

# Raman spectroscopy-based detection of RNA viruses in saliva: A preliminary report.

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Raman spectroscopy based assay have been developed for rapid and sensitive detection of several bacterial and viral pathogens like HIV, Dengue and other diseases, that can be carried out at the point-of-care (POC) with no associated recurrent costs for reagents in a a non-invasive manner.

Different biomolecules, such as DNA, RNA, proteins, lipids, from intact viruses are known to display unique Raman spectra vibrations based on their structural heterogeneity. We asked if these biomolecules can form the basis to detect the presence of viruses in human saliva. We analysed 201 spectra generated from 131 viral negative and 54 viral positive saliva samples, spiked with lentiviral RNA particles. Using Principal Component Analysis and Linear Discriminant Analysis (LDA), an unbiased statistical prediction model was generated based on 65 of 1200 spectral features, with prediction accuracy to 91.6% (92.5 % sensitivity and 88.8 % specificity) to detect viral positive samples. The surprise came in when we realized that majority of bands defining the range of 65 deterministic features are attributable to different RNA moieties, such as the nitrogenous uracil base, ribose-phosphate and A/G ring, suggesting RNA molecule as primal to detect the presence of RNA virus by Raman spectroscopy. Interestingly, the prediction was significantly reduced when the viral positive saliva samples were treated with RNase, but not with DNase or Proteinase —suggesting a novel concept of using RNA spectra obtained from intact virion to form the basis for the detection of RNA viruses in human sputum.

Furthermore, to minimize variability and automate the downstream analysis we developed a GUI based freely available analytical tool RNA Virus Detector, RVD that can be downloaded through a website.

In summary, we describe a string proof of concept to detect the RNA viruses in saliva with high sensitivity. This provides the essential framework for field application of Raman Spectroscopy based RVD in monitoring and responding to the COVID-19 pandemic. Positive tests could be validated by follow-up testing with molecular biology laboratory-based diagnosis to confirm the status of SARS-CoV-2, while epidemiological containment measures are implemented for the individual.

