

Task

Molecular Information from 500 nL Droplets

Droplet based microfluidics is a powerful tool for high throughput experimentation in life science or material science research. Usually information about the droplet based processes are recorded by non-contact spectroscopic means. Unfortunately, absorbance or fluorescence based read-outs are unspecific parameters and require dedicated assays to gain molecular information in an indirect way.

Option

Surface Enhanced Raman Spektroskopie - SERS

Surface enhanced Raman spectroscopy allows the determination of compound specific Raman-bands at very low concentrations. Preconditions are the contact of the specimen specific compounds with a SERS-active surface. To interface the pipe based serial droplet processing technology and the array based SERS-substrates we developed a robotic system for I) the sensor triggered transfer of droplets onto SERS-substrates and II) SERS-Measurement of the array. In Figure 1 the overall system is shown.

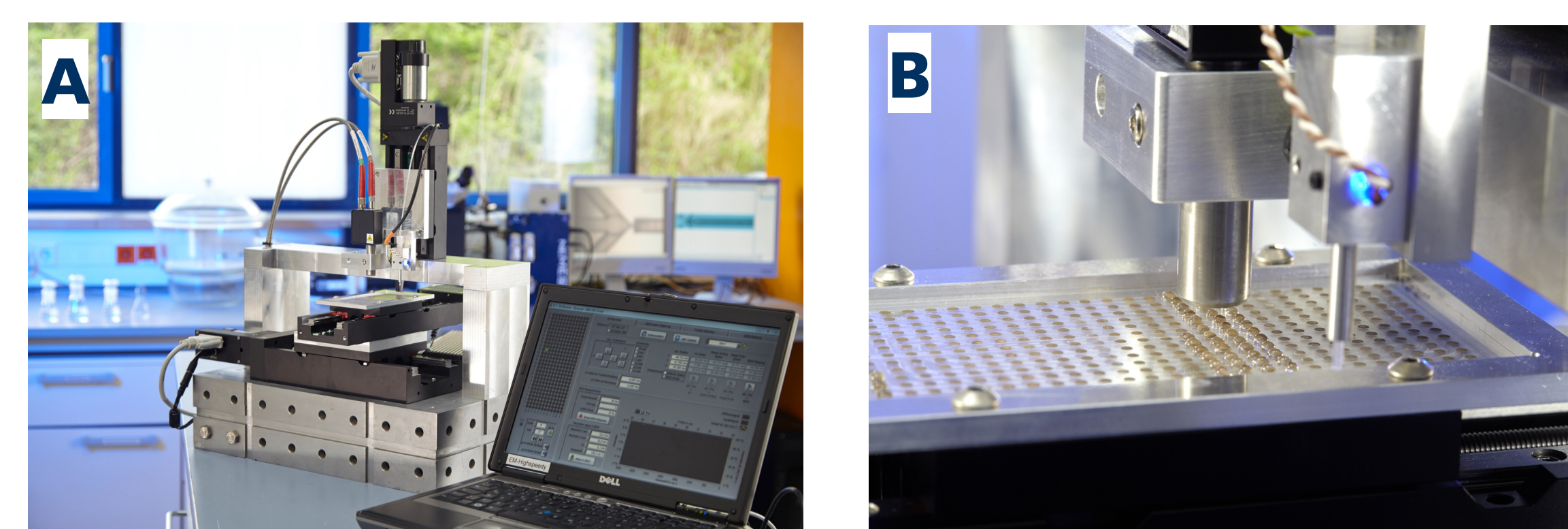


Figure 1: **A)** Image of the robotic system for sensor-triggered droplet transfer from pipe based processes onto SERS-active substrates. **B)** SERS-Probe measuring wetted SERS-Spots.

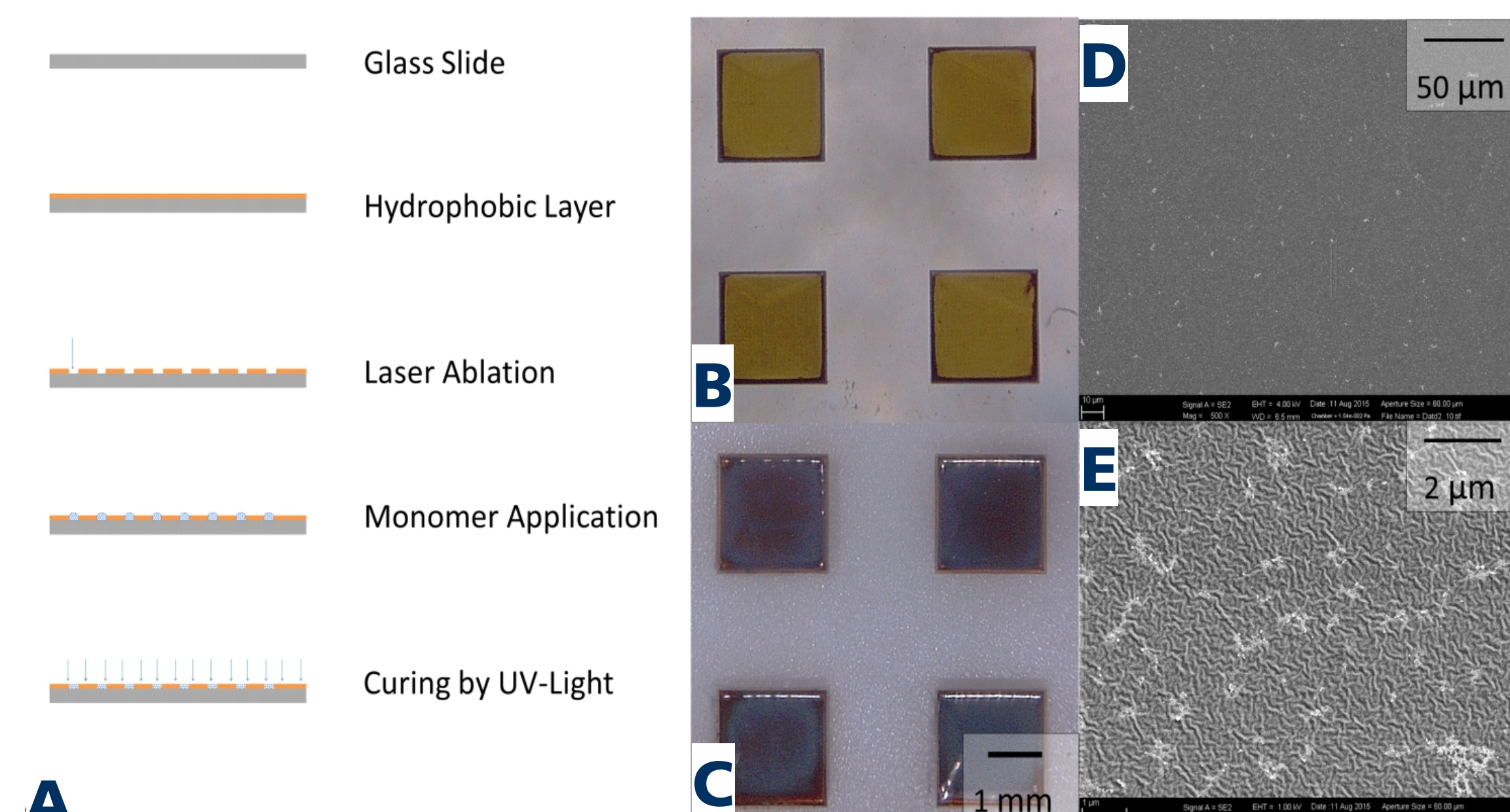


Figure 2: **A)** Formation process scheme for glass-based spott arrays **B)** Image of polymer spots with embedded Ag-nanoparticles **C)** Image of the spots after Ag-reinforcement **D + E)** SEM-Images of the appropriate spot surfaces.

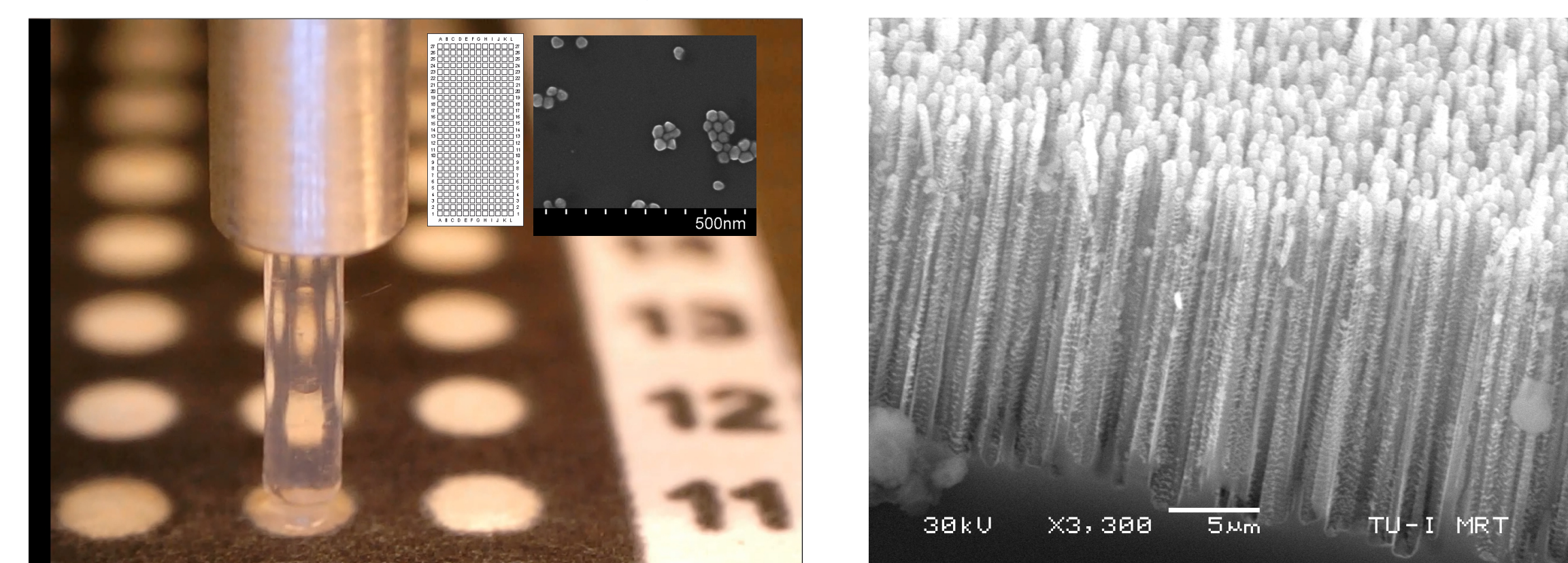


Figure 3: **A)** Image of a paper based SERS-Substrate made by solid-ink printing, termpering and colloidal Ag-nanoparticle coating during droplet transfer. Inlet: SEM-Image of Ag-Nanoparticles **B)** Image of a black silicon substrate coated with Ag.

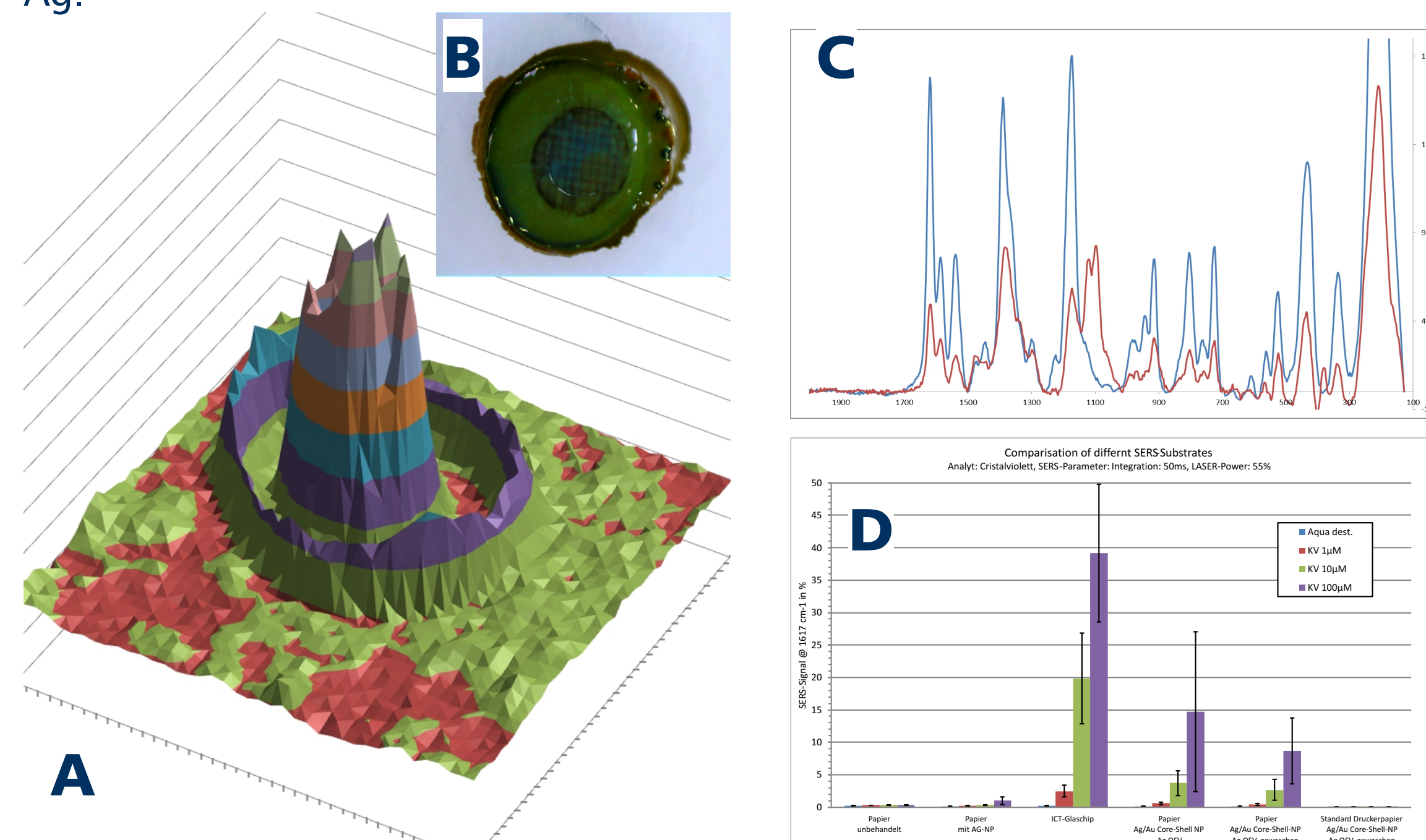


Figure 5: **A)** 2D-Scan of a single SERS-Spot after transfer of a 500 nL, 10 µmol Cristalviolet solution droplet. **B)** Image of the analyzed spot after SERS measurement **C)** Exemplary SERS-Spectra of spot with background measurement **D)** Comparison of the different SERS-Substrates (Ag@black-Si was not investigated).

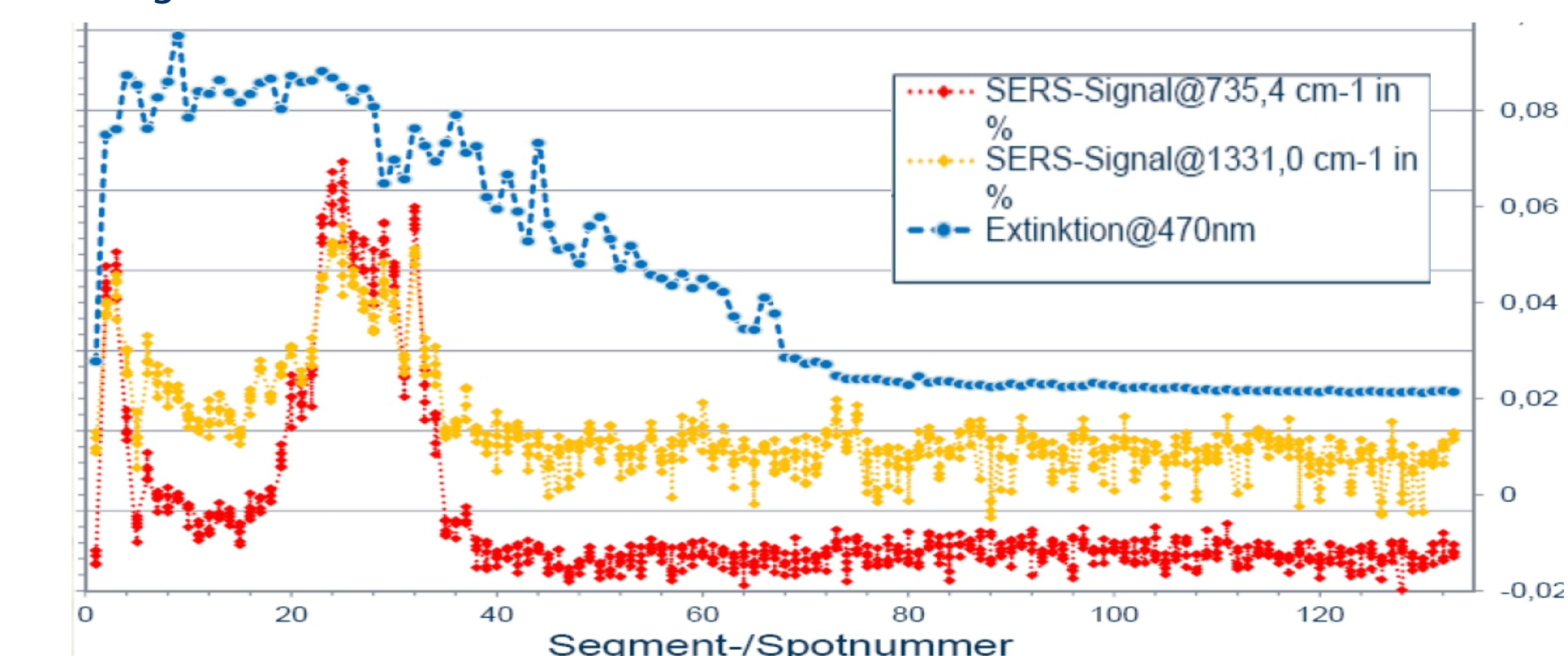


Figure 6: Application to soil microorganism. The sub lethal stimulation of a soil bacteria consortium on Cu-ion stress was observed clearly in the droplets 20-40.

Development

SERS - Substrates

We compared three different SERS-active substrates: a) RIE-etched black silicon substrates sputtered with Ag B) Solid-Ink printed paper substrates, coated with colloidal Ag-nanoparticles and C) Polyacrylate based spots on laser structured glass substrates. In Figure 2 and 3 the different substrate examples are shown.

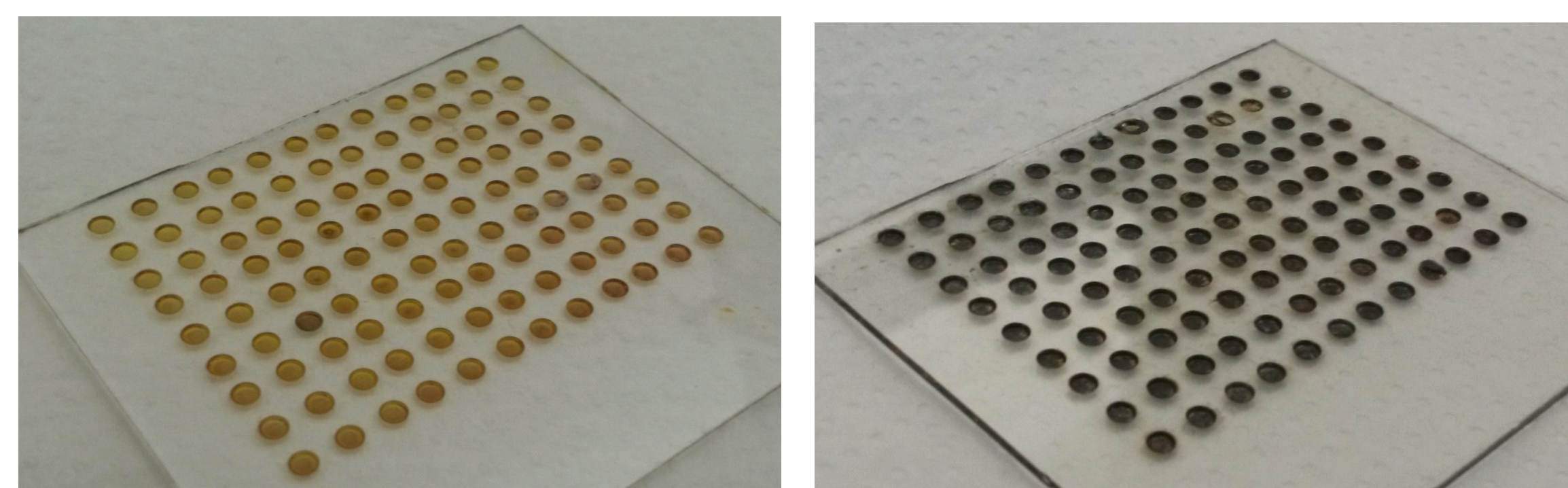


Figure 4: **A)** Image of a laser structured glass substrate, dip-coated with a monomer mixture containing a photoinitiator and AgNO₃ **B)** After UV-excitation.

Results

SERS-Analysis of Droplets

Optimized SERS-active polymeric spot arrays were made by laser ablation of hydrophobic glass substrates followed by selective wetting of the spots and photopolymerization of nanoparticle doped acrylate monomers. Process scheme and received spots are shown in figure 2 A-E. However, the second approach of paper based wicking substrates yield less intensive SERS-signals but with a much less demanding technological approach. Unfortunately, the Black-Si substrates did not show reproducible SERS-results and were not further investigated.

In figure 5 different results are summarized. To minimize the deviation caused by spot inhomogeneity (Fig. 5 A+B) as well as background variation, the development of a powerful data analysis software was required.

The response of soil microorganisms on heavy metal stress was investigated during the project BactoCat (FKZ: 031A161A). Therefore, a droplet sequence of microorganisms with increasing Cu-ion concentration was incubated for about one week and transferred onto the SERS-array. As a result the sub lethal stimulation of the microorganisms against the toxic Cu-ions was observed at different SERS-bands.