Analysis of lipopolysaccharides by coupling microscale solid-phase extraction with capillary electrophoresis-laser induced fluorescence

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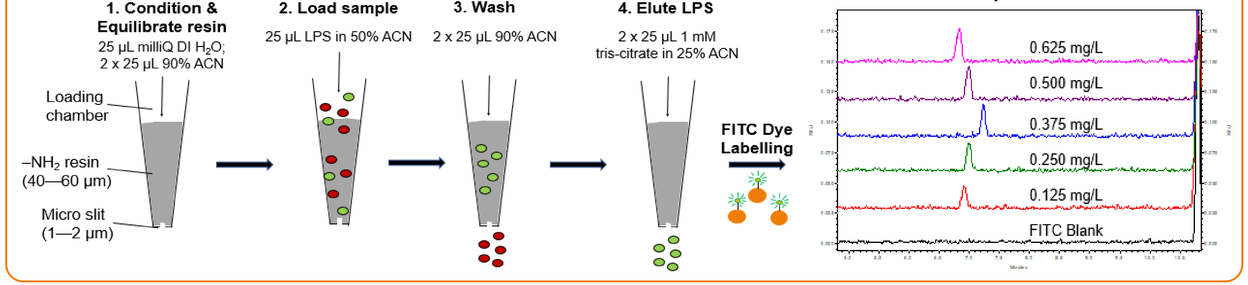
Introduction

- Lipopolysaccharides (LPS) are released when the cell wall of gram-negative bacteria breaks during bacterial growth, cell division and lysis
- LPS influences many cellular and humoral immune mediated systems
- Boiling water alone does not eliminate LPS
- Recognized by US FDA, Limulus Amebocyte Lysate detects intact LPS → False positives with (1-3)-β-D-glucans, batch variation, expensive and endanger horseshoe crabs

Project Objectives

- To develop and optimize a laboratory-made microscale solid-phase extraction (μSPE) device coupled with capillary electrophoresis with laser induced fluorescence (CE-LIF) for LPS purification, enrichment and quantification
- To investigate the effectiveness of the method by determining the limits of detection (LOD), preconcentration factor and recovery rate
- To apply the developed μSPE with CE-LIF in the analysis of real water samples

Experimental Procedure



Results

Klebsiella pneumoniae (KP); *Escherichia Coli O55:B5* (O55:B5)

μSPE-CE-LIF Sensitivity

KP: **LOD** = 1.32 ng/mL
O55:B5: **LOD** = 0.88 ng/mL

μSPE Preconcentration Factor

KP: (22.65 ± 0.02)
O55:B5: (26.20 ± 1.57)

μSPE Recovery Rate

KP: (97.6 ± 2.4)%
O55:B5: (87.5 ± 11.0)%

Analysis of Water Samples

Secondary Effluent: 4.8—7.3 ng/mL
Raw Sewage: 8.2—12.5 ng/mL

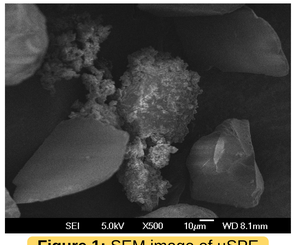


Figure 1: SEM image of μSPE –NH_2 sorbent loaded with LPS

Conclusion

- Monitoring of water quality from source to tap → μSPE with CE-LIF allows sensitive, selective LPS detection and is portable for on-site LPS extraction → making it accessible to remote and resource-limited areas during contaminations, disasters or pandemic to ascertain if the water is safe for consumption → presence of pathogens and inflammatory agents can be detected to contain spread of infections and diseases → UN SDGs 3 and 6

Reference

R. R. X. Lim, F. M. Fung, H.-T. Feng and S. F. Y. Li, Microchemical Journal, 2021, 161, 105771.

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