Distribution and Variability of Per- and Polyfluoroalkyl Substances (PFAS) Across three Categories of Wastewater Treatment plants in Kaohsiung, the industrial hub of Taiwan

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Supporting information

A biplot presented in **Fig S1(a)** was developed to examine specific PFAS loadings for individual PFAS compounds to understand their contribution to the factor plane defined by the two principal components. For WWTP 1–4, the factor-variable correlations for PCA1 and PCA2 in wastewater treatment plants (WWTP) 1-4 indicate varying relationships among different per- and polyfluoroalkyl substances (PFAS). For PCA1, variables like PFHxA, PFBS, and 6:2 FTS have strong negative correlations, suggesting they contribute negatively to this principal component, while PFDA and PFNA show positive correlations. For PCA2, PFHpA, PFOA and PFNA are strongly negatively correlated, whereas PFOS and PFDS show positive correlations. Overall, the data reveal distinct patterns in how these substances load onto the two principal components, which could reflect differences in their environmental behavior or sources in wastewater systems.

As seen in **Fig S1(a)**, PFAS compounds cluster into distinct groups based on their contributions to PCA1 and PCA2. **Short-chain PFAS** compounds, such as **6:2 FTS**, **8:2 FTS**, **PFDS**, and **PFDoA**, tend to cluster on the left side of the plot. This suggests these compounds

behave similarly in the treatment process, possibly because of their shared chemical properties. In contrast, **long-chain PFAS** compounds, including **PFOS**, **PFOA**, **PFHxS**, and **PFHpA**, cluster on the right side of the plot, indicating a different pattern of behavior compared to their short-chain counterparts. This separation reflects differences in how these PFAS compounds respond to treatment processes and may be driven by their resistance to removal or persistence in the environment. Previously, it has been shown that L-C PFAS, fragment during the wastewater treatment process to form S-C PFAS, which are more resistant to the treatment processes.

The length and direction of the vectors **in Fig S1(a)** provide important information about the contribution of each PFAS compound to the principal components. Compounds with longer vectors, such as **PFHpA**, **PFHxS**, and **PFOA**, are more strongly correlated with PCA1 and contribute significantly to the variation explained by this component. These compounds are key drivers of the differences in PFAS concentrations across the samples and are likely important in differentiating the behavior of PFAS within the wastewater treatment process. Conversely, compounds like **PFDS** and **FOSA**, which have shorter vectors, contribute less to the overall variation, suggesting that they may be less influential in explaining the variability in PFAS profiles. Small angles between vectors, such as those for PFHxS and PFHpA, indicate a positive correlation, meaning these compounds tend to increase or decrease together across the samples. In contrast, angles close to 180 degrees suggest an inverse relationship between the concentration of the PFAS compounds, while right angles indicate that the compounds are uncorrelated, suggesting no direct relationship between their behavior in the dataset.

In WWTP5, the factor-variable correlations in this dataset reveal that most PFAS compounds have strong negative correlations with PCA1, particularly 6:2 FTS, PFOA, PFHpS, and PFDA, indicating they load heavily in a similar direction on the first principal component, as seen in **Fig. S1b**. In contrast, PCA2 shows positive correlations with PFPeA, PFBS, PFHxA, PFHpA, and PFTrA, suggesting these variables contribute differently in this component. Notably, PFTeA has

the highest positive correlation with PCA2, while 6:2 FTS remains nearly neutral in PCA2. These patterns highlight potential groupings or shared sources and behaviors among PFAS compounds in WWTP5.

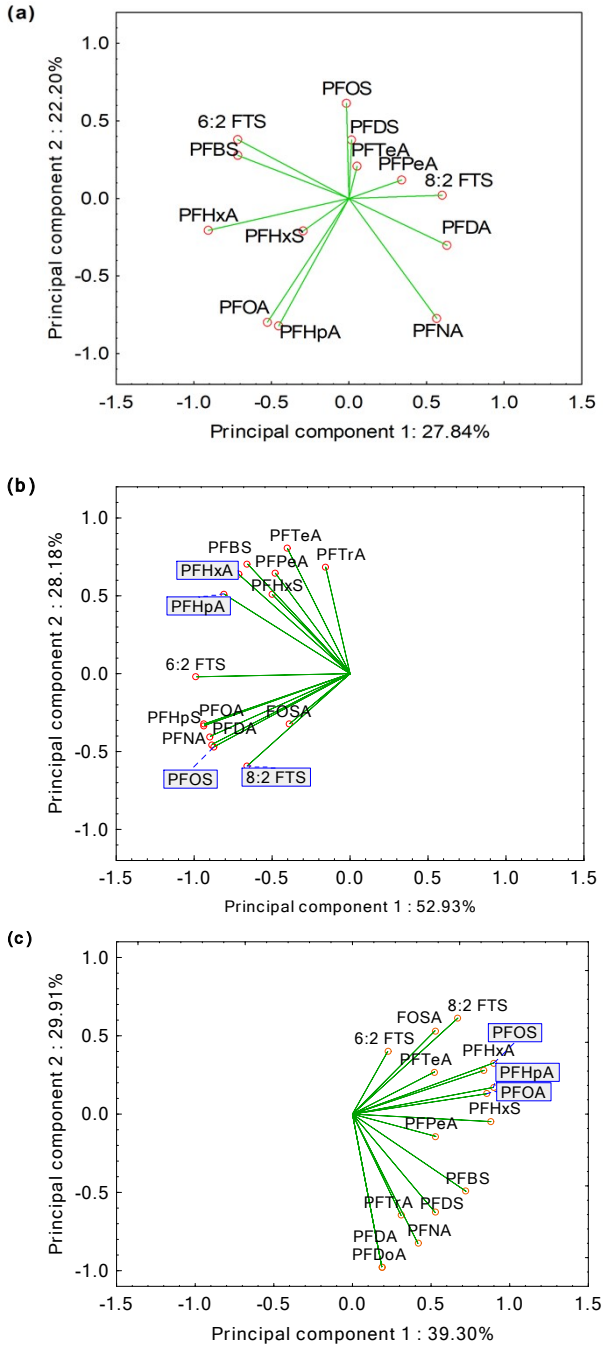

Fig S1. Biplots for the PFAS compound distributions in WWTP1-4, WWTP5, and (c) WWTP 5-6

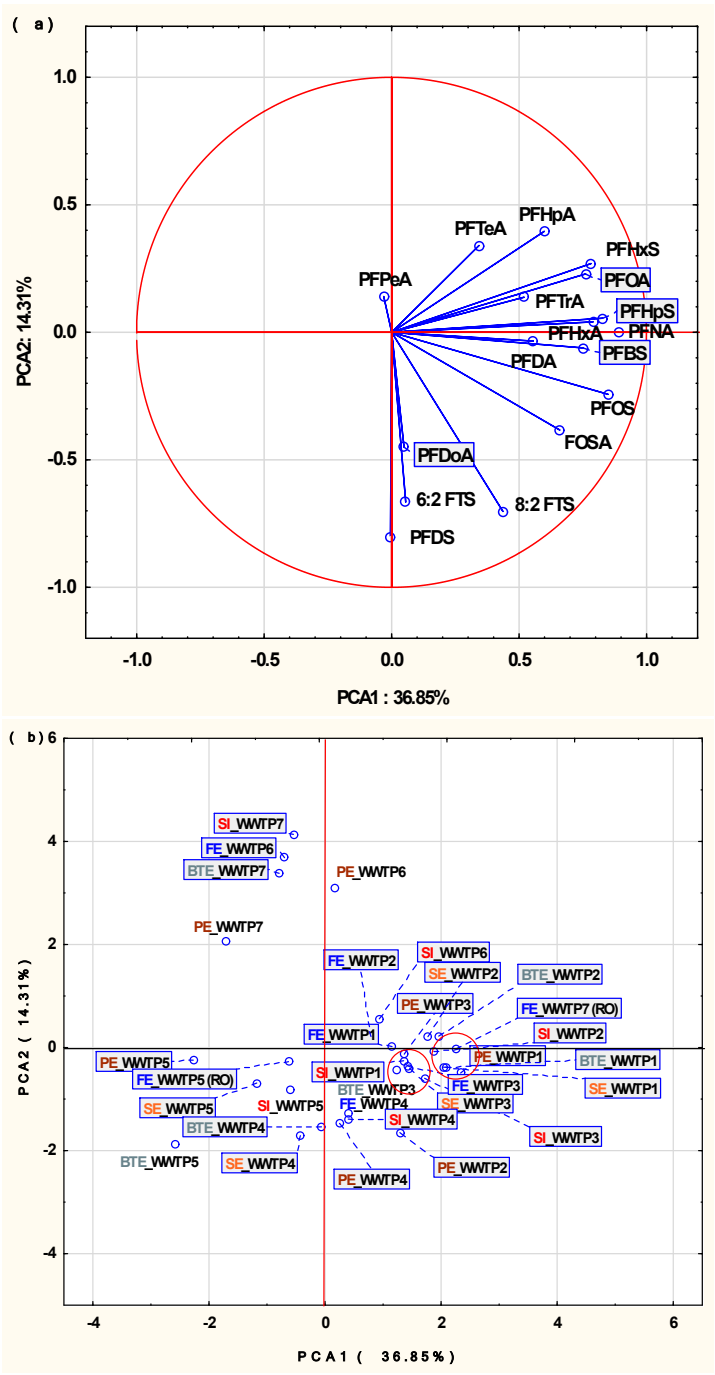

Fig S2. Showing the bipolar plots and the PCA analysis results for the PFAS compound concentrations in 7 wastewater treatment plants

The PCA biplot (**Fig. S2a**) illustrates the distribution and correlation of 17 PFAS compounds across the seven WWTPs, with PCA1 and PCA2 explaining 36.85% and 14.31% of the total variance, respectively. The loading vectors indicate that compounds such as PFHpS, PFHxS, PFOA, and PFNA are closely correlated, suggesting similar sources or behavior within the treatment processes. In contrast, PFDoA, PFDS, and 6:2 FTS are positioned separately, indicating distinct distribution patterns possibly due to different physicochemical properties or removal

efficiencies. The separation of long-chain PFAS (e.g., PFDoA, PFDS) from shorter-chain compounds (e.g., PFHxA, PFBS) suggests a difference in adsorption or degradation mechanisms within the WWTPs.

The PCA score plot (**Fig. S2b**) displays the spatial distribution of sampling points from the seven WWTPs, showing clustering patterns corresponding to different treatment stages (e.g., SI, PE, FE, BTE, and SE). Notably, WWTP6 and WWTP7 are positioned separately along PCA1, indicating unique PFAS profiles possibly due to variations in influent characteristics or treatment technologies, such as RO (reverse osmosis). The clustering of effluent (FE) samples near the origin suggests a reduction in PFAS concentrations across most WWTPs, whereas sludge and influent (SI, PE) samples are more dispersed, reflecting higher variability in PFAS inputs. These results provide valuable insights into the distribution patterns and removal efficiencies of PFAS compounds, highlighting the impact of treatment processes and influent characteristics on PFAS fate in wastewater treatment systems.

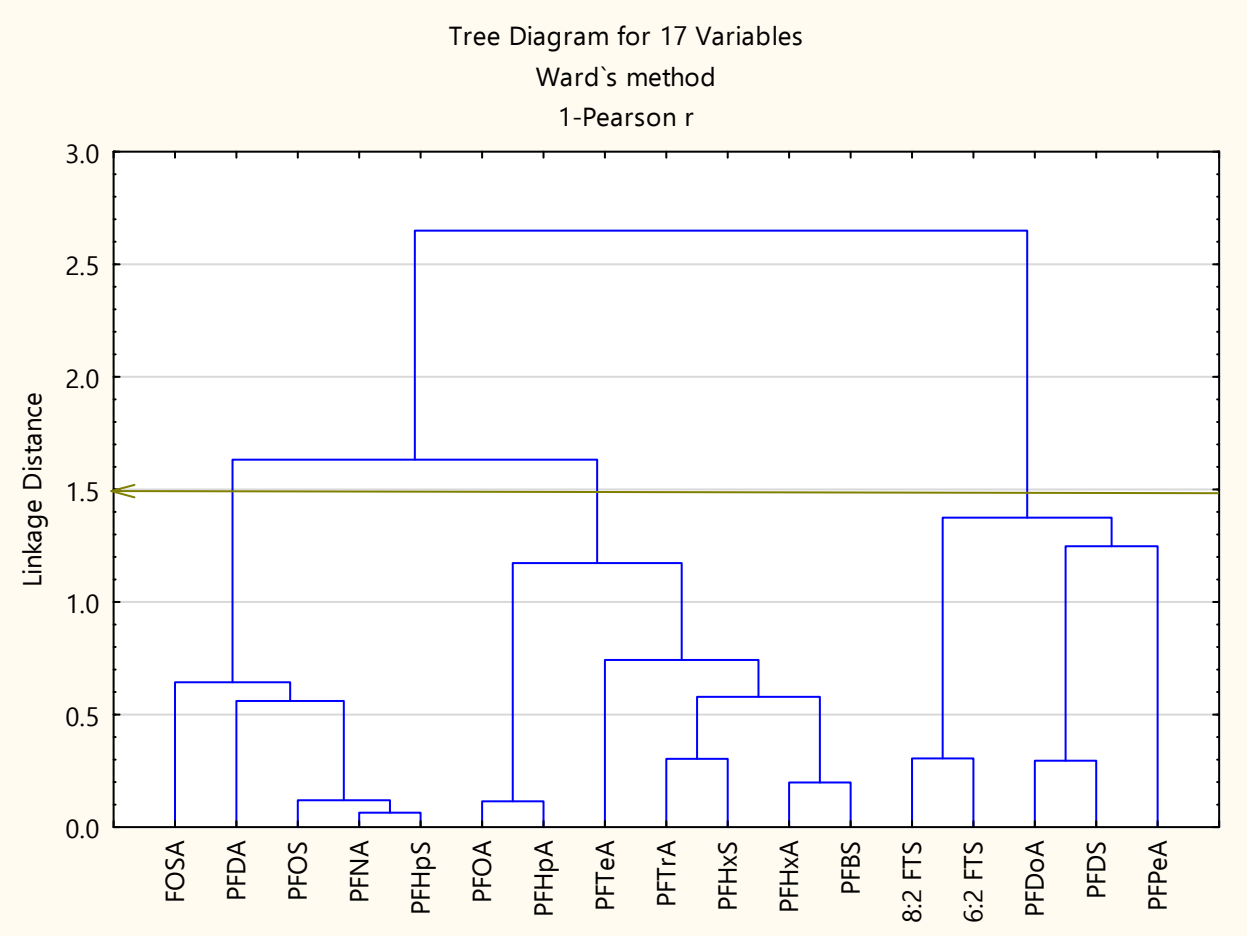

Fig. S3. Hierarchical cluster plot for standardized data with 1-Pearson r.

The Hierarchical Cluster Analysis (HCA) in **Fig. S3** uses Ward's method and Pearson correlation groups to classify 17 PFAS compounds into distinct clusters, revealing patterns that

reflect their behavior and fate across different stages of WWTPs. Overal, the compounds exhibit non-linear removal trends, calling for further investigation into their behavior under different hydraulic retention times and sludge compositions.

PFAS compounds in cluster 1 including FOSA, PFDA, PFOS, PFNA, PFHpS have shorter linkage distances compared to the other clusters. They are predominantly long-chain PFASs such as PFDA, PFNA and sulfonamide -FOSA compounds. These compounds are generally more hydrophobic, leading to stronger sorption to sludge and lower concentrations in the aqueos phase in WWTPs. The grouping of PFOS and FOSA suggests a common source or similar behavior, possibly due to their legacy usage in industrial applications and persistence in the environment. The inclusion of PFHpS in this cluster indicates similar partitioning behavior, likely influenced by their sulfonic acid groups, which possess strong resistance to degradation. Cluster 2 contains the following compounds; PFOA, PFHxA, PFHxS, PFHpA, and PFBS. This cluster groups shorter-chain carboxylic acids such as PFHxA, PFHpA and sulfonates such as PFHxS and PFBS, which are more water-soluble and relatively lower adsorption to sludge. Consequently, they exhibit lower removal patterns rates from the aqueous phase compared to LC-PFAS. Their clustering suggests common sources, possibly from consumer products or industrial emissions, and similar transport and partitioning mechanisms. Ultra-long chain carboxylic acids in this cluster including, PFTeA and PFTrA suggest distinct partitioning behavior. Cluster 3 contains the following PFAS compounds including: 6:2 FTS, 8:2 FTS, PFDoA, PFDS. This grouping highlights the precursor-product relationships between fluorotelomer sulfonates (FTSs) and long-chain perfluorinated compounds. The co-clustering of 6:2 FTS and 8:2 FTS suggests biotransformation pathways. The separation PFDoA and PFDS from other long and ultra-long chain PFAS in clusters 1 suggests distinct partitioning behavior, potentially due to higher hydrophobicity and affinity for sludge. The clustering of shorter-chain PFPeA with long chain PFASs, is unexpected and suggests unique interactions with WWTP matrices, possibly influenced by specific influent characteristics or

treatment conditions. This cluster emphasizes the influence of chain length and functional groups on removal efficiencies, particularly in advanced treatment processes.