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Hydrazide-based Reactive Matrices for the Sensitive Detection of Aldehydes and Ketones by MALDI MSI **



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INTRODUCTION

A one-step MALDI matrix for the detection enhancement of amine and phenolic compounds has previously been developed by our group (Shariatgorji et al., 2019), but there is currently no equally useful matrix available for carbonyl compounds. Three in-house developed reactive matrices showed an equal or higher detection efficiency than Girard's reagent P, enabling improved imaging of brain metabolites without the need of an additional co-matrix.

RESULTS

Ionization efficiency: In-house matrix candidates vs Girard's reagent P

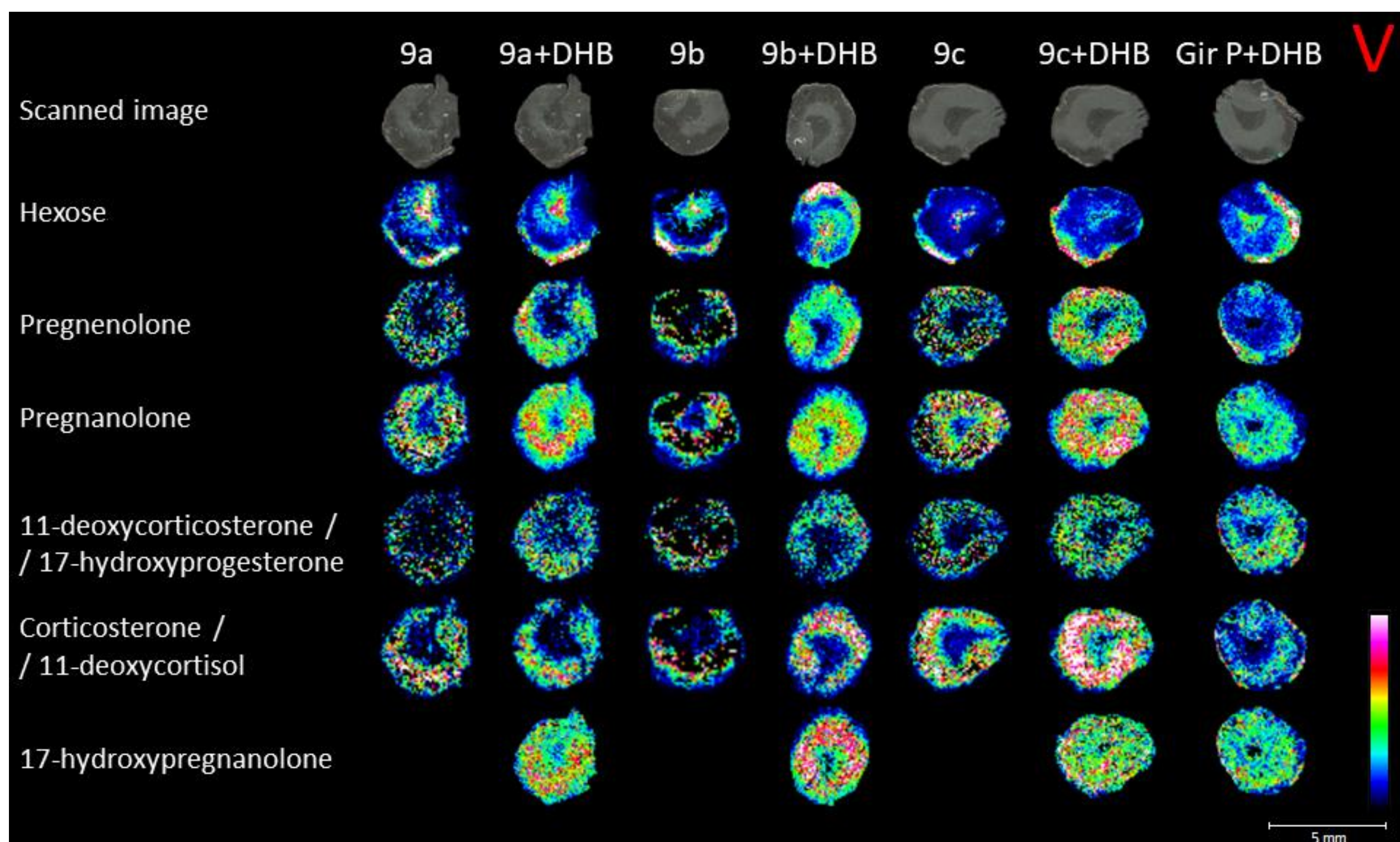


Figure 1. Imaging of endogenous compounds in rat adrenal cortex with reactive matrices (9a-c) vs. Girard's reagent P (Gir P) without and with DHB as co-matrix. Spatial resolution: 80 μ m. Intensity set individually to visualize the best possible distribution of the compounds.

- Improved or equal ionization efficiency was obtained with the in-house matrices (9a-c) compared to the more established Girard-P reagent (Girard and Sandulesco, 1936). For some compounds even without co-matrix (DHB).
- In-house matrix 9b was selected for further evaluation based on the results in Figure 1 and from standards spotted on control brain tissue (not shown).

Fatty aldehydes and monosaccharides in rat brain

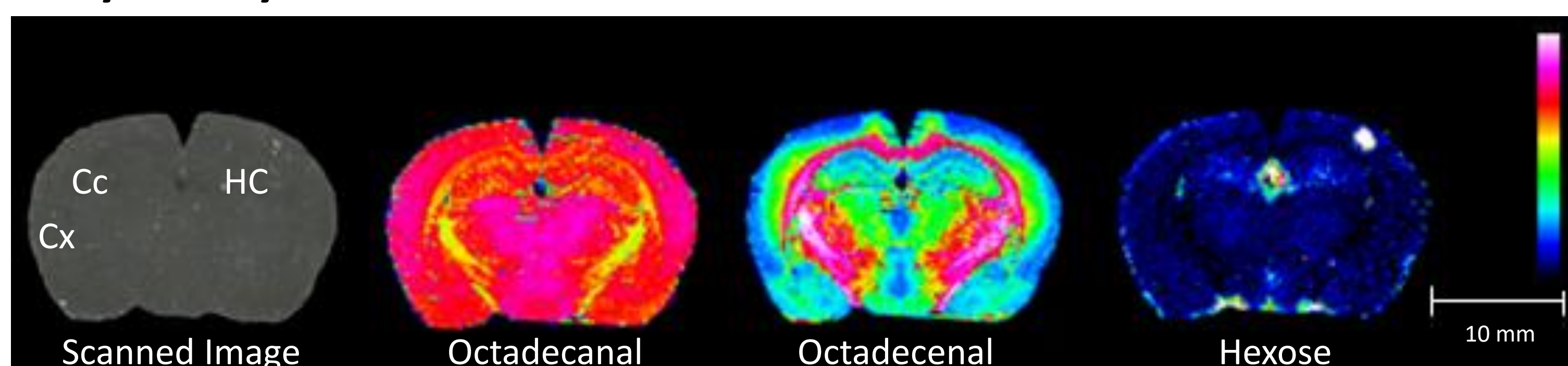


Figure 2. Imaging of fatty aldehydes and monosaccharides in rat brain with reactive matrix 9b (without co-matrix). Spatial resolution: 150 μ m. Intensity set individually to reveal the best possible distribution of the compounds. Cx: cortex; cc: corpus callosum; HC: hippocampus.

METHODOLOGY

Matrix development

Candidate MALDI matrices (9a to 9c) were designed and synthesized aiming to incorporate a UV-chromophore, an ionizable moiety and a reactive hydrazide group, respectively, into their structures.

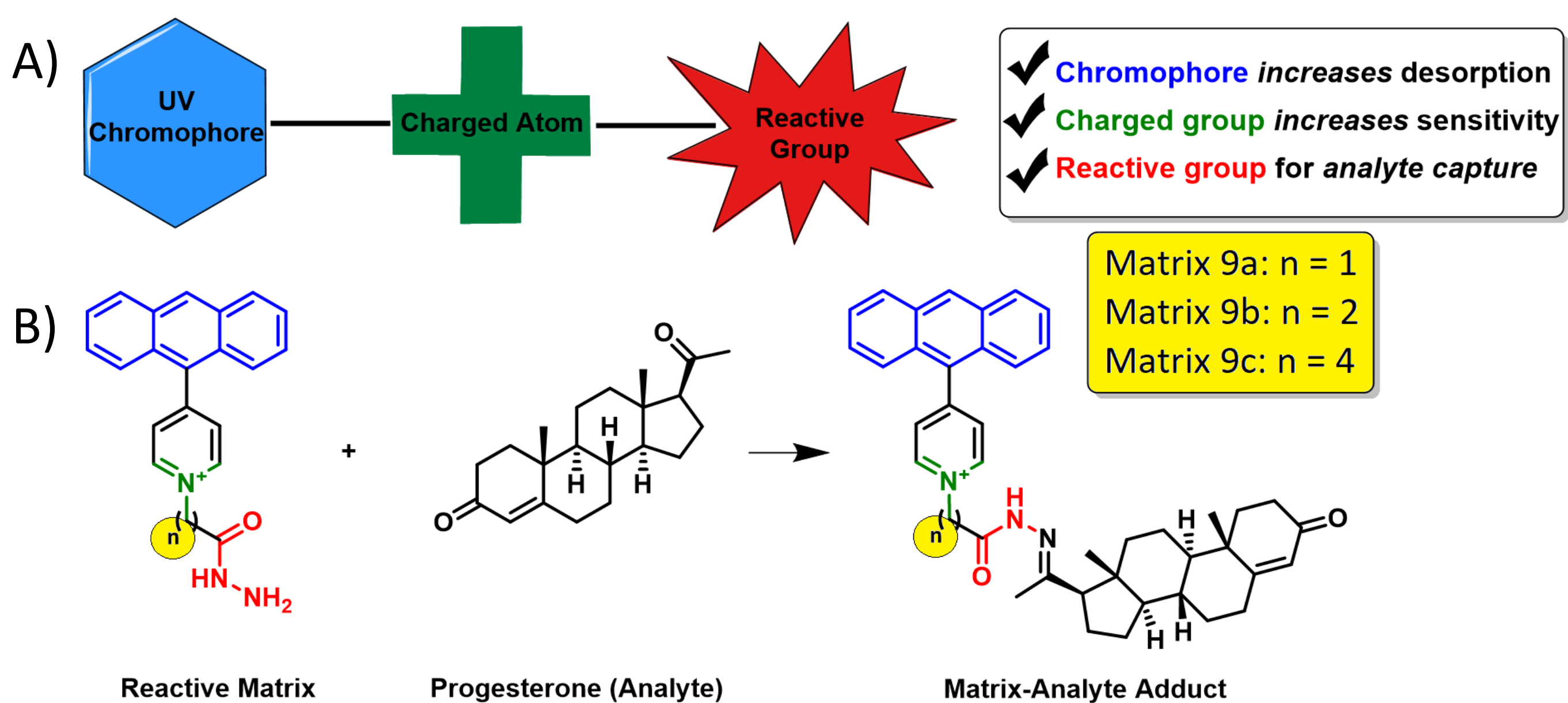


Figure 4. A) Design aspects for the novel carbonyl-targeting reactive matrices. B) Reaction scheme

The hydrazide group of the matrices reacts selectively with analyte carbonyl groups, forming covalent matrix-analyte adducts.

Endogenous compounds in mouse brain

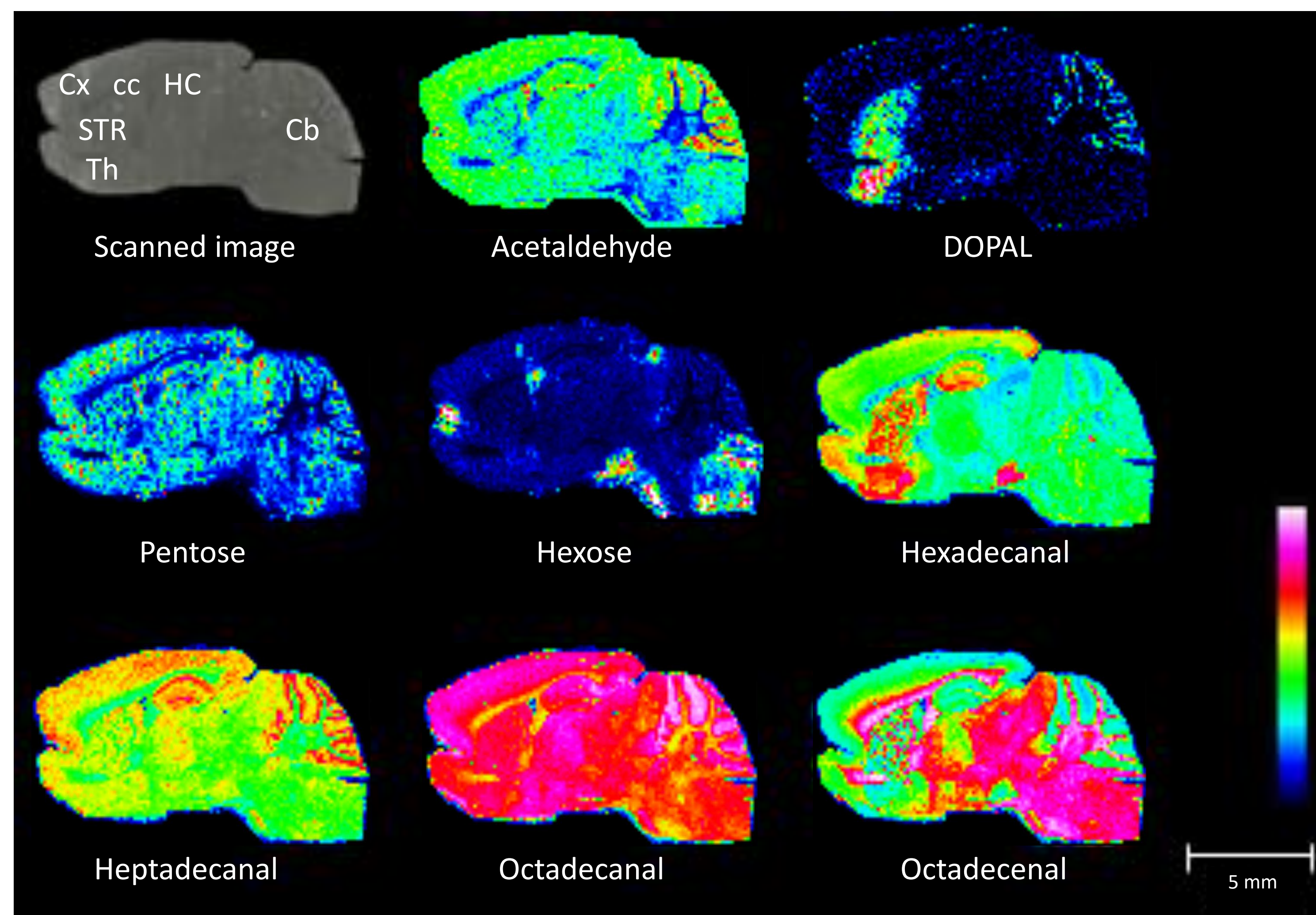


Figure 3. Imaging of a metabolites and endogenous compounds in mouse brain using reactive matrix 9b (without co-matrix). Spatial resolution: 120 μ m. Intensity set individually to visualize the best possible distribution of the compounds. Cx: cortex; cc: corpus callosum; STR: striatum; Th: Thalamus; HC: hippocampus; Cb: cerebellum.

- The spatial distribution of a number of endogenous aldehydes together with some monosaccharides were imaged using in-house matrix 9b without the need of additional DHB co-matrix (Figures 2 and 3).

CONCLUSIONS

- A one-step, on-tissue chemical derivatization method has been developed for MALDI MSI of carbonyl containing endogenous compounds.
- The in-house developed reactive matrices offered higher or equal MALDI-MSI ionization efficiency compared to Girard's reagent P for the tested carbonyls.
- In many cases, this was obtained without additional application of DHB, which is a requirement for achieving sufficient ionization with the Girard-P reagent.

Tissue preparation

Rat and mouse brain, respectively, and rat adrenal cortex were used for optimization of the derivatization conditions. Cryosections of 12 μ m thickness were thaw-mounted onto indium tin oxide coated glass slides which were stored at -80 $^{\circ}$ C. The slides were thawed in a vacuum desiccator for at least 20 minutes before matrix application.

Conditions for derivatization and application of co-matrix

Matrix application was performed using a TM-sprayer (HTX Technologies). 10 mg of reactive matrices in 5800 μ L 70% MeOH + 200 μ L TFA were applied in 20 passes (nozzle temp. 90 $^{\circ}$ C, 1100 mm/min, flow rate 80 μ L/min). The co-matrix (35 mg/mL 2,5-dihydroxybenzoic acid, DHB) in 50% ACN with 0.2% TFA was applied in 6 passes (criss-cross): nozzle temperature 95 $^{\circ}$ C, 1100 mm/min, flow rate 70 μ L/min.

MALDI-MSI

The obtained intensities of the model compounds and endogenous carbonyl containing compounds were evaluated following MALDI FTICR MSI (solariX 7T 2w, Bruker Daltonics) using flexImaging (Bruker Daltonics).

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