SUPPLEMENTARY

Analytical Methods

Assessment of Paraffin Removal from FFPE Sections using FTIR-FPA Imaging



Supp. Fig. 1: Experiment setup outline of sections 1 and 2 that were floated onto CaF_2 substrates and section 3 was processed and stained with H&E. The square with 4 boxes represents the region of interest zone, encompassing a 2x2 infrared mosaic image, sampling ~1418 μ m of tissue.



Supp. Fig. 2:Optical images from imaging system with tissue sections1 and 2 in paraffin (a) and after 5 minutes of solvent incubation (b) with xylene (section1) and hexane (section2).



Supp. Fig. 3 : Typical spectrum of paraffin from a non-tissue region of a sample prior to solvent washing



Supp. Fig. 4: Examples of unprocessed single point spectra from Sample 2 prior to solvent incubation. The paraffin wax ranged in thickness across the sample, hence any incomplete removal of paraffin would show elevated methyl and methelyne absorbance peaks, resulting in random classicisation of pixel data during *K*-Means clustering. A+B are comparative examples of a 'paraffin only' and 'sample-plus-paraffin' spectrum on an extreme absorbance level respectively; C+D demonstrate examples on the typical absorbance scale.