Supplementary Information (SI) for Environmental Science: Water Research & Technology. This journal is © The Royal Society of Chemistry 2025

## The synergy of particles and biofilms behind drinking water discoloration processes in PVC pipes of a full-scale laboratory facility.

Artur Sass Braga, Yves Filion, Ben Anderson

## **Supplementary Data**

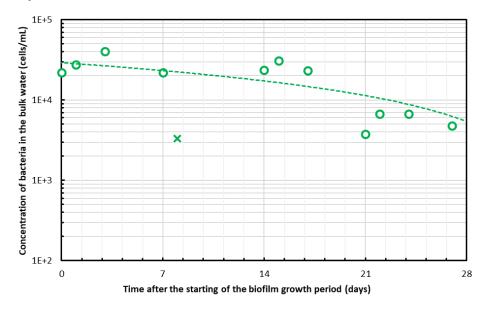


Figure S1 – Concentration of bacteria cells in the bulk water of the Biofilm experiment along the biofilm growth period measured through flow cytometry. Sharp decrease can be observed between the datapoints of the days 16 and 21.

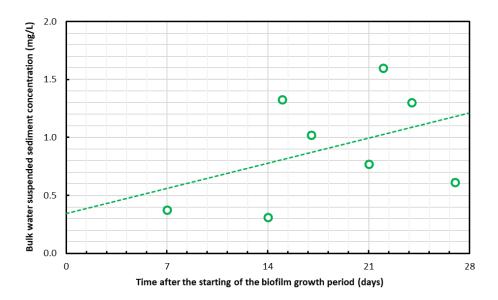


Figure S2 – Concentration of suspended sediments (SSC) in the bulk water of the Biofilm experiment along the biofilm growth period measured using the gravimetric method. Only volatile SSC were detected for this period.

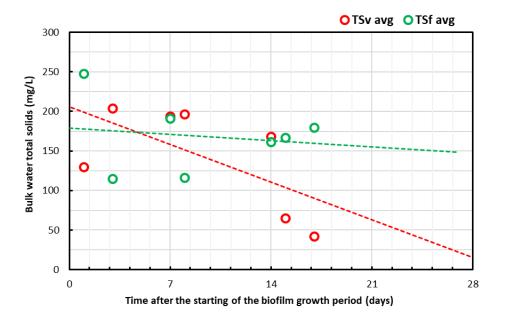


Figure S3 – Concentration of volatile (TSv) and fixed (TSf) total solids fractions in the bulk water of the Biofilm experiment along the biofilm growth period measured using the gravimetric method. Data indicates that TSv was depleted around the 3<sup>rd</sup> week of the biofilm preconditioning period.

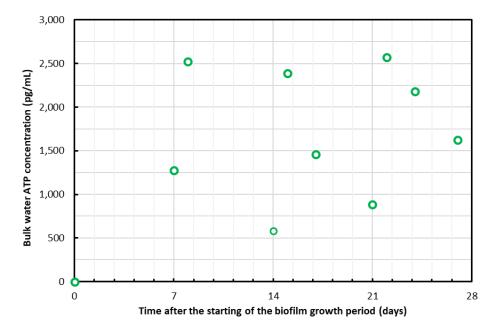


Figure S4 – Concentration of adenosine thiophosphate (ATP) in the bulk water of the Biofilm experiment along the biofilm growth period measured using a LuminaUltra luminometer. ATP was observed to quickly increase after the weekly addition of nutrients and progressively decrease along each week, suggesting a fast assimilation of nutrients by microorganisms.

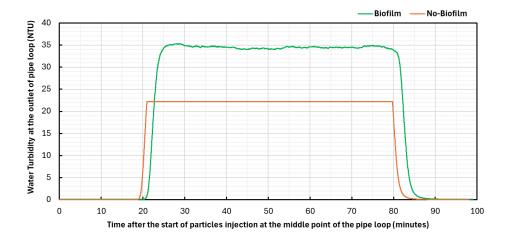


Figure S5 – Water turbidity at the outlet location of the pipe loop for the Biofilm and No-Biofilm experiments during the particle injection phase.



Figure S6 – Large loose iron oxide particles observed on a) pipe wall samples collected from the invert position of the pipes; b) filtered SSC samples collected from flushing phase of the experiments; and c) bottom of the bottles used to collect bulk water samples during the flushing phase of the experiment.

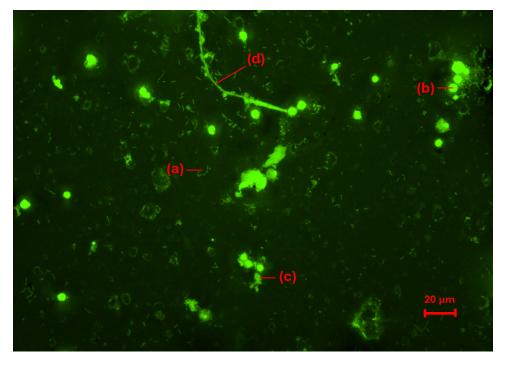


Figure S7 – Fluorescence microscopy image taken at magnification of 400X from pipe wall samples collected from the Biofilm experiment and stained with a DNA label dye – SYTO9, highlighting different components of early stage of biofilm colonization on the PVC pipe surface: a) bacilli bacteria; b) large cocci bacteria; c) small cluster of small cocci bacteria; and d) filamentous structure connecting multiple bacteria cells.