Supporting Information

*Christina Meisenbichler, Florian Kluibenschedl and Thomas Müller**

A 3-in-1 Hand-Held Ambient Mass Spectrometry Interface for Identification and 2D Localization of Chemicals on Surfaces

Chemicals and Samples. (3-Aminopropyl)trimethoxysilane, 4-Bromacetophenone, 2-Heptanone, Adenosine, Allyl phenyl sulfide, Bis(trimethylsilylmethyl) sulfide, Chlorpyrifos, Citric acid, Cyclohexanone oxime, Cytidine, Dimethyl sulfoxide, Ethyl bromoacetate, Ibuprofen, N-Methyl-2-pyrrolidone, o-Xylene, D-L-Phenylalanine, Polypropylene glycol (PPG) 1000, Pyridine, and Tetraethylammonium chloride were purchased from Sigma-Aldrich (St. Louis, USA) and Toluene from VWR (Fontenay Sous Bois, France). Anthracene, Ascorbic acid, Citronellal, Dimethylacetamide, Dimethylformamide, Glycerol, Myrcene and Reserpine were purchased from Fluka (Buchs, Switzerland), Benzoic acid from Acros Organics (Geel, Belgium).

Determination of limits of detection.

Limit of detection (LOD) for LTP mode: standard solutions of 2-heptanone in methanol were prepared (2 mM, 10 mM, 20 mM and 200 mM); 1 µl of a standard solution was spotted onto a glass plate. LOD was found to be 20 nmol of 2-heptanone spotted onto a glass surfaces. LOD for DESI/EASI mode: standard solutions of PPG 1000 in methanol were prepared (1 µM, 5 µM, 10 µM, 20 µM and 200 µM); 1 µl of a standard solution was spotted onto silica TLC. LOD was found to be 5 pmol of PPG 1000 spotted onto a TLC plate.

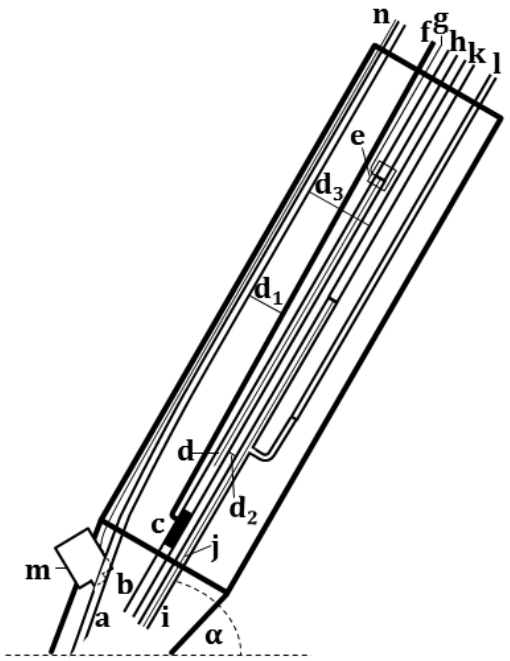


Figure S-1. Schematic representation of the hand-held, pen-like ambient mass spectrometry interface comprising a DESI/EASI as well as an LTP ion source. The components and materials, which were used for (a) – (n), are listed in table S-1; the angle α was kept between 40° - 65°; distances preferably were d_1 = 5 mm, d_2 = 2mm, and d_3 = 8mm.

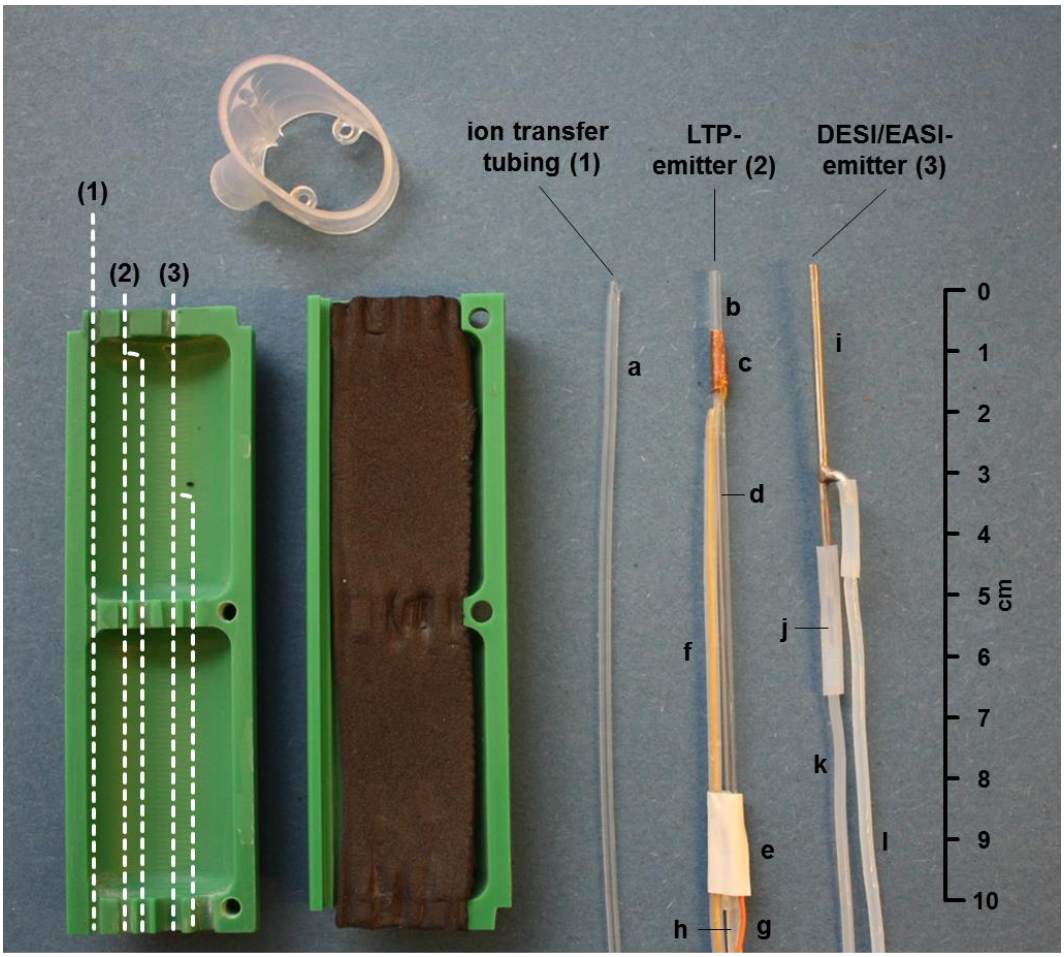


Figure S-2. Teardown of the hand-held, pen-like ambient mass spectrometry interface comprising a DESI/EASI as well as an LTP ion source. The two halves of the housing as well as the transparent tip are shown on the left side; the dashed lines mark the positions of the ion transfer tubing (1), the LTP emitter (2) and the DESI/EASI emitter (3). On the right side, ion transfer tubing (1), LTP emitter (2) and DESI/EASI emitter (3) are depicted together with ruler. All components and materials used for (a) – (l) are listed in Table S-1.

Figure S-3. The flexible ion transfer tubing of the DESI/EASI/LTP source was directly connected to the heated capillary of a LTQ-Orbitrap XL mass spectrometer (Thermo Fisher Scientific GmbH) using a PTFE sleeve.

Table S-1. Materials and components used for the construction of the hand-held interface

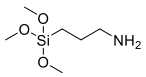
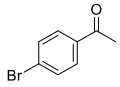
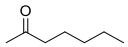
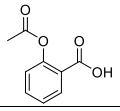
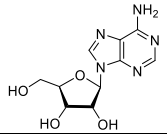
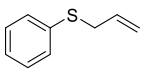
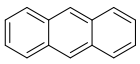
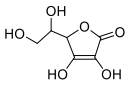
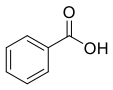
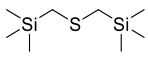
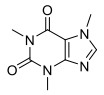
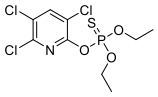
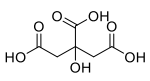
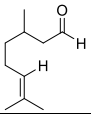
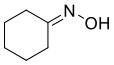
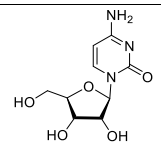
Symbol	Material	Description	Value
Droplet/ion transfer tubing			
a	PEEK tubing Polyether ether ketone or FEP tubing Fluorinated ethylene propylene	flexible tubing	ID: 0.8 mm OD: 1.6 mm length: 60 cm
LTP-emitter			
b	glass capillary	dielectric barrier	ID: 0.6 mm OD: 1.4 mm length: 8.5 cm
c	copper tape	outer electrode	width: 1.0 cm
d	copper wire	grounding electrode	OD: 100 µm length: 5.0 cm
e	PTFE tubing Polytetrafluoroethylene	junction	width: 1.0 cm
f	isolated cable	live wire from the AC high voltage generator	2-5 kV_{pp} 22.5 kHz
g	isolated cable	connection to ground	
h	PE tubing Polyethylene	He gas supply	ID: 1.0 mm OD: 1.8 mm gas stream: < 1 bar
DESI/EASI-emitter			
i	metal capillary	sprayer tip	ID: 1.6 mm OD: 0.8 mm length: 5.0 cm
j	fused silica tubing	sprayer tip	ID: 50 µm OD: 360 µm length: 5.5 cm
k	PE tubing Polyethylene	solvent supply MeOH, EtOH or IPA	ID: 500 µm OD: 1.6 mm flow rate: 10-20 µL/min
l	PE tubing Polyethylene	N_2 gas supply	ID: 1.0 mm OD: 1.8 mm gas stream: ≥ 2 bar (DESI) ≥ 4 bar (EASI)
Camera module			
m	Raspberry Pi camera	camera module	
n	ribon cable	connecting cable	length: 30 cm

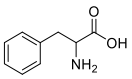
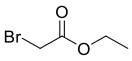
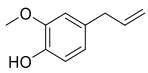
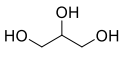
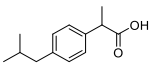
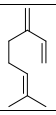
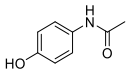
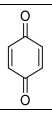
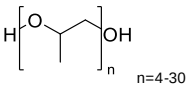
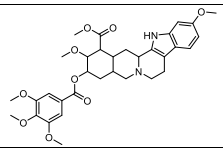
Separate Control Unit

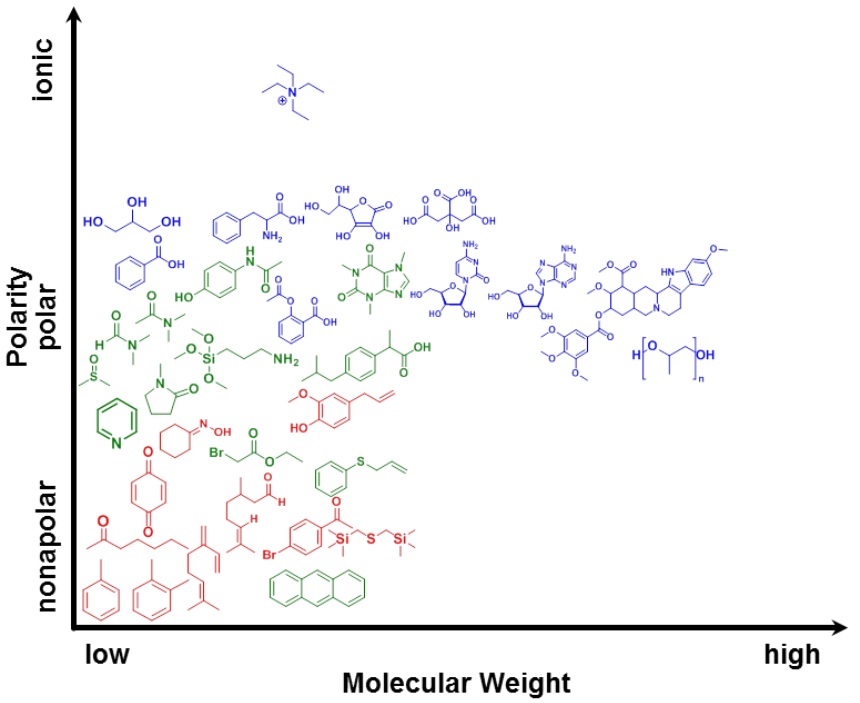
Hardware: A Raspberry Pi 4 (Model B) with a 1.5 GHz 64-bit quad core ARM Cortex-A72 processing unit and 4 GB random-access memory was utilized for switching between the three ionization modes LTP, DESI and EASI. A miniature camera for a Raspberry Pi (Type OV5647) with a 5 MegaPixel sensor and 70° degree field of view in length of 30 cm, was mounted at the frontend of the pen-like device, extended using a flex cable and connected to the 15 pin Camera Slot Interface (CSI) of the Raspberry Pi 4. Furthermore, a Seeed Relay Board v1.0 for Raspberry Pi was used to (i) switch two solenoid gas valves to control Helium and Nitrogen gas flows, (ii) switch a reed relay to interconnect the high voltage potential of the mass spectrometer, and (iii) power on the AC high voltage generator for the LTP ion source.

Software: All software was written in Python running on the common Raspberry Pi Operating System (Raspbian Buster, 2019) as well as on the Windows 7 OS of the workstation of the mass spectrometer. The separate control unit of the hand-held interface and the workstation were connected via Ethernet. A basic version of the supplemental software displayed a live stream from the camera module and showed a control window with buttons to switch between DESI, EASI and LTP mode.

Table S-2. Comprehensive list of studied compounds: names, chemical structures, molecular formulae and the exact masses of the neutral compounds, types of molecular ion which could be detected in LTP or DESI/EASI mode.

Name	Structure	Exact mass and Molecular Formula	LTP-MS	DESI- / EASI-MS
(3-Aminopropyl)-trimethoxysilane		179.0978 $C_6H_{17}O_3NSi$	$[M+H]^+$	$[M+H]^+$
4-Bromoacetophenone		197.9674 C_8H_7OBr	$[M+H]^+$	
2-Heptanone		114.1039 $C_7H_{14}O$	$[M+H]^+$	
Acetylsalicylic acid		180.0423 $C_9H_8O_4$		$[M+Na]^+$
Adenosine		267.0962 $C_{10}H_{13}N_5O_4$		$[M+H]^+$
Allyl phenyl sulfide		150.0503 $C_9H_{10}S$	$[M+H+O]^+$	$[M+H+O]^+$ $[M+Na]^+$
Anthracene		178.0783 $C_{14}H_{10}$	$[M-H]^+$	$[M+2H]^+$
Ascorbic acid		176.0321 $C_6H_8O_6$		$[M-H]^-$
Benzoic acid		122.0368 $C_7H_6O_2$		$[M-H]^-$
Bis(trimethylsilylmethyl) sulfide		206.0975 $C_8H_{22}SSi_2$	$[M+H]^+$ $[M+H+O]^+$	
Caffeine		194.0804 $C_8H_{10}N_4O_2$	$[M+H]^+$	$[M+Na]^+$
Chlorpyrifos		348.9257 $C_9H_{11}Cl_3NO_3PS$	$[M+H]^+$	$[M+K]^+$
Citric acid		192.0270 $C_6H_8O_7$		$[M-H]^-$
Citronellal		154.1358 $C_{10}H_{18}O$	$[M-H]^+$	
Cyclohexanone oxime		113.0841 $C_6H_{11}NO$	$[M+H]^+$	
Cytidine		243.0850 $C_9H_{13}N_3O_5$		$[M+H]^+$

Dimethyl sulfoxide		78.0139 C_2H_6OS	$[M+H]^+$	$[M+Na]^+$
Dimethylacetamide		87.0684 C_4H_9NO	$[M+H]^+$	$[M+Na]^+$
Dimethylformamide		73.0528 C_3H_7NO	$[M+H]^+$	$[M+Na]^+$
D-L-Phenylalanine		165.0790 $C_9H_{11}NO_2$		$[M-H]^-$
Ethyl bromoacetate		165.9629 $C_4H_7BrO_2$	$[M+H]^+$	$[M+Na]^+$
Eugenol		164.0837 $C_{10}H_{12}O_2$	$[M+H]^+$	
Glycerol		92.0473 $C_3H_8O_3$		$[M+Na]^+$
Ibuprofen		206.1307 $C_{13}H_{18}O_2$	$[M+H]^+$	$[M-H]^-$
Myrcene		136.1252 $C_{10}H_{16}$	$[M+H]^+$	
N-Methyl-2-pyrrolidone		99.0684 C_5H_9NO	$[M+H]^+$	$[M+H]^+$ $[M+Na]^+$ $[M+K]^+$
o-Xylene		106.0777 C_8H_{10}	$[M-H]^+$	
Paracetamol		151.0633 $C_8H_9NO_2$	$[M+H]^+$	$[M+H]^+$ $[M+Na]^+$
p-Benzochinone		108.0211 $C_6H_4O_2$	$[M+H]^+$ $[M+2H]^+$	
Polypropylene glycol		58.0419 C_3H_6O		$[M+Na]^+$ $[M+K]^+$
Pyridine		79.0422 C_5H_5N	$[M+H]^+$	$[M+H]^+$ $[M+Na]^+$
Reserpine		608.2734 $C_{33}H_{40}N_2O_9$		$[M+H]^+$
Tetraethylammonium chloride		130.1590 $C_8H_{20}N^+$		$[M-Cl]^+$



Toluene		92.0626 C_7H_8	$[M-H]^+$

Figure S-4. The major advantage of combining complementary ionization techniques in a single hand-held device is an explicit extension of the operating range of the ion source. Structures in green represent compounds which were detected in DESI, EASI as well as LTP mode, while compounds drawn in red required LTP ionization and structures drawn in blue could only be detected using DESI or EASI.

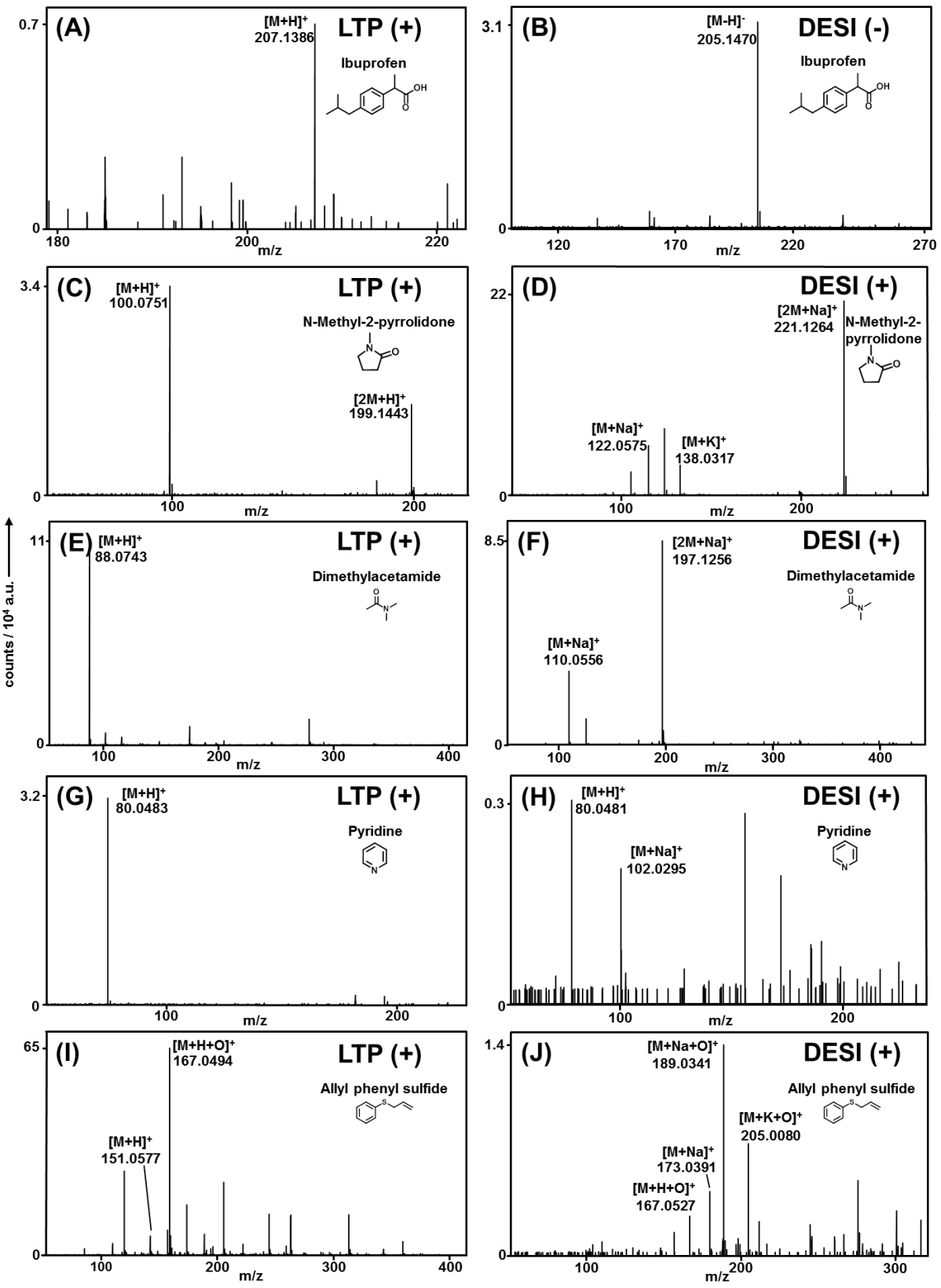


Figure S-5. Mass spectra obtained using the LTP ion source (A, C, E, G, I) as well as using the DESI ion source (B, D, F, H, J): commercial Ibuprofen (A and B), N-Methyl-2-pyrrolidone (C and D), Dimethylacetamide (E and F), Pyridine (G and H), and Allyl phenyl sulfide (I and J).

Interactive Mass Spectrometry Imaging (IMSI)

Imaging data acquisition was performed with an in-house developed python program, which utilizes OpenCV for digital image processing and to track the motion of the pen. In order to spawn a reference point for a virtual 2D coordinate system, a small green dot was plotted on the surface of interest using a textmarker pen (BIC® Brite Liner®). OpenCV algorithms were then utilized to recognize the reference point within every single frame of the live video stream from the tip of the pen (Figure S-6A) and to calculate the pen's motion relative to the surface of interest (Figure S-6B). The tracking software simultaneously sent a start signal to the MS instrument, which continuously recorded full MS spectra.

In order to generate ion images the obtained .raw file was converted to a .mzXML file using the ReAdW converter. With the help of a second python script every single full MS spectrum was then assigned to a certain position within the 2D coordinate system. Finally, a contour plot was used to visualize relative signal intensities of selected ions (Figures S-6C and S-6D).

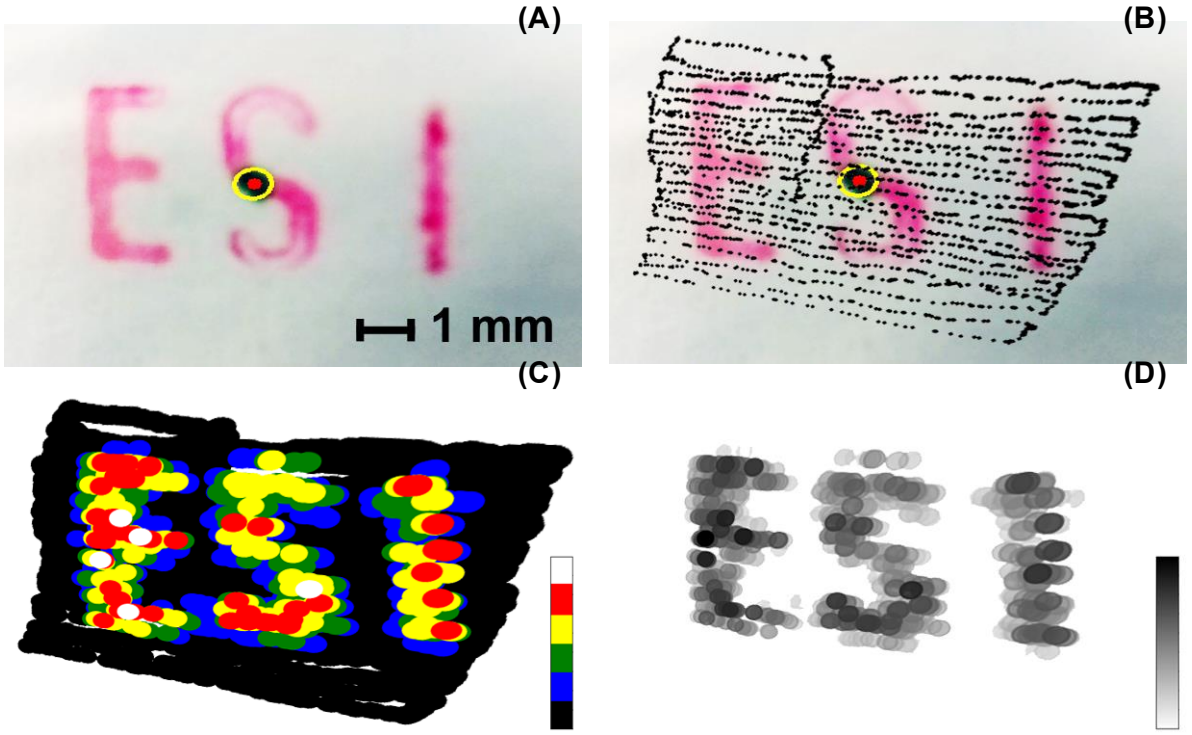


Figure S-6. IMSI experiment with the 3-in-1 hand-held pen utilizing EASI in positive ion-mode. (A) Three letters were imprinted on white paper using a stamp and red ink of a Staedler® Lumocolor permanent pen diluted with methanol; a green reference spot was plotted at the center of the letter "S"; the tracking software automatically recognized the spot and marked it with a red dot and a yellow circle. (B) The dotted line visualizes the tracked motion of the pen (i.e. the area of desorption) during the IMSI experiment; black dots mark coordinates where full MS spectra were recorded. (C) False color ion image representing the relative signal intensities found at m/z = 429 (from black = 0 % to white = 100 %). (D) Monochrome ion image representing the relative signal intensities found at m/z = 429 (from white = 0 % to black = 100 %).

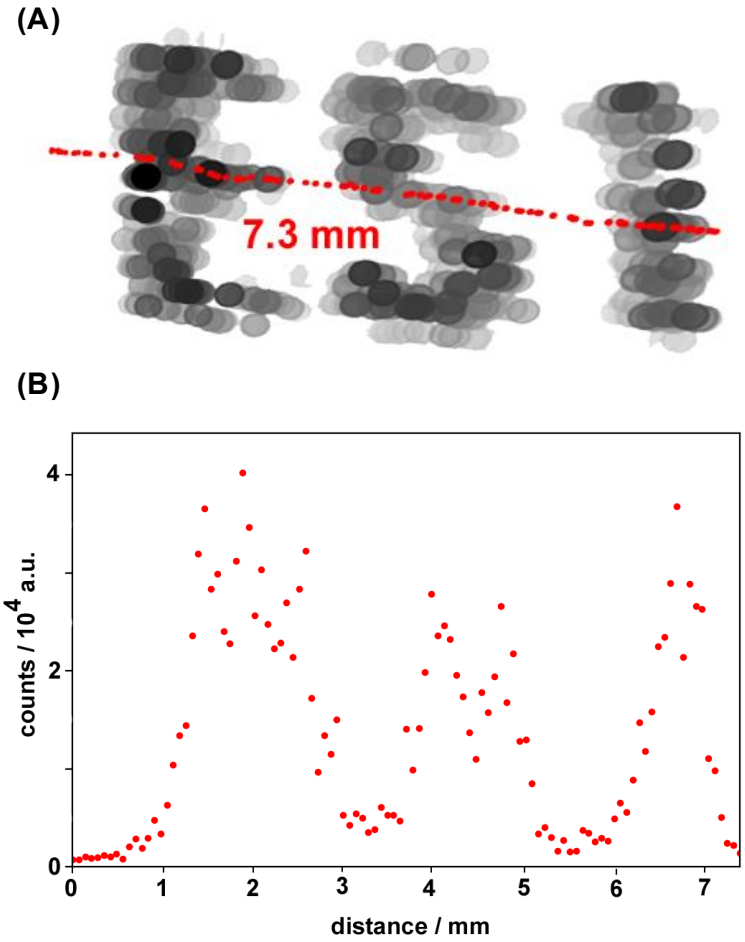


Figure S-7. Signal intensities of analyte ions (m/z = 429) observed along the pen's move across the sample: (A) The 7.3 mm long path of the area of desorption is highlighted in red; every dot marks a recorded full MS spectrum. (B) Covered distance plotted against observed signal intensities at m/z 429.