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RAMAN SPECTROSCOPY CHARACTERIZATION OF SERUM-DERIVED EXTRACELLULAR VESICLES FROM TUBERCULOSIS PATIENTS

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Raman spectroscopy has been increasingly applied in disease diagnosis and appears to be a promising tool. Tuberculosis (TB) continues to be a chaotic infectious disease that needs urgent medical attention upon diagnosis. According to our knowledge, no studies to date have investigated the diagnostic use of Raman spectroscopic analysis of serum-derived extracellular vesicles (EVs) concerning tuberculosis. In this study, a total of 46 serum-derived EV samples (Active tuberculosis (ATB, n=14), latent tuberculosis (LTB, n=15), household contacts (CT, n=10), and healthy controls (HC, n=7)) were isolated from patients attending the chest clinic in Kandy. These samples were biochemically investigated using Raman spectroscopy with a laser wavelength of 785 nm. Raman spectra were collected from 100 to 3,200 cm-1 wavelengths for all patient samples. Spectra were normalized using the WiRE 3.4 software. The mean spectra, peak analysis, and principal component analysis (PCA) were performed using Origin Pro V10.5.88. The results show intensity variations in five main Raman peak positions from 756-1425, 1440-1740, 1750-2400, 4-2400-2800, and 2900-3100 cm^{-1}, and may play a major role in TB detection. In addition, higher Raman intensities in these regions are mainly assigned to lipids (Region 2, 3, and 4), proteins (Amide bands 1 and 2), and carbohydrates (Region 2, 3, 4, and 5). The highest Raman peak was observed around 2127-2135 cm^{-1}. The mean peak intensities (Arbitrary unit: a.u.) of the highest Raman peak showed a gradual decrease from ATB to HC (Mean Raman peak intensities for ATB: 3909.71, LTB: 3294.75, CT: 2733.27 and HC: 1963.14 a.u.). The PCA results from this study confirmed that the first three principle components (PC1, PC2, and PC3) contribute more than 99.5% to the variance of the four clinical groups. According to the score plot, all four clinical groups showed a clear differentiation among groups. Furthermore, each clinical cohort showed characteristic Raman peaks that need further analysis and validation to investigate the contribution towards biochemical pathways involved in TB pathogenesis. Overall, our preliminary results demonstrate that Raman spectroscopy could be used to differentiate between serum-derived EVs from ATB, LTB, CT, and healthy individuals.

Keywords: Extracellular vesicles, Raman spectroscopy, Serum, Tuberculosis.