4.1.3 Scientific and technical results and foregrounds

During the first months of the project, the consortium has focussed primarily on developing the reflection mode (S11, the scattering parameter for reflection) of the current scanning microwave microscopy (SMM) technology and further extending the capabilities of the SMM to enable S21 transmission measurements. The team demonstrated the capability of the SMM to measure calibrated capacitances and dopant densities at the nanoscale. In particular the groups at Keysight and CNR-Rome have shown a noise level of 1 aF for capacitance measurements and a dynamic range of 10^{14}-10^{20} atoms/cm^3 for dopant profiling. Furthermore, they have shown a lateral resolution of 10 nm using standard electrical conductive cantilevers and semiconductor samples.

In parallel the consortium, to support the development of the overall system, designed, produced and used calibration kits to enable:

1. Quantification of the measured signals
2. Extraction of the intrinsic electromagnetic properties of the materials under investigation.

The team at MC2 technologies designed and manufactured calibration samples for complex permittivity measurements (**Figure 1**). The kits have been distributed to Keysight, IBEC, and CNR-IMM. Each calibration sample has several (>20) calibration spots (i.e. Schottky diodes). The partners decided that rather than a patent a scientific paper will generate greater impact to the scientific SMM community, so based on this deliverable a draft scientific paper is currently being worked upon and will be submitted for peer review in due course.

In collaboration with Keysight, these calibrations kits enabled the measurement of complex permittivity in the microwave range with the SMM equipment. In particular, complex impedance measurements have been performed on both the metal-oxide-semiconductor based (MOS) capacitors as well as the Schottky diodes (gold pads directly placed on the doped silicon without oxide). The calibration sample has been used for complex permittivity measurements of high-k oxides and complex permittivity measurements of cells and nanoparticles.

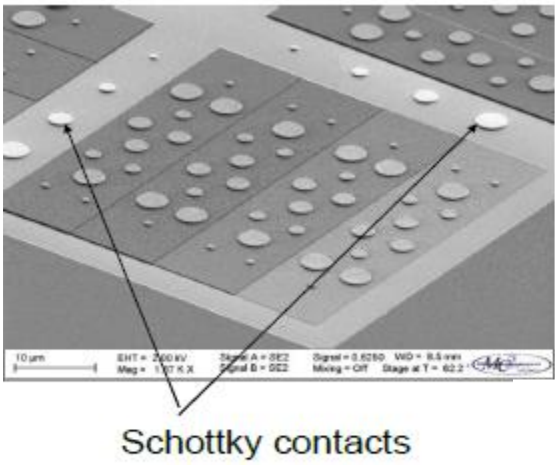

Figure 1. Scanning electron microscope micrograph of the kit for calibration of complex permittivity measurements

These experiments laid the groundwork for the VSMMART-NANO project regarding SMM capacitance calibration and demonstrated that the SMM delivers enough sensitivity both for capacitance measurements and lateral resolution.
These experimental measurements have been supported throughout by utilising state-of-the art computational modelling tools (EMPro and COMSOL), which have aided the validation of the system.

In particular, the consortium performed 2D and 3D advanced finite element analysis of the tip-sample interaction of a SMM. We investigated the effect of variations of the basic system variables, such as tip-angle, tip-radius, tip-sample distance and the operational frequency on the impedance. Also we studied the impedance variation when spherical inclusions of different sizes, permittivity and positions are present both beneath and onto the surface of a SiO_2 sample. Furthermore, we estimated the capacitive contributions of stray electromagnetic field between the cantilever and the sample. We studied the effect of variation in the conductivity of the bulk silicon substrate and the inclination angle of the probe on the impedance. Also we investigated the effect of variation in sample size on the system impedance. Our findings underline the sensitivity of the SMM system to small variations in both the probe and the sample parameters, which has important experimental implications as they shed new light on the sensitivity and performance of the SMM system.
Additionally, the team developed a new algorithm for the calibration of near-field scanning microwave microscopes, which enabled the simultaneous measurement of the topography, capacitance, and the resistance of a sample with standard AFM cantilevers that have a tip radius below 10 nm.

The team at CNR-Rome lead the design and manufacturing of a tool box allowing SMM measurements in transmission. The team performed the initial measurements in transmission by using the tool box connected with a Microwave Precision Network Analyzer (PNA) and a Scanning Microwave Microscopy (SMM). A broad-band prototype, made by a flange connector properly mounted on a dielectric sample holder, has been used in conjunction with the SMM to perform preliminary transmission.
Currently, imaging was possible including both the contact mode and a small separation between the SMM tip and the sample surface. In particular, dielectric structures can be imaged in greater details compared to metal ones, probably because of the higher losses induced by the signal back reflection. This is actually the first step towards the volumetric scanning technique. The obtained results are very important for demonstrating that measurements can be performed:

(i) in transmission mode, trespassing the limit of one-parameter only, and with the future possibility
to image both sides of the investigated sample, and
(ii) in non-contact mode, thus opening the possibility for measurements not affected by the tip wearing, which influences the calibration of the full setup.

The team at BNC continued the theoretical simulation of the performance of the VSMM in transmission mode

In particular, it developed an analytical model to calculate and relate the total impedance of the Transmission mod Scanning Microwave Microscope (Tx-SMM) to the scattering parameters and investigated the sensitivity of the system. Also it performed 3D advanced finite element analysis of the electromagnetic interactions between the probes and the sample in the Tx-SMM. The team compared the analytical and modelling results and studied the effect of variations in the conductivity of the bulk silicon substrate on the system characteristics. Furthermore, it investigated the effect of variations in the permittivity of both the subsurface constituents and the covering material of the sample on the scattering parameters. The numerical simulations showed that the Tx-SMM leads to better sensitivity (from twofold to as much as 5 times increase) than the system sensitivity in the reflection mode operation. In particular, we found the sensitivity of the phase of the S-parameter to be 3 times better than that of its magnitude. These findings underlined the increased sensitivity of the Tx-SMM system against small variations in the sample parameters, which s shed new light on the overall performance of the Tx-SMM system.

4.1.3.1 Prototypes of VSMM cantilever probes

A very important component of the VSMM instrument is the cantilever probes, which defines the lateral resolution of the device. The team at Nanoworld developed novel probes suitable for VSMM measurements where the key issue is the integration of a shielded signal transmission line into support chip, cantilever and tip of a SPM probe to reduce parasitic capacitances and electric stray fields.
Based on the application-specific requirements of Keysight and IBEC –strongly supported by electrical high
frequency simulations of BNC – the team in Erlangen developed a suitable probe design and a batch fabricating process to manufacture cost-effective shielded SPM probes (**Figure 2**).
Using the fabrication scheme we could realize different demonstrators of monolithic VSMM-probes and have performed first functionality tests as topography – and SMM measurements.

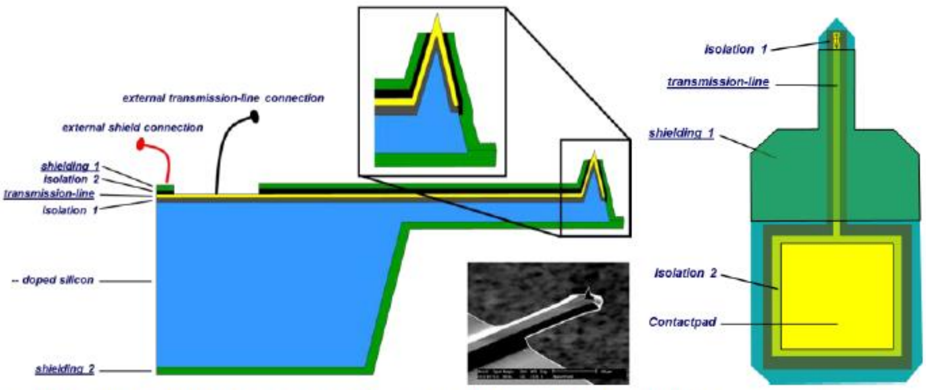

Figure 2. Sketch of the VSMM shielded probe. Left: cross section. Right: top view.

In parallel the team at CNR following the encouraging results obtained during the development of the transmission mode hardware, continued the development of the S21-Toolbox, using numerical simulations to evaluate the expected change in the overall performance of the Toolbox.

As a result, the team concluded that the best microwave emitter for providing a wideband response is a commercial flange connector whose RF launcher parameters can be easily tuned. Moreover, no significant improvement in the signal-to-noise ratio is obtained when narrow diameter connectors or cone-shaped pins are used. Finally, the transmitted power can be drastically reduced if too narrow pins are used as emitters.
Thus the team made some mechanical adjustments on the original sample holder to improve both the alignment and the interaction between the SMM tip and the sample "illuminated" by the bottom placed pin, but maintaining the same wideband radiating element.

All the efforts from the consortium allowed the development of an integrated VSMM instrument that can be used in several different modes allowing for subsurface imaging. The first group of new modes is operating at different frequencies, including dual frequency dC/dV, interferometric sweep SMM design, simple PNA frequency changes, and time domain gating experiments. The second group of new modes is working at different tip-sample distances including constant height imaging, lift mode and backwafer imaging. The latter group is also efficiently supported by 3D FEM numerical solvers including EMPro that was adapted for those workflows.
Some of the frequency domain modes (time domain gating and the simple PNA frequency approach) allow getting insights into the 3D layer structure and thicknesses of the individual layers. However, most of those techniques can be only used for semiconductors that have free carriers and depletion zones and can't be applied simply to bio-samples. In contrast, most of the non-contact imaging modes can be applied both to semiconductor and "soft" samples, such as cells. The non-contact imaging modes have been successfully used to measure the samples with either air, oxide or silicon in between tip and the sample. In all cases, roughly 300-500nm thickness is allowed to get proper signal-to-noise. The detailed value depends on the dielectric properties which can be modelled with EMPro. The 300-500 nm is currently the limit for shallow subsurface imaging using S21 and/or S11.
A scorecard based evaluation has been done and different aspects of the methods are evaluated. There are
two winner approaches, one in frequency domain (the interferometric sweep mode) and one in noncontact imaging (constant height imaging mode). EMPro modelling can currently be used to have semi-quantitative evaluation of the 3D geometry. In future, 3D calibration samples (in collaboration with MC2) and shielded cantilevers (in collaboration with NWS) will complement the portfolio for calibrated and quantitative 3D imaging.

4.1.3.2 Application of VSMM imaging in bioscience

Having developed and validated all the components of the VSMM prototype, the team at IBEC analysed the ability of the VSMM instrument to characterize the dielectric properties of cells and single nanoparticles. To this end they have carried complex permittivity measurements on single bacteria cells (Escherichia Coli, **Figure 3**) and single gold nanoparticles (~40 nm radius). Results show that the SMM instrument is able to quantitatively measure the intrinsic dielectric properties of single bacteria cells in the GHz frequency range. Furthermore, they also show the possibility to detect the dielectric response of single gold nanoparticles in remarkable agreement with theoretical predictions. These results provided the basis for the analysis of the interaction of metallic nanoparticles with living cells.

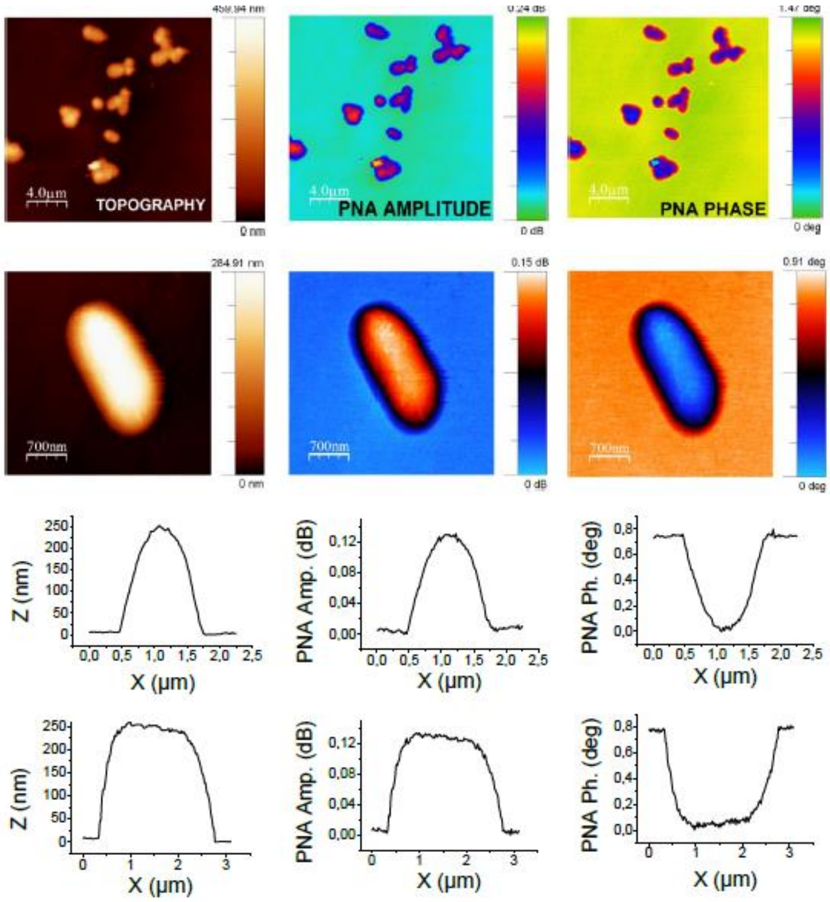

Figure 3. First column: topographic images of *e. coli* bacteria on HOPG in ambient conditions with two different magnifications and the corresponding transversal and longitudinal profiles. Second and third column: raw PNA amplitude and phase images and their corresponding cross-section profiles.

The team at IBEC lead the application of scanning microwave imaging of fixed cell samples for imaging of fixed cells in air conditions. Sample preparation protocols have been optimized by BNC in order to facilitate its imaging with the scanning microwave microscope.

The team at BNC selected iHBECs cells for imaging because they represent a good model for further studies of nanotoxicology. These cells are primary human epithelial cells immortalized by serial transfection using retroviral constructs containing cyclin-dependent kinase 4 (Cdk4) and human telomerase reverse transcriptase (hTERT). Basal cells are considered the stem/progenitor cell population in the human airway and when grown in a culture, they have basal characteristics.

Similarly, BNC team considered a variety of sample substrates with different coatings and electrical properties to elucidate the most suitable for SMM imaging. SMM images on fixed cells have been recorded in the typical reflection mode imaging of the current SMM configuration (Figre 4) , showing clearly some of the nanoscale structural features of cells.

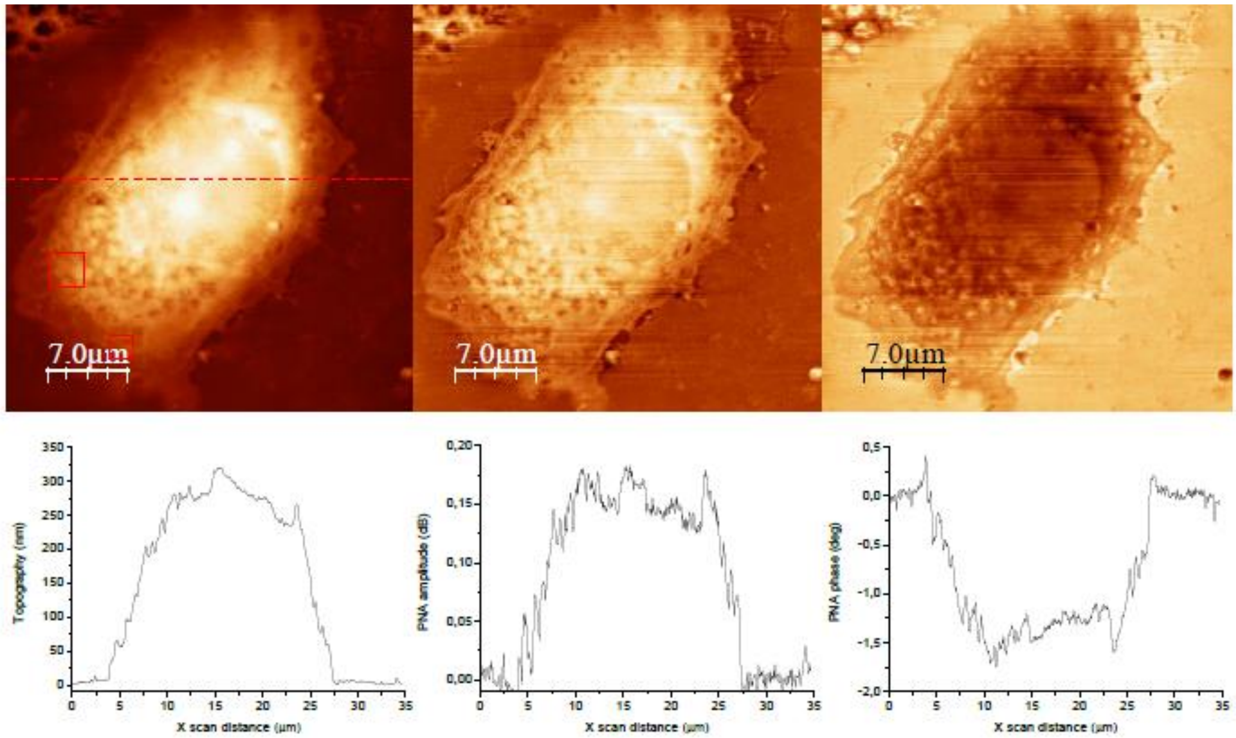

Figure 4. Top row: (left) Topographic, (middle) PNA amplitude and (right) PNA phase images simultaneously obtained on an isolated CHO cell on a silicon substrate. Bottom row: corresponding profiles taken along the red line marked in the topographic image

Then the consortium employed the VSMM to image fixed cell samples with internalized gold nanoparticles of 100 nm in diameter. Sample preparation protocols have been optimized in terms of nanoparticle solution concentration. Samples have been prepared both on conducting substrates for scanning microwave imaging, as well as, on glass substrates for optical microscopy inspection. SMM images have been recorded in the reflection mode imaging of the current SMM configuration. Small structures down to 100 nm have been imaged, although they could not be unambiguously identified as nanoparticles. The analysis of the results indicate that living cell imaging in liquid media may offer better conditions for imaging of internalized nanoparticles since a smoother surface will be displayed by the cells.

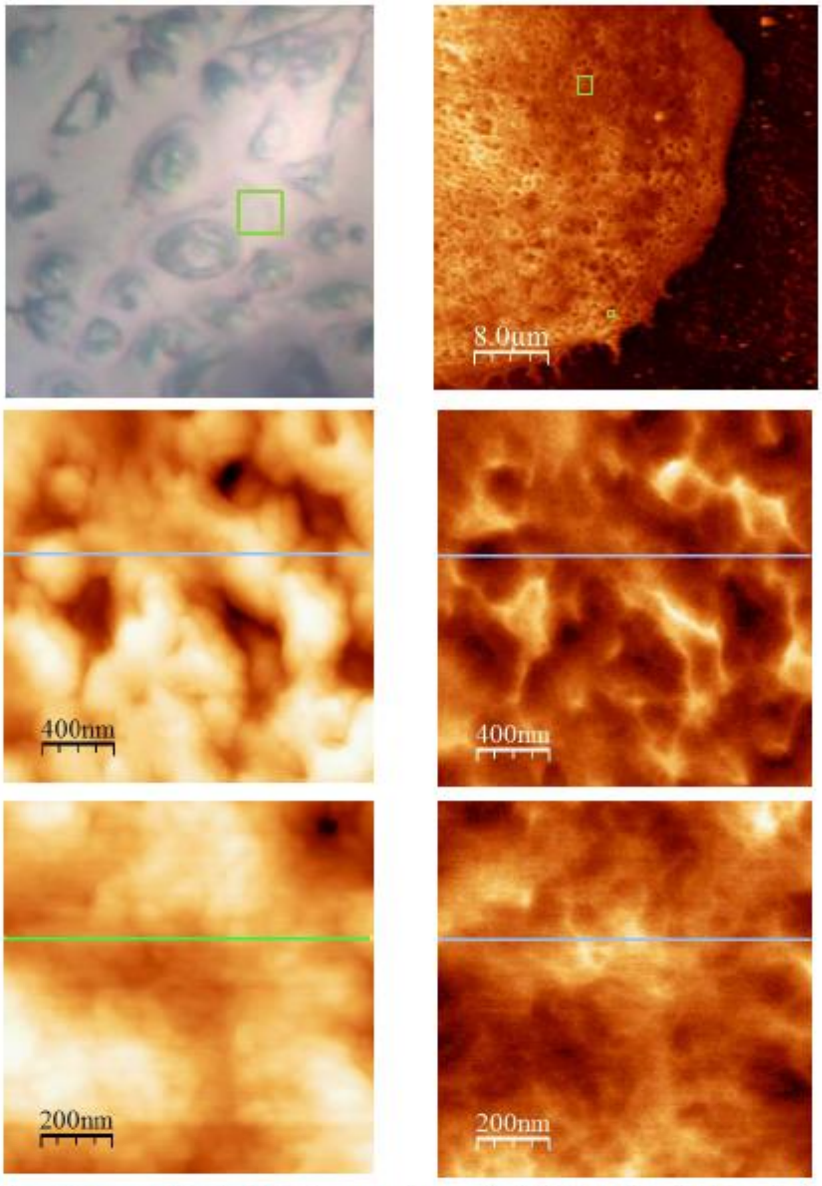

Figure 5. Top row: (left) Optical microscopy image of an iHBEC sample with internalized nanoparticles and (right) large scale topographic image obtained in the highlighted region in the optical image. Middle and bottom rows: Topographic (left) and electrostatic force microscopy image (right) obtained at higher magnification (highlighted in green).

4.1.3.4 3D Calibration Kits

The team at MC2 was involved on the design and manufacturing of calibration samples for standardized 3D SMM measurement. The layout of the calibration sample was designed and an optimized layout was achieved. The process workflow has been developed. To achieve this, a number of process tests have been performed in the cleanroom. Two process solutions have been considered. In one case, metallic nanoparticles have been included in Silicon Oxide. In the second case, dielectric nanoparticles have been included in Silicon Oxide. In collaboration with Partners (Keysight, CNR-IMM, IBEC and NWS) these calibrations kits have been measured, analysed and demonstrated to be a very promising 3D tomographic microwave imaging calibration kit at nanoscale.These new calibration kits have been sent to all the partners of the consortium for evaluation, in total more than 10 samples have been distributed to Keysight, IBEC, and CNR-IMM. Each calibration sample has several calibration spots.

These Calibration kits (10 samples) for standardized 3D SMM measurements calibration samples have been sent also to the US NIST laboratory for evaluation (Pavel Kabos).

Figures 6 and 7 present the Si_yN_x pellets measurement at 19.31 GHz where a reflexion coefficient of S11 -18dB have been obtained with the reflectometer SMM.

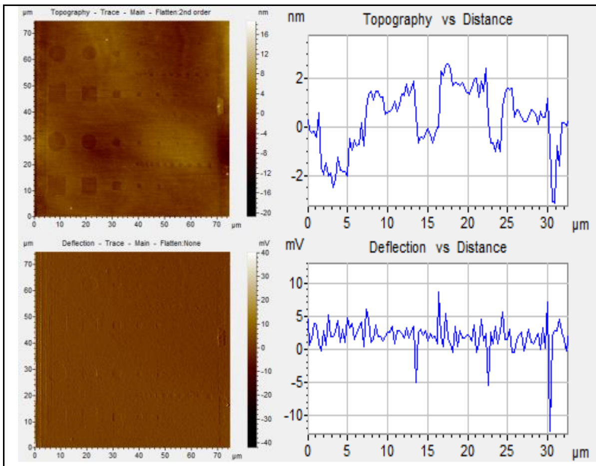

Figure 6: Profile AFM measurements of the 3D calibration kit Si_xN_y pellets.

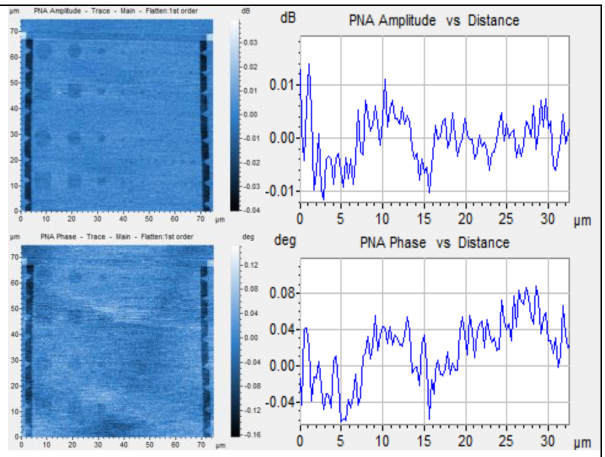

Figure 7: Magnitude and phase of the S11 reflexion coefficient following the position of the cantilever on the 3D calibration kit.

The SMM measurements were repeated at lower frequencies using the electrostatic force microscope (EFM). Figures 8 and 9 present the gold pellets measurement EFM measurements.

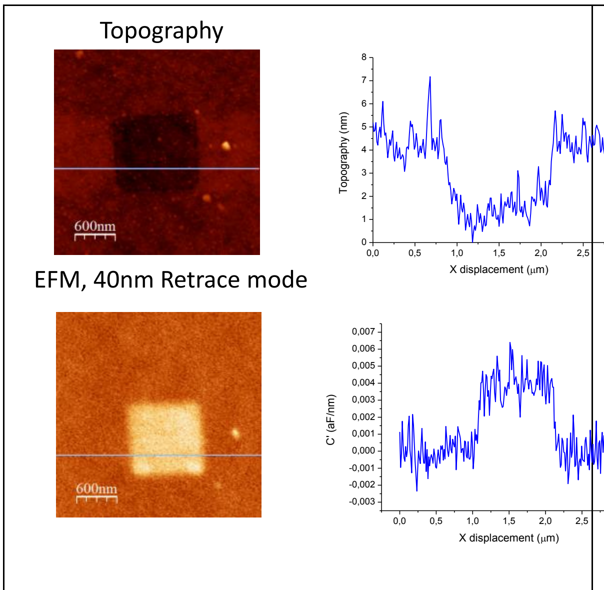

Figure 8 : Profile AFM measurements of the 3D cal kit Au pellets and the EFM associated of a square of 1µm.

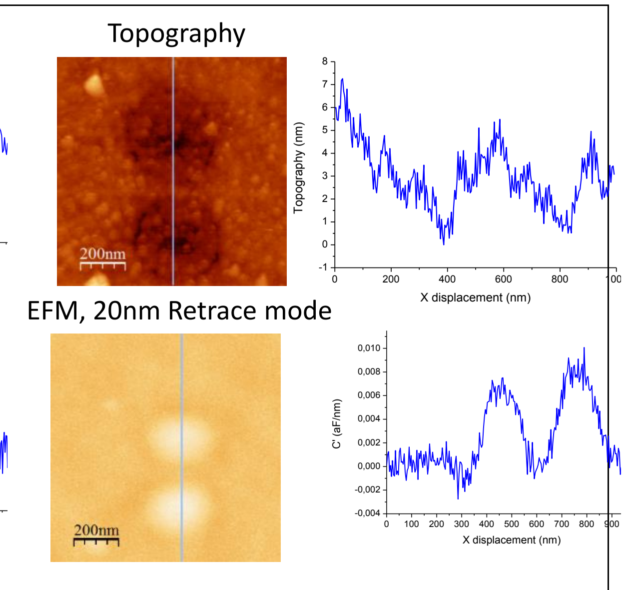

Figure 9: Profile AFM measurements of the 3D cal kit Au pellets and the EFM associated of a close disc 200nm.

The 1µm square buried structures and the close disc 200nm buried can be clearly detected in EFM. The images shows a minimal cross-talk with topography which needs to be taken into account for quantitative measurements. The same studies have been done regarding the 3D calibration kit of the Si_xN_y pellets. Figures 10 and 11 present the Silicon Nitride pellets EFM measurements.

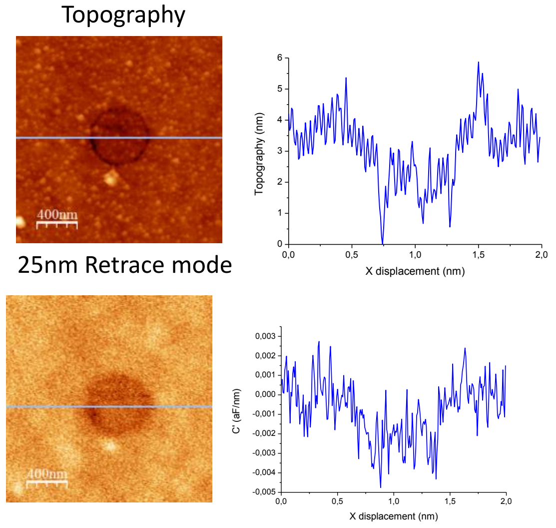

Figure 10: Profile AFM measurements of the 3D calibration kit Si_xN_y pellets and the EFM associated of a circle of 400nm.

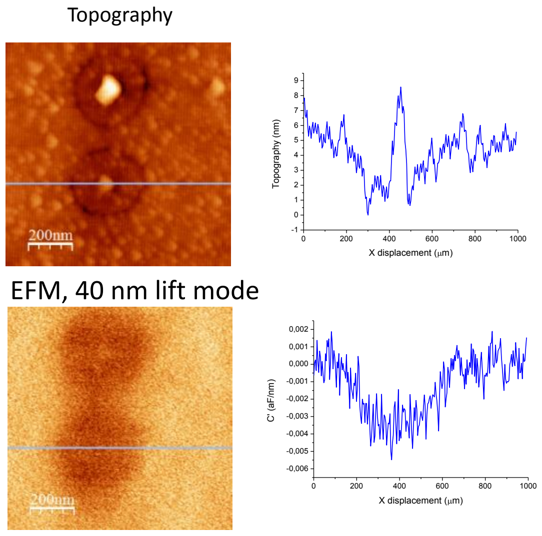

Figure 11: Profile AFM measurements of the 3D cal kit Si_xN_y pellets and the EFM associated of a close disc 200nm.

By comparing the results obtained with the same EFM setup on the two 3D calibration kits (gold pellets and Si_xN_y pellets), we observed that the signal obtained with the gold pellets is higher. Overall, the high frequency (GHz) SMM measurements and the low frequency (kHz) EFM measurements agree very well in the topographical images as well as in the electrical images. The 3D cal-kit can be properly used for both frequency regimes and it will be used to calibrate the measurements for subsequent application work.

4.1.3.5 Software for Advanced VSMM imaging

The team at Keysight implemented integrated software for advanced VSMM imaging. In particular they developed software in the following four domains:

1. VSMM control software to run novel measurement modes including transmission mode imaging
2. Data analysis software for complex impedance imaging including real time implementation into PicoView.
3. 3D modelling software EMPro was combined with 2D ADS circuitry VSMM models to allow a full VSMM model for data interpretation.

4. 3D reconstruction and superposition capabilities are shown for topography and capacitance overlay and 3D data representation. Using differential dC/dV also spectroscopic tip-bias DC voltage superposition can be done.

Part of the software is running as stand-alone scripts (eg implemented in Matlab, Python, and C) while most of the developed software is already included in the VSMM control software PicoView, in the 3D/2D modelling software EMPro/ADS, and in the 3D post-processing software PicoImage. Most of the software is not released yet as product (only the EMPro VSMM model is officially available). However, for the R&D work the software is available and implemented in the VSMM prototypes together with the corresponding hardware. **The consortium delivered a complete hardware and software prototype VSMM which, for the moment, is only available to the consortium members**. The software is easy to use for beginners but also allows advanced users to have full control of the parameters.

Figure 12 shows the software for S21 transmission mode measurements integrated in PicoView (left panel) and available in the PNA software (right panel). The information from the PNA is included in the AFM software and the most important parameters can be selected within PicoView. The software allows switching between reflection (S11) and transmission (S21) measurements without changing the hardware and cables. The AFM and the PNA communicate properly on all channels.

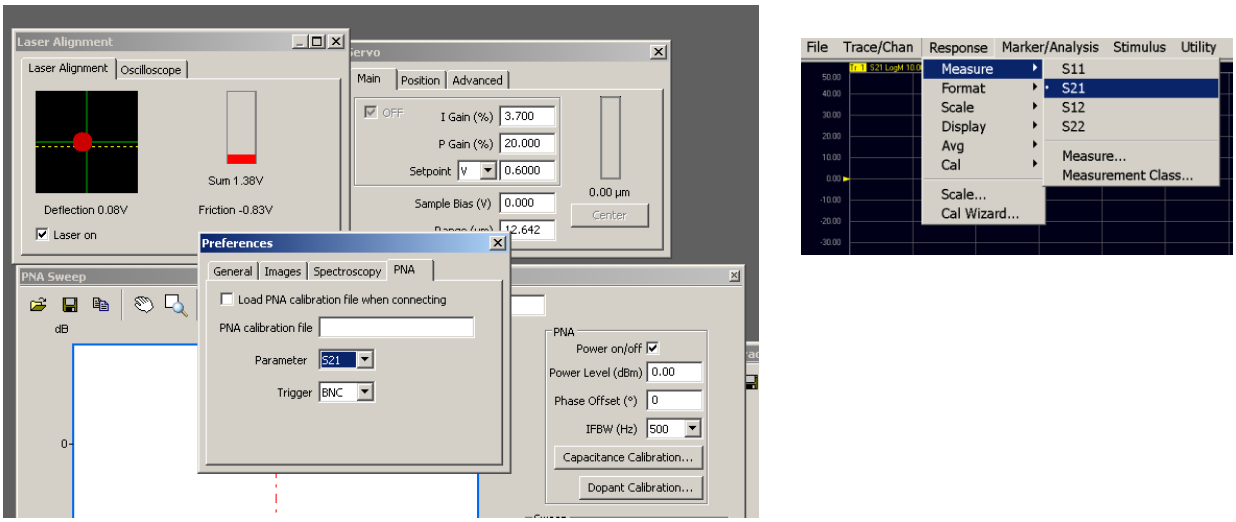

Figure 12: Software S21 to switch between transmission and reflection mode integrated in PicoView (left) and on the PNA (right).

Two different complex impedance calibration workflows were implemented in the software and partly integrated in PicoView. The two workflows are based on the two corresponding scientific publications from Gramse et al (Nanotechnology, 2014) and Hoffmann et al. (IEEE-NANO,2012) Figure (left panel) shows the GUI of the Gramse et al workflow. In particular the dC/dz, conductance and capacitance data channels are shown as well as the calibration parameters and additional SMM parameter information. Figure 13 (right channel) shows the Tanbakuchi et al (IEEE 2013) GUI where the three error-parameters are included to calibrate the conductance and capacitance data. Both software workflows result in calibrated capacitance and conductance images as shown in Figure 14. A detailed manual was established with a step-by-step procedure for the experimental data acquisition and the use of the scripts.

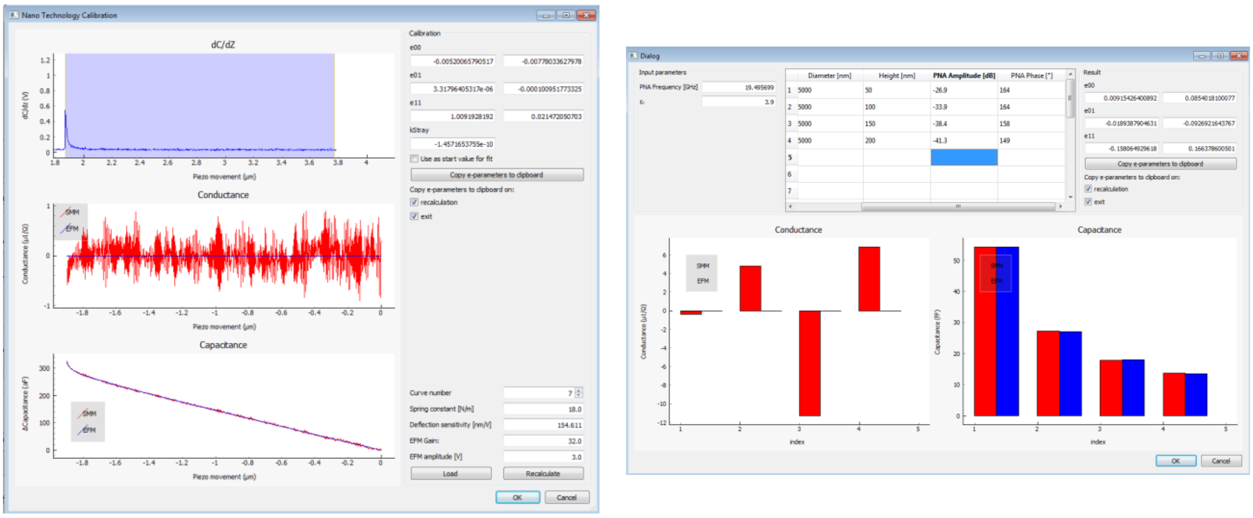

Figure 13. Left panel: GUI for calibrated complex impedance based on the Nanotechnology 2014 workflow. Right panel: GUI for the Tanbakuchi et al (IEEE 2013) calibration workflow.

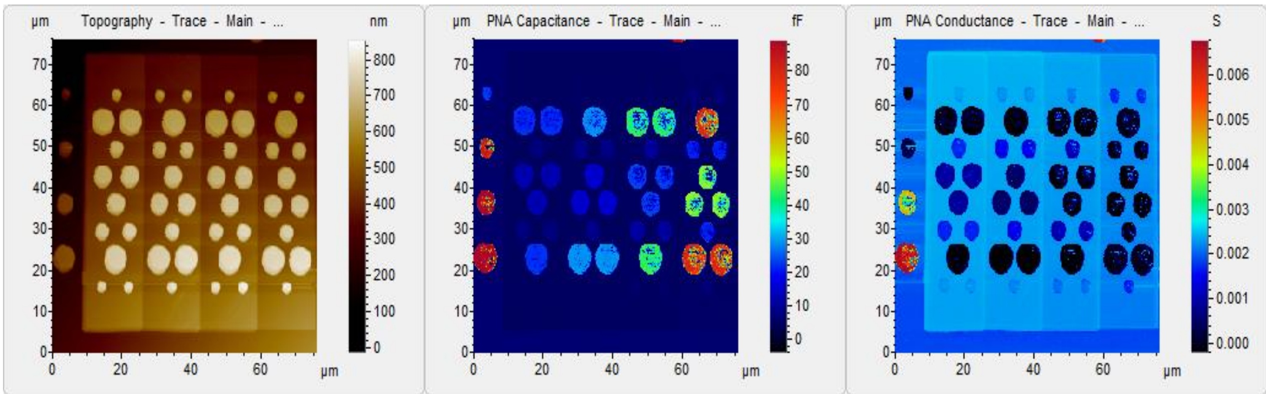

Figure 14. Real-time complex impedance images in PicoView. Topography is shown in the left panel, the capacitance in the center, and the conductance on the right. All z-scales are calibrated in nm, fF, and S, respectively. Shown is the MC2 calibration sample.

The team at Keysight developed a SMM model for the 3D field solver EMPro and showed integration with the 2D circuitry design software ADS. The models were used for the scientific investigation of the experiments and used in several projects in the VSMMART consortium. For the modelling a 3D-field solver (Keysight Electromagnetic Professional software EMPro) and 2D circuitry design software (Keysight Advance Design Software ADS) were used for qualitative interpretation of reflection and transmission mode measurements. The integration capability of ADS and EMPro enables the simulation of the microwave hardware circuit and the electromagnetic interaction of the tip-sample in a single model. Figure 15 shows the combined EMPro/ADS model. A full description of the model and the EMPro/ADS simulation details are given in deliverable 4.5 (entitled 'modelling for data interpretation'). The SMM model embedded in EMPro is commercially available as well as the integration in ADS. This model has been used by several teams already to simulate different SMM tip-sample geometries including nanoparticles and single layer graphene (Figure 16 as done by the Keysight Linz team).

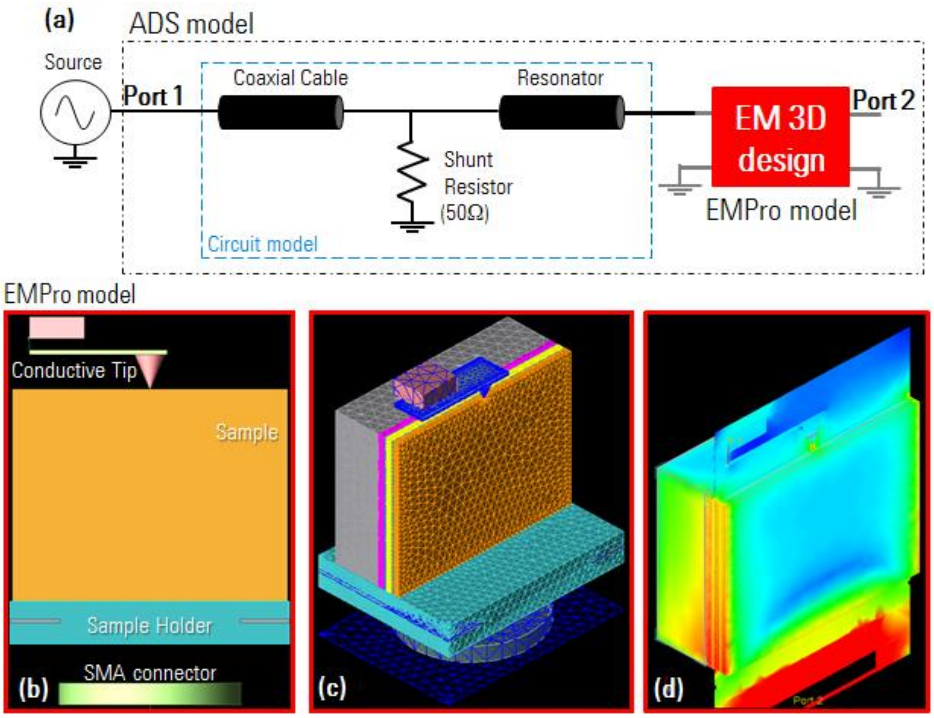

Figure 15: Simulation model of the SMM system for transmission mode and reflection mode. (a) Integrated 2D circuitry model (ADS) and 3D EM design block (EMPro model). (b) 3D CAD model of the cantilever, sample and EM transmitter/SMA connector. (c) Meshed view in EMPro advanced mode. (d) E-Field distribution at 10 GHz on the sample and the cantilever when the tip is contact with a doped silicon layer.

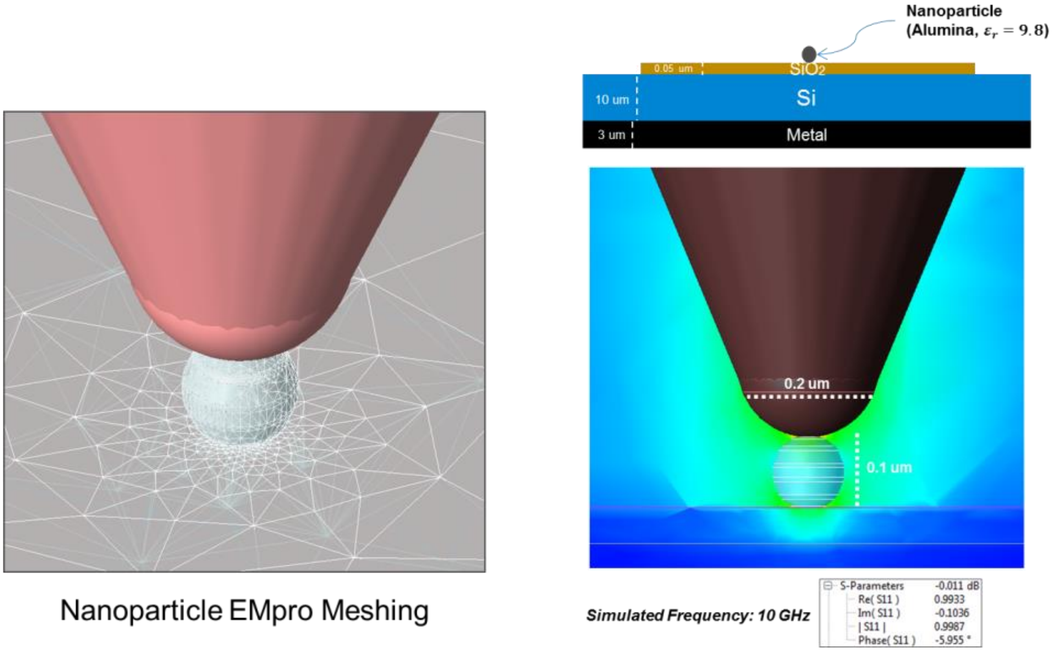

Figure 16. EMPro model applied to single nanoparticle SMM S11 reflection mode studies.

Figure 17 shows how individual images can be stacked and analysed together using the combined PicoImage/PicoView capabilities. Different image channels can be loaded and modified with the history of changes saved to the workflow. 3D overlay functions allow then the superposition of individual images, for instance topography and capacitance as shown in Figure 7. While the topography of the sample is given in 3D the color gives the calibrated capacitance information. The superposition can be done also with the dC/dV image acquired at different tip-bias DC voltages.

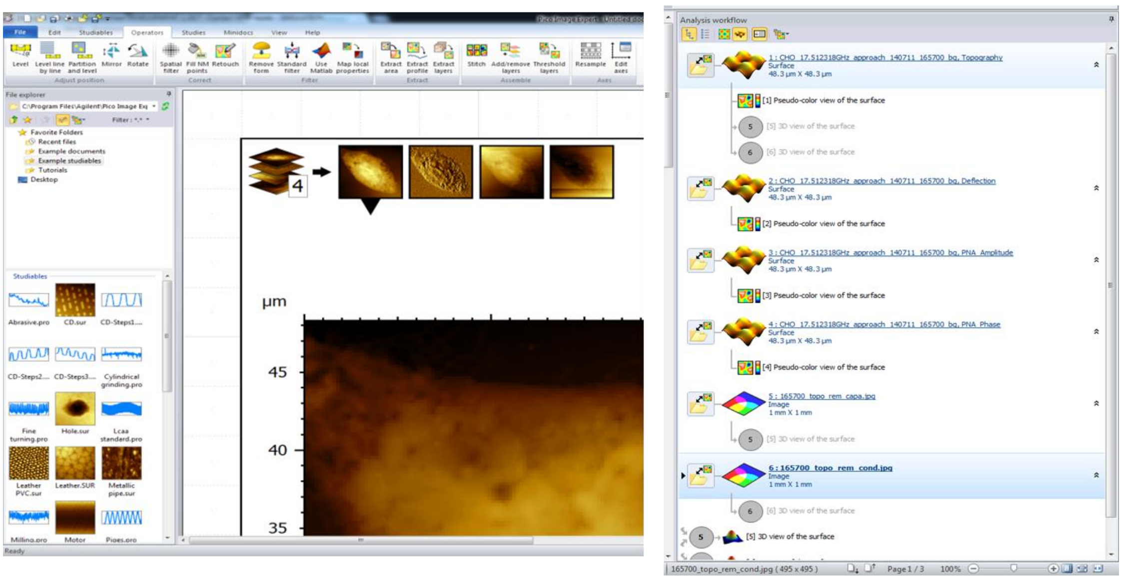

Figure 17. Image analysis and modification procedures. Different image channels can be selected (left) and put together with the history of actions saved in the workflow (right channel).

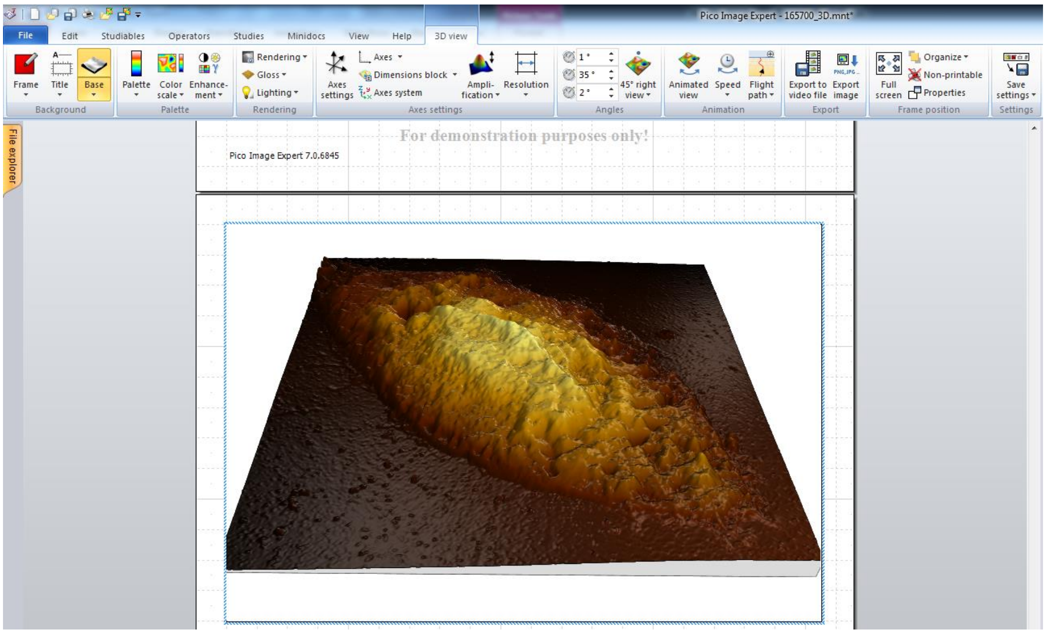

Figure 18. Superposition of topography and capacitance (respectively dC/dV at different bias voltages) of a single cell in air.