

REVOLUTIONARY DIAGNOSTICS – NONTRADITIONAL APPROACHES FOR DEVELOPING BREAKTHROUGH CAPABILITIES AGAINST EMERGING THREATS

Proteomics An Agnostic Approach To Detect Emerging Viruses

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The COVID-19 pandemic, but also MERS, SARS-1, monkeypox and other emerging biological threats have shown the pitfalls of diagnostics targeting only one or, at best, a limited number of pathogens. To overcome this, a next generation diagnostic platform that is causative-agent agnostic is developed. Moreover, the method is able to identify 1895 different virus species including strain variants, based on a single proteome analysis. To this end, samples are subjected to proteolytic cleavage using trypsin to produce smaller peptide sequences. These peptides are then subjected to liquid chromatography-tandem mass spectrometry (LC-MS/MS) on a Thermo Scientific Orbitrap instrument coupled to a nano-LC, in which the masses of the peptides and fragments thereof are measured. The obtained data is processed in PEAKS X using a combination of de novo sequencing and database searching. Next, the identified peptides are searched against a database with 1895 viruses, including strain variants, using a web application (<https://www.proteome2infection.com/>).

This diagnostics pipeline has been applied to analyze 35 samples (mainly supernatant of virus cultures) until now. Thereby, all 18 different viral species (Betacoronavirus OC43, Enterovirus A, Human coronavirus 229E, Human mastadenovirus F, Human metapneumovirus, Human orthopneumovirus (RSV A and RSV B), Human rubulavirus 4, Influenza A virus (H1N1 and H3N2), Influenza B virus, Mamastrovirus 1, Measles morbillivirus, Norwalk virus, Parechovirus A, Rhinovirus A, Rotavirus A, SARS-CoV-2, Vaccinia virus and West Nile virus) were correctly identified.

Our proteomics based approach identifies a virus based on several (≥ 3) species specific peptide sequences. This resulted in a 100% (35/35) specificity of the samples analyzed.

Advantages of our proteomics based approach are that in a single analysis 1895 virus species including strain variants can be identified within eight hours. The assay is executed without virus specific reagents. Finally, a few mutations in the virus aren't affecting the detection of the virus.

In conclusion, the presented proteomics based approach demonstrates that a variety of clinically relevant viral species can be identified through a single shotgun proteomic analysis, indicating that mass spectrometry is a promising agnostic tool to identify viruses. The implementation of this diagnostic platform will help ensure that we are not surprised by emerging biological threats in the future.

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