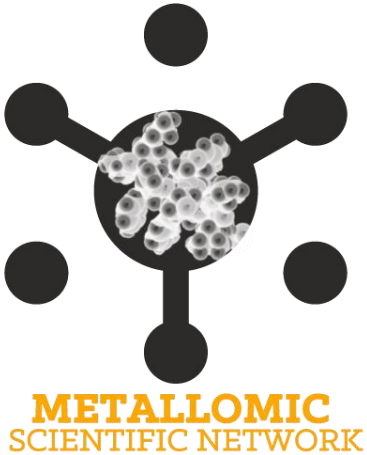

Metallomic Scientific Network No. 11440027

Název : **MALDI Imaging of MT in tissue slices of chicken embryos**

Jméno: Roman Guráň

Datum: 14. 8. 2015

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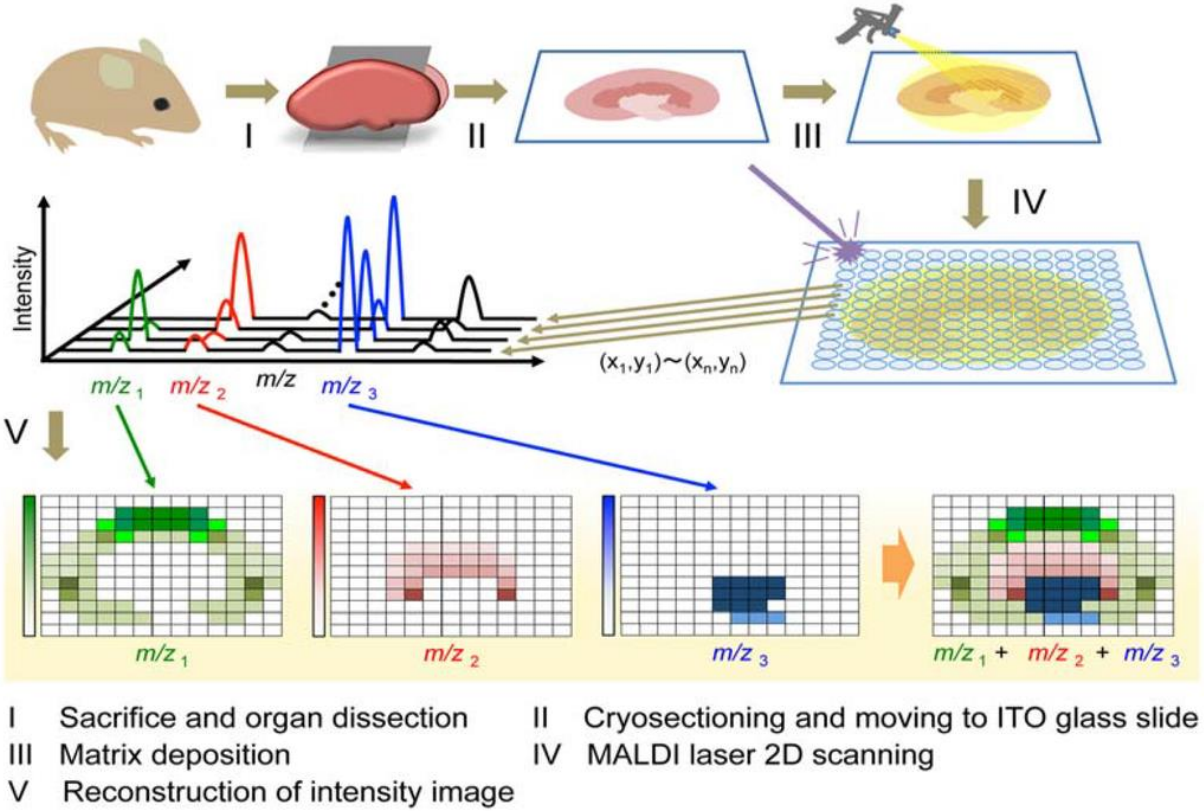
MALDI MSI



I Sacrifice and organ dissection
II Cryosectioning and moving to ITO glass slide
III Matrix deposition
IV MALDI laser 2D scanning
V Reconstruction of intensity image

Fujimura, Y.; Miura, D. MALDI Mass Spectrometry Imaging for Visualizing *In Situ* Metabolism of Endogenous Metabolites and Dietary Phytochemicals. *Metabolites* **2014**, 4, 319-346.

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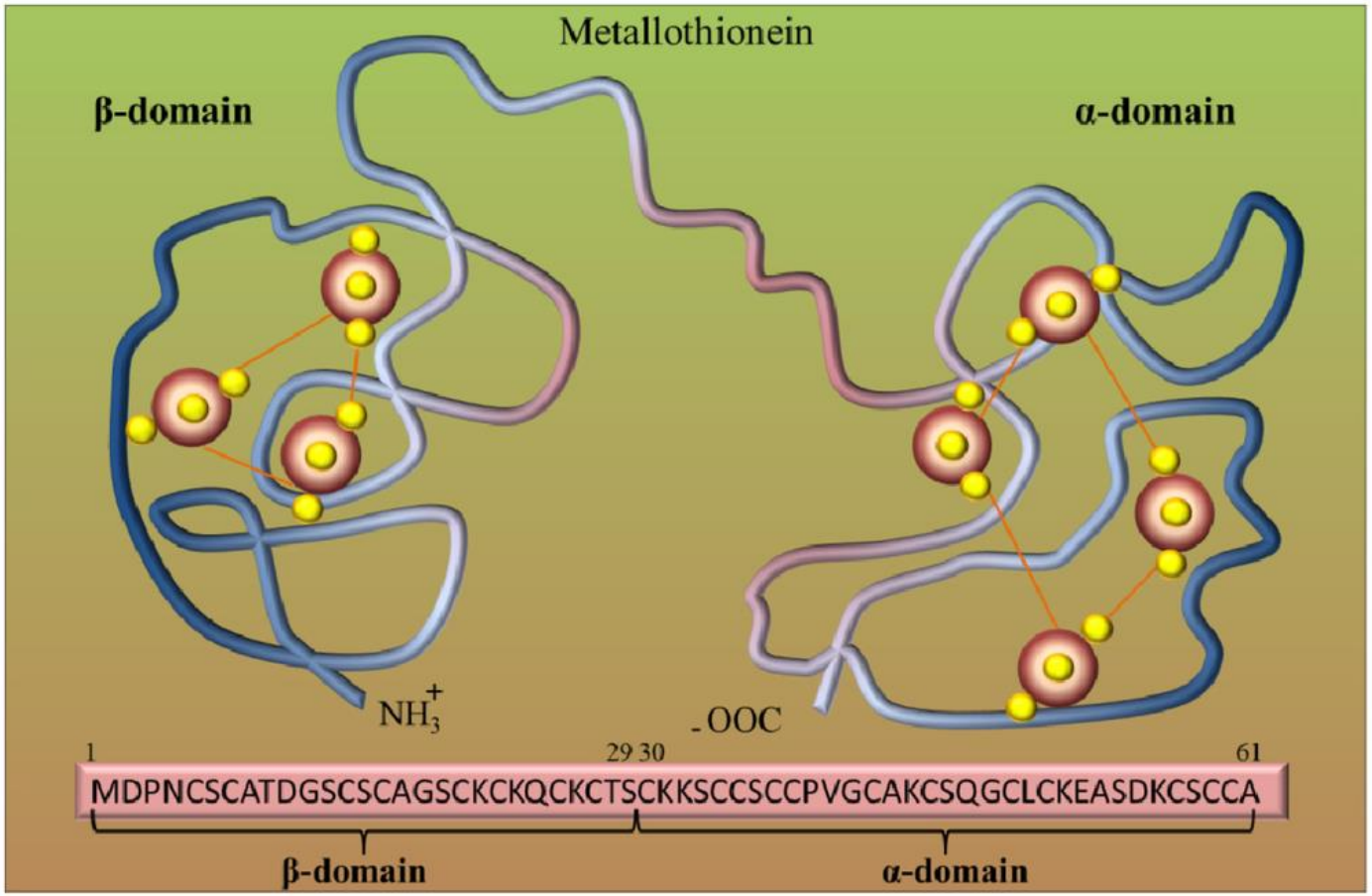
Metallothionein



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Our aims

- Optimization of deparaffination and antigen retrieval

- MALDI MSI of metallothionein in chicken embryo

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Deparaffination procedure

1) Xylene (3 min)
2) Xylene (3 min)
3) 100% ethanol (1 min)
4) 100% ethanol (1 min)
5) 95% ethanol (1 min)
6) 70% ethanol (1 min)
7) Washing the slide with Milli-Q water (3 min)
8) Washing the slide with Milli-Q water (3 min)

Casadonte, R. and R. M. Caprioli (2011). "Proteomic analysis of formalin-fixed paraffin-embedded tissue by MALDI imaging mass spectrometry." *Nat. Protocols* 6(11): 1695-1709.

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Antigen retrieval

1) Submerge ITO glass slides into 10 mM Tris buffer (pH 9) in Coplin jar.
2) Add 500 ml of dH_2O to the decloaking chamber pan and place there the Coplin jar.
3) Set the program of decloaking chamber: Set point 1 – temp. 95 °C, 20 min; Set point 2 – temp. 90 °C, cca 10 s.
4) Start the chamber and wait cca 5 min for reaching 95 °C. Then the above program will start. Pressure should remain below 5 psi (cca 34.5 kPa).
5) At the end of program, chamber will „beep“ and Coplin jar can be removed with hot-hand protector.
6) Let the jar to cool on the bench for 10 min. Then exchange five times Tris buffer with Milli-Q H_2O, pouring out half of the liquid each time.
7) Stand ITO slide vertically on a paper filter until it is completely dry.
8) Acquire optical image of the ITO glass slide and place it in a bench vacuum desicator (vacuum approx. 12 psi) until needed.

Casadonte, R. and R. M. Caprioli (2011). "Proteomic analysis of formalin-fixed paraffin-embedded tissue by MALDI imaging mass spectrometry." *Nat. Protocols* 6(11): 1695-1709.

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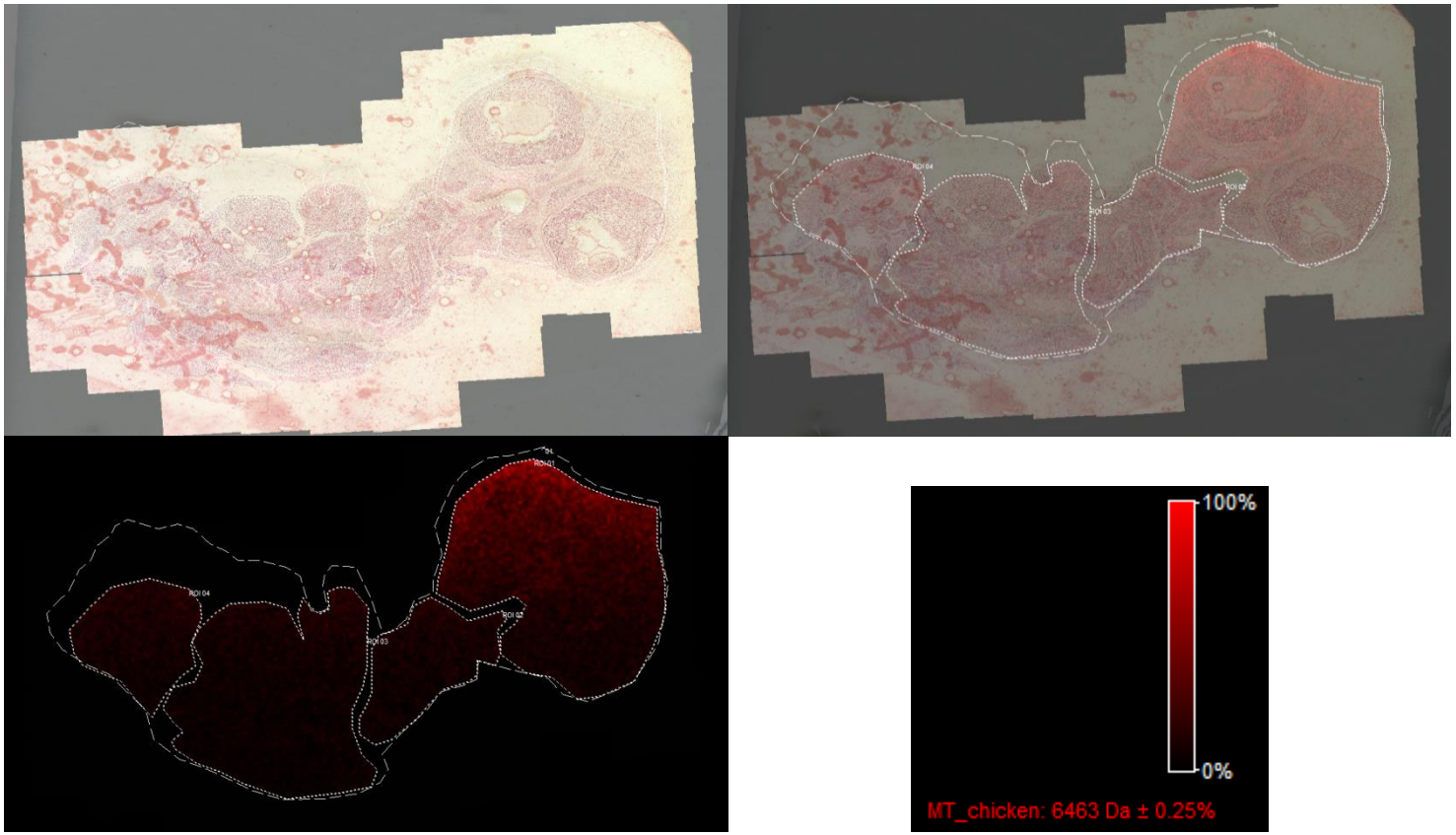
Results from chicken embryo



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Acknowledgments

The project is supported by International Visegrad Fund, www.visegradfund.org

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Thank you for attention!