Multi-compartment chemical characterization and risk assessment of chemicals of emerging concern in freshwater systems of western Kenya

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Research

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Abstract

Background: Within the last decades, there has been increasing research on the occurrence of chemicals of emerging concern (CECs) in aquatic ecosystems due to their potential adverse effects on freshwater organisms and risk to human health. However, information on CECs in freshwater environments in sub-Saharan countries is very limited. Here, we investigated the occurrence of CECs in snails and sediments collected from 48 sites within the Lake Victoria South Basin, Kenya, which have been previously investigated for water contamination. Samples were analyzed by liquid chromatography coupled to high resolution mass spectrometry (LC-HRMS) with a target list of 429 compounds.

Results: In total, 30 compounds have been detected in snails and 78 in sediment samples, compared to 79 previously identified compounds in water. By extending the monitoring of CECs to snails and sediments, we found 68 compounds that were not previously detected in water. These compounds include the anti-cancer drug anastrozole, detected for the first time in the Kenyan environment. Individual compound concentrations were detected up to 480 ng/g wet weight (N-ethyl-o-toluenesulfonamide) in snails and 110 ng/g organic carbon (pirimiphos-methyl) in sediments. Higher contaminant concentrations were found in agricultural sites than in areas not impacted by anthropogenic activities. Crustaceans were the organisms at greatest toxic risk from sediment contamination (toxic unit (TU) up to 0.99) with diazinon and pirimiphos-methyl driving this risk. Acute and chronic risks to algae were driven by diuron (TU up to 0.24), whereas fish were found to be at low to no acute risk (TU up to 0.007).

Conclusions: The compound classes present at highest frequencies in all matrices were pesticides and biocides. This study shows substantial contamination of surface water in rural western Kenya. By filling data gaps on contamination of sediments and aquatic biota, our study reveals that CECs pose a substantial risk on environmental health in Kenya demanding for monitoring and mitigation.

Background

Chemicals of emerging concern (CECs) such as pesticides, pharmaceuticals, and personal care products (PPCPs), surfactants and other industrial chemicals are of global environmental concern due to their toxic risk on ecosystems and human health. The occurrence of CECs in freshwater ecosystems has been shown to cause adverse effects including long-term changes in the aquatic community composition (Liess & Von Der Ohe, 2005). Several studies have documented the global occurrence of CECs in freshwater resources (Luo et al. 2014; Sui et al. 2015; Aus der Beek et al. 2016; Li et al. 2017). However, most research is focused on high-income countries. Although many African laboratories are increasing their efforts to monitor the occurrence of CECs in African aquatic ecosystems (Mzukisi et al. 2017), this research field is yet to be fully explored. Many previous studies from Kenya focused largely on urban areas with a limited number of compounds investigated (Bagnis et al. 2020; Getenga et al. 2004; K’oreje et al. 2018; Kairigo et al. 2020; Kimosop et al. 2016; Ngumba et al. 2016; Otieno et al. 2015). Among the studies on pesticides in the environment, previous work largely focused on organochlorine and organophosphates compounds (Wandiga et al. 2002; Musa et al. 2011; Osoro et al. 2016) although the
next generation of pesticides including phosphoric esters and neonicotinoids are currently in use and have been found in Kenyan surface water (K’oreje et al. 2018). There is need to increase the number of compounds under research and expanding to rural areas.

When CECs are discharged into the aquatic ecosystem, they undergo equilibrium partitioning between water and sediments and may accumulate in biota according to their hydrophobicity and toxicokinetics in the organism (Di Toro et al. 1991; Mayer and Holmstrup 2008; Ashauer et al. 2011; Inostroza et al. 2017). In this context, sediments and biota act as passive samplers reflecting long and mid-term exposure, respectively, and allow also for the detection of compounds that are hardly detectable in the water phase. Detection of contaminants in biota proves their bioavailability and bioaccumulation potential (Mackay et al. 2011; Yin et al. 2017). Mollusks such as snails and mussels are particularly suitable as passive samplers due to their low metabolism competence (Nhan et al. 2001; Oehlmann and Schulte-Oehlmann 2003). Persistent organic pollutants (POPs) with a high octanol-water partitioning coefficient (logK\textsubscript{ow}) are particularly prone to accumulate in sediments and lipid-rich biological tissues. This results in long-term exposure particularly of benthic species and poses a substantial risk to the aquatic ecosystem (Li et al. 2017). Thus, a large body of literature on the bioaccumulation of legacy POPs including organochlorine pesticides, polychlorinated biphenyls (PCBs) and polycyclic aromatic hydrocarbons (PAHs) exist (Heim & Schwarzbauer, 2013; Merhaby et al. 2019). However, currently used organic chemicals including modern pesticides are designed to be more polar and less persistent. These chemicals may still accumulate in sediments and may even pose higher toxic risks to aquatic ecosystems due to their higher bioavailability. Many CECs can be considered as pseudo-persistent with emission exceeding degradation rates and have been shown to accumulate in sediments and biota (Masiá et al. 2013; Ccanccapa et al. 2016). These chemicals are in the focus of this present study.

The objectives of this study were: (1) to investigate the occurrence and distribution of CECs in aquatic ecosystems in snails and sediments in the Lake Victoria South Basin and to reveal whether analyzing these matrices can substantially increase the number of chemicals detected compared to the water analysis only, (2) to determine the spatial distribution of contaminants dependent on land use within the Lake Victoria South Basin, and 3) to assess risks of sediment-associated chemicals on fish, crustaceans and algae by calculating equilibrium water concentrations from sediment concentrations. For the water phase, data were obtained from our previous study (Kandie et al. 2020) and used for comparison of compound concentrations in snail and sediments.

**Materials And Methods**

**2.1 Chemicals**

Analytical standards were purchased from various suppliers with purity of above 97%. The 429 target compounds included pesticides and biocides (PaBs), PPCPs, industrial compounds and transformation products which have been detected in surface waters and sediments. More information on the target compounds and internal standards is presented in the Supplementary Information (SI Table SI-1 and
Table SI-2 respectively). Methanol (LC-MS grade), ethyl acetate, dichloromethane (DCM) and acetone (all LC grade) were obtained from Sigma Aldrich (Germany), while LC-MS grade water was purchased from Thermo-Fisher (Germany). Primary Secondary Amine (PSA), Sodium chloride and magnesium sulfate were supplied by Sigma-Aldrich.

2.2 Description of the study area and sampling

The study area was located within the Lake Victoria South Basin (LVSB) in western Kenya covering Kisii, Nyamira, Migori, Homabay, Kericho, Kisumu and Narok counties. Sampling sites have been described in detail in Kandie et al. (2020) and Becker et al. (2020). Forty-eight sites were sampled for sediments and snails between September and October 2017 in parallel to water samples analyzed previously Kandie et al. (2020). For rivers or drainage canals, 50-m transects were sampled with four sampling spots distributed equally along the transect, with bias on suitable snail habitats. For reservoirs, the four edges were taken as sampling spots in each site. Snails were sampled using a handheld stainless-steel snail catcher. Approximately 200 g of sediment were sampled using a pre-cleaned stainless-steel scoop, homogenized, sieved (2 mm) and transferred to a zip lock bag. Sediment samples were covered with aluminum foil to avoid photo degradation and immediately put into a portable freezer (-4°C) and transferred to -20°C in the laboratory. Snails were kept alive in falcon tubes for identification for up to 15 hours. After identification in the laboratory, the samples were stored at -20°C prior to extraction.

2.3 Sample preparation and extraction

2.3.1 Snail extraction

Snails were found in 20 out of the 48 sites sampled and separated according to sites and species. The snails sampled were from the genera Bulinus, Biomphalaria, Melanoides, Lymnaea, Physa and Ceratophallus. The snails were found in shallow slow-moving water. Except for Physa which were much larger (approximately 3 cm), all the other genus had an average size of 5mm. Most of the snails were attached to aquatic vegetation within streams and drainage canals. The snails from each site belonging to the same species were pooled. Chemicals were extracted from the snail tissues by applying a modified QuEChERS (Quick, Easy, Cheap, Effective, Rugged and Safe) method as described by Inostroza et al. (2016). Briefly, 1 g of crushed snails was homogenized in 5 mL acetonitrile (LC-MS grade) using a Stuart handheld homogenizer (SHM1/UK) for 1 minute. For salting out, 2 g of anhydrous magnesium sulfate and 500 mg of sodium chloride were added, vortexed immediately on highest intensity for 2 minutes and centrifuged at 3000 x g for 5 minutes. An aliquot (1 mL) of the acetonitrile phase was transferred into a pre-assembled 2-ml QuEChERS tube containing 150 mg anhydrous magnesium sulfate and 50 mg Primary Secondary Amine (PSA) for clean-up, vortexed on highest intensity for 1 minute and centrifuged for 5 minutes at 17,000 x g. The supernatant extract was then vacuum filtered (0.2 µm PTFE Samplicity system filter, Merck Millipore, Germany) into a 1.5 mL autosampler vial. The filtered extract (400 µL) was
transferred into amber autosampler vials and stored at -20°C until instrumental analysis. A method blank was prepared using water from a pristine reference stream within the Harz Mountains (Wormsgraben, Germany) and processed similar to the samples. From recovery experiments carried out in a pre-study, 80% of the compounds had recoveries between 70% and 120% showing a good overall performance of the QuEChERS method.

2.3.2 Sediment extraction and clean up

Sediment samples were freeze-dried (SP Scientific, Advantage EL-85) and the sediment mass extracted was adjusted according to the total organic carbon (TOC) contents to obtain a comparable matrix load of the extracts (Massei et al. 2018). The TOC content of the sediments was determined on a Flash 200 organic elemental analyzer coupled to a Delta V Advantage Isotope Ratio mass spectrometer (Thermo). The analyzer was operated at 1000°C to ensure complete combustion. Details of the TOC procedure are described in SI SI-3.

Compounds were extracted from sediments according to the method described by Massei et al. (2018) with minor modifications. Briefly, pressurized liquid extraction (PLE, ASE 200 device, Dionex) was performed by taking approximately 5 to 10 mg of freeze-dried sediment (corresponding to 100 mg of TOC content) and mixed with pure diatomaceous earth (25% of sediment weight, Hydromatrix, Restek). This mixture was transferred to stainless steel PLE cells fitted with 27 mm glass fibre filters (Dionex) and extracted at 100°C with ethyl acetate and acetone (50:50, v/v) in two static cycles at a pressure of 100 bar. A method blank containing only hydromatrix was prepared for each batch of samples to evaluate instrument background contamination. The extract was concentrated to 0.5 mL using a gentle stream of nitrogen (Xcelvap, Thermo), and the solvent was exchanged for DCM. For extract clean-up, flash chromatography was performed using a pre-packed chromatography column (Chromabond Flash RS 4 SiOH, 4 g, Macherey-Nagel) and an Agilent 1260 binary pump. Conditioning of the column was done using DCM prior to clean up. A corresponding volume of internal standard (1 µg/mL) was added to the concentrated sample extract in the vials (final concentration 50 ng/mL). Using a glass pipette, the extract was transferred to the flash column for clean up using DCM and methanol at 5 mL min⁻¹ (SI-4) and collected in separate vials. The DCM and methanol fractions were combined (50:50, w/w) and concentrated to 1 mL using a gentle stream of nitrogen while rinsing the vial walls with methanol and blowing down to 0.5 mL to exchange the solvent completely for methanol.

2.4 Instrumental analysis

Aliquots of 100 µL snail extracts were transferred into 2 mL vials with insert together with 10 µL of internal standard mix (1 µg/mL) containing 40 isotope-labelled compounds (Table SI-2). Eleven method-matched calibration levels were prepared ranging from 0.05 to 500 ng/mL in vial. For the calibration samples, an appropriate volume of the standard solution was added to 1 mL of water and processed in the same way as the samples.
Sediment extracts were analyzed in 100-µL aliquots transferred to a 2-mL vial with insert. Method-matched calibrations were prepared at twelve concentration levels corresponding to 0.2 to 2000 ng/mL in vial by adding standard solutions to 5 mL of ethyl acetate:acetone (50:50 v/v) to mimick the PLE extracts. The solvent was evaporated using a gentle stream of nitrogen (Xcelvap, Thermo) and exchanged to DCM. Calibration solutions were subjected to clean-up with flash chromatography after adding 50 µL of internal standard (50 ng/mL).

All the samples were analysed using liquid chromatography (Thermo Ultimate 3000 LC) coupled to high resolution mass spectrometry (QExactive Plus, Thermo) equipped with a heated electrospray ion source. Snail and sediment extracts were analysed by injecting 10-µL and 5-µL aliquots into the instrument, respectively. Instrument settings and the chromatographic conditions have been described by Kandie et al. (2020). A water (A) and methanol (B) solvent gradient, both with 0.1% v/v formic acid, was applied in separate positive and negative ionization analytical runs. A combination of full scan (m/z range 100-1500) at a nominal resolving power of 70,000 (referenced to m/z 200) and data-independent MS/MS fragmentation at a resolving power of 35,000 was used for both positive and negative mode. The mass spectrometer settings are described in Kandie et al. (2020).

2.5 Risk assessment based on Toxic Units (TU)

The ecotoxicological relevance of the obtained compound concentrations was evaluated by performing risk assessment based on toxic units (TU). Equilibrium water concentrations ($C_{ewsed}$) were derived from concentrations in sediment organic carbon ($C_{sed}$) assuming equilibrium partitioning (Equation 1). Organic carbon-water partitioning coefficients ($K_{oc}$) were derived using linear solvation energy relationships (LSER) applying the open access UFZ-LSERs database (Ulrich et al. 2017) as described by Inostroza et al. (2017).

Equation 1: see equation 1 in the supplementary files.

The TU was calculated for each chemical by normalizing the equilibrium water concentrations ($C_{ewsed}$) to the effect concentrations (EC) for fish, crustaceans and algae according to Equation 2. Effect concentrations were derived from Busch et al. (2016). The EC values were retrieved from United States Environmental Protection Agency’s (USEPA) ECOTOX database and were based on the 5th percentile of measured acute values (Busch et al. 2016), predicted read-across or ECOSAR.

Equation 2: see equation 2 in the supplementary files.

The calculated TUs were compared to acute and chronic risk threshold values as proposed by Malaj et al. (2014). The acute risk thresholds (ART) for all organisms is 0.1 TUs while chronic risk thresholds (CRT) for fish (0.01 TUs), Daphnia (0.001 Tus) and algae (0.02 TUs) were applied.
To predict mixture toxicity, individual TU values were summed up (TU\textsubscript{sum}) based on the concentration addition (CA) model (Loewe & Muischnek, 1926) designed for compounds with similar mode of action (Altenburger et al., 2003; de Almeida et al., 2017) but being also a reasonable estimate for mixtures of environmental compounds without knowing their individual modes of action (Backhaus and Faust 2012).

2.6 Data analysis

Peak detection and annotation of target compounds was performed using MZmine (Version 2.38, Pluskal et al., 2010) and detected target compounds were further confirmed and quantified using TraceFinder 4.1 (Thermo). The MZmine and TraceFinder settings were applied as shown in Kandie et al. (2020). Jchem was used for structure-based determination of compound properties including molecular formula and exact mass. The method detection limits (MDLs) were determined using calibration standards based on USEPA (2011) guidelines. Graphs and statistical analysis were performed using Microsoft Excel 2013 and SigmaPlot 13.0.

Results And Discussion

3.1 Body burden of pollutants in snails

Out of the 429 compounds targeted, 30 compounds including PaBs, PPCPs and industrial compounds were detected in snail tissues (Figure 1). Concentrations detected in snail tissues on wet weight basis (ng/g ww) are given in the SI (Table SI-5 and Figure SI-1).

Figure 1: Frequency of detection for compounds in snail tissue samples from the Lake Victoria South Basin. TCEP: Tris(2-chloroethyl)phosphate; NBBS: N-Butylbenzenesulfonamide; NETS: N-Ethyl-o-toluenesulfonamide; BTSA: 2-Benzothiazolesulfonic acid.

Almost two thirds of the chemicals detected in snails were PaBs with 19 out of 30 compounds (Figure 1). Atrazine, a pre-emergence and post-emergence herbicide still in use in Kenya for the control of broadleaf weeds and grasses, was detected in 95% of the snail samples followed by the insect repellant diethyltoluamide (DEET) with 93%. The frequent detection of DEET may be explained by the high mosquito infestation and the high prevalence of malaria infecting 20 to 40% of the population in counties within the lake endemic region of western Kenya (Bashir et al., 2019). DEET is used as a topical insect repellant for the control of mosquitoes. It could be released into the water when performing water-related activities such as taking a bath in the rivers and reservoirs (Thomas et al. 2014), a common practice which was observed during sampling in the region. Individual pesticide concentrations ranged from 0.2 ng/g ww to 375 ng/g ww with maximum concentrations found for the herbicide atrazine at 375 ng/g ww, the fungicide bupirimate at 97 ng/g ww and the insecticide diazinon (37 ng/g ww). The neonicotinoids, acetamiprid and imidacloprid were present in the snail tissues in concentrations up to 27 ng/g ww and 21 ng/g ww, respectively.
Among PPCPs, temazepam (98%) and 2-hydroxyquinoline (78%) had the highest detection frequencies (Figure 1). Individual compound concentrations ranged from 0.8 ng/g ww to 137 ng/g ww. Maximum concentrations were recorded for efavirenz (137 ng/g ww) and 2-hydroxyquinoline (115 ng/g ww). This highest concentration for efavirenz was found in PS28 (Figure SI-1) located within Kisumu County and could be attributed to the high HIV/AIDS prevalence (16.3%) and the access to antiretroviral therapy (ART) with a coverage of 90% (National AIDS Control Council NACC 2018) in the region. Efavirenz is a non-nucleoside reverse transcriptase inhibitor (NNRTI) used in combination with other medications as antiretroviral treatment for HIV/AIDS (Kreitchmann et al. 2019).

Plasticizers and flame retardants were also present in snail tissues with highest detection frequencies reported for tris(2-chloroethyl)phosphate (100%) and N-butylbenzenesulfonamide (NBS, 80%). Individual compound concentrations ranged from 2.8 ng/g ww to 481 ng/g ww. The highest concentration was found for N-ethyl-o-toluene sulfonamide (NETS, 481 ng g\(^{-1}\) ww), a compound with wide-spread use in industrial products and as an ingredient to pesticide formulations. In addition, cotinine was present in all the snails sampled (detection frequency 100%) with concentrations up to 311 ng/g ww (Figure SI-1). Due to the compound being the most abundant metabolite of nicotine, its high stability and half-life, cotinine has been suggested to be the ideal biomarker of tobacco exposure and smoking status (Petersen et al. 2010). Cotinine is excreted by human mainly through the urine and may end up in surface water through effluent discharge of domestic waste.

Variability in compound concentrations was observed in the different snail species sampled from the same site. Example sites include PS17 and PS28 (Figure SI-1) where total concentrations in *Ceratophallus sp.* and *Melanoides tuberculata* were lower than those in other species. A plausible explanation for this observation may be different lipid contents and age which could not be considered during sample preparation. A study carried out by Duncan et al. (1987) noted considerable variation in total lipid for *Biomphalaria glabrata* (5%) and *B. alexandrina* (2%) snails and, in addition, intraspecific differences in total lipids (1-10%) in *B. glabrata*. Since most pollutants accumulate primarily in lipid tissue, variation in lipid content may lead to differences in the body burden of contaminants.

The predators of the snails investigated in this study include cray fish (Procambarus alleni), water bug (Belostoma flumineum), waterfowl and cichlid fishes (Monde et al. 2016; Halstead et al. 2018). These predators could biomagnify and bioaccumulate certain contaminants during feeding resulting in elevated levels of contaminants in the food chain. The consumption of contaminated food is a major source for xenobiotics in predating birds and mammals (Nendza et al. 1997). Effects of pesticide exposure in birds have been linked to neurotoxicity and endocrine disruption (Köhler and Triebskorn 2013). Other effects of pesticides include impaired foraging and chick rearing, eggs shell thinning and reproductive failure (Köhler and Triebskorn 2013). In a study carried out by Guo et al. (2016) on compound prioritization based on potential of secondary poisoning in fish-eating birds and mammals, diazepam was ranked the highest (risk score 0.1–1) among the pharmaceuticals tested. In addition, gregarious animals including fish and birds could biomagnify and bioaccumulate contaminants then migrate becoming a predominant pathway for contaminants in the environment (Blais et al. 2007).
3.2 Occurrence and distribution of pollutants in sediments

Out of the 429 targeted compounds analyzed in the sediment samples from the LVSB, 78 compounds were detected (Figure 2 and SI Table SI-6).

Figure 2: Frequency of detection for compounds in sediment samples from the Lake Victoria South Basin. 3,4,5-TCP: 3,4,5-trichlorophenol; 2-MBT: 2-morpholinothiobenzothiazole; 2_IPT: 2-Isopropylthioxanthone; 6:2-FTSA: 6:2 fluorotelomer sulfonic acid

Similar to snail samples, PaBs were the dominant chemical class with 71% of compounds detected in the sediments. Compounds frequently detected included DEET (98%), triclocarban (71%), diuron (65%), pirimiphos methyl (58%) and diazinon (56%) (Figure 2). High detection frequencies of diazinon and pirimiphos-methyl are in line with a study performed by Musa et al. (2011) who noted that diazinon and pirimiphos-methyl were among the commonly used pesticides in the Nyando catchment area which is within the LVSB. Individual compound concentrations reached up to 111 ng/g organic carbon (OC) with highest concentrations recorded for pirimiphos-methyl (111 ng/g OC), diuron (93 ng/g OC) and DEET (68 ng g⁻¹ OC) (Figure 3). Pirimiphos-methyl is a broad spectrum insecticide used for the control of pest during storage. In addition, it is approved as an insecticide for indoor spraying against mosquitoes, cockroaches and houseflies (Pest Control Products Board 2018). Diuron is a selective herbicide for the control of weeds in sugarcane plantations. Concentrations reported in this study are within the range reported in a review by K’oreje et al. (2020) on the occurrence of pesticides in African river sediments.

Figure 3: Compound concentrations and spatial distribution in sediment samples

A total of 19 PPCPs were detected with frequencies ranging from 6% (bezafibrate) to 56% (efavirenz). We also found the preservative propylparaben (33%) and the anti-cancer drug anastrozole (8%). To the best of our knowledge, this is the first study to report anastrozole occurrence in Kenyan aquatic ecosystems. The detection of anastrozole could be linked to the rising diagnoses and treatment of breast cancer in the continent and particularly in Kenya (Ekpe et al., 2019). Anastrozole is applied for hormone therapy during breast cancer treatment. Highest PPCP concentrations were detected for efavirenz with up to 29 ng/g OC. Other compounds detected at higher concentrations include crotamiton (1.8 ng/g OC) and diazepam (1.5 ng/g OC). The concentrations of the antibiotic sulfamethoxazole measured in this study (0.21 ng/g OC) fall below the concentrations reported by Kairigo et al. (2020) in sediments from Mwania river in Kenya by about one order of magnitude. This is probably due to differences in consumption patterns and the impact of municipal waste in the study area since their study was performed in an urban setting.

3.3 Comparison of the incidence of CECs in different environmental matrices of western Kenya

The chemical data from the present study were compared with the compounds found in the water phase in Kandie et al. (2020). In total, 142 compounds were detected in the study area with 79 compounds in
3.4 Impact of land use on contamination patterns in different environmental compartments

Among the 48 sampling sites, 17 sites could be clearly connected to specific land uses (Figure SI-3), including agricultural areas (i.e., sugarcane, tea and rice) and reservoirs characterized by low anthropogenic inputs. The other sites showed mixed land use patterns and were not taken into consideration. Overall, sugarcane growing areas showed the highest concentrations of PaBs of all investigated types of land use (Figure 3, Figures SI-1, SI-2 and SI-3).
Sites located in areas without evident anthropogenic influence (PS21, PS39, PS46, PS47 and PS58) were generally less contaminated although total CEC concentrations (1 µg/L) in water from PS 39 indicates hidden wastewater impact as a source for the pharmaceuticals acetyl-sulfamethoxazole (A-SMX) and diphenhydramine, and the industrial compound triethycitrate (Figure 3, Figures SI-1, SI-2 and SI-3). Total CEC concentrations were up to 2 ng/g OC (mainly triclocarban and DEET) and 228 ng/g ww in sediments and snails, respectively. Cotinine, efavirenz, atrazine, N-ethyl-o-toluene sulfonamide and N-butylbenzenesulfonamide were the compounds contributing to the pollution found in snails. Low contaminant concentrations were observed in sites such as in PS46 located within Homabay County with no immediate impact from anthropogenic activities. In addition, a lot of the farmers in this region practice subsistence farming for household consumption, therefore less pesticide inputs may be used in the agricultural practices. The site PS 47 is located closely downstream of the Ruma national park, a wildlife reserve with limited human activities.

Agricultural sites such as PS17 and PS18 within sugarcane plantations had high contaminant concentrations in all the three matrices (Figure 3, Figures SI-1, SI-2 and SI-3). Total compound concentrations were up to 11 µg/L in water, 555 ng/g ww in snails, 304 ng/g OC in sediments. Notably, high concentrations were obtained for the PaBs atrazine in snails and pirimiphos-methyl, DEET and triclocarban in sediments, while 2,4-dichlorophenoxyacetic acid (2,4-D) and hexazinone predominated in water (Kandie et al. 2020). The high concentrations of pesticides are in agreement with sampling during the spraying period in sugarcane plantations (September-October). Other compounds that contributed to pollution included efavirenz and NETs (in both snails and sediments) while the hydrophilic CECs A-SMX, the sweetener acesulfame and triethycitrate were found only in water samples from these two sites. The presence of Awendo town nearby could contribute as an important source of these compounds into the aquatic system.

In rice plantation fields (PS25, PS26, PS27, PS28 and Uli15), high total compound concentrations of up to 3.3 µg/L in water, 598 ng/g ww in snails, 22 ng/g OC in sediments were detected. Similar to sugarcane sites, PaBs contributed most to the overall pollution with atrazine predominating in snails and triclocarban in sediments. Carbendazim and bendiocarb contributed most to water contamination (Kandie et al. 2020).

Among agricultural areas, sites within tea plantations (PS56, PS57, Uli1, Uli3) were least impacted with pollutants. Total CEC concentrations reached up to 6 ng/g OC in sediments with the insect repellant DEET as major contributor, while no snails were found at these sites. A plausible explanation for the low pesticide contamination is the mismatch of sampling (September and October) and the spraying period in tea plantations (June). Additionally, the streams in tea growing areas are protected by wide buffer zones while in other agricultural areas the farmland is much closer to the waterbodies leading to higher run-off potential.

In general, the occurrence of PPCPS in agricultural sites shows evidences of municipal waste impact on the local water bodies. For example, the pharmaceuticals efavirenz and 2-hydroxyquinoline were found in
high concentrations in snails collected from rice fields. A plausible explanation for the high concentrations found could be the direct discharge of untreated domestic wastewater from residential areas, lack of sanitation facilities and effluent discharge from wastewater treatment facilities into the river.

### 3.5 Risk assessment based on sediment concentrations

Based on equilibrium water concentrations calculated from sediment, toxic risks were estimated for fish, crustacean and algae. For fish, the $\text{TU}_{\text{sum}}$ ranged from $9.1 \times 10^{-8}$ to $1.4 \times 10^{-2}$ (Table SI-7, Figure SI-4). Pirimiphos-methyl and imidacloprid-urea were identified as risk drivers for fish exposed to sediments in PS17, while diuron is predominating in PS59. Ethyl azinphos contributed greatly to the risk in PS42 and PS43. Maximum TUs were observed for pirimiphos-methyl ($\text{TU} = 0.007$) and the transformation product imidacloprid urea ($\text{TU} = 0.005$), but did not exceed the acute and chronic risk thresholds. However, with this low toxic risk on fish observed, it should be mentioned that natural and synthetic estrogenic hormones were not measured in this study. These hormones have been shown to drive effects on fish reproduction in the ng/L range resulting in the collapse of whole fish populations (Kidd et al. 2007). Estrogens are often emitted with untreated wastewater (König et al. 2017).

Cumulative TUs for crustaceans were generally higher than those obtained for fish (range: $9.6 \times 10^{-8}$ to 1.1) with diazinon, fipronil sulfone and pirimiphos-methyl driving the overall risk (Table SI-8, Figure 5). Maximum $\text{TU}_{\text{sum}}$ was reported for the site PS17 ($\text{TU}_{\text{sum}} = 1.1$) impacted by the large agro-industrial sugarcane plantation with pirimiphos-methyl ($\text{TU} = 0.99$) and diazinon ($\text{TU} = 0.11$) driving the risk, both exceeding the ART ($\text{TU} > 0.1$) for crustaceans. The chronic risk threshold of $\text{TU} > 0.001$ was exceeded at 20 sites for diazinon, at 14 sites for pirimiphos-methyl and at four sites for fipronil sulfone.

Figure 5: Distribution of risk of toxicity for crustaceans from compounds present in sediment extract based on equilibrium water concentrations. 345-TCP:3,4,5 trichlorophenol

For algae, the cumulative risk was higher than for fish but lower compared to crustaceans. The $\text{TU}_{\text{sum}}$ ranged from $8.7 \times 10^{-7}$ to 0.24, with the photosynthesis inhibitor diuron driving the risk to algae at most sites (Table SI-9, Figure 6). Diuron is a selective herbicide for the control of weeds in sugarcane plantations (Pest Control Products Board 2018). Highest TU values were obtained in sediments from PS59 ($\text{TU} = 0.24$) exceeding ART and PS17 ($\text{TU} = 0.03$) exceeding CRT.

Figure 6: Distribution of risk of toxicity for algae from compounds present in sediment extract based on equilibrium water concentrations. 345-TCP:3,4,5 trichlorophenol; 6:2-FTSA: 6:2-fluorotelomer sulfonic acid

### Conclusions

The present study bridges substantial data gaps on contamination of freshwater habitats in rural areas within western Kenya with CECs. The study demonstrates that complementing water monitoring with the
analysis of sediments and biota may strongly increase the number of detectable contaminants allowing for a more comprehensive assessment of pollution. In this study 66 chemicals were detected only in snails and sediments while 51 compounds were found exclusively in water. Although common hot spots of contamination were identified in all three matrices, sediments and snails provide a picture that is rather independent from water concentrations measured in randomly taken grab samples. These findings support the complementary monitoring of biota and sediments as time-integrated samplers of pollution.

Although partly confounded by complex land-use structures, contamination of sediments and biota could be linked to specific agricultural production. Pesticide application in sugarcane plantations and rice fields were important sources of contamination and toxic risks to aquatic organisms. Poorly treated and untreated municipal wastewater and a lack of sanitation may be seen as another highly relevant source of pollution with pharmaceuticals and personal care products.

Very high toxic risks due to contaminated sediments were found for crustaceans in some of the sites, with diazinon and pirimiphos-methyl driving this risk. Substantial acute and chronic risk was also observed for algae mainly driven by diuron, while fish suffered only low toxic risk. Sublethal effects such as endocrine disruption and other specific effects by natural and synthetic steroids and pharmaceuticals discharged with untreated wastewater could not be considered in this study but might be more relevant for fish populations than acute toxicity. Future studies should focus on potential toxicological risk to humans and wildlife resulting from bioaccumulation and biomagnification of certain contaminants due to consumption of contaminated food.

Overall, this study indicates substantial contamination of rural areas in western Kenya and promotes systematic monitoring and assessment of CECs in different matrices in order to characterize and mitigate risks to ecosystems but also human health.

Declarations

Ethics approval and consent to participate

Compliance with relevant national and institutional guidelines, and appropriate permissions were obtained and followed during this study.

Consent for publication

Not applicable

Availability of data and material

The datasets supporting the conclusions of this article are included within the article and its additional files.
Competing interests

The authors declare that they have no competing interests

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Authors’ contributions

FK, MK, AG, BT, UL, JB, ML and WB designed the research; FK and AG conducted the research; FK, MK and RM analyzed and interpreted the data; FK drafted the initial version; MK, RM and WB edited and reviewed; FK, MK, RM, AG, BT, UL, JB, ML and WB contributed to the final version. All authors read and approved the final manuscript.

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Abbreviations

ART: acute risk threshold; CRT: chronic risk threshold; CECs: chemicals of emerging concern; $C_{ew}$: equilibrium water concentrations; $C_{sed}$: sediment organic carbon; DCM: dichloromethane; EC: effect concentrations; $K_{oc}$: organic carbon-water partitioning coefficients; LC-HRMS: liquid chromatography - high resolution mass spectrometry; LVSB: Lake Victoria South Basin; PLE: pressurized liquid extraction; PPCPs: pharmaceuticals and personal care products; PSA: Primary Secondary Amine; QuEChERS: Quick, Easy, Cheap, Effective, Rugged and Safe; TOC: organic carbon contents; TU: toxic units

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Figures

Figure 1

Frequency of detection for compounds in snail tissue samples from the Lake Victoria South Basin. TCEP: Tris(2-chloroethyl)phosphate; NBBS: N-Butylbenzenesulfonamide; NETS: N-Ethyl-o-toluenesulfonamide; BTSA: 2-Benzothiazolesulfonic acid.
Figure 2

Frequency of detection for compounds in sediment samples from the Lake Victoria South Basin. 3,4,5-TCP: 3,4,5-trichlorophenol; 2-MBT: 2-morpholinothiobenzothiazole; 2-IPT: 2-Isopropylthioxanthone; 6:2-FTSA: 6:2 fluorotelomer sulfonic acid
Figure 3

Compound concentrations and spatial distribution in sediment samples
Figure 4

Numbers of detected compounds in water, sediment and snail samples within the study area.

Figure 5
Distribution of risk of toxicity for crustaceans from compounds present in sediment extract based on equilibrium water concentrations. 345-TCP: 3,4,5 trichlorophenol

Figure 6

Distribution of risk of toxicity for algae from compounds present in sediment extract based on equilibrium water concentrations. 345-TCP: 3,4,5 trichlorophenol; 6:2-FTSA: 6:2-fluorotelomer sulfonic acid

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