

Supplementary Information

Figure S1 | Alternative display of Figure 2A, Principal component analysis loadings plots of the first 4 PC loadings showing 11 results in each plot with an increasing HAP amount of HAP in inclusions moving from the top to the bottom, i.e. blue 0 mg purple ~50 mg.



ure S2| A, Raman spectra of pure hydroxyapatite (HAP) (blue solid line) and Polytetrafluoroethylene (PTFE) (black dashed line). B, loadings data (PC1) when guided with incorrect reference spectra (PTFE) of 11 TRS Raman maps with HAP ranging from 0 mg to 50 mg buried in porcine tissue . C-E, PC1 Scores of 11 maps vs arbitrary spatial location when guided with no guiding spectra, PTFE and HAP respectively.

Fig

A comparison of HAP and PTFE (Figure S2 A) have been provided to show how there is no overlap between the respective spectra. Introducing PTFE into the matrix prior to PCA calculation

induces the loadings to show a feature corresponding to PTFE main peak, similar to as was shown in Figure 3B with HAP. However, unlike when HAP was used, the inclusion of PTFE does little to change the pattern seen in the scores when compared to conventional PCA i.e. no guiding spectra (Figures S2 C&D), while the scores for GPCA with HAP, shows a clear evolution of signal with increasing HAP, which is localised to the spatial location of where HAP was in the Raman map (Figure S2E).