Direct Detection and Identification of Viruses in Saliva in Real-Time Using a SpecID™ Mass Spectrometer

Patented mass spec technology capable of detecting any virus in saliva with no sample prep in real-time

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Abstract

The COVID-19 (SARS-CoV-2) pandemic has led to a significant number of deaths globally. Accurate early diagnosis, surveillance, and prophylaxis are considered essential measures to reduce the spread of SARS-CoV-2. There remains a need for a reliable, fast, high-throughput screening method to identify individuals infected with SAR-CoV-2. Since respiratory viruses are typically present in nasal and oral secretions, saliva could be a good target for testing. Polymerase chain reaction (PCR) remains the most sensitive detection method of SARS-CoV-2 infection in biological samples at down to 500 virions per ml, and is widely used by hospitals. However, PCR has a false-negative rate of 15–20%. Although ELISA tests are now relatively common, they are less sensitive and associated with even higher rates of false negatives. Saliva testing has slowly gained popularity in the diagnostic market for testing, based on biomarkers and other constituents ranging from organic compounds (*e.g.*, food additives) and peptides to microorganism and viral pathogens, including SARS-CoV-2. In this study, the SpecID mass spectrometer was evaluated for the detection of viruses in saliva at very low levels, in seconds, and with no sample preparation. The main goal of the study was to investigate the ability of a portable, high-throughput, rapid, modified mass spectrometer to detect viruses in saliva. The platform is not limited to SARS-CoV-2, but can potentially detect any virus in saliva and other body fluids. While work is being done to expand the utility of the platform, the portability and speed could make it useful in the field for screening patients, such as in hospital triage areas, or public health laboratories.

Background

SpecID:

- ►**FDA NCTR patented mass spectrometry ionization method**
- ►**Enables real time pathogen detection**
- ►**Detects viruses directly in saliva and other matrices**
- ►**No sample preparation and no reagents required**
- ► **Sensitive – to detect asymptomatic carriers and beyond**
- ► **Portable – for use in the field, hospitals, clinics, field labs, etc.**
- ►**Other uses: pathogens (bacteria, fungi, etc.), mycotoxins, pesticides, clinical disease testing, and more**

Objective

Development of a portable real-time mass spectrometer technology to:

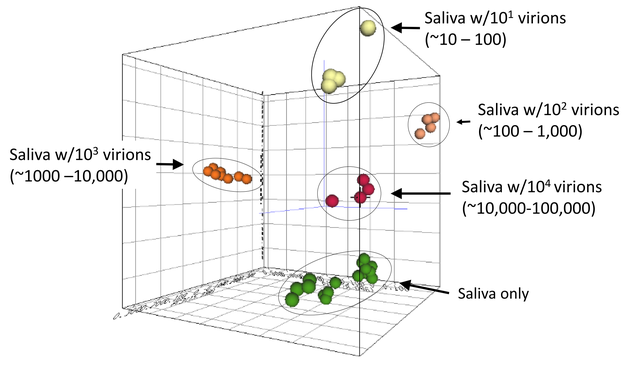
- ► **Detect pathogens in food safety, environmental, clinical, and other applications**
- ► **Detect viruses in saliva and other d matrices**
- ► **Determine the limit of detection (LOD) for viruses in saliva**
- ►**Assess the ability to distinguish viruses with close genetic homology**
- ►**Demonstrate the ability to detect and distinguish SARS-COV-2 variants**

Specific features

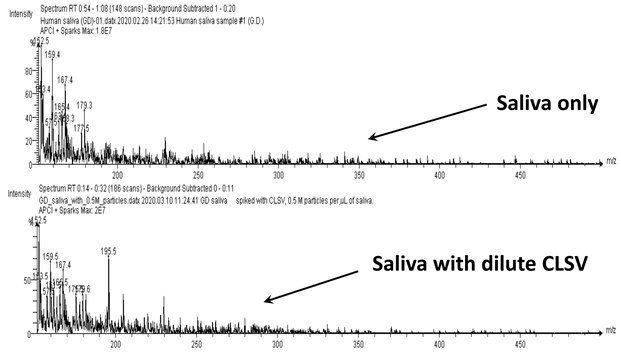
- ► **Mass spectrometry ionization source - gas plasma sparks ionize samples**
- ► **For use with atmospheric pressure ionization mass spectrometers**
- ► **Portable and rugged**
- ► **Sensitive – tested to an LOD of 10 ppb for chemical detection**
- ► **Fast, real-time detection (8 s per sample)**
- ► **Direct analysis in body fluids, cream medications, gels, foods, etc.**

Limit of detection (LOD)

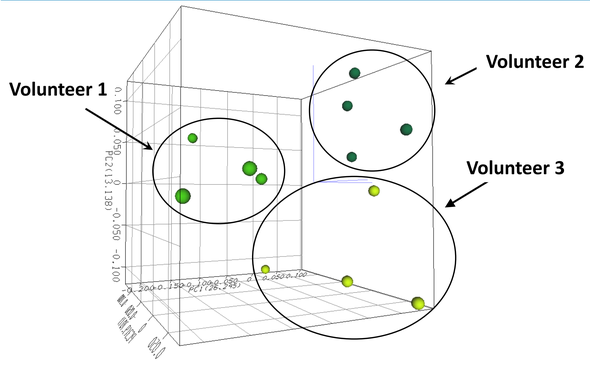
- ► **LOD in saliva as determined using cucumber leaf spot virus (CLSV)**



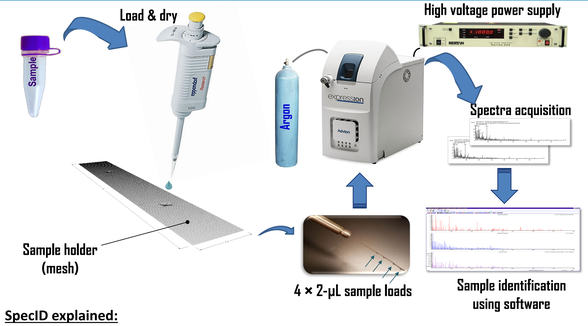
Spectral results



Volunteer saliva only samples



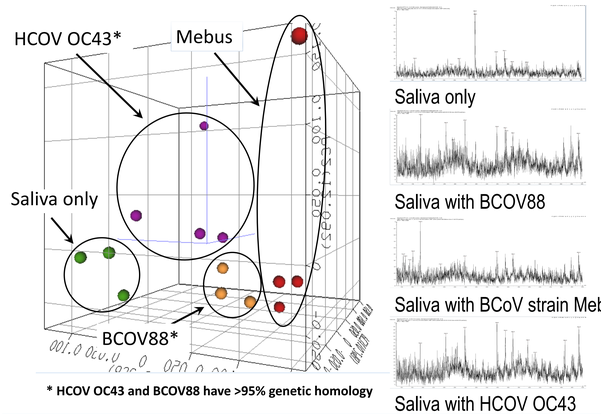
SpecID workflow schematic



SpecID explained:

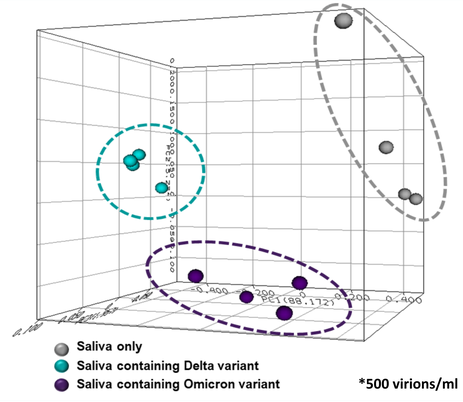
- Samples loaded (up to 4 per mesh) on a steel mesh strip
- Mesh placed in the instrument using a special sample holder
- Plasma sparks from needle ionize samples deposited on mesh
- Vaporized samples flow into the instrument for analysis

Volunteer saliva spiked with Coronaviruses



* HCOV OC43 and BCOV88 have >95% genetic homology

SARS-CoV-2 variants in saliva vs saliva only



Conclusions

1) **Research shows SpecID is able to detect and distinguish different coronaviruses, SARS-CoV-2 strains, and CLSV in saliva at low levels**
2) **The LOD was found to be 10-50 virions/2 µL sample**
3) **Reagent-less detection with spectral acquisition takes 8s**
4) **ANNs and other algorithms will be implemented for dynamic database development of different viruses**
5) **Potential to detect unknown viruses and add~~ing~~ to the database after genetic identification for novel outbreaks**
6) **With software automation, analysis for 4 samples is < 1 min**