/g fat			ng/g fat			
Bangun	BAN-E-1	3	9.45	9.5	5.1	14.6	2.2	3.6	97	NA
	Bangun 1	3	13	10.8	3.1	13.9	1.10	2.7	153	NA
Bantar Gebang	B-EEG-1	6	17.80	10.0	4.3	14.3	1.25	3.9	309	2,345
	B-EGG-4	6	21.50	3.8	2.0	5.80	0.36	0.89	<50	<100
	BEK-EGG-03/22	2	16.1	20	7.4	27.4	0.97	41	NA	NA
	BEK-EGG-04/22; B-EGG-3	4	17.90	34	20	54	3.1	5.3	<50	1,921
	BEK-EGG-05/22	2	15.5	20	8.4	28.4	0.74	1.96	NA	NA
	EGG-DUMPSITE	2	14.8	14.6	7.8	22.4	2.1	3.7	NA	NA
Karawang	KAR-EGG-01/22	3	14.90	3.2	1.1	4.3	0.85	1.79	<50	732
	KAR-EGG-1	4	13.80	11	1.7	12.7	0.70	4.6	<50	NA
	KAR-EGG-2	4	13.50	178	34	212	6.1	16	69	NA
	KAR-EGG-3	4	16.70	109	10	119	3.5	4.8	237	NA
Kendalsari	KEN 01	9	27.38	49	35	84	1.07	1.5	NA	NA
	KEN-E-1/19	6	14.29	41	20	61	1.28	2.5	160	NA
Morowali	MWL-EGGS-1	3	11.21	5	1.1	6.1	NA	NA	<50	<100
Tangerang	SEM-E-1	3	16.22	54	18	72	3.6	6.1	153	NA
	TAN-ESIN-01	5	13.69	20.4	7.4	27.8	NA	NA	NA	NA
Sumberw.	SUM-E-1_2	6	14.12	11	2	13	0.26	0.58	50	NA
Tropodo	TROP-E-1	6	13.89	140	32	172	1.67	4.1	97	NA
	Tropodo 1	3	15	200	32	232	1.90	5.5	65	NA
Minimum	-	-	-	3.2	1.1	4.3	0.26	0.58	<50	<100
Maximum	-	-	-	200	35	232	6.1	41	309	2,345
EU/Indonesia limit[24,25,27]	-	-	-	2.5	-	5.0/2.5	-	-[4]	-	-
Reference samples	JAK-SUP	6	9.53	0.0012	0.0020	0.0032	<0.1	<0.1	136	NA
	KAR-EGG-R	3	17.40	0.23	0.02	0.25	5	1.9	151	NA

Table 2: Results of the POPs analyses in pooled egg samples from various locations in Java and from Morowali, Sulawesi – second set of chemicals (flame retardants and PFASs). Levels exceeding limit values set for eggs as food in EU are marked in yellow.

Locality	Sample ID (eggs)	sum HBCD	sum of PBDEs	nBFRs	DP	PFASs	PFOA	PFOS	PFHxS	EFSA-PFASs
-	Units	ng/g fat				ng/g ww				
Bangun	BAN-E-1	538	1,457	124	NA	97	0.05	92	0.048	93
	Bangun 1	5.19	91	NA	NA	26	0.39	17.7	0.058	19
Bantar Gebang	B-EEG-1	<4.2	8	<LOQ	<0.3	1.18	0.012	0.305	0.079	0.44
	B-EGG-4	<4.2	6.3	3.62	<0.3	1.49	<0.006	0.863	0.019	0.92
	BEK-EGG-03/22	<4.2	28	5.46	2.32	4.2	0.014	1.90	0.007	2
	BEK-EGG-04/22; B-EGG-3	<4.2	10	<LOQ	<0.3	2.8	0.031	1.74	0.024	1.86
	BEK-EGG-05/22	<4.2	3.2	<LOQ	<0.3	1.28	0.032	0.376	0.018	0.47
	EGG-DUMPSITE	5.0	31	1.01	2.72	6.4	0.11	4.5	0.046	4.8
Karawang	KAR-EGG-01/22	<4.2	10	<LOQ	1.99	0.67	<0.006	0.359	0.006	0.39
	KAR-EGG-1	48	<LOQ	<LOQ	NA	0.85	0.014	0.14	<0.006	0.18
	KAR-EGG-2	39	73.00	18.39	NA	4.2	0.032	1.05	0.021	1.20

[4] There is a maximum residues level set in EU for HCB as 20 ng/g ww, none of the samples, exceeded this level.

	KAR-EGG-3	36	<LOQ	<LOQ	NA	2.3	0.014	0.31	<0.006	0.37
Kendal-sari	KEN 01	<4.2	6.19	<LOQ	NA	NA	NA	NA	NA	NA
	KEN-E-1/19	<4.2	149.58	12.23	NA	0.35	<0.01	0.14	<0.01	0.16
Morowali	MWL-EGGS-1	<4.2	10.8	<LOQ	<0.3	NA	NA	NA	NA	NA
Tangerang	SEM-E-1	844	320.77	33	NA	6.2	0.27	2.5	0.03	3.3
Sumberw.	SUM-E-1_2	4.47	8.21	0.87	NA	0.46	0.01	0.26	<0.01	0.29
Tropodo	TROP-E-1	<4.2	27,159	2,166	NA	0.30	<0.01	0.14	<0.01	0.16
	Tropodo 1	<4.2	65	NA	NA	2.7	0.10	1.04	<0.01	1.3
Minimum	-	<4.2	<LOQ	<LOQ	<0.3	0.30	<0.006	0.14	<0.006	0.16
Maximum	-	844	27,159	2,166	2.72	97	0.39	92	0.079	93
EU limit[26]	-	-	-	-	-	-	0.30	1.0	0.30	1.7
Reference samples	JAK-SUP	<4.2	1.39	<LOQ	NA	0.1	<0.01	<0.01	<0.01	<0.01
	KAR-EGG-R	<4.2	<LOQ	<LOQ	NA	0.05	0.007	<0.006	<0.006	0.007

Results are compared with limit values set for POPs in eggs as food in Tables 1 and 2 as well. All free-range eggs samples exceeded an EU limit for PCDD/Fs of 2.5 pg WHO-TEQ/g fat[25] and also Indonesian limit for PCDD/Fs/dl PCBs of 2.5 pg WHO-TEQ/g fat[24]. One sample exceeded limit of 0.3 ng/g ww set in the EU for PFOA[26], eight samples exceeded the limit value of 1 ng/g ww set for PFOS[26], and six samples exceeded limit of 1.7 ng/g ww set for 4 PFASs[26] evaluated by the European Food Safety Authority (EFSA)[28]. Only the limit values of 0.3 ng/g ww for PFHxS[26] was not exceeded in any of analyzed pooled free-range egg samples in this study.

SCCPs were analyzed in 14 out of 20 free-range egg samples in this study, and in both reference samples. MCCPs were analyzed in 4 free-range egg samples. SCCPs and MCCPs ranged from levels below the limit of quantitation (LOQ) (50 ng/g fat) to 309 ng/g fat and from levels below LOQ (100 ng/g fat) to 2,345 ng/g fat, respectively.

For PBDD/F the highest level of 12.8 pg WHO-TEQ/g fat was measured in sample KAR-EGG-01/22, where it contributed to 75% of the overall dioxin-like toxicity. The second-highest level of PBDD/Fs (6.93 pg WHO-TEQ/g fat) was found in egg sample SEM-E-1 from Tangerang, which has been discussed in previous studies. Levels >LOQ of 0.57 and 0.33 pg WHO-TEQ/g fat were measured in two other egg samples KEN-E-1/19 and TROP-E-1, respectively. The PBDD/F congeners pattern in free-range egg samples is in graph at Figure 1.

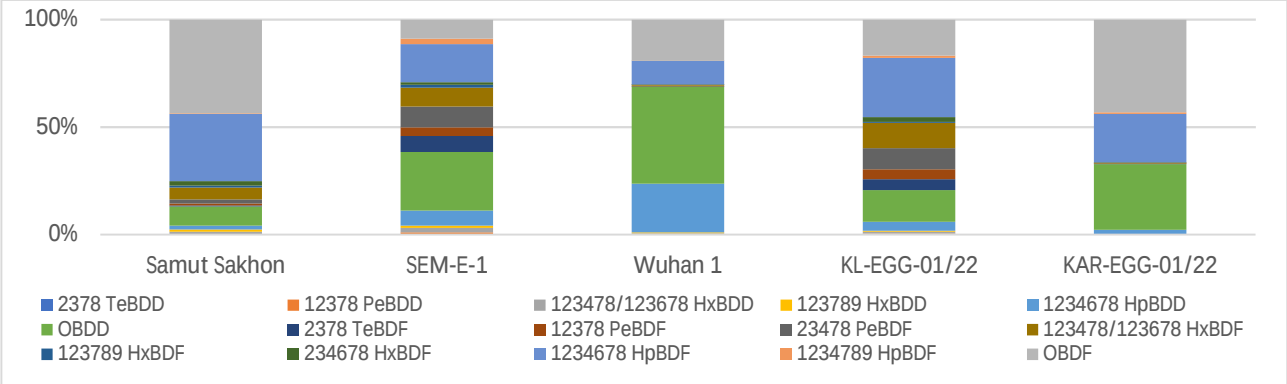

Figure 1: PBDD/F congeners pattern in free-range eggs samples from Indonesia (SEM-E-1 and KAR-EGG-01/22) in comparison with samples from Thailand (Samut Sakhon and Kalasin: KL-EGG-01/22), China (Wuhan 1).

4 Discussion

Higher levels of PCDD/Fs and dl PCBs were observed in free-range chicken eggs from localities affected by combustion processes such as plastic waste incineration in Tropodo and Karawang or aluminium smelters in Kendalsari and galvanization or metallurgical plants in Bantar Gebang (sample BEK-EGG-04/22; B-EGG-3).[7,8,10] However, levels in eggs from sites potentially affected by open burning of waste (Bangun , Bantar Gebang), are also high and exceed the EU and Indonesian standards by two or more and by four or more fold respectively.

In the free-range chicken egg samples presented in this study, four (Tropodo-1, KAR-EGG-2, TROP-E-1, and KAR-EGG-3) were measured as the fourth, fifth, sixth, and seventh highest levels of PCDD/Fs in eggs from Asia and seventh, eighth, ninth, and eleventh highest levels globally, respectively, in comparison with other samples included in the global review[3]. They exhibit the same level of contamination from both localities contaminated by incineration of plastic waste used as fuel in tofu (Tropodo) and lime (Karawang) production facilities, respectively. Dioxin concentrations in these eggs exceeded the EU standard of 2.5 pg TEQ/g fat by approximately 80, 71, 56, and 44-fold, respectively. Higher levels

of dioxins in free-range chicken eggs from Asia were observed only in Bien Hoa, a former US Army base in Vietnam contaminated by Agent Orange [29].

The contribution of the highest level of PBDD/Fs of 12.8 pg WHO-TEQ/g fat in eggs in this study, to the overall dioxin-like toxicity from 75% is very rare compared to other sites, where PBDD/Fs typically contribute only one-tenth to the total TEQ level. The PBDD/Fs level in eggs from Karawang and Tangerang ranks sixth and tenth highest, respectively, among free-range eggs globally.[3,30] The level in sample KAR-EGG-01/22 is comparable to the 15.8 pg WHO-TEQ/g fat measured in eggs from Samut Sakhon, Thailand[31].

HCB levels in two egg samples from Bantar Gebang (BEK-EGG-03/22) and Karawang (KAR-EGG-2) rank as the second and fifth highest measured in Asian countries, respectively.[8,30]

SCCPs concentrations are often quite high in non-free-range egg samples from supermarkets, which originate from large farms where hens don't have access to soil or grass outside. A source of contamination can be, for example, packaging for feed[32]. MCCPs reached much higher levels than SCCPs in eggs from Bantar Gebang, ranging from <LOQ to 2,345 ng/g fat, and are comparable to levels observed in eggs near waste disposal sites in Tanzania[33].

The highest levels of PBDEs in this study were measured in the eggs from Tropodo and Bangun sampled in October/November 2019[7]. The level in the sample from Bangun is higher than levels observed in eggs from Agbogbloshie, an e-waste scrapyard site, or from Wuhan, in the vicinity of a municipal solid waste incinerator. It is also the seventh-highest ever measured level of PBDEs in free-range eggs. The level exceeding 27,000 ng/g fat of PBDEs measured in eggs from Tropodo is the second-highest ever measured level in eggs globally. The PBDEs in eggs from Tropodo and Bangun were in the same range as in egg samples from e-waste sites in China.[9]

Eggs from Bangun were most contaminated with PFASs, exceeding the limit value of 1 ng/g ww set in EU for PFOS in eggs[26] almost 18- and 92-times respectively. They were followed by free-range egg samples from Bantar Gebang, and Tangerang. All three sites are heavily affected by waste dumping/landfilling. Level for PFOS was exceeded also in some eggs from Karawang and Tropodo which are mainly affected by incineration of plastic waste, which contributed also to contamination of some eggs in Bantar Gebang.[8] In Bangun, Bantar Gebang, and Tangerang the highest rates of exceedance of the Tolerable Daily Intake (TDI) by consuming just half an egg per day were estimated based on measured levels of four PFASs evaluated by EFSA in 2020 in a previous larger study focused on POPs in free-range eggs from Java.[8]

5 Conclusions

Severe environmental and food chain contamination with POPs resulting from using plastic waste as fuel in both tofu factories in Tropodo and lime kilns in Karawang Regency has been confirmed by analyzing free-range chicken egg samples in this study. The contamination of eggs with PCDD/Fs/dl PCBs/PBDD/Fs represents the highest levels recorded in Asia and globally. Furthermore, the contamination of eggs with PFASs is particularly concerning, notably in Bangun, Bantar Gebang, and Tangerang, where the highest rates of exceedance of the TDI by consuming just half an egg per day were estimated based on measured levels of 4 PFASs evaluated by EFSA in 2020.

It is imperative to avoid using plastic waste as fuel in facilities such as lime kilns or tofu factories and to cease uncontrolled plastic waste imports and disposal at large plastic waste yards like Bangun, Tangerang. More stringent limits for defining POPs waste are necessary to regulate disposal options for waste produced by waste incineration, including facilities like lime kilns burning plastic waste.

By consuming a normal average portion of eggs, i.e. half an egg from free-range hens in one of the locations sampled in this study, an adult Indonesian can reach or exceed the tolerable daily intake of dioxins and/or dioxin-like compounds by up to 48 times.

For the first time, PBDD/Fs were found to contribute significantly to the overall dioxin-like toxicity, accounting for three quarters of the total compared to PCDD/Fs/dl PCBs in a pooled egg sample from hens foraging at a site with ash from incinerated plastic waste in Karawang.

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