Predicting Lymph Node Metastasis in Endometrial Cancer by multi-modal mass spectrometry imaging

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Overview
• Determine lymph node metastasis (LNM) in endometrial cancer (EC) using MS1.
• Complementary MALDI MSI MS platforms used to generate metabolomic + proteomic profiles, providing more signals to evaluate as markers for EC with LNM.

Introduction
• One reliable prognostic factor in EC is the presence of LNM.
• Clinicians currently face challenge that radiological imaging unreliable in determining if the EC has metastasized.
• Molecular markers could provide insight into the tumour biology and the process that leads to metastasis and may immediately serve as diagnostic markers to guide surgeons.

Methods
• Single sections (from one patient – 60B) or formalin-fixed paraffin embedded EC tissue microarrays (TMAs) containing 2 cores from 43 EC patients (16 classified as LNM being present, 27 classified as no LNM) were sectioned for analysis.
• For peptide MSI, antigen retrieval and matrix deposition was carried out using two different described methods (Ref. 1,2).
• For small molecule MSI, sample preparation and matrix was carried out as previously described (Ref. 3).
• Prepared TMA’s were analysed using two different MALDI MS platforms:
  o Bruker UltraflexXTreme MALDI-TOF MS
  o Bruker MALDI-TOF-Qq-TOF MS platform
• Peptid imaging measured in positive ion mode, small molecule imaging in negative ion mode. Data assessed using SCILS lab software.

Results
• It was observed in Fig. 1 that MALDI-tims-Qq-TOF Skyline Spectra exhibited more peaks, with higher intensity, compared to MALDI-TOF MS Skyline Spectra.

Figure 2: Distribution of m/z 599.32 in EC in single section (A), comparison of signal to simulated PI (B) and (C) ROC assessment to discriminate LNM presence (Green boxes) and absence in representative EC TMA using MALDI-tims-Qq-TOF MS analysis.

Figure 3: No distribution of Annexin A2 (m/z 1542.83), alpha actinin 4 peptide (m/z 1429.76 Da) and Annexin A1 peptide (m/z 1099.29 Da) in representative EC TMA using MALDI-tims-Qq-TOF MS analysis.

Conclusions
• MALDI-tims-Qq-TOF MS provided new potential small molecule markers for LNM.
• Peptides from previous MSI analysis (Ref. 4) were not observed using MALDI-tims-Qq-TOF MS, but were observed using MALDI-TOF MS. This may be due to different sample preparation methods measured on each system, but also indicates high complementary nature between systems.
• Future work aims to combine datasets using canonical correlation analysis (Ref. 2) and neural networks of signals from small molecule and peptide MSI.

References