Emerging Contaminants 11 (2025) 100580

Contents lists available at ScienceDirect

Emerging Contaminants

journal homepage: www.keaipublishing.com/en/journals/emerging-contaminants

Novel and legacy per- and poly-fluoroalkyl substances in major wastewater treatment plants within the Lake Victoria basin, East Africa

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ARTICLE INFO

Article history:
Received 3 July 2025
Received in revised form
7 September 2025
Accepted 22 September 2025
Available online 22 September 2025

Keywords:
Per- and poly-fluoroalkyl substances
Wastewater treatment plants
Mass loadings
Ecological risks
Lake victoria

ABSTRACT

Over the past two decades, rapid urbanization and industrialization in Uganda have generated wastewater containing emerging contaminants including per- and poly-fluoroalkyl substances (PFASs). This study assessed PFASs contamination of wastewater from Bugolobi (Kampala) and Kirinya (Jinja) wastewater treatment plants (WWTPs) by analyzing 80 influent and effluent samples for 15 PFASs using LC-MS/MS. We quantified 10 PFASs, with levels ranging from non-detectable (n.d) up to 372.4 ng/L (mean: 20.94 ± 0.42 ng/L). At Bugolobi WWTP, influent levels ranged from n.d to 190.01 ng/L (60.85 ± 1.03 ng/L) while effluents varied from n.d to 372.4 ng/L (237.91 ± 7.06 ng/L). At Kirinya WWTP, influent levels ranged from n.d to 29.37 ng/L (17.58 ± 3.54 ng/L) and effluents up to 30.21 ng/L (7.79 ± 0.85 ng/L). Short-chain PFASs (PFBS, PFBA) were more predominant, suggesting their possible use or degradation of the long-chain PFASs. Total mass loadings were higher at Bugolobi WWTP (5353.56 mg/day), serving the more densely populated Kampala, than at Kirinya WWTP (93.62 mg/day). PFSAs exhibited higher removal (72.45 % Bugolobi; 36.45 % Kirinya) than PFCAs (−127.38 % Bugolobi; −20.50 % Kirinya), which could be attributed to their stronger hydrophobic adsorption and partial biodegradation. Bugolobi, with ~82.59 % total removal outperformed Kirinya (~25.19 %) due to its advanced conventional treatment. Ecological risk assessment revealed higher risks at lower trophic levels at Bugolobi compared to Kirinya, likely due to lower influx and partial mitigation by its pond-based system. These findings highlight the role of WWTPs as critical point sources of PFASs, posing ecological risks to aquatic ecosystems.

1. Introduction

Per- and poly-fluoroalkyl substances (PFASs) are synthetic chemicals extensively used in various industries, including

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Peer review under the responsibility of KeAi Communications Co., Ltd.

https://doi.org/10.1016/j.emcon.2025.100580

textiles, firefighting foams, and non-stick cookware. These chemicals are desirable due to their unique physicochemical properties such as thermal stability and water repellence [1,2]. However, their widespread use and extreme persistence have led to global environmental concerns. PFASs resist chemical, thermal, and biological degradation due to the strength of their carbon-fluorine bonds, resulting in their accumulation in aquatic systems and biota [3,4]. The environmental persistence of PFASs is compounded by their transformation under environmental conditions via hydrolysis, photolysis, or microbial degradation [5,6]. For instance, partially fluorinated precursors degrade into highly stable perfluoroalkyl substances, contributing to the dominance of short-chain PFASs in wastewater matrices [7]. Though less bioaccumulative than their long-chain counterparts, these short-chain variants exhibit higher mobility and water solubility, raising concerns about their potential ecotoxicological impacts [8].

Studies have shown that PFASs exposure adversely affects aquatic organisms by causing developmental delays, genotoxicity, reduced growth, and reproductive impairments in fish, in addition to bioaccumulation in tissues that threatens higher trophic organisms, including humans [9–11]. These toxicological risks coupled with their persistence underscore the urgent need for stringent monitoring and mitigation efforts. Moreover, more than 4730 PFASs have been identified globally, but more attention has been given to the legacy compounds such as perfluorooctanoic acid (PFOA) and perfluorooctanesulfonic acid (PFOS) [1]. Regulatory measures, such as the voluntary phase-out of PFOS production by 3M in 2000 and its inclusion in the Stockholm Convention on Persistent Organic Pollutants (POPs) in 2009, have significantly reduced the production and use of these compounds in developed countries [12,13]. However, this shift has driven the increased use of short-chain PFASs as substitutes, especially in developing regions where regulatory frameworks for managing PFASs remain inadequate [14].

In Africa, research on the occurrence of PFASs is sparse, with studies conducted in only 11 countries to date, primarily focusing on wastewater, surface water, and biota [15]. Wastewater treatment plants (WWTPs), commonly established in urbanized regions to manage diverse waste streams, play a significant role in the discharge of PFASs into aquatic ecosystems [16]. These facilities receive wastewater inputs from industrial, commercial, and domestic sources, making them hotspots for PFASs contamination [17]. Although studies on mass loading of PFASs in WWTPs have been conducted in Kenya [18] and South Africa [19], such reports remain generally scarce across the African continent. The present study, therefore, provided a case study on regional mass loadings from WWTPs into the Lake Victoria basin, which contributes to addressing this broader continental knowledge gap.

In Uganda, urbanization and industrialization within the Lake Victoria basin, particularly in cities such as Kampala and Jinja, have significantly increased wastewater generation, placing immense pressure on the existing WWTPs [20]. Lake Victoria, the largest freshwater body in Africa and a critical resource for over 40 million people across Uganda, Kenya, and Tanzania, serves as a vital water source, supporting agriculture, fisheries, and domestic use [21]. Despite its substantial dilution capacity, the lake is increasingly vulnerable to contamination from anthropogenic activities, including untreated and partially treated effluents discharged from urban areas and WWTPs.

Although PFASs have been detected in Lake Victoria and its tributaries [22], the role of WWTPs as point sources of PFASs within Uganda remains poorly understood. Earlier investigations in the basin have reported PFOS as the predominant compound in fish muscle (0.067–0.136 ng/g ww) and PFOA <0.20 ng/g ww in Nile perch and <0.13 ng/g ww in Nile tilapia, with liver concentrations ranging up to 1.95 ng/g ww [23]. In surface waters, average PFAS concentrations ranged from 0.08 to 23.8 ng/L in the Nakivubo Channel, and from n.d–1.46 ng/L in open lake water, with PFHxA and PFOS dominant across multiple matrices, including atmospheric deposition and municipal tap water [22]. A study by Dalahmeh et al. [20] measured $\sum$PFASs of 8.5–12 ng/L in Nakivubo Channel, 1.0–2.5 ng/L in lake water, 1700–7900 pg/g dw in soils, and 160–380 pg/g dw in crop tissues, identifying WWTPs as probable point sources.

Nonetheless, little is known about PFASs levels, mass loadings, or transformations in WWTPs, highlighting the need to assess their contribution to PFASs pollution in Lake Victoria, and its downstream ecosystems, such as the River Nile, which serves over 11 African countries and 300 million people [15]. Given the growing use of PFAS-containing products, this knowledge gap poses ecological risks, especially in aquatic ecosystems that are exposed to effluents from the WWTPs. This study aimed to bridge this knowledge gap by providing a comprehensive assessment of PFASs in wastewater from Uganda's largest WWTPs of Bugolobi: conventional (Kampala) and Kirinya: non-conventional (Jinja). The study objectives were to: (i) determine PFASs levels in influent and effluent samples, (ii) evaluate PFASs mass loadings into the receiving water bodies, (iii) assess the removal efficiency of WWTPs, and (iv) evaluate potential ecological risks to aquatic life in the receiving aquatic ecosystem.

2. Materials and methods

2.1. Study area

This study was conducted at Bugolobi WWTP in Kampala and Kirinya WWTP in Jinja, Uganda (Fig. 1). Kampala and Jinja are among the most densely populated, urbanized, and industrialized cities in Uganda [24]. The urban population in Kampala and Jinja frequently uses PFAS-relevant consumer products such as packaged food containers, textiles, lubricants, adhesives, and paints. Although our study did not quantify their relative contributions, research findings indicate that such consumer goods represent major pathways of PFAS entry into the wastewater systems [2,25]. Bugolobi WWTP, the largest treatment facility in East Africa, has a design capacity of about 45 million L/day, and it employs the conventional treatment configuration: primary sedimentation, biological trickling filters for secondary treatment, and final chlorination [26]. Kirinya WWTP, the largest stabilization pond system in Uganda, is located along Lake Victoria and is operated by National Water and Sewerage Corporation. It comprises a series of facultative and maturation ponds that provide extended retention for natural treatment. Both plants discharge treated effluent into streams that drain into Lake Victoria, the largest freshwater lake in Africa ([27]; see Table S1a for full WWTP performance and operational details).

2.2. Sample collection

A total of 80 wastewater samples (1 L each) were collected in August 2024 from Bugolobi WWTP (n = 40) and Kirinya WWTP (n = 40). Uganda has a bimodal rainfall regime with wet seasons in March–May and September–November, and drier periods in December–February and June–August. Sampling was scheduled at the late dry–early wet transition to capture storm-influenced inflows that are most relevant for PFAS transport, thereby improving representativeness despite limited duration [28]. At Bugolobi WWTP, samples were collected sequentially over three days from three treatment stages: initial influent (IIF), primary effluent (PEF), and final effluent (FEF). The plant operates at ~750 m^3/h, with a short hydraulic retention time (HRT) of ~3 h [26]. To account for

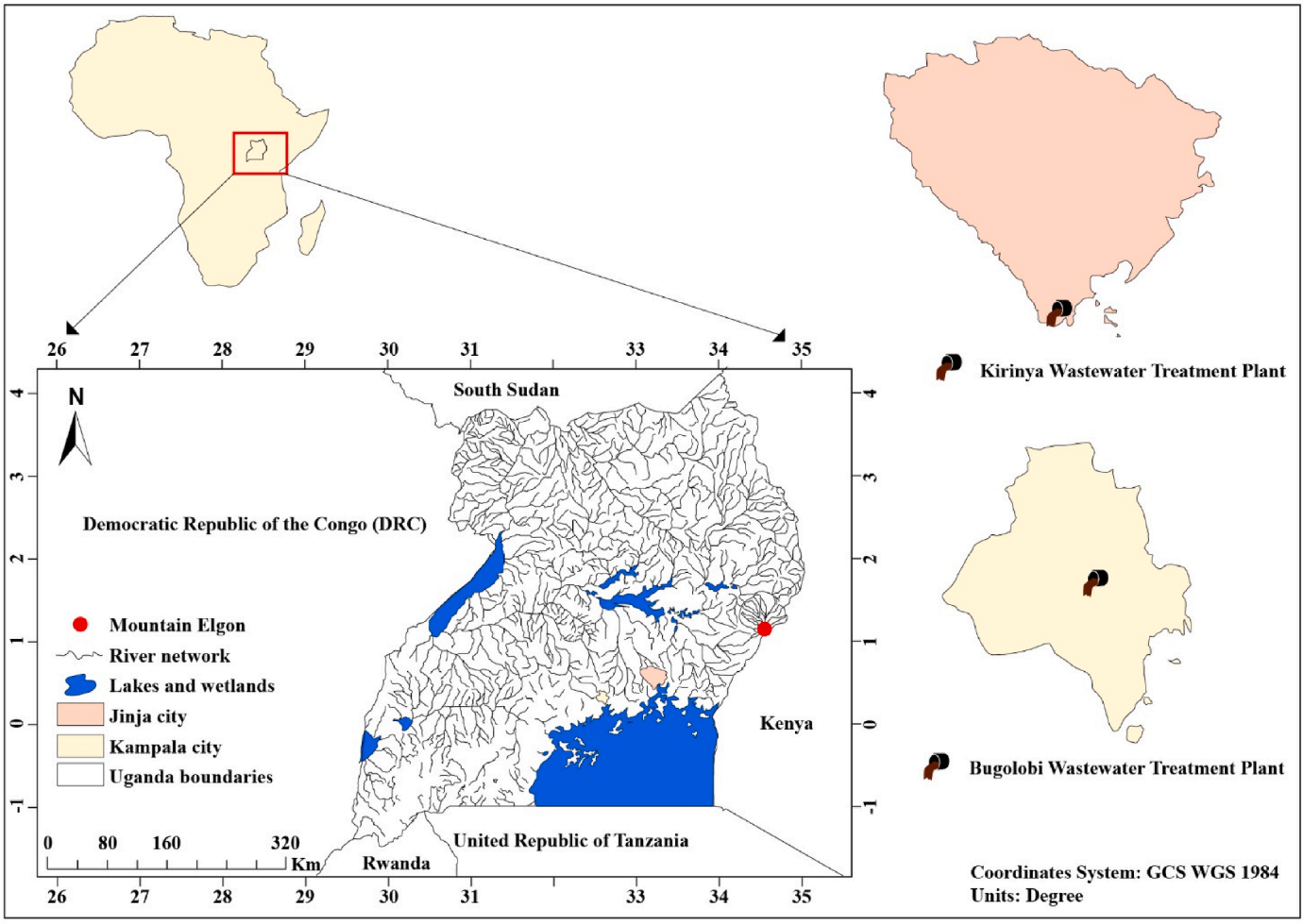

Fig. 1. A map showing the sampling locations.

operational variability, influent and effluent samples were matched and collected at 3 to 4-h intervals each day. Both WWTPs operate continuously thus interval-led sampling was designed to span each diurnal operating cycle under continuous flow.

At Kirinya WWTP, sampling covered facultative ponds (Fac), maturation ponds (Mat 1 and Mat 2), and the final effluent release point (Dis). Given its pond-based configuration, slower flow rates (1.3–12 cm/day) and longer HRT necessitated a five-day sampling period, with 24-h intervals between collections to track the progression of PFASs through the treatment system. Although the plant is designed at 4400 m^3/day, it currently serves a population exceeding 300,000 [27], necessitating this extended sampling to ensure representative data. As a background control, surface water samples (n = 3, collected in triplicate) were collected from a remote crater lake on Mt. Elgon, far from urban/industrial sources, to verify the absence of the local PFAS inputs and contextualize WWTP-derived concentrations (see results in Table S5). All samples were transported to the Pesticides Laboratory at the Department of Chemistry, Makerere University, and frozen at −20 °C to prevent microbial degradation prior to extraction and analysis.

2.3. Target analytes, reference standards, and reagents

This study analysed 15 PFASs, which included 4 perfluoroalkyl carboxylates (C_4, C_6, C_8, C_9 PFCAs: PFBA, PFHxA, PFOA, PFNA), 3 perfluoroalkyl sulfonates (C_4, C_6, C_8 PFSAs: PFBS, PFHxS, PFOS), 3 fluorotelomer alcohols (FTOHs: 6:2 FTOH, 8:2 FTOH, 10:2 FTOH), 2 perfluorooctanesulfonamides (FOSAs: N-EtFOSA, N-MeFOSA), 1 perfluoro-octane sulfonamide ethanol (FOSE: N-EtFOSE), 1 ammonium GenX salt, and the emerging replacement PFAS (HFPO-DA) (see Table S1b for full names of abbreviations and purity). These 15 PFASs were selected because of their known presence in environmental systems and significant ecotoxicological effects on aquatic environments, as highlighted in a study by Ankley et al. [29], as well as for their potential impact on environmental pollution and human exposure, especially within the wastewater systems in Uganda.

PFAS reference standards were purchased from AccuStandard (New Haven, CT, USA), with purities >99 %. Additionally, internal standards, $^{13}C_8$-PFOS and $^{13}C_8$-PFOA (≥99 % isotopic purity), were used to quantify sulfonated PFASs (PFSAs) and carboxylated PFASs (PFCAs), respectively, and were bought from Wellington Laboratories (Ontario, Canada). Methanol (≥99.5 %) and ammonium hydroxide (30 %) used were of analytical grade and purchased from Sigma-Aldrich. Stock and working standard solutions were prepared in methanol and stored at −20 °C (**Text S1**: Supporting information on standards preparation).

2.4. Extraction of wastewater samples for PFASs

Wastewater samples were extracted following a method described by Ahrens et al. [30].

Briefly, wastewater sample (500 mL) was filtered through glass microfiber filters (0.7 μm, Ø 47 mm, GE Healthcare, Life Science, Whatman™). The filtrate was then spiked with 100 μL of isotopically labelled standard (IS) mixture and loaded onto a solid phase extraction (SPE) cartridge that had been pre-conditioned with 4 mL of 0.1 % ammonium hydroxide in methanol, then followed by methanol (4 mL) and Millipore water (4 mL). After percolation, the cartridges were washed with 4 mL of 25 mM ammonium acetate buffer (pH = 4) in Millipore water and centrifuged at 3000 rpm for 2 min. PFASs were then eluted from the cartridges using methanol (4 mL), then followed by 0.1 % ammonium hydroxide in methanol (4 mL). The eluate was then concentrated to 1 mL under a gentle stream of pure nitrogen and transferred to amber vials for instrumental analysis.

2.5. LC-MS/MS analysis

PFASs were quantified using an Agilent 1290 Infinity LC system coupled to a Triple Quadrupole Mass Spectrometer (Agilent Technologies). Optimized LC-MS/MS parameters, including chromatographic and mass spectrometric conditions, are provided in **Text S3**, and the full SRM transition data for all target PFASs and isotopically labelled standards are summarized in Table S3.

2.6. Quality control and quality assurance

The limit of detection (LOD) and limit of quantitation (LOQ) for the PFASs analytes were estimated as analyte concentrations equal to three times the signal-noise-ratio and ten times the signal to noise ratio, respectively. The LOD values ranged from 0.01 ng/L (PFBA) to 0.49 ng/L (PFOS) while the LOQ values varied from 0.04 ng/L (PFBA) to 1.64 ng/L (PFOS). Recoveries were assessed at three spiking levels (0.05 ng/L - low; 10 ng/L - medium; and 25 ng/L - high) in triplicates. The recoveries ranged from 65 ± 0.63 % (PFHxA) to 98 ± 2.6 % (PFBS) at 0.05 ng/L; 67 ± 2.0 % (N-EtFOSA) to 100 ± 3.0 % (PFHxS) at 10 ng/L; and from 71 ± 0.5 % (N-EtFOSA) to 100 ± 0.99 % (PFBS) at 25 ng/L). The LOD and LOQ values and the recovery efficiencies for all target PFASs are summarized in Table S4a.

In addition, the recoveries of isotope-labelled internal standards were also monitored and remained within the acceptable ranges of 85–95 % (M8PFOA), and 87–96 % (M8PFOS) across all samples, while blanks showed 90–98 % recoveries, confirming method stability. The procedural blanks, consisting of the Milli-Q water processed alongside the samples showed no detectable PFASs, confirming the absence of blank interference. As a result, no blank corrections were made to the generated PFASs data.

2.7. Mass loading of PFASs into the neighbouring aquatic environment

The daily discharge load from WWTPs was calculated using equation (1), assuming that effluent concentrations remain constant throughout the day [21]. During the sampling campaign, Bugolobi WWTP operated at an average of 22,500 m^3/day, which represents its current operational capacity due to limited sewer connectivity, while Kirinya WWTP operated at its full design capacity of 4400 m^3/day (Table S1a).

$$Dd = C_W \times C_{WWTP} \times 1000 \times 10^{-6}\ (mg/day) \quad (1)$$

Where: Dd = Daily discharge of PFASs (mg/day), C_w = Concentration of individual PFASs in the effluents (ng/L), C_{WWTP} = Capacity of WWTP (m^3/day).

2.8. Removal efficiency of PFASs in the sampled WWTPs

The removal efficiencies of the conventional processes at Bugolobi were assessed against the wastewater stabilization ponds at Kirinya. Additionally, the treatment efficiencies for the individual PFASs were evaluated. Furthermore, the treatment efficiencies at each sampling point per WWTP were calculated using Equation (2) (Kairigo et al., 2020).

$$\text{Percent Removal Efficiency} = \left(\frac{C_{Influent} - C_{effluent}}{C_{Influent}}\right) \times 100\% \quad (2)$$

Where $C_{influent}$ is the concentration of PFASs in raw sewage, and $C_{effluent}$ is the concentration of PFASs in the treated sewage.

2.9. Ecological risks of PFASs-contaminated effluents

The potential ecological risks posed to aquatic organisms via PFASs exposure was estimated using risk quotients (RQs), calculated as the ratio of the maximum measured environmental concentration (MEC) to the Predicted No Effect Concentration (PNEC) (Equation (3)). This study considered three trophic levels for chronic toxicity assessment: algae (*Chlorella vulgaris*), crustaceans (*Daphnia magna*) and fish (*Danio rerio*), and bluegill sunfish (*Lepomis macrochirus*) for acute toxicity.

$$RQ = \left(\frac{MEC}{PNEC}\right) \quad (3)$$

Risk assessment served as the criterion against which the impact of PFASs was evaluated [31]. A PNEC was considered a concentration below which an unacceptable effect was less likely to occur and was computed using equation (4). In cases where the long-term no observed effect concentration (NOEC) was not available, lowest short-term concentration causing 50 % death (LC_{50}), or the lowest observed effect concentration (LOEC), or NOEC value was applied. The appropriate assessment factor (AF) accounted for uncertainty in extrapolating laboratory toxicity test data to the natural environment.

$$PNEC = \left(\frac{(NOEC(or\ L(OE)C_{50}}{AF}\right) \quad (4)$$

PFASs with known NOEC/LD_{50}/EC_{50} values were assigned AF values based on the number of trophic levels represented. A lower AF was applied for long-term tests, reflecting minimal uncertainty in extrapolation [32]. An AF of 100 was used for PNEC calculations for species representing one trophic level, while AFs of 50 and 10 were used for species representing two and three trophic levels, respectively. Additionally, an AF of 1000 was applied for acute toxicity data [31]. For each PFASs, RQ was calculated based on the worst-case scenario, considering the maximum concentration detected across all sites and the lowest NOECs or E(L)C_{50}. RQ values > 1 indicated high ecotoxicological risk, while values between 0.1 and 1, and <0.1, represented possible medium and low risks, respectively [33].

2.10. Statistical data analysis

Both descriptive and inferential statistical analyses were conducted to evaluate PFAS concentrations, mass loadings (Dds), removal efficiencies (REs), and risk quotients (RQs). Descriptive statistics included the calculation of means, standard deviations, and concentration ranges. The Shapiro-Wilk test was used to

assess for normality in PFAS data. Non-normally distributed datasets were analysed using the Kruskal-Wallis test as a non-parametric alternative, allowing robust comparisons for groups that were not normally distributed. A one-way ANOVA was applied to examine the mean differences for normally distributed data, while MANOVA was used to assess the relationships between the multiple dependent variables (PFAS analytes).

All statistical analyses were performed at a 5 % significance level ($p < 0.05$, 95 % confidence level) using TIBCO Statistica™ 14.0.0 for Windows (Palo Alto, CA: TIBCO® Software Inc.). Graphical representations, including correlations, were generated by using GraphPad Prism (v8.0.2; GraphPad Software, San Diego, CA, USA). The data from the LC-MS/MS instrument were initially recorded in RAW format and processed using MZmine software, then exported to Microsoft Excel for cleaning and preliminary calculations.

3. Results and discussion

3.1. Levels of PFASs in wastewater from Bugolobi and Kirinya WWTPs

The study analysed 15 PFASs analytes, and only 10 compounds (PFBS, PFHxA, PFBA, PFHxS, PFOA, PFOS, PFNA, N-MeFOSA, N-EtFOSA, and HFPO-DA [detected via its characteristic CO_2-loss fragment, HFPO-DA-CO_2]) were identified in influent and effluent samples from Bugolobi and Kirinya WWTPs. Short-chain PFASs (PFBS and PFBA) were the dominant compounds (contributing 63.3 % and 24.0 % to $\sum_{10}$ PFASs, respectively). Their prevalence was likely due to their increased use in consumer products, their persistent nature in the environment and/or degradation of long-chain PFASs into shorter-chain counterparts [8,34].

The levels of the analysed PFASs at different treatment stages at Bugolobi and Kirinya WWTPs are presented in Table 1. At Bugolobi WWTP, there was a reduction in $\sum$PFASs concentrations between the initial influent (106.89 ng/L) and the primary effluent (18.61 ng/L). The reduction in concentration was likely due to their sorption onto the primary sludge [35]. However, an increase in concentration was observed in the final effluent (96.26 ng/L), suggesting potential desorption from biosolids or transformation of precursors during secondary treatment. A similar pattern was observed at Kirinya WWTP, where PFAS concentrations decreased from the facultative pond (17.30 ng/L) to maturation pond 1 (16.99 ng/L) and further to maturation pond 2 (12.71 ng/L), but then slightly increased from the discharge point (15.38 ng/L) which was 10 m from the maturation pond 2. These findings suggest that while biological and sedimentation processes contribute to PFASs removal, the persistence of PFASs in the final effluents remains evident. The detection of PFNA in the effluents (4.35 ng/L) despite its absence in the influents further supported potential precursor transformation or release from the accumulated biosolids, with N-MeFOSA and N-EtFOSA being possible precursors [36].

3.1.1. Levels of PFASs in influent and effluent from Bugolobi and Kirinya WWTPs

Significant differences in the PFASs levels were observed between the influent samples from Bugolobi and Kirinya WWTPs ($H < 0.0001$, $p < 0.05$; Kruskal-Wallis test). Bugolobi exhibited higher PFASs levels ($\sum_{10}$PFASs: mean: 60.85 ± 1.03 ng/L) compared to Kirinya ($\sum_{10}$PFASs: mean: 17.58 ± 3.54 ng/L), likely associated with the differences in wastewater sources, such as industrial discharges, population density, and urban runoff, as reported in the previous studies [8,37]. The low levels at Kirinya could be attributed to reduced wastewater influx from Jinja city which is less urbanized compared to Kampala metropolitan area [26]. This observation was further supported by evidence that no target PFAS compounds were detected in water samples from Mountain Elgon (a control site for this study; Table S5).

Furthermore, the levels of PFASs in effluent water samples were significantly higher than those in influents at both WWTPs ($p < 0.05$; Kruskal-Walis) (Fig. S1). The influent levels of PFASs at Bugolobi ranged from non-detectable (n.d) to 190.01 ng/L (mean: 60.85 ± 1.03 ng/L), while effluent levels varied from n.d to 372.4 ng/L (mean: 237.91 ± 7.06 ng/L). At Kirinya, the levels of PFASs in influent samples ranged from n.d to 29.37 ng/L (mean: 17.58 ± 3.54 ng/L), and effluents varied from n.d to 30.21 ng/L (mean: 7.79 ± 0.85 ng/L). The elevated levels of PFASs in the final effluents could be due to desorption from accumulated biosolids or sediments within the treatment system since adsorption –desorption equilibria influence PFAS partitioning during the treatment process [38,39]. Furthermore, processes such as biodegradation, hydrolysis, or photolysis can lead to the formation of new PFAS species during wastewater treatment [36].

3.1.2. Trends in PFAS distribution across treatment stages

3.1.2.1. By PFAS group. In this study, perfluoroalkyl carboxylic acids (PFCAs) exhibited the highest concentrations among the PFAS groups (trend: PFBA > PFHxA > HFPO-DA > PFOA > PFNA) followed by perfluoroalkyl sulfonates (PFSAs; PFBS > PFHxS > PFOS) and perfluorooctanesulfonamides (FOSAs; N-MeFOSA > N-EtFOSA). At Bugolobi, PFCA levels were up to 39.04 ± 0.65 ng/L (PFBA in influent), whereas the levels at Kirinya were up to 11.46 ± 1.97 ng/L (PFBA in the facultative effluent). For PFSAs, Bugolobi recorded

Table 1
Mean levels of PFASs (ng/L) at different treatment stages from Bugolobi and Kirinya WWTPs.

PFAS analytes	Bugolobi WWTP			Kirinya WWTP			
	IIF	PEF	FEF	Fac	Mat 1	Mat 2	Dis
PFBS	7.96 ± 0.12	25.10 ± 0.63	206.58 ± 6.97	0.96 ± 0.24	1.17 ± 0.46	0.80 ± 0.21	0.86 ± 0.38
PFHxA	3.33 ± 0.34	3.85 ± 0.38	9.99 ± 0.99	2.14 ± 0.19	0.42 ± 0.05	2.65 ± 0.39	4.75 ± 0.84
HFPO-DA-CO_2	2.53 ± 0.52	2.74 ± 0.87	4.51 ± 0.27	n.d	n.d	n.d	n.d
PFBA	39.04 ± 0.65	51.86 ± 1.68	9.99 ± 1.10	11.46 ± 1.97	11.67 ± 0.92	4.98 ± 0.59	8.17 ± 0.90
PFHxS	5.08 ± 0.47	4.51 ± 0.74	1.26 ± 0.29	n.d	n.d	n.d	n.d
PFOA	0.03 ± 0.01	0.19 ± 0.03	1.56 ± 0.14	0.33 ± 0.08	n.d	0.65 ± 0.06	0.79 ± 0.08
PFOS	0.41 ± 0.05	0.78 ± 0.07	1.24 ± 0.12	0.16 ± 0.04	0.06 ± 0.01	0.91 ± 0.09	0.81 ± 0.10
PFNA	n.d	n.d	4.35 ± 0.03	n.d	n.d	n.d	n.d
N-MeFOSA	1.30 ± 0.13	1.54 ± 0.55	1.33 ± 0.17	1.29 ± 0.11	1.37 ± 0.23	1.53 ± 0.50	n.d
N-EtFOSA	1.17 ± 0.05	1.37 ± 0.43	1.18 ± 0.07	1.24 ± 0.14	1.26 ± 0.11	1.30 ± 0.23	n.d
$\sum$PFASs	106.89 ± 1.92	18.61 ± 2.63	96.26 ± 4.68	17.30 ± 2.31	16.99 ± 2.23	12.71 ± 1.75	15.38 ± 1.30

Mean levels are arithmetic means ± standard deviation. IIF - Initial Influent, PEF - Primary Effluent, FEF - Final Effluent, Fac - Facultative Pond, Mat 1- Maturation Pond 1, Mat 2 - Maturation Pond 2, Dis - discharge point to the environment is 10 m from Mat 2, n.d. - non-detectable.

levels between 0.41 ± 0.05 ng/L (PFOS in influent) and 206.58 ± 6.97 ng/L (PFBS in effluent), while the levels for Kirinya were up to 0.96 ± 0.24 ng/L (PFBS in facultative effluent). The wider variability observed among PFSAs likely reflected the preferential use and substitution of PFOS with short-chain PFBS, following the phase-out of legacy long-chain PFSAs [8]. Among FOSAs, N-MeFOSA and N-EtFOSA levels at Bugolobi varied from 1.17 ± 0.05 ng/L (N-EtFOSA in influent) to 1.41 ± 0.27 ng/L (N-MeFOSA in effluent), whereas Kirinya recorded values ranging from 1.24 ± 0.14 ng/L (N-EtFOSA in facultative effluent) to 1.54 ± 0.55 ng/L (N-MeFOSA in final effluent). Across both sites, PFCA levels were up to 39.04 ± 0.65 ng/L, while PFSA levels were up to 206.58 ± 6.97 ng/L, demonstrating considerable variations among compound classes and wastewater treatment stages.

The dominance of PFCAs could be attributed to the physico-chemical properties governing their environmental fate and persistence. PFCAs, particularly short-chain homologues such as PFBA and PFHxA, exhibit higher water solubility and lower adsorption potential to organic matter, facilitating their transport through WWTPs with minimal retention in sludge [38]. Conversely, PFSAs (PFOS and PFHxS) exhibit greater sorption affinity due to their amphiphilic nature, leading to higher retention in biosolids and reduced mobility in aqueous phases [1]. Additionally, the elevated PFBS levels in effluents suggested potential precursor degradation pathways or inefficient removal in WWTPs.

3.1.2.2. By chain length. Short-chain PFASs were more abundant than long-chain analogues in both WWTPs, with PFBA and PFBS dominating the profiles. At Bugolobi, PFBA and PFBS reached up to 39.04 ± 0.65 ng/L and 206.58 ± 6.97 ng/L, respectively, while corresponding levels at Kirinya were markedly lower. Long-chain PFASs such as PFHxS, PFOS, and PFOA occurred at comparatively lower concentrations, often near or below detection limits, particularly at Kirinya. These patterns suggested differential source inputs and removal efficiencies, with Bugolobi's industrialized catchment contributing to higher overall PFAS burdens. The observed chain-length-dependent distribution underscores the need for site-specific management strategies targeting both short- and long-chain PFASs.

3.1.3. Key PFAS contributors to ecological risk

This study identified key PFAS derivatives and precursors, including HFPO-DA, N-MeFOSA, and N-EtFOSA. The detection of HFPO-DA, a derivative of hexafluoropropylene (HFP), an industrial fluoropolymer precursor, highlights potential environmental transformation processes of short-chain PFASs, challenging the assumption of their stability and emphasizing their persistence in aquatic ecosystems [40]. Furthermore, the presence of N-MeFOSA and N-EtFOSA, known precursors to PFSAs (e.g., PFOS), suggested ongoing degradation pathways contributing to environmental burden of persistent and toxic PFASs [41]. This was corroborated by the observed increase in PFOS levels post-biotreatment, concurrent with a decrease in FOSAs (Table 1), suggesting microbial or oxidative transformation of FOSAs into PFOS during wastewater treatment [38,42].

3.1.4. Comparison with global PFAS levels in WWTPs

The levels of PFASs in Bugolobi and Kirinya WWTPs were lower than those reported in the WWTPs from the developed regions (Table S6). For instance, Gallen et al. [43] detected PFHxA as predominant in Queensland, Australia, ranging from 73 to 25,000 ng/L, which is up to about 130 times higher than the highest levels for the dominant PFBS (372.4 ng/L) recorded in this study. Similarly, effluent PFAS levels reported in North America (14–520 ng/L) [44] exceeded the levels observed in Uganda. While Coggan et al. [45] reported PFOA, PFOS, PFBS, PFNA, and 6:2 FTS as predominant PFASs in Australian WWTPs, this study found PFBS and PFBA dominant in wastewater samples, with low PFOA and PFOS levels. This difference was likely due to the 2009 global restrictions on PFOA and PFOS [12], which have led to declining trends, especially in regions where production was historically lower, such as Africa [14]. Furthermore, long-chain PFASs degrade into shorter-chain alternatives, reducing their persistence [8]. A study by Moneta et al. [46] reported PFBS dominance in Italian WWTPs (13400–107000 ng/L), which is up to 288 times higher than the maximum effluent levels observed in Uganda. This trend reinforces the global shift toward shorter-chain PFASs due to their preferred use in industry [47].

In South Africa, Morethe et al. [28] recorded influent levels as high as 3327 ng/L and effluent levels reaching up to 7699 ng/L across different WWTPs which was an order of magnitude higher than the levels detected in this study. The difference could be attributed to South Africa's higher industrialization levels and the extensive use of the PFAS-containing products. In contrast, a study by Chirikona et al. [18] in Kenya reported effluent levels of PFASs as low as 0.9–9.8 ng/L, suggesting lower contamination sources and potential dilution effects in the wastewater streams. Similarly, in the United States, Pétré et al. [37] reported effluent PFAS levels in Burlington (26–742 ng/L) and Fayetteville (40–377 ng/L), which were about 2–16 times higher than those observed in this study. These variations highlighted the influence of urbanization on PFASs use patterns. PFAS contamination for WWTPs in Uganda found in this study was comparable to the levels reported in other African countries with similar socio-economic and regulatory frameworks.

3.2. Mass loading of PFASs into the environment

Daily discharges (Dds) of PFASs are presented in Table 2. The Dds exhibited variability between Bugolobi and Kirinya WWTPs ($H = 62.24$, $p < 0.0001$; Kruskal-Wallis test). PFBS showed high variability relative to the overall variance ($H = 70.15$, $p < 0.0001$) at Bugolobi.

The daily PFAS loads varied between the two WWTPs based on pollutant type, plant capacity and location. Bugolobi WWTP (capacity: 18000–23000 m^3/day), treating domestic sewage and urban runoff in densely populated Kampala exhibited higher PFAS mass loadings than Kirinya WWTP (4400 m^3/day), which processes only domestic waste. PFBS had the highest mean load at Bugolobi (4648.03 ± 2.5 mg/day), indicating substantial PFAS discharge into receiving waters, with potential contamination of surface and drinking water sources. While studies on PFAS mass loadings remain limited in Africa, some assessments have been conducted in South Africa [19]. However, more research is needed across the continent since the synergistic effects of multiple PFASs remain unknown. The total PFAS mass loads at Bugolobi (5353.56 mg/day) and Kirinya (93.62 mg/day) were lower than the calculated equivalents of the European Union's effluent limits (1000 ng/L for total PFASs and 300 ng/L for PFOA/PFOS combined), which correspond to 10924.96 mg/day and 395.01 mg/day, respectively [48]. Although these levels do not pose immediate ecological risks, the persistence and bio-accumulative nature of PFASs necessitates continuous monitoring.

The PFAS mass loadings for WWTPs in Uganda were lower than those reported in highly industrialized regions elsewhere (Table S7). In Japan, WWTPs with capacities between 16400 and 615530 m^3/day were reported to discharge 124.95 g/day of PFASs, while in South Africa, WWTPs in Gauteng Province, treating up to 209564 m^3/day, reported 35.8–224.4 g/day [19]. These differences emphasize the role of industrial activity and WWTPs in

Table 2
Daily discharge of PFASs to the environment through the effluents.

PFASs	Site	Minimum (mg/day)	Maximum (mg/day)	Mean ± S.D (mg/day)
PFBS	Kirinya	2.75	8.28	3.78 ± 0.02
	Bugolobi	162.29	8379.16	4648.03 ± 2.5
PFHxA	Kirinya	n.d	125.35	20.91 ± 0.37
	Bugolobi	n.d	636.68	224.82 ± 0.22
HFPO-DA-CO_2	Kirinya	8.82	28.77	13.16 ± 0.63
	Bugolobi	52.42	262.11	101.53 ± 0.61
PFBA	Kirinya	n.d	132.91	35.94 ± 0.39
	Bugolobi	n.d	869.65	224.84 ± 0.25
PFHxS	Kirinya	n.d	n.d	NA
	Bugolobi	n.d	144.88	28.91 ± 0.49
PFOA	Kirinya	n.d	10.98	3.46 ± 0.04
	Bugolobi	n.d	89.60	35.20 ± 0.28
PFOS	Kirinya	n.d	13.64	3.55 ± 0.04
	Bugolobi	n.d	95.33	27.90 ± 0.28
PFNA	Kirinya	n.d	n.d	NA
	Bugolobi	n.d	19.4650	2.32 ± 0.06
N-MeFOSA	Kirinya	5.06	12.6540	6.79 ± 0.02
	Bugolobi	26.23	47.7180	31.65 ± 0.60
N-EtFOSA	Kirinya	4.99	10.9060	6.02 ± 0.01
	Bugolobi	25.13	36.8890	28.36 ± 0.36
$\sum_{10}$PFASs	Kirinya	93.62 mg/day = 0.034 kg/yr		
	Bugolobi	5353.56 mg/day = 1.95 kg/yr		

NA - data not available, n.d - non-detectable.

determining PFAS mass emissions. The WWTPs in Uganda that process mainly domestic wastewater, likely receive lower PFAS inputs compared to industrialized regions where effluent contains industrial discharges. Despite lower total PFAS loads at Bugolobi (5.353 g/day) and Kirinya (0.093 g/day) than elsewhere in the world (South Africa, Japan, and USA), their persistence and bioaccumulation necessitate continued monitoring, especially in transboundary waters like Lake Victoria and the Nile River.

3.3. Removal efficiencies of PFASs in wastewater by Bugolobi and Kirinya WWTPs

The removal efficiencies (REs, %) of PFASs in wastewater from Bugolobi and Kirinya WWTPs are presented in Table 3. Average removal efficiencies, calculated across all valid stages while excluding non-detectable (n.d.; replaced with NA) values, followed the order: PFHxS (41.65 %) > PFBA (7.86 %) > N-EtFOSA (−2.0 %) > N-MeFOSA (−5.68 %) > HFPO-DA (−36.45 %) > PFHxA (−140.98 %) > PFBS (−187.22 %) > PFOS (−298.46 %) > PFOA (−425.3 %). Short-chain PFCAs exhibited generally low removal due to their high solubility and mobility, limiting adsorption and retention in biological and filtration processes. Conversely, long-chain PFASs displayed relatively higher removal owing to stronger solid-phase affinity, although PFOS and PFOA showed poor removal, likely reflecting desorption or precursor transformation.

Bugolobi demonstrated higher removal efficiencies (IIF → PEF: 82.59 % > IIF → FEF: 6.05 %) than Kirinya (Mat 1 → Mat 2: 25.19 % > Fac → Dis: 1.95 %), likely attributable to its more advanced treatment technology. At Bugolobi, PFBS and PFOS showed limited removal in primary sedimentation, possibly due to desorption or release from sludge [38]. Similarly, PFHxA and PFOA were poorly removed, consistent with their persistence and resistance to biodegradation. Later stages enhanced short-chain PFAS removal, with PFBA reaching 80.7 %, likely due to microbial degradation and reduced adsorption resistance [38]. PFBS levels increased in later stages, likely from degradation of longer-chain sulfonates such as N-MeFOSA and N-EtFOSA, consistent with known biotransformation pathways [34]. Breakdown of precursors like 6:2 FTS into short-chain PFCAs, including PFBS and PFBA, may also contribute to this trend [38].

Table 3
Percent (%) removal efficiency of PFASs at specific treatment stages.

Site PFASs	Bugolobi		Kirinya		
	IIF → PEF	PEF → FEF	Fac → Mat 1	Mat 1 → Mat 2	Mat 2 → Dis
PFBS	−215.3	−723.0	−21.9	31.6	−7.5
PFHxA	−15.6	−159.5	80.4	−530.9	−79.3
HFPO-DA-CO2	−8.30	−64.6	NA	NA	NA
PFBA	−32.8	80.7	−1.8	57.3	−64.1
PFHxS	11.2	72.1	NA	NA	NA
PFOA	−533.3	−721.1	NA	NA	−21.5
PFOS	−90.2	−58.9	62.5	−1416.7	11.0
PFNA	NA	NA	NA	NA	NA
N-MeFOSA	−18.4	13.6	−6.2	−11.7	NA
N-EtFOSA	−17.1	13.9	−1.6	−3.2	NA
∑PFASs[a]	82.6	−417.3	1.8	25.2	−21.0

[a] Represents % aggregate removal efficiency for total PFAS levels at each treatment stage, NA = Data not available to calculate because one or both stage concentrations were below the LOD, negative removal indicates increased concentrations. IIF → PEF: Raw wastewater (Initial Influents) to Primary Effluent (after primary sedimentation), PEF → FEF: Primary Effluent to Final Effluent (after trickling filters, biofilters, and secondary sedimentation), Fac → Mat 1: Facultative Pond to Maturation Pond 1 (biological treatment), Mat 1 → Mat 2: Maturation Pond 1 to Maturation Pond 2 (settling and polishing), Mat 2 → Dis: Maturation Pond 2 to Final Effluent (treated water discharge).

At Kirinya, PFHxA (80.4 %) and PFOS (62.5 %) showed notable removal between the facultative pond and Maturation 1, likely due to adsorption onto organic matter. In contrast, PFBS (−21.9 %) and PFBA (−1.8 %) increased, suggesting precursor degradation and limited adsorption. PFBA decreased by 57.3 % from Maturation 1 to 2, while PFOS and PFHxA showed negative removal, likely reflecting incomplete degradation or desorption from bound phases, consistent with observed transformation behavior [49].

Overall, sulfonated PFASs (∑PFSAs) exhibited higher removal (72.45 % at Bugolobi; 36.45 % at Kirinya) than the carboxylates (∑PFCAs; −127.38 % at Bugolobi; −20.50 % at Kirinya), reflecting stronger adsorption even at comparable chain lengths. This is consistent with the enhanced hydrophobic interactions and microbial degradation associated with the sulfonate group, whereas carboxylates are more prone to solvation, limiting adsorption [1,50].

The observed REs aligned with the global trends, highlighting persistent challenges in PFAS removal. Stabilization ponds in the U. S. have reported REs as low as −1467 % (Houtz et al., 2018), while conventional systems ranged from −200 % to 50 % (Chen et al., 2018). Poor removal of short-chain PFASs is generally linked to high solubility, whereas long-chain PFASs show removal inefficiencies due to desorption or precursor transformations. Similar patterns were observed in Spain (PFHxA: 157 % to −2067 %) (Lorenzo et al., 2019) and France (PFOS and PFOA: 300 %–~70 %) due to desorption from sludge [4]. In low-industrial regions such as Sweden (sand filters: < −20 %–59 %) and Denmark (lagoons: 20 % to −30 %), poor PFOS and PFHxS removal is attributed to hydrophobicity, consistent with the trends observed at Kirinya. Detailed comparisons are provided in Table S8.

3.4. Potential ecotoxicological risks to the receiving aquatic ecosystem

Three trophic levels were selected to assess the ecotoxicological risks associated with the PFASs-contaminated effluents discharged into surface waters. Kruskal-Wallis's test showed significant variation in risk quotients (RQs) across trophic levels for both Kirinya ($p = 0.003$) and Bugolobi ($p < 0.0001$), with more pronounced differences in Bugolobi. The calculated RQs presented in Table 4 based on worst-case scenario, considered the maximum detected concentrations and the lowest NOECs or $E(L)C_{50}$ endpoint values. A comprehensive toxicity profile and a summary of the ecotoxicological endpoints are shown in Table S9. PNEC values (Table S10) were calculated following methods detailed in Section 2.9.

At Bugolobi WWTP, RQ values for PFASs ranged from 0.43 to 186.2 for algae, 0.0017 to 0.72 for crustacean, and 0.0021 to 0.91 for fish, with PFBS consistently showing the highest RQs across these groups. Notably, PFOS, despite its legacy risk profile, exhibited relatively low risk in this study, with a maximum RQ of 0.0082, far below the high-risk threshold (RQ > 1). Similarly, PFOA did not present any notable acute toxic effects, reinforcing the notion that emerging PFASs, such as PFHxA and PFBS, may pose more immediate ecological threats than some legacy compounds under current contamination profiles.

At Kirinya WWTP, PFBA and PFHxA, which are both PFCAs, exhibited high RQ values across trophic levels, suggesting considerable ecological risk. The pond-based system may facilitate a potential PFCA biotransformation [51]. The authors reported oxygenase-expressing bacteria converting FOSA precursors into stable terminal PFCAs. At Kirinya, FOSA precursors were detected in influents suggesting possible biotransformation process. This transformation likely contributed to the observed ecological risks, as newly formed PFCAs might accumulate in aquatic organisms over time. Further studies may assess potential degradation pathways of PFASs by microbes at Kirinya pond-based technology.

Notably, correlation analysis showed a weak, non-significant negative relationship between chain length and RQs at Kirinya (Spearman $\rho = -0.424$, $p = 0.222$), while a strong, significant negative correlation was found at Bugolobi (Spearman $\rho = -0.890$, $p = 0.0006$ Fig. S2). These findings suggested that chain length did not strongly influence RQs at Kirinya while longer-chain PFASs exhibited lower RQs at Bugolobi. Furthermore, PFSA RQs were higher than PFCA in Bugolobi (PFSA mean RQ = 10266.68 ± 94.3, PFCA mean RQ = 6082.41 ± 55.2), whereas PFCA RQs dominated in Kirinya (PFSA mean RQ = 128.32 ± 7.3, PFCA mean RQ = 1046.99 ± 33.3). This suggests that RQs vary significantly based on site-specific factors and molecular structure, which can inform the ecological risk assessments and management strategies.

3.5. Novelty of this study

To the best of our knowledge, this study represents the first attempt to calculate the risk quotients (RQs) for PFASs in the aquatic ecosystems in Uganda, specifically evaluating the ecotoxicological risks across the trophic levels. Previously, Ekperusi et al. [52] conducted a related study assessing the potential risks of PFOS exposure to aquatic life in the Gulf of Guinea. They reported estimated daily intake (EDI) values of 0.26 and 0.0014 ng kg^{-1} day^{-1} for fish and shrimp, respectively, with hazard ratios below the safety threshold of 1.00 (0.40 for fish and 0.0023 for shrimp), indicating low immediate risk. Consistent with their findings, the

Table 4
Calculated risk quotients for Bugolobi and Kirinya WWTPs.

Site	Bugolobi				Kirinya			
Trophic levels	1T (Algae)	2T (Daphnia)	3T (Fish)	Acute toxicity	1T (Algae)	2T (Daphnia)	3T (Fish)	Acute toxicity
AFs	AF100	AF50	AF10	AF1000	AF100	AF50	AF10	AF1000
PFBS	186.20	0.72	0.91	114938.30	0.94	0.0037	0.0046	580.56
PFHxA	14.15	0.055	0.069	8734.57	14.25	0.055	0.070	8793.21
HFPO-DA-CO_2	5.83	0.023	0.029	3595.68	3.27	0.013	0.016	2018.21
PFBA	174.35	0.67	0.85	107623.50	15.11	0.058	0.074	9324.07
PFHxS	10.94	0.042	0.054	6753.09	0.00	0.00	0.00	0.00
PFOA	1.99	0.0077	0.0098	1229.01	1.25	0.0048	0.0061	770.06
PFOS	2.12	0.0082	0.0104	1307.72	1.55	0.0060	0.0076	956.79
PFNA	0.43	0.0017	0.0021	267.00	0.00	0.00	0.00	0.00
N-MeFOSA	1.06	0.0041	0.0052	654.63	1.44	0.0056	0.00704	887.65
N-EtFOSA	0.82	0.0032	0.0040	506.17	1.24	0.0048	0.0061	765.12

AFs – Assessment factors, 1T – 1st trophic level, 2T – 2nd trophic level, 3T – 3rd trophic level.

current study also identified PFOS as having one of the lowest risk quotients among the assessed PFASs, suggesting relatively low ecological threats from this legacy compound for aquatic ecosystems in Africa compared to other continents, possibly due to lower usage or contamination levels.

Furthermore, this study evaluated RQs rather than EDIs, encompassing a broader spectrum of PFASs across the different trophic levels. The results highlight those organisms at lower trophic levels, particularly primary producers such as algae, face significant immediate risks from short-chain PFASs like PFBS and PFBA. These findings underscore the importance of PFAS risk assessments in African aquatic ecosystems and provide foundational insights for regulatory and mitigation strategies tailored to the continent's unique environmental conditions.

3.6. Study limitations and future direction

This study utilized toxicity values, including LOECs, LD_{50}, EC_{50}, and NOECs, derived primarily from data in the ECOTOX database and Norman Network. These sources predominantly reflect ecosystems in Europe and other non-African regions. Given Africa's unique environmental conditions and potential variations in species sensitivities, these values may not fully represent the ecological risks specific to African aquatic ecosystems [53]. Consequently, the calculated RQs should be interpreted with caution. To improve accuracy and contextual relevance, future research should focus on determining PFAS-specific toxicity thresholds, such as LD_{50} and NOEC values, directly within African ecosystems such as Lake Victoria and River Nile. The priority endpoints may include those species of higher consumer importance in the region such as Nile tilapia, silver fish, Nile perch and lung fish. Such data would enable the establishment of region-specific PNECs and enhance the reliability of risk assessments for local environments.

Furthermore, this study investigated only wastewater influent and effluent samples. Future studies may consider analysing wastewater in parallel with the biosolids and sediments to provide direct evidence of PFAS adsorption and desorption processes, thereby enabling a more complete mass balance assessment of PFASs within WWTPs. Further investigations applying source apportionment methods to quantify the contributions of PFASs from various sources are also essential to draw more definitive conclusions.

4. Conclusions

This study provided the first comprehensive assessment of 15 PFASs in wastewater from Bugolobi and Kirinya WWTPs in Uganda. PFASs were detected in all influent and effluent samples, with levels reaching up to 372.4 ng/L and 30.21 ng/L at Bugolobi and Kirinya, respectively. Short-chain PFASs (PFBS, PFBA, and PFHxA) dominated across the samples, likely due to their high solubility and mobility. Novel compounds, including HFPO-DA, N-EtFOSA, and N-MeFOSA were detected in Uganda for the first time.

Mass loading estimates confirmed WWTPs as important sources of PFASs to receiving waters, with Bugolobi discharging 5353.56 mg/day and Kirinya 93.62 mg/day. Removal efficiencies varied by chain length, with long-chain PFASs like PFOS moderately removed (up to 87.1 %) at Bugolobi, whereas short-chain PFASs remained persistent or showed negative removal, likely from precursor biotransformation. Kirinya WWTP's stabilization ponds achieved better PFHxA removal (80.4 %) compared to Bugolobi's trickling filters.

Ecological risk assessments showed that short-chain PFASs (PFBS, PFBA, PFHxA) pose significant risks, particularly to algae, with 8 PFASs exhibiting RQs >1 at Bugolobi. Legacy PFASs like PFOA and PFOS showed minimal immediate risks. Our findings underscored the persistence, mobility, and ecotoxicological concerns of PFASs in treated effluents and highlighted the need for targeted management strategies to reduce PFAS discharge into the aquatic ecosystems.

CRediT authorship contribution statement

Ashirafu Miiro: Writing – original draft, Methodology, Investigation, Formal analysis, Conceptualization. **Edward Mubiru:** Writing – review & editing, Supervision, Conceptualization. **Oghenekaro Nelson Odume:** Writing – review & editing, Supervision, Conceptualization. **Silver Odongo:** Writing – review & editing, Methodology. **George William Nyakairu:** Writing – review & editing, Conceptualization. **Henry Matovu:** Writing – review & editing, Methodology. **Charles Drago Kato:** Writing – review & editing, Conceptualization. **Ivan Špánik:** Writing – review & editing, Conceptualization. **Mika Sillanpää:** Writing – review & editing, Conceptualization. **Douglas Sifuna:** Writing – review & editing. **Liudmyla Khvalbota:** Writing – review & editing. **Patrick Ssebugere:** Writing – review & editing, Supervision, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

This study was supported by the Makerere University Research and Innovation Fund (Grant No. RIF/080/22, MAKRIF/CH/01/21, MAKRIF/CH/02/21); Slovak Research and Development Agency (Grant No. APVV-21-0178); Alexander von Humboldt Foundation (Grant No. UGA-1185413-GF-E); Austrian Partnership Programme in Higher Education and Research (APPEAR) under project 249 - Environmental Chemistry for Sustainable Development (ECSDevelop); Erasmus+ International Credit Mobility programme (Grant No. 2022-1-SK01-KA171-HED-000077735); African Water Resources Mobility Network (Grant No. 2019–1973/004-001); and the European Strategy Forum on Research Infrastructures (ESFRI) for project EIRENE (Grant No. 101079789).

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.emcon.2025.100580.

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