MALDI Imaging Mass Spectrometry for the study of cardiovascular pathology

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Overview

MALDI-IMS, Cardiovascular disease, Endomyocardial biopsy, FFPE

Introduction

Endomyocardial biopsy (EMB) is performed on a regular basis following each transplant for the diagnosis of cardiac allograft rejection (CAR) based on the criteria established by International Society for Heart and Lung Transportation (ISHLT). Here we have applied a comprehensive Matrix-assisted laser desorption/ionization (MALDI) Mass spectrometry (IMS) approach to formalin-fixed paraffin-embedded (FFPE) tissue samples that have been acquired from the EMB conducted after the transplantations at Hôpital Européen Georges-Pompidou in Paris, France.

Methods

Tissue Preparation: FFPE tissue samples were transferred from Georges - Pombidou European Hospital, (GPEH) Paris, France. Tissue sections were prepared from FFPE tissue samples according to modified procedures as follows: 1) Dewaxing 2) On-tissue digestion using trypsin as a protease 3) Deposition of MALDI matrix with Image Prep and HTX TM-sprayer with or without manual sprayer for better ionization.

Immunohistochemistry: Anti TTR antibody and anti AL antibody (Santa Cruze Biotechnology,Inc.) were used for IHC. Amyloid staining were performed with Sirius red.

Mass Spectrometry: Matrix coating: DHB in 50% MeOH, ImagePrep and or manually sprayed with airbrush; Standard trypsin deposition protocol; MALDI: Rapiflex; flexImaging 5.0 software. (Bruker Daltonics). Tissues were stained by H&E. PCA and pLSA analysis of mass spectra was performed with SCiLS Lab 2016B software. (Bruker Daltonics).

Cardiac Amyloidosis represents a growing number of diverse and devastating diseases which comprise cerebral forms such as Alzheimer's disease and cerebral amyloid angiopathy (Kakuda et al., 2017). Peripheral disorders including senile systemic, dialysis-related, primary (AL) and secondary (AA) amyloidosis. These diseases are characterized by the aggregation of normally soluble, natural proteins or their fragments into highly ordered amyloid fibrils that deposit in target organs leading to dysfunction.

Results and Discussions

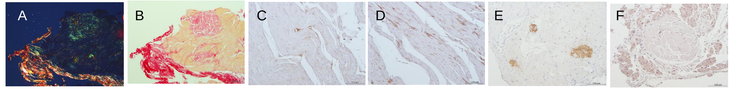
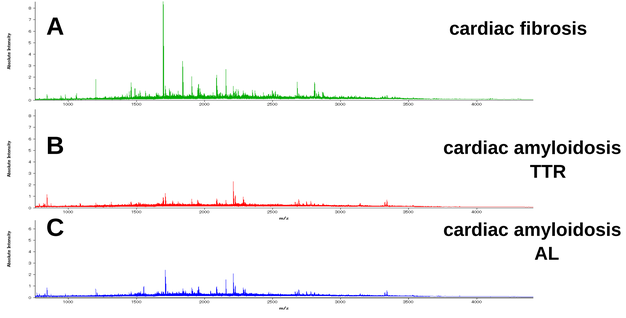


Figure.1 Histochemistry of cardiac amyloidosis. A,B: Sirius Red staining of cardiac amyloidosis. C,D: Cardiac fibrosis, E: TTR amyloidosis, F: AL amyloidosis. C,E: anti TTR antibody, D,F: anti AL antibody.



Mean spectra from each FFPE tissue samples with a novel protocols.

Figure.2 Mass spectrometry obtained from FFPE samples. Mean spectra from EMB samples from patients with cardiac amyloidosis and fibrosis was shown. A: Cardiac fibrosis, B: TTR amyloidosis, C: AL amyloidosis.

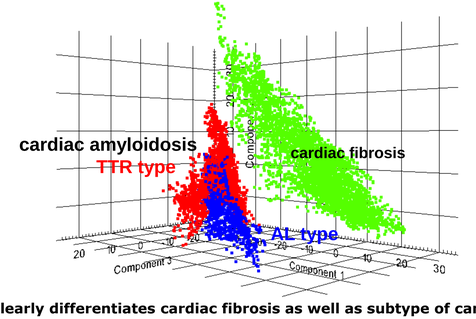


Figure.3 PCA clearly differentiates cardiac fibrosis as well as subtype of cardiac amyloidosis.

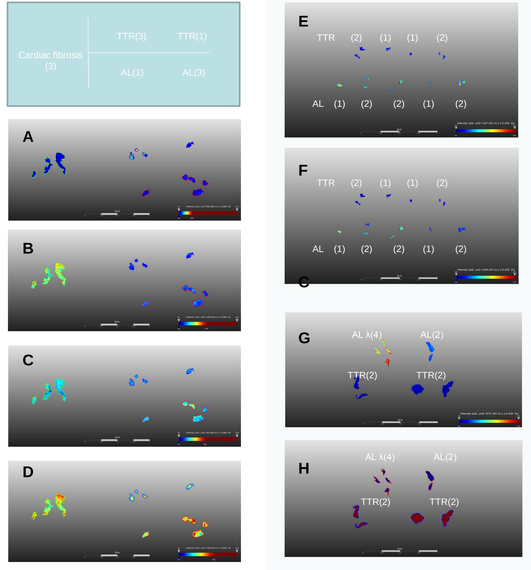


Figure.4 MS ion image of each peptide in biopsied samples. From A to D, A. TTR type. B. Fibrosis. C. AL type D. AL + Fibrosis dominant single peak distribution was shown. From E to H: matrix spraying with HTX TM-sprayer. G. m/z 1075.3 derived from YASWYQQK, from 48th to 56th positions of a tryptic digest of human immunoglobulin λ. Ref.3

Summary

- MALDI-IMS protocol for FFPE samples of human cardiac biopsy was established
- Typing cardiac amyloidosis from FFPE samples with MALDI-IMS
- Future studies using cardiac biopsy samples from cardiac allograft, atherosclerosis and cardiomyopathy is ongoing

Reference

1) Circulation. Lupy A. Et al., 2017
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Acknowledgements

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