Serial block face scanning electron microscopy for three dimensional analysis in materials science

A Zankel[1,2], H Reingruber[1,2], H Schroettner[1,2] and P Poelt[1,2]

1. Institute for Electron Microscopy, Graz University of Technology, Steyrerg. 17, A-8010 Graz, Austria
2. Center for Electron Microscopy, Graz, Steyrerg. 17, A-8010 Graz, Austria

armin.zankel@felmi-zfe.at
Keywords: tomography, ESEM, 3D-reconstruction

Serial block face scanning electron microscopy (SBFSEM) combines the slicing technique of an ultramicrotome and the imaging capabilities of an environmental scanning electron microscope (ESEM). Automated slicing and imaging of the blockface of a specimen can be performed using backscattered electrons (BSE), requiring sufficient compositional contrast of the investigated material. This technique was originally developed for neuronal science and is, up to now, mostly used in life sciences [1]. However, first results in the field of materials science were published in [2] and [3].

The results presented here were carried out using an ESEM Quanta 600 FEG (FEI, Eindhoven, the Netherlands) and the serial block face sectioning and imaging tool, 3View™ of Gatan, Inc. (Pleasanton, CA, USA). For BSE imaging a specially designed detector from Gatan was used, providing enhanced performance at low electron energies. The 3View™ system is controlled by the software Digital Micrograph™.

SBFSEM is operated in the low vacuum mode of the ESEM, enabling the investigation of electrically nonconductive materials without the necessity of a conductive coating. It gives three dimensional information about the structure of materials like polymeric membranes, paper samples, particle filled polymers and interfaces [2,3]. Aside from three dimensional representations of special volumes, software programs like amira® or Avizo® deliver additional data (volume fraction of phases particle filled polymers, porosity and specific surface area of membranes etc.), which are fundamental for further simulations. In the left image of Fig. 1 the surface of a polyethersulfone membrane was recorded in a scanning electron microscope. The plotted rectangle indicates the dimensions of a region of interest which was three dimensionally reconstructed after embedding the specimen into resin and performing automated slicing and imaging with SBFSEM (see right image of Fig. 1).

Additionally to the three dimensional structural information it is often desirable to get information about the chemical composition of various phases. For this purpose energy dispersive x-ray spectroscopy (EDS) can be used [4, 5]. The recording of the respective spectrum maps was carried out with an X-Max® Silicon Drift Detector (Oxford Instruments Analytical Ltd., UK) with an 80 mm² active detection area. With this system the investigation of the distribution of the chemical phases in an aluminum-copper alloy of the type EN AW 2024 T351 (by AMAG, Ranshofen, Austria) was performed. Due to the electrical conductivity of the specimen the high vacuum mode of the microscope could be used [4].

As the parallel operation of the Gatan BSE detector and the X-Max detector is not possible at our system up to now, additionally to the spectrum maps instead of backscattered electron images secondary electron images were recorded (E_0 = 5 keV). A total of 200 slices with a thickness of 100 nm, corresponding to a total volume of (26 × 22 × 20) µm³, were cut. After each cut an image and an spectrum map were recorded (total measurement time: about 22 hours). Subsequently the spectrum maps were quantified with the INCA™ software.

Fig. 2 shows the resulting 3D reconstructions of the distribution of the different phases. The left image of Fig. 2 shows the reconstruction of the major S-phase (Al_2CuMg), the right image further minor phases like $Al_{20}Cu_2Mn_3$ (T-phase, violet), Al_2Cu (blue) Al_7Cu_2Fe (grey), $Al_{12}(Mn,Fe)_3Si$ (green) and Mg_2Si (red). Whereas generally a specimen is mounted in the SBFSEM by gluing it to a rivet, in the case of the investigated aluminium sample a monolytic rod was tailored out of a sample

directly fitting to the specimen stage of the system. So a site specific three dimenionally chemical analysis of the material could be performed.

The great variety of materials already investigated using SBFSEM demonstrates that this method, with a resolution in the µm and even sub-µm range, is complementary to other tomographic methods (e.g. transmission electron microscopical tomography, nuclear magnetic resonance etc.).

References

[1] W. Denk & H. Horstmann PLoS Biol **2** (2004): e329.
[2] A. Zankel, B Kraus, P Poelt, M Schaffer, E Ingolic, J. Microsc. **233** (2009) p140.
[3] H. Reingruber, A Zankel, C Mayrhofer, P Poelt, J. Membr. Sci. **372** (2011) p66.
[4] A. Zankel, R Reingruber and H Schroettner , Imaging and Microscopy **2** (2011) 35.
[5] This work was financially supported by Austrian Cooperative Research (Project No. BMWFJ-98.175/0043-C1/11/2009). The EDS detector was kindly provided by Oxford Instruments GmbH, Wiesbaden, Germany.

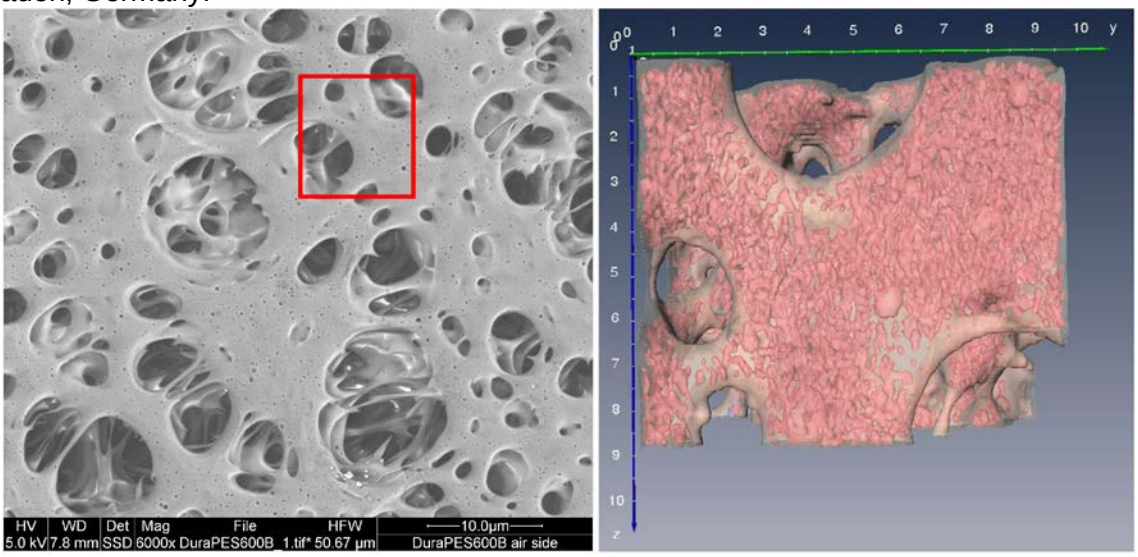

Figure 1. *Left: Image of the surface of a polyethersulfone membrane. The rectangle indicates the dimensions of the region of interest of the 3D reconstruction in the right image. Right: 3D reconstruction of a part of the poplyethersulfone membrane revealing even small isolated pores within the membrane material. Unit: microns*

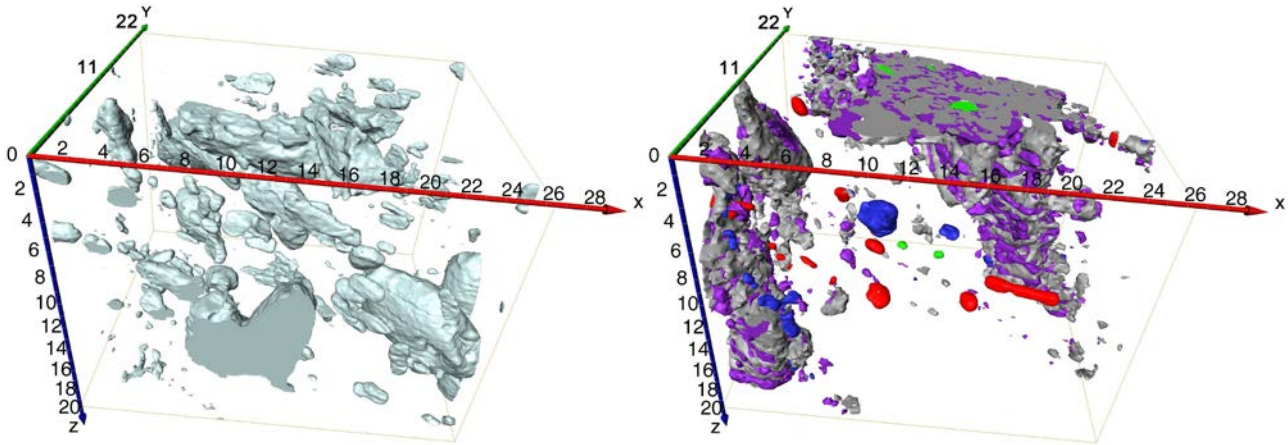

Figure 2. *Aluminium-copper alloy: Left: reconstruction of the major S-phase (Al_2CuMg). Right: reconstruction of the minor phases like $Al_{20}Cu_2Mn_3$ (T-phase, purple), Al_2Cu (blue) Al_7Cu_2Fe (grey), $Al_{12}(Mn,Fe)_3Si$ (green) and Mg_2Si (red). Unit: microns*