## Proteomics: Revolution in bacteria identification and AMR detection in one

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## Introduction

Treatment of bacterial infections is being complicated by the prevalence of AMR bacteria, while rapid detection of AMR bacteria and prescription of effective antibiotics continues to be a significant challenge. Identifying an infectious agent and simultaneously determining whether it expresses a high-risk AMR phenotype enables fast and targeted patient care. The proteome2pathogen (P2P) system, a proteomic analysis approach, revolutionizes testing by not only identifying bacteria but also determining if they are high-risk AMR strains. This allows a rapid optimization of therapy and timely control of AMR spread.

## Methods

The P2P-system was evaluated using 9 bacterial strains. 3 *Escherichia coli (E. coli* negative, *E. coli* TEM-52 ESBL+, *E. coli* CTX-M-15 ESBL+ and TEM-1 Beta lactamase + ), a *Acinetobacter baumannii* producing PER-1-like and ADC ESBLs and OXA-23-like and OXA-51-like carbapenemases. These isolates were cultured in TSB/BHI medium with 1 µg/ml cefuroxime. A methicillin resistant and a methicillin susceptible *Staphylococcus aureus* were cultured with oxacillin added to the growth medium (Muller-Hinton agar). Finally, a VanA, a VanB and a vancomycin susceptible *Enterococcus faecium* were analyzed that were cultured with vancomycin added to Muller-Hinton agar. Cultures were processed with a fast sample preparation protocol, which resulted in a trypsin digest from the cultured bacteria. The digests were subjected to liquid chromatography-tandem mass spectrometry, where the masses of the peptides and their fragments were measured. The obtained data were processed in PEAKS X, utilizing a developed database containing 498 bacterial entries for peptide identification. Retrieved peptides lists were subsequently analyzed in the in P2P application, which assess the identity of the bacteria and whether it is expressing a high-risk AMR phenotype. **Results** 

In this pilot study, the 9 isolates were correctly identified and relevant antimicrobial resistance proteins (ARPs) were detected. Moreover, bacteria were identified to species level and ARPs to the subfamily level based on multiple peptides.

## Conclusion

By using the P2P-system, bacteria are quickly identified and high-risk AMR phenotypes demonstrated. Enabling the detection of all kinds of high-risk AMR bacteria at once within 24 hours. This allows rapid effective antibiotics prescription serving the patient health and public health.