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A Novel Automated Water Pretreatment System for Microbial Preconcentration and DNA Extraction Using Monolithic Adsorption Filtration (MAF)

Moyseos M[1*], Demosthenous P[1], Seidel M[2]

[1]CyRIC – Cyprus Research and Innovation Center Ltd, Cyprus
[2]Technical University of Munich (TUM), Germany

*corresponding author:
e-mail: Maria.moyseos@cyric.eu

Abstract. Nowadays, there is an urgent need for rapid and efficient microbial monitoring in water resources. This need has become especially critical due to new EU directives mandating zero pathogen presence in drinking and reuse water. This work introduces an innovative, automated pretreatment system that integrates high efficiency preconcentration with direct DNA extraction of pathogens, utilizing Monolithic Adsorption Filtration (MAF) technology. The system demonstrates strong potential for in-line deployment within water reuse and safety management plans. This paper outlines the core methodology and presents key performance metrics validating its potential applicability in real-world scenarios.

Keywords: *DNA extraction, Monolithic Adsorption Filtration (MAF), Pathogen Preconcentration, Water Monitoring*

1. Introduction

The spread of waterborne indicator microorganisms such as *Escherichia coli*, coliphages, and emerging pathogens like *Helicobacter pylori*, represents a significant public health challenge, particularly under climate change scenarios that exacerbate microbial contamination of water resources. While *H. pylori* is traditionally associated with gastrointestinal infection, increasing evidence supports its potential waterborne transmission, highlighting the need for versatile monitoring platforms capable of detecting a broad range of microbial threats. EU Directives 2020/2184 and 2020/741 mandate stringent microbial monitoring, including 0 cfu/100 mL *E. coli* and 6-log reduction (10^6) of total coliphages in water reuse. Traditional laboratory-based detection methods are inadequate for real-time or field use due to their time-consuming nature. The proposed system addresses this gap by delivering a rapid, field-deployable, automated solution for microbial pre-concentration and DNA extraction (Nwabor et al., 2016; Vesga et al., 2018).

2. System Overview

The suggested system integrates two major processes in a single automated unit: (1) a two-step filtration/preconcentration of pathogens, and (2) MAF-based DNA extraction. Preconcentration relies on cross-flow ultrafiltration followed by adsorption on custom made DEAE - functionalized MAFs. Subsequent DNA adsorpion & extraction is performed directly on MAF - OH monoliths, enabling rapid transition from sample to analyte for downstream qPCR detection (Peskoller et al., 2009; Göpfert et al., 2020).

3. Results

MAF - Based Preconcentration and Validation

Validation of the system's preconcentration module was performed using *E. coli* spiked into 10 L water samples at concentrations of 10^5 to 10^7 cells. The system achieved recovery rates of 62.1% and 63.2% for 10^5 and 10^6 cells respectively, and 35.7% at 10^7 cells. The MAF module, operated in tangential flow mode, effectively minimized clogging while ensuring efficient bacterial capture. Cleaning procedures were validated to remove residual chlorine and microbial contamination, confirming the system's suitability for repeated operation (Demosthenous et al., 2025).

MAF – Based DNA Extraction and Validation

Key procedures were validated in a master's thesis at TUM. The study optimized lysis and DNA purification directly on MAF columns, demonstrating successful direct lysis of *E. coli* using 0.075 M GuSCN at 56 °C for 10 minutes, followed by DNA binding and elution on MAF-OH using ultrapure water (Peskoller et al., 2009). A concentration factor of 100x was achieved, with reproducibility comparable to commercial extraction kits. However, challenges with matrix inhibition (e.g., humic acids) in wastewater highlight areas for further optimization (Göpfert et al., 2020).

Table 1 summarizes the system's performance against key targets. Preliminary results show high efficiency in pathogen concentration and DNA extraction, with processing times suitable for real-time water monitoring.

Table 1. Summary of the system's performance

Metric	Target Value	Achieved Value (Preliminary)
Volume processed	10–100 L	up to 10 L
Preconcentration efficiency	80–90%	~63% for *E. coli*
DNA extraction efficiency	75–85%	~80%
Concentration factor	≥ 100×	100x
DNA extraction time	≤ 15 min	10 min
Total operation time	≤ 30 min	~30 min
Automation level	Fully automatic	Semiautomatic

4. Automation and Integration

The automation component, currently under development and optimisation, focuses on the seamless exchange and operation of single-use MAF discs, ensuring sample integrity and minimizing cross-contamination. The system is designed to handle volumes up to 100 L and deliver results within 30 minutes. Real-time operation without human intervention makes it ideal for in-line integration into water reuse systems and safety plans. Figure 1 is a schematic worklflow of the system, illustrating the sequential steps from raw water intake to DNA elution and detection via q-PCR, integrating preconcentration and extraction through MAF-based columns.

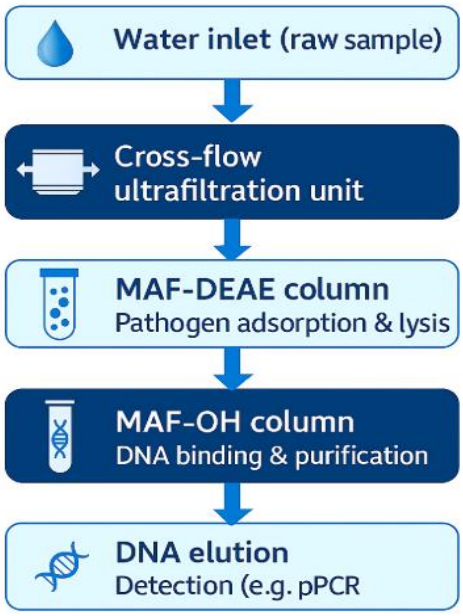

Figure 1. System Workflow.

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