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Urinary concentration levels and health risk assessment of phthalates, parabens, benzophenone-3, bisphenols and triclosan in the adult population of Kinshasa: a cross-sectional study in Democratic Republic of Congo (DRC)

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Abstract

Background Phthalates, parabens, benzophenone-3, bisphenols and triclosan are among the contaminants suspected of being involved in several hormone-related pathologies. In developing countries, weak regulations and lack of a precise monitoring plan lead to exposures that could be much worse than in developed countries. The objectives of this study were to evaluate the level of exposure of the adult population of Kinshasa in the Democratic Republic of the Congo to these compounds and to assess the health risk induced by these pollutants.

Methods Concentrations of four parabens, nine phthalate metabolites, two non-phthalate plasticizers, benzophenone-3, three bisphenols and triclosan were assessed in the urine of 145 volunteers recruited between November 2022 and January 2023 in Kinshasa. Measurements were performed using a liquid or a gas chromatography coupled to a mass spectrometer.

Results Methylparaben (MeP: 62.6 µg/L), mono-n-butyl phthalate (MnBP: 78.1 µg/L) and bisphenol A (BPA: 1.54 µg/L) were detected in over 95% of urine samples, with exposure levels exceeding those observed in Western populations. For BPA and MnBP, 100% and 15% of the study samples, respectively, exceeded human biomonitoring guidance values (HBM-GV) proposed by the European Human Biomonitoring Initiative (HBM4EU) consortium, meaning that their exposure levels are associated with health concerns.

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Conclusion The exposure of the population of Kinshasa to these pollutants merits consideration, as it constitutes a serious public health concern. To enhance protection, regulatory measures must be implemented and large-scale studies and awareness campaigns must be conducted.

Keywords Urinary Biomarkers, Human Biomonitoring, Phthalates, Parabens, Bisphenols, Triclosan, Benzophenone-3, General Population, Kinshasa

Text box 1. Contributions to the Literature

- We performed a human biomonitoring study measuring various non-persistent organic pollutants in urine from adult volunteers recruited in Kinshasa, DRC;
- This study contributes towards addressing the lack of data on human exposure to non-persistent organic pollutants in sub-Saharan Africa;
- The compounds investigated are widely used in the production of everyday items and are currently being investigated for their potential negative impact on human and environmental health;
- The study provides reliable environmental data which highlights the urgent need for regulations to be implemented in order to protect human health and environment.

Background

For decades, humans have been synthesizing a growing number of chemicals used in agriculture, chemical or plastic industries, food production, pharmaceutical and cosmetics, and more. These chemicals have beneficial effects on human life, including increased agricultural yield, protection against or treatment of diseases, and the production of inexpensive and durable consumer products. Phthalates, for example, are a group of compounds that make plastics more flexible and help certain cosmetics to penetrate the skin [1]. Parabens are widely used as preservatives in perishable products (i.e., food or cosmetic products) [2]. Benzophenone-3 (BP-3), an ultraviolet filter, is added to body care products to enhance skin protection, prevent photoaging, and reduce the risk of skin cancer [3]. Bisphenols are essential monomers used to manufacture polymerized plastics (polycarbonates) and epoxy resins. They are found in cans, baby bottles, plastic toys and clear water bottles. Triclosan (TCS) is an antimicrobial agent that is often added to personal care products [4].

However, despite the benefits these compounds provide to human health and well-being, many of these compounds are suspected of being harmful to humans by acting as endocrine disruptors. They are thought to be implicated in the rising prevalence of various hormone-related pathologies including, but not limited to, fertility disorders [5–7], metabolic pathologies [8–10], and cancers of the endocrine glands [11, 12]. Consequently, regulatory agencies such as the European Food Safety Authority (EFSA) and the United States Environmental Protection Agency (US EPA) have classified some of these molecules, including phthalates and bisphenols, as substances of very high concern [13, 14].

On the other hand, the number of morbidity and mortality cases linked to chronic hormonal diseases is constantly rising in the Democratic Republic of the Congo (DRC). For example, cases of thyroid cancer have doubled in recent decades [15, 16]. According to the International Diabetes Federation, the incidence of type 2 diabetes in the DRC has increased from 3.1% of the adult population in 2011 to 7.7% in 2024, and would be one of the causes of underdevelopment [17, 18]. In light of the rising incidence of these endocrine-related pathologies, the question of the implication of the population's exposure to toxicants in the DRC could be reasonably raised. To answer to this question, the first step is to accurately evaluate the exposure of the population of the DRC to potentially toxic compounds.

Over time, a reduction in human exposure to some of these compounds has been observed in Europe and the United States, following the enhancement of regulatory measures implemented since the 2000s [13, 19–21]. However, in developing countries such like DRC, weak regulations and a lack of a precise monitoring plans result in the unrestricted addition of these compounds to many daily-use items. This leads to exposures that could be much worse than in developed countries [22, 23]. Additionally, the absence of an effective waste management policy would facilitate the accumulation of these substances in various environmental compartments, threatening ecosystems and human health.

Unlike some other African countries such as South Africa, Egypt, Nigeria, Tunisia or Ghana, which have data on non-persistent pollutants in environmental and biological matrices, the DRC lags behind in this area [24–26] although the use of these chemicals in daily-use products is expected to be extensive. There are several reasons for this delay, including a lack of specialized laboratories in the country, a failure to address this issue in national health priorities and the limited national and international funding.

To begin to fill this gap, we conducted in 2019 a pilot study based on 15 volunteers recruited among the adult population in Kinshasa, and we observed particularly high levels of parabens, BP-3, TCS, bisphenol A (BPA), and some phthalate metabolites in their urine [27]. Based on these observations, we recruited, between 2022 and 2023, a larger number of adult volunteers ($n=145$) in Kinshasa in order to measure in their urine the concentrations of 4 parabens, 9 phthalate metabolites, 3

bisphenols, BP-3 and TCS as well as 2 non-phthalate plasticizers added to the menu. The aim of the present study is to obtain more data concerning the exposure of the adult population of Kinshasa to these toxicants and to assess the associated health risk. This is the first large-scale biomonitoring study of non-persistent organic pollutants in the general adult population of Kinshasa, in the DRC.

Methods

Study population

The study was carried out in Kinshasa, the capital of the DRC, and targeted adults aged 18 and over. The participants were stratified into five age groups: 18–29, 30–39, 40–49, 50–59, and ≥ 60 years. Outreach sites and recruitment process were previously described in Bayebila et al., 2025 [28]. Between November 2022 and January 2023, 145 volunteers, ranging in age from 18 to 80, were recruited from across the city (Fig. 1). The sociodemographic characteristics of the population under study were presented in detail in Table 1. Overall, males were overrepresented, accounting for 64.1% of the sample. The mean age was 36.9 years (range 18–80 years) and the mean BMI was 25.6 kg/m² (range 15.6–43.0 kg/m²). High participation rates were seen among adults aged 18–29 (42.8%), drivers (13.8%) and residents of the Funa district (33.1%).

Urine sample collection

Volunteers were asked to provide a random urine spot in the morning, either fasting or not. The sample was collected in a polypropylene vial after the volunteer completed a form providing information on their lifestyle and anthropometric measurements. The samples were then placed in an isothermal box and transported to the Physical Chemistry and Biopharmaceutics Laboratory at the Faculty of Pharmaceutical Sciences of the University of Kinshasa. There, the samples were stored at -20°C , before being sent to the Clinical, Forensic and Environmental Toxicology Laboratory at the University of Liège (Belgium) for chemical analysis.

Chemical analysis

The concentrations of four parabens, namely, methylparaben (MeP), ethylparaben (EtP), propylparaben (PrP) and n-butylparaben (nBP), nine phthalate metabolites, namely, monoethyl phthalate (MEP), mono-n-butyl phthalate (MnBP), mono-isobutyl phthalate (MiBP), mono-2-ethylhexyl phthalate (MEHP), mono-2-ethyl-5-hydroxyhexyl phthalate (5-OH-MEHP), mono-2-ethyl-5-oxohexyl phthalate (5-oxo-MEHP), 7-carboxy octyl phthalate (7-cxOP), 6-hydroxypropylheptyl phthalate (6-OH-PrHpP), and monobenzyl phthalate (MBzP), two non-phthalate plasticizers (substitutes for phthalates),

namely, cyclohexane-1,2-dicarboxylate-mono-(7-hydroxy-4-methyl)octyl ester (OH-MINCH) and cyclohexane-1,2-dicarboxylate-mono-(7-carboxylate-4-methyl)heptyl ester (cx-MINCH), and BP-3 were measured in urine according to the method extensively described in Dewalque et al., 2014 [29] and slightly modified (to allow the measurement of OH-MINCH and cx-MINCH) as reported in Pirard et Charlier 2022 [19]. In brief, after enzymatic beta-glucuronidase hydrolysis of the samples, the samples were extracted on a solid-phase extraction Bond Elut Certify cartridge 10 cc (130 mg) from Agilent. After evaporation and reconstitution in a mixture of mobile phases (acetic acid (0.1%) in water/acetic acid (0.1%) in acetonitrile), the samples were analyzed on a liquid chromatography Acquity UPLC system coupled to a mass spectrometer Quattro Premier XE (Waters, Milford, MA, USA).

Bisphenols (namely, BPA, bisphenol F (BPF), bisphenol Z (BPZ)) and TCS urinary concentrations were assessed using an Agilent 7890 A GC/ 7000 A GC Triple Quad mass spectrometer (Agilent Technologies, California, USA). The sample preparation encompassed a simultaneous double enzymatic hydrolysis with beta-glucuronidase and sulfatase, followed by an extraction on solid phase cartridge (Oasis HLB, 60 mg, 3 cc from Waters) and subsequent a liquid/liquid extraction. After the evaporation of the organic phase, extracts were derivatized using N-methyl-N-trimethylsilyl trifluoroacetamide before injection on GC-MS/MS apparatus. This method was extensively detailed in Pirard et Charlier 2022 [19].

Quality assurance

The analytical methods used for this study were previously validated according to the total error approach [30, 31]. The limits of quantification (LOQ) for each molecule are gathered in Table 2 and were defined as the smallest concentrations measurable in samples with a total error not exceeding 30%. In the present study, we did not use the limit of detection and we considered measurements below the LOQ to be non-detected. In addition to the unknown samples, each batch analyzed contained a calibration curve (spiked LC-MS grade water), one procedural blank and several quality controls (QC) extracted as real samples. Quality controls consisted in materials from previous external quality controls from previous German External Quality Assessment Scheme (G-EQUAS) provided by the Out-Patient Clinic for Occupational, Social and Environmental Medicine of the University Erlangen-Nuremberg (for phthalates and parabens), and from External Quality Assessment Scheme for Organic Substances in Urine (OSEQAS) provided by the Institut national de santé publique du Québec (for bisphenols and TCS). For a measurement to be considered satisfactory, the measured concentration in these QCs must fall

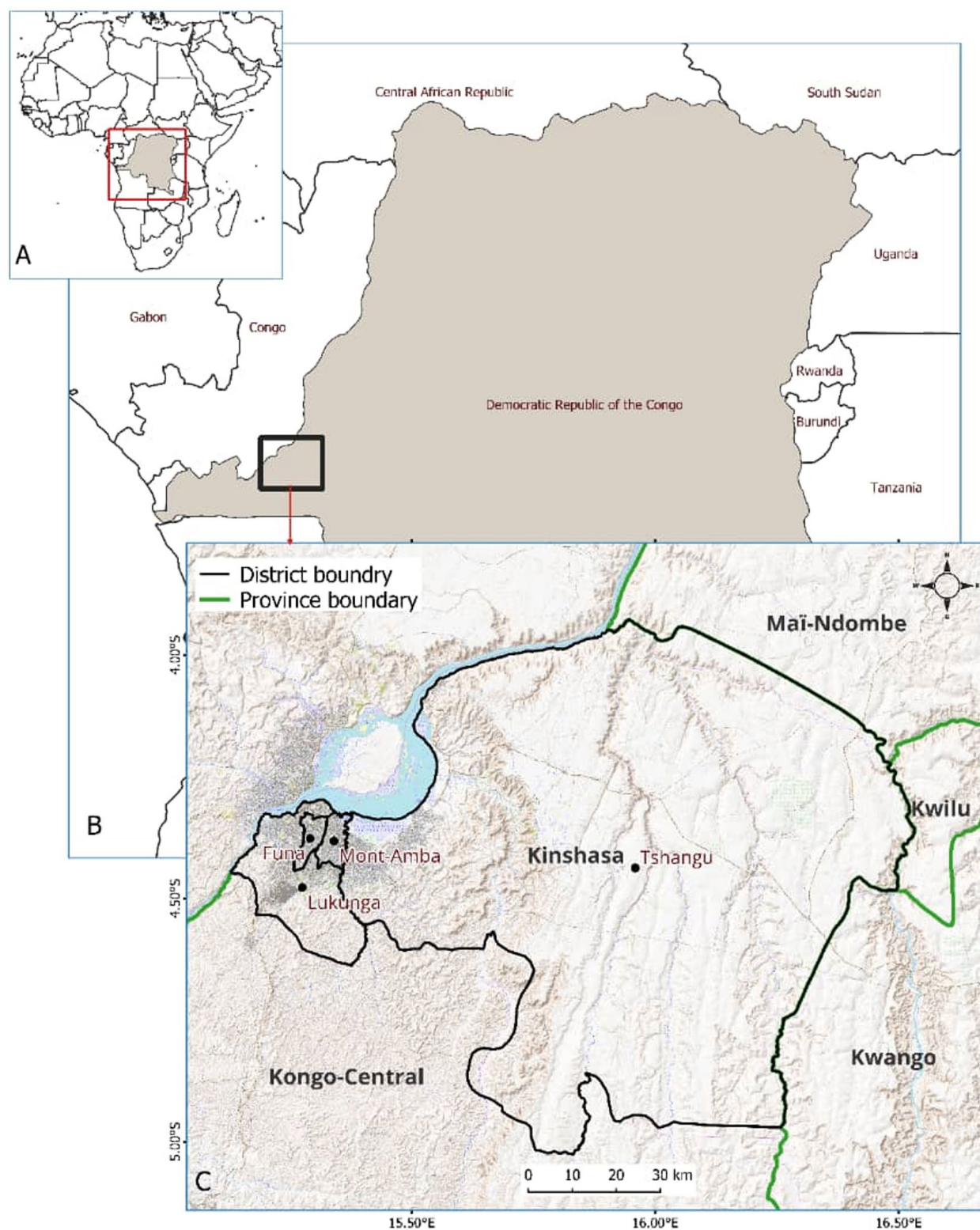


Fig. 1 Geographical location of study area (C) in DRC (B) and Africa (A)

Table 1 Socio-demographic characteristics of the studied population

	Total (%)	Men (%)	Women (%)
All	145 (100)	93 (64.1)	52 (35.9)
Age (years)			
Mean	36.9	36.4	37.9
SD	15.7	15.7	15.7
Median	33.0	32.0	34.0
P25	24.0	23.0	25.8
P75	46.0	46.0	48.5
Range	18.0–80.0	18.0–74.0	18.0–80.0
Age categories			
18–29 years	62 (42.8)	41 (28.3)	21 (14.5)
30–39 years	28 (19.3)	19 (13.1)	9 (6.20)
40–49 years	23 (15.8)	14 (9.65)	9 (6.20)
50–59 years	14 (9.66)	6 (4.14)	8 (5.52)
≥ 60 years	18 (12.4)	13 (8.96)	5 (3.45)
BMI (kg/m ²)			
Mean	25.6	25.0	26.6
SD	4.92	4.59	5.35
Median	25.3	25.0	25.7
P25	22.1	21.9	23.3
P75	28.0	26.9	29.1
Range	15.6–43.0	15.6–39.6	16.0–43.0
< 18.5	7 (4.83)	6 (4.14)	1 (0.69)
18.5–24.9	63 (43.4)	41 (28.2)	22 (15.2)
25.0–29.9	52 (35.9)	34 (23.5)	18 (12.4)
≥ 30	23 (15.9)	12 (8.27)	11 (7.59)
Activities			
Drivers	20 (13.8)	20 (13.8)	0 (0.0)
Teachers	19 (13.1)	14 (9.7)	5 (3.4)
Traders	16 (11.0)	9 (6.2)	7 (4.8)
Students	16 (11.0)	11 (7.6)	5 (3.4)
Medical staff	16 (11.0)	6 (4.1)	10 (6.9)
Others	58 (40.0)	33 (22.8)	25 (17.2)
Feed 24 h before sampling			
No	8 (5.5)	5 (3.4)	3 (2.1)
Vegetables	24 (16.6)	14 (9.7)	10 (6.9)
Vegetables and meat	69 (47.6)	46 (31.7)	23 (15.9)
Meat	35 (24.1)	22 (15.2)	13 (8.9)
Others	9 (6.2)	6 (4.1)	3 (2.1)
Area of residence			
Tshangu	29 (20.0)	19 (13.1)	10 (6.9)
Mont Amba	47 (32.4)	36 (24.8)	11 (7.6)
Funa	48 (33.1)	25 (17.2)	23 (15.9)
Lukunga	21 (14.5)	13 (9.0)	8 (5.5)

BMI Body Mass Index

within the acceptable range provided by the supplier. Since several chemicals were not covered by these commercial QCs (namely the four parabens, BP-3, 7-cxOP and 6-OH-PrHpP), we also included two homemade QCs (LC-MS water spiked at low and high concentrations) and two samples from two different pools of urine, where the chemicals were measured ten times before use

as a QC. For batch validation, the concentrations of the chemicals measured within these internal QCs must fall within $\pm 30\%$ of the target concentration.

Our laboratory is regularly (twice a year) participating to G-EQUAS and OSEQAS. Moreover, since we successfully participated to several HBM4EU QA/QC programs, our laboratory was qualified as “HBM4EU lab” for the analyses of BPA and BPF, and for all phthalate metabolites measured during this study.

Statistical analysis

For each analyzed chemicals, we reported in Table 2, detection frequency (DF), arithmetic and geometric means, standard deviation, minimum, maximum and percentiles 25, 50, 75 and 95. Descriptive statistics for socio-demographic variables were reported in Table 1. Statistical analyses were performed to identify parameters associated with chemicals urinary concentrations. For the descriptive and inferential analyses, contaminant levels below the LOQ were replaced by the LOQ multiplied by the DF of the compound in our population [32, 33]. Contaminants with a DF below 70% were dichotomized (detected vs. not detected). Pollutant concentrations were not normally distributed (Shapiro-Wilk test < 0.05), therefore for quantitative variables, non-parametric tests (Kruskal-Wallis, Mann-Whitney) and Spearman's rank correlation were used to assess the difference in contamination across the study sub-populations, as well as the patterns of co-exposure within the study population. For categorical variables and compounds with DF $< 70\%$, chi-squared test was used. P-value ≤ 0.05 was considered significant. RStudio (version 2023.09.1), Rcmdr software (version 3.6.3., CRAN) and Excel 2013 (Microsoft, Redmond, WA) were used for statistical processing of data.

Health-based risk assessment

One of the main objectives of this study was to evaluate the health risks associated with exposure to chemical substances among the population of Kinshasa. To accomplish this, we compared urinary pollutant levels measured in our population with reference levels provided by scientific authorities and working groups. The European Human Biomonitoring Initiative (HBM4EU) provided human biomonitoring guidance values (HBM-GVs) for BPA, MnBP, MiBP and the sum of 5-oxo-MEHP and 5-OH-MEHP [34–36]. These HBM-GVs are defined as the concentration in a biological matrix (i.e., urine) at and below which negative health effects are not expected based on the current knowledge. We then compared urinary concentrations of these compounds in our study sample to the corresponding HBM-GVs proposed by Apel et al., 2023 [34]. A human biomonitoring value I (German HBM-I value are similar to HBM-GV from

Table 2 Descriptive statistics of non-persistent organic pollutants in urine

Pollutant	LOQ (µg/L)	DF (%)	Mean	SD	GM	P25	P50	P75	P95	Range
<i>Parabens</i>										
MeP	2.00	99	282	1021	59.9	15.2	62.6	216	853	< LOQ – 11,442
EtP	0.30	44	2.80	8.60	0.401	< LOQ	< LOQ	1.20	12.5	< LOQ – 66.1
PrP	0.36	95	84.7	194	10.1	1.50	7.50	52.8	439	< LOQ – 1365
nBP	6.00	6	< LOQ	< LOQ	< LOQ	< LOQ	< LOQ	< LOQ	< LOQ	< LOQ – 6.51
<i>Phthalates</i>										
MEP	0.94	99	373	1,053	74.8	25.4	61.3	229	1,591	< LOQ – 7,194
MnBP	0.99	100	105	105	70.1	41.8	78.1	143	270	2.31–815
MiBP	1.23	99	26.4	24.5	17.7	10.6	17.1	32.2	79.9	< LOQ – 115
MEHP	0.62	95	7.10	25.1	3.20	1.40	3.10	5.90	16.4	< LOQ – 294
5-oxo-MEHP	0.53	99	15.5	79.3	6.40	3.50	6.30	10.8	21.9	< LOQ – 955
5-OH-MEHP	0.43	100	30.1	175	11.2	6.01	11.4	19.4	44.7	0.601–2,108
Sum of DEHP metabolites*			52.8	278	21.9	12.6	21.5	36.3	81.5	1.74–3,357
MBzP	0.61	51	1.40	5.90	0.701	< LOQ	< LOQ	1.10	3.20	< LOQ – 69.9
7-cxOP	2.00	68	4.10	3.80	3.10	1.40	2.90	5.10	9.20	< LOQ – 31.7
6-OH-PrHpP	2.00	6	< LOQ	2.80	< LOQ	< LOQ	< LOQ	< LOQ	2.50	< LOQ – 30.4
OH-MINCH	2.00	6	< LOQ	1.50	< LOQ	< LOQ	< LOQ	< LOQ	2.15	< LOQ – 12.2
cx-MINCH	2.00	6	< LOQ	1.20	< LOQ	< LOQ	< LOQ	< LOQ	2.30	< LOQ – 10.2
<i>Benzophenone</i>										
BP-3	0.20	96	2.60	4.30	1.50	0.701	1.40	2.80	7.01	< LOQ – 37.5
<i>Bisphenols</i>										
BPA	0.29	98	2.91	6.48	1.63	0.971	1.54	2.32	8.33	< LOQ – 55.9
BPF	0.07	69	0.265	0.789	0.134	< LOQ	0.130	0.235	0.667	< LOQ – 8.95
BPZ	0.06	10	< LOQ	0.06	< LOQ	< LOQ	< LOQ	< LOQ	0.0919	< LOQ – 0.483
<i>Triclosan</i>										
TCS	0.33	100	158	379	53.2	24.6	50.2	125	499	1.55–3,152

*Sum of DEHP metabolites = MEHP + 5-oxo-MEHP + 5-OH-MEHP; LOQ Limit of Quantification, DF Detection Frequency, SD Standard Deviation, GM Geometric Mean

HBM4EU) was also proposed by the German Human Biomonitoring Commission for TCS [37].

To the best of our knowledge, no HBM-GV or associated threshold is available for parabens. Nevertheless, in 2004, the EFSA established an Acceptable Daily Intake (ADI) of 0–10 mg/kg body weight per day (mg/kg bw/day) for the sum of MeP and EtP. Therefore, for each volunteer in our study, we calculated an estimated daily intake (EDI) based on the formula proposed by Ma et al., 2013 [38]:

$$EDI = \frac{50 \times C_i \times V}{BW}$$

where C_i is the sum of the urinary concentrations of MeP and EtP (µg/L), V (L/day) is the daily urine excretion rate (a value of 1.7 L/day was used for adults) and BW (kg) is the body weight of each participant. We then compared the EDIs with the ADI provided by the EFSA.

Results and discussion

Phthalate metabolites and non-phthalate plasticizers

All phthalate metabolites were detected in more than 95% of the samples, except for MBzP (51%) and 7-cxOP (68%). Non-phthalate plasticizers were detected in only 6% of

the samples, with concentrations ranging from below the LOQ to 12.2 µg/L, unlike in countries with established regulations on legacy phthalates, where detection of non-phthalate plasticizer alternatives in biological fluids is constantly increasing [19, 21], this indicates a very low level of these alternatives in items sold in Kinshasa. The highest median concentrations (Table 2) were measured for MnBP (median: 78.1 µg/L, range: 2.31–815 µg/L), followed by MEP (median: 61.3 µg/L, range: < LOQ–7,194 µg/L), and the sum of the di-2-ethylhexyl phthalate (DEHP) metabolites (sum of MEHP + 5-OH-MEHP + 5-oxo-MEHP) (median: 21.5 µg/L, range: 1.74–3,357 µg/L). These levels are consistent with those reported in our 2019 pilot study [27], suggesting that these molecules are still present in personal care products, as well as in the food packaging, pharmaceutical and cosmetic products sold in the city although they are banned or restricted in several countries [19, 39].

We reviewed the literature to highlight studies measuring urinary levels of phthalate metabolites in populations recruited in 2015 or later. Over the past decade, phthalate metabolites urinary levels have been assessed in several adult populations worldwide, some examples are reported in Table 3. As shown in Table 3, the MEP levels measured in our study population are two to three times

Table 3 Levels of urinary non-persistent organic pollutants in different populations worldwide

Population	Year of collection	Pollutants median values (µg/L)						Reference
		Phthalates						
		MEP	MnBP	MiBP	MEHP	5-OH-MEHP	5-oxo-MEHP	
Belgium, adult population, N=92, 46.7% men/53.3% women	2018	20.4	11.8	-	< LOQ	2.89	1.92	[19]
Danish, young population, N= 100, 100% men	2017	23.9	20.9	23.1	1.09	5.64	3.81	[13]
Korea, adult population, N= 3782, 43.6% men/56.4% women	2015–2017	-	25.3	-	-	13.7	10.7	[9]
USA, adult population, N= 1862, 49.5% men/50.5% women	2017–2018	29.9	9.6	7.4	0.9	4.6	3.1	[40]
German, children and adolescents, N= 2256, 52% boys/48% girls	2014–2017	23.1	21	26.2	1.5	11.1	7.7	[20]
Kinshasa (DRC), adult population, N = 145, 64.1% men/35.9% women	2022–2023	61.3	78.1	17	3.1	11.4	6.3	This study
		Parabens						
		MeP	EtP	PrP				
Belgium, adult population, N=92, 46.7% men/53.3% women	2018	3.56	0.58	< LOQ				[19]
Korea, adult population, N= 3787, 43.6% men/56.4% women	2015–2017	34.6	36.2	2.1				[9]
Sichuan (China), non-occupational population, N=986, 36.8% men/63.2% women	2021	10.1	0.7	0.55				[41]
Nantong city (eastern China), general population, N= 104, 60.6% men/39.4% women	2020	3.28*	0.45*	0.19*				[42]
Suizhou and Beijing (China), general population, N= 203, 45.8% men/54.2 women	2018–2019	7.83	0.248	0.614				[43]
China, Guangzhou, general adult population, N= 299, 39.1% men/60.9% women	2018–2019	24.5	3.31	2.05				[44]
Spain, breastfeeding mothers, N= 180, 100% women	2015	17.7*	-	-				[45]
India, young adult women from Assam, N= 52, 100% women	2020	108	0.11	37.1				[46]
Kinshasa (DRC), adult population, N = 145, 64.1% men/35.9% women	2022–2023	62.6	< LOQ	7.5				This study
		Benzophenone						
		BP-3						
Belgium, adult population, N= 261, 47.1% me/52.9% women	2013	1.3						[47]
Tunisia, women, N= 34, 100% women	2012	1.73						[48]
Sichuan (China), non-occupational population, N= 986, 36.8% men/63.2% women	2021	0.1						[41]
Nantong city (eastern China), general population, N= 104, 60.6% men/39.4% women	2020	0.103*						[42]
Kinshasa (DRC), adult population, N = 145, 64.1% men/35.9% women	2022–2023	1.4						This study
		Bisphenols						
		BPA	BPF	BPZ				
Belgium, adult population, N= 92, 46.7% men/53.3% women	2018	0.79	0.12	< LOQ				[19]
Korea, adult population, N= 3782, 43.6% men/56.4% women	2015–2017	1.3	-	-				[9]
Nantong city (eastern China), general population, N= 104, 60.6% men/39.4% women	2020	0.398*	0.584*	0.024*				[42]
Shenyang, (northeastern China), women, N= 111, 100% women	2019–2020	5.84	0.54	< LOQ				[49]
Spain, breastfeeding mothers, N= 180, 100% women	2015	0.927*	0.042*	-				[45]

Table 3 (continued)

Population	Year of collection	Pollutants median values (µg/L)			Reference
India, young adult women from Assam, <i>N</i> = 52, 100% women	2020	< LOQ	< LOQ	0.4	[46]
USA, adult population, <i>N</i> = 1829, 47.3% men/52.7% women	2015–2016	1.1	0.14	-	[50]
Nigeria, Ota University students, <i>N</i> = 73, unknown men/women ratio	?	2.052	-	-	[51]
Kinshasa (DRC), adult population, <i>N</i> = 145, 64.1% men/35.9% women	2022–2023	1.54	0.13	< LOQ	This study
		<i>Triclosan</i>			
		TCS			
Sichuan (China), non-occupational population, <i>N</i> = 986, 36.8% men/63.2% women	2021	10.58			[41]
China, Shenzhen, general adult population, <i>N</i> = 1163, 97.9% men/2.1% women	2017	3.67			[52]
China, Guangzhou, general adult population, <i>N</i> = 299, 39.1 men/60.9% women	2018–2019	0.737			[44]
Spain, Barcelona, pregnant women, <i>N</i> = 546, 100% women	2018–2021	0.8			[53]
USA, adult population, <i>N</i> = 1829, 47.3% men/52.7% women	2015–2016	3.1			[50]
Iran, Tehran, pregnant women, <i>N</i> = 189, 100% women	2019–2020	14.08			[54]
Mexico, low- or middle-income adult women, <i>N</i> = 91, 100% women	2017–2019	58.8			[55]
Belgium, general population, <i>N</i> = 131, 49.6% men/50.4% women	2011	2.24			[4]
Kinshasa (DRC), adult population, <i>N</i> = 145, 64.1% men/35.9% women	2022–2023	50.2			This study

*: Geometric mean, LOQ Limit of Quantification

higher than those reported in developed countries over the past 10 years and the MnBP levels are also several times higher in our population than in other recent studies [9, 13, 19, 20, 40]. This suggests the presence of their respective parent compounds, diethyl phthalate and di-n-butyl phthalate, in perfumes, cosmetics and nail polishes sold in Kinshasa. The levels of MiBP and DEHP metabolites are similar to the high levels reported in Denmark, Korea and Germany. Overall, our study participants are more exposed to phthalates than individuals from Western countries. This is likely due to the lack of Congolese legislation on these products, leaving the population exposed.

Table 4 shows the significant differences in urinary concentration according to the variables studied in our population. Men, particularly drivers (all drivers in our population were men), showed higher levels of DEHP metabolite (Table 4). DEHP (a high-molecular-weight phthalate) was commonly used in food and beverage packaging, as well as in plastics in interior car coatings. The use of this compound is now restricted in Western countries, but not in DRC [56, 57]. Therefore, food packaging in DRC are probably contaminated. Furthermore, most vehicles in Kinshasa are old, secondhand

cars imported from Western countries or Asia. Drivers are likely exposed to DEHP through their vehicles, which were produced in countries and/or at times when DEHP was not restricted [58, 59]. Drivers, students, and people aged 18–49, showed relatively high concentrations of MiBP and MnBP, likely due to their frequent use of cosmetics and nail polishes as they belong to a category that includes young people [60], a potential user group for body care products (Table 4; *p*-value < 0.05). Furthermore, overweight and obese adults showed higher levels of MEP (Table 4; *p* < 0.05), this observation is quite surprising since diethylphthalate is usually associated to cosmetic use and not eating habits [61].

As women are the primary users of cosmetic products, they are therefore usually more exposed to low-molecular-weight phthalates such as diethyl phthalate, di-n-butyl phthalate and di-iso-butyl phthalate. However, this study found that exposure levels to these compounds were the same for men and women (see supplementary material: Table S1), a finding that warrants further investigation in the Kinshasa market.

Table 4 Contamination of the Kinshasa population according to some studied variables

Variable	Pollutant	Median (µg/L)						P-value
Sex		Males	Females					
	MeP	35.5	136					0.002
	PrP	5.14	16.1					0.010
	MEHP	3.99	2.45					0.016
	5-OH-MEHP	13.1	8.89					0.003
	BP-3	1.16	2.31					0.002
Area of residence	BPA	1.66	1.22					0.029
		Tshangu	Mont Amba	Funa	Lukunga			
	EtP	0.84	0.22	0.13	0.13			0.016
	7-cxOP	3.80	2.24	3.77	2.13			0.037
BMI	BPF	0.0968	0.188	0.0897	0.130			0.017
		< 18.5	18.5–24.9	25.0–29.9	≥ 30			
	MEP	10.1	37.9	90.2	87.6			0.029
Activities		Drivers	Teachers	Traders	Student	Medical staff	Others	
	MiBP	28.1	7.47	23.4	24.3	15.7	17.6	< 0.001
	MnBP	98.9	27.3	75.7	132	87.3	80.5	< 0.001
	5-OH-MEHP	19.4	4.16	13.5	12.1	12.7	9.86	< 0.001
	5-oxo-MEHP	11.1	2.80	6.45	6.65	8.35	5.42	< 0.001
Age		18–29 years	30–39 years	40–49 years	50–59 years	≥ 60 years		
	MiBP	17.0	15.9	26.6	13.5	9.12		0.011
	MnBP	84.6	84.8	82.1	55.3	46.5		0.006

BMI Body Mass Index

Parabens

More than 95% of the samples tested positive for MeP and PrP, with median concentrations of 62.6 µg/L and 7.50 µg/L, respectively. Conversely, nBP and EtP were detected in 6% and 44% of the samples, respectively (Table 2). The levels of MeP and PrP observed in this study are significantly lower than those highlighted in the 2019 pilot study [27]. However, the sample size of the present study is almost tenfold higher than in the pilot study (145 vs. 15), the current estimates are likely better indicators of the exposure of the population of Kinshasa.

Table 3 reports the median levels of parabens measured in some study populations recruited after 2014. The MeP and PrP exposure observed in our study population is several times higher (up to one order of magnitude) than in Belgian [19], Korean [9], Spanish [45], and Chinese [41–44] populations. On the other hand, the level of parabens measured in Indian young adult women were significantly higher than those highlighted in our study [46]. In Europe, the use of parabens is strictly regulated. PrP use in food is banned, and maximum concentrations of MeP and EtP are established (Regulation (EU) 1129/2011). MeP, EtP, PrP, and nBP are authorized for use in personal care products, but the European Commission has imposed maximum thresholds (Regulation (EU) 1004/2014). Such legislation is unavailable in the DRC; therefore, cosmetics, food and pharmaceutical products sold in Kinshasa may contain high amounts of these toxic substances, which probably explains the high levels measured in the urine of our volunteers.

Paraben urinary concentrations was consistent across all studied variables (see supplementary material: Tables S1–S3), except for gender and area of residence (Table 4). Women were found to be relatively more contaminated than men by MeP and PrP, potentially due to the presence of these molecules in personal care products (Table 4; P-value < 0.05), as observed in several worldwide studies [47, 62]. In our study, participants from Tshangu showed relatively higher exposure to EtP than those from other districts. This observation is surprising and warrants further investigation.

Benzophenone-3

Benzophenone-3 was detected in 96% of the samples, with a median concentration of 1.40 µg/L and concentration values ranging from <LOQ to 37.5 µg/L (Table 2). The current median level measured in our sample of Kinshasa inhabitants is one order of magnitude higher than in the Chinese population [41, 42], though it is similar to levels in the Belgian adult population [47] and Tunisian women [48]. Similar to what has been observed for parabens, women had higher exposure levels to BP-3 than men (Table 3; P-value < 0.05), this observation is likely due to women's more frequent use of cosmetics and hair products [63]. The BP-3 contamination observed in this study is also significantly lower than that highlighted in the 2019 pilot study, but the present study's estimates are likely better than those in the previous study which involved a limited number of volunteers.

Bisphenols

Bisphenols Z, F, and A were detected in 10%, 69%, and 98% of the samples, respectively. The highest median value was found for BPA (1.54 µg/L, range: < LOQ–55.9 µg/L), followed by BPF (0.13 µg/L, range: < LOQ–8.95 µg/L) (see Table 2). BPA is one of the most studied endocrine-disrupting compounds [53]. Exposure to this compound has been assessed in many populations worldwide. For example, Acevedo et al., 2025 [64] conducted an extensive review of epidemiological studies performed in Asia, Latin America, Australia, and Africa. Table 3 shows some examples of the median (or geometric mean) BPA concentrations measured in the urine of recently recruited adult study samples. Our study adult population exhibited slightly higher BPA exposure levels than the Belgian [19], Spanish [45], US [50], and Korean [9] populations. The median BPF level in our samples is similar to those observed in Belgium [19] and higher than those reported in Spain [45]. In China, urinary levels of BPA measured in different study populations vary greatly: Xu et al., 2022 [42] reported a geometric mean of 0.398 µg/L in a general population sample recruited in Nantong (eastern China). This level is four times lower than those reported in the present study. Conversely, Olawole et al., 2022 [51] and Li et al., 2024 [49] observed respectively a far higher median concentration in a population of students from Ota University of Nigeria (2.052 µg/L) and of adult women from Shenyang (northeastern China) (4.33 µg/L). The BPF concentrations in these two Chinese populations were very similar (0.584 µg/L in Xu et al., 2022, and 0.54 µg/L in Li et al., 2024) and were several times higher than those in our study population. There is a lower difference in bisphenol urinary concentrations between our samples of Kinshasa inhabitants and that of developed countries than we observed for phthalate metabolites and parabens. However, it will be interesting to observe the trend in the coming years. In Western countries, the use of BPA is declining due to increasingly restrictive regulations, while alternatives like BPF are being used more frequently. This is reflected in the decline of concentrations of BPA measured in the urine of volunteers in Europe and the US [19, 64]. Conversely, an increasing trend in urinary BPA concentration has been observed in Australia, Asia, and Latin America, while any trend could be identified in Africa due to the too few existing data from this continent [64]. To our knowledge, only seven studies assessed the urine concentration of bisphenols in study populations recruited in Africa: our pilot study, three performed in Egypt [65–67], one in Tunisia [48], one in Ghana [68] and one in Nigeria [51]. All these studies involved a relatively low number of individuals (from 15 to 73 volunteers). In the study with the largest sample [51], the median BPA concentration was 2.052 µg/L but it

was determined with an ELISA kit which limits comparison with our results obtained using a GC-MS apparatus. The small number of studies, involving limited number of volunteers and the use of less reliable analytical methods in some works underscores again the urgent need for studies in low- and middle-income countries in Africa and highlights the relevance of the data collected in the present study.

In our study population, men showed significantly higher levels of BPA than women (Table 4; $p < 0.05$), and Mont Amba inhabitants had higher BPF levels (Table 4; $p < 0.05$) than those in other districts. The first observation could be explained by dietary habits, as men are more likely to consume food and beverages stored in contaminated packaging [69]. The median BPA level in the present study is similar to those highlighted in our pilot study (1.36 µg/L).

Triclosan

Triclosan was detected in all analyzed samples, with a median value of 50.2 µg/L and urinary levels ranging from 1.55 to 3,152 µg/L (see Table 2). We reviewed the literature for studies measuring TCS in the urine of adult volunteers recruited in 2015 or later. Some examples of study populations are presented in Table 3. The levels measured vary greatly from one study sample to another, even within the same country. For example, the median concentrations measured in China range from 0.737 µg/L in Guangzhou [44, 52] to 10.58 µg/L in Sichuan [41]. Except for the Mexican population recruited by Zamora et al., 2023 [55], the highest level of TCS measured in the last decade was 14.08 µg/L in Iranian pregnant women [54]. This suggests that TCS exposure among the participants studied in Kinshasa is several times higher than in most other populations worldwide (our median concentration is 50.2 µg/L) [4, 50, 53]. The only other study that highlights a similar (and slightly higher) level of exposure is the work of Zamora et al., 2023 [55], which explored the concentrations of several non-persistent pollutants in the urine of low- or middle-income Mexican women. The exceptional nature of exposure to TCS measured during our study indicates that self-care products sold in the city are likely highly contaminated with this compound. This underscores the urgent need for strict regulations in the DRC. Regardless of the variable studied, no difference in TCS concentration was observed between the groups (Table 4; $p > 0.05$). The high levels of TCS highlighted in the present study were not surprising regarding the concentrations previously measured during our pilot study (median level: 40.1 µg/L).

Health risk assessment

Our results demonstrate that the volunteers of our study are highly exposed to non-persistent organic pollutants,

especially compared to inhabitants of developed countries. All of the compounds measured in our study are potential endocrine disruptors; therefore, the associated health risks of this high exposure could be significant in the capital of the DRC. To evaluate this risk, we compared the concentrations measured in our volunteers to thresholds proposed in the literature.

First, the HBM4EU consortium proposed an HBM-GV of 190 µg/L for MnBP [34]. In our study sample, 15% of individuals had a urinary MnBP concentration above this threshold (Table 5). This HBM-GV is derived from a rat study highlighting the negative impact of di-n-butyl phthalate (the parent compound of MnBP) exposure on sexual system development. Moreover this compound is known to interfere with the activation of sexual steroid hormone receptors and could thus disrupt reproductive system homeostasis [70]. Furthermore, one volunteer exceeded the HBM-GV proposed for the sum of 5-oxo-MEHP and 5-OH-MEHP (500 µg/L) (see Table 5). This threshold was also determined based on a study that examined the effects of DEHP on the reproductive system [34]. This compound is also capable of interfering with sexual steroid receptors [70]. Therefore, based on these results, we can conclude that a significant percentage of our study population is susceptible to reproductive disorders due to exposure to phthalates. However, none of the volunteers exceeded the HBM-GV proposed for MiBP (230 µg/L) [34].

In Apel et al., 2023 [34], a HBM-GV of 230 µg/L was proposed for BPA. Nevertheless, this HBM-GV was based on the tolerable daily intake (TDI) proposed by the EFSA in 2015 and recently, EFSA proposed a 20,000-fold lowering of TDI for BPA to 0.2 ng/kg bw/day. Therefore we recalculated a new HBM-GV based on this actualized TDI [36, 71]. With this recalculated HBM-GV of 0.0115 µg/L, all individuals, in our study sample but also in virtually every population worldwide will be considered at risk. This would be consistent with the ample evidences of BPA toxicity, notably due to its estrogenic potential [72].

Table 5 Summary of the exposure risk assessment to pollutants

Chemical compounds	Reference levels for adults	Number of participants exceeding the threshold
<i>Urine concentration</i>		
MnBP	190 µg/L	22 (15%)
Σ5-OH-MEHP, 5-oxo-MEHP	500 µg/L	1 (0.7%)
MiBP	230 µg/L	0 (0%)
BPA	0.0115 µg/L	145 (100%)
TCS	3,000 µg/L	1 (0.7%)
<i>Tolerable daily intake</i>		
ΣMeP, EtP	10 mg/kg bw/day	1 (0.7%)

Although the levels of TCS measured in our study sample are very high compared to other populations worldwide, only one of our volunteers exceeds the HBM-I value of 3,000 µg/L proposed by Apel et al. 2017 [37], HBM value established based on the hematotoxicity of TCS (Table 5).
Finally, we calculated the EDI for parabens (the sum of MeP and EtP). Only one volunteer exceeded the TDI of 10 mg/kg bw/day proposed by the EFSA and thus should be considered at risk for impaired reproductive function (Table 5).

Several volunteers had urine concentrations of some compounds that exceeded the thresholds associated with health risks, though most individuals were below these limits (except for BPA). Nevertheless, we must consider that we are all simultaneously exposed to numerous potentially toxic compounds that can act in an additive or synergistic manner. Therefore, although already significant, the percentage of our volunteers exposed to toxic levels of non-persistent pollutants could be worse than if we consider the pollutants individually.

Correlations between pollutants

In the previous sections, we suggested some exposures sources for the studied pollutants. It is important to emphasize that these suggested specific exposure sources (e.g., cosmetics, food packaging) are purely speculative. To date, no studies have investigated the contamination of Congolese consumer products by these toxic compounds. The administration of a more detailed questionnaire on lifestyle and consumption habits would have allowed highlighting the potential sources of contamination in our population. Nevertheless, since these two sources of information are unavailable, investigating the correlation patterns of the pollutants could help us test our hypothesis about the sources of exposure, even if this approach is limited compared to two others. The Spearman's correlation coefficients for pollutant urinary concentrations are reported in (Table 6).

Bisphenol F is not correlated with any of the other chemicals, including BPA, while BPA is poorly correlated ($r<0.33$) with other compounds. This suggests that these two plasticisers have independent sources of exposure. Several compounds (TCS, BP3, parabens and low-molecular-weight phthalates) are usually associated with the use of personal care products [19]. Nevertheless, TCS is poorly correlated with other compounds ($r<0.22$). The correlation between MeP and PrP is strong ($r=0.82$), however, the correlation with other compounds is once again low or null ($r<0.35$), suggesting that TCS and parabens have their own exposure sources. Higher correlation coefficients were observed for BP3 and low-molecular-weight phthalate metabolites (e.g. MEP, MiBP and MnBP), suggesting potential common exposure sources.

Table 6 Spearman's rank correlations between urinary phthalate metabolites, parabens, bisphenols, BP3 and TCS

	MEP	MiBP	MnBP	MEHP	5-oxo-MEHP	5-OH-MEHP	MeP	PrP	BPA	BPF	BP-3	TCS
MEP	—											
MiBP	0.42***	—										
MnBP	0.39***	0.77***	—									
MEHP	0.18*	0.47***	0.37***	—								
5-oxo-MEHP	0.28***	0.56***	0.59***	0.64***	—							
5-OH-MEHP	0.27***	0.53***	0.52***	0.63***	0.93***	—						
MeP	0.29***	0.11 ^{ns}	0.13 ^{ns}	-0.018 ^{ns}	0.017 ^{ns}	-0.028 ^{ns}	—					
PrP	0.34***	0.17*	0.19*	0.021 ^{ns}	0.025 ^{ns}	-0.016 ^{ns}	0.82***	—				
BPA	0.17*	0.28***	0.30***	0.27**	0.32***	0.28***	0.076 ^{ns}	0.071 ^{ns}	—			
BPF	0.12 ^{ns}	0.017 ^{ns}	0.026 ^{ns}	-0.058 ^{ns}	0.11 ^{ns}	0.14 ^{ns}	-0.069 ^{ns}	-0.056 ^{ns}	0.047 ^{ns}	—		
BP-3	0.31***	0.51***	0.41***	0.27**	0.30***	0.25**	0.19*	0.14 ^{ns}	0.26**	-0.0027 ^{ns}	—	
TCS	0.027 ^{ns}	0.10 ^{ns}	0.20*	0.18*	0.21*	0.17*	0.12 ^{ns}	0.086 ^{ns}	0.061 ^{ns}	0.11 ^{ns}	0.084 ^{ns}	—

*P < 0.05; **P < 0.01; ***P < 0.001; ^{ns}: not significant.

As expected, correlations between DEHP metabolites are high, since they share the same precursor. However, we also observed moderate to good correlations between these phthalate metabolites and some low-molecular-weight phthalate metabolites (MiBP and MnBP). DnBP and DiBP are commonly used in personal care products, as well as in plastics, so a moderate correlation between their metabolites is not surprising and suggests that these compounds could be present in the same consumer products. Our results confirmed that individuals are exposed simultaneously to many chemicals and the exploration of correlation patterns suggest that exposure sources are also multiple. The simultaneous exposure of individuals to this chemical mixture could have adverse effects due to the additive, synergistic or antagonistic effects that are known to occur with these toxic compounds, a phenomenon commonly referred as 'cocktail effect' in the literature [73, 74].

Strengths and limitations of the study

Except our 2019 initial pilot study which involved only 15 volunteers, this study provides the first data in DRC on the exposure of population to several different environmental non-persistent pollutants. However, despite the selection strategy used for the recruitment, the population could still not be considered as representative of the city's entire population, let alone that of the DRC. A study sample of 145 volunteers could hardly be expected to be representative of a city with a population of around 17.7 million inhabitants, spread over a large geographical area and with very different socio-demographic characteristics. Moreover, in our study population men are somewhat overrepresented compared to women (64.1% vs. 35.9%), which is a limitation given that exposure to many of the chemicals studied in this work are gender-related. This could bias comparisons with other study populations. To improve comparability between studies, we have reported the male-to-female ratio for each population in Table 3. In addition, supplementary files provide

a table (Table S4) similar to Table 3, with gender stratification where possible.

We could not compare the concentration of each substance with HBM or TDI values because these reference levels were missing from the literature. Furthermore, no scientific organization has proposed a health risk threshold that accounts for simultaneous exposure to multiple toxicants. Therefore, our evaluation of the health risks associated with non-persistent pollutants in Kinshasa is incomplete.

Furthermore, the absence of chemical analyses of consumer products sold in the Kinshasa market and external environmental media (such as food, drinks cosmetics and dust) in the DRC, and the lack of detailed lifestyle questionnaires for the recruited volunteers, constitute another limitation of our study.

Another limitation of our study is that we only used one random urine sample. Due to the short half-life of the studied compounds and the significant temporal variability in urinary concentrations, using 24-hour urine samples or repeated sampling would be more suitable options for accurately assessing the population's exposure to these compounds in Kinshasa [75].

Conclusion

Despite the limitations of our study, the results of our work shed light on the potential high exposure of the population of Kinshasa to non-persistent pollutants. If these high levels detected do not accurately reflect the population's current exposure, it could be considered as the first indicators of the current exposure to these environmental pollutants among the population in Kinshasa and, eventually, the DRC. The presence of these various compounds in our study sample merits particular attention from the Congolese people and authorities, as they are implicated in various hormonal pathologies. These substances are present in many common-use items, including packaging, cosmetics, food and pharmaceuticals. To scientifically confirm these suspected sources of

contamination, it is urgent that future studies collect and analyse samples of consumer products sold in the city.

Although the exposure to some endocrine disruptors was most often below certain toxicological reference values, it always poses a serious public health risk due to the potential additive or synergic effects of these compounds present simultaneously in the organism. Thus, eliminating or replacing these toxic products with healthier alternatives in the Congolese market will contribute to achieving the Sustainable Development Goals by 2030, which are a set of 17 global objectives adopted by the United Nations to strike a better balance between humans and the ecosystem [76]. This will require good synergy between public authorities, companies, and university researchers for continuous monitoring and the implementation of regulatory measures, as in Western countries. Consumers must also be committed to choosing safe products.

Abbreviations

5-OH-MEHP	Mono-2-ethyl-5-hydroxyhexyl phthalate
5-oxo-MEHP	Mono-2-ethyl-5-oxohexyl phthalate
6-OH-PrHP	6-hydroxypropylheptyl phthalate
7-cxOP	7-carboxy octyl phthalate
ADI	Acceptable Daily Intake
BMI	Body Mass Index
BP-3	Benzophenone-3
BPA	Bisphenol A
BPF	Bisphenol F
BPZ	Bisphenol Z
BW	Body weight
Cx-MINCH	Cyclohexane-1,2-dicarboxylate-mono-(7carboxylate-4-methyl)heptyl ester
DEHP	Di-2-ethylhexyl phthalate
DF	Detection Frequency
DRC	Democratic Republic of Congo
EDI	Estimated Daily Intake
EFSA	European Food Safety Authority
EtP	Ethylparaben
G-EQUAS	German External Quality Assessment Scheme
GM	Geometric Mean
HBM	Human Biomonitoring
HBM4EU	European Human Biomonitoring Initiative
HBM-GV	Human Biomonitoring Guidance Value
LOQ	Limit of Quantification
MBzP	Monobenzyl phthalate
MEHP	Mono-2-ethylhexyl phthalate
MeP	Methylparaben
MEP	Monoethyl phthalate
MiBP	Mono-isobutyl phthalate
MnBP	Mono-n-butyl phthalate
nBP	N-butylparaben
NHANES	National Health and Nutrition Examination Survey
OH-MINCH	Cyclohexane-1,2-dicarboxylate-mono-(7-hydroxy-4-methyl) octyl ester
OSEQAS	External Quality Assessment Scheme for Organic Substances in Urine
PrP	Propylparaben
SD	Standard Deviation
TCS	Triclosan
TDI	Tolerable Daily Intake
US EPA	United States Environmental Protection Agency

Supplementary Information

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Supplementary Material 1.

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Authors’ contributions

TB and PD drafted the manuscript and performed the statistical analysis. CP, CI, AM, MM, JN, RM, JM and CC conceived of the study and participated in its design and coordination and helped to draft the manuscript. All the authors read, commented the draft versions and approved the final manuscript.

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Data availability

Availability of the data is guaranteed by the authors upon request.

Declarations

Ethics approval and consent to participate

Ethical approval for this study was conducted by the National Health Ethics Committee in the Democratic Republic of Congo (401/CNES/BN/PMMF/2022). The study was conducted in accordance with the ethical principles outlined in the Declaration of Helsinki. All participants were informed about the purpose of the study, assured of confidentiality, and provided written consent prior to participation. Participation was voluntary, and respondents could withdraw at any time without consequence.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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