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Technical note

Three-dimensional scanning electron microscopy of maxillofacial biomaterials☆

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Recent studies have used conventional scanning electron microscopy (SEM), micro-computed tomography (μCT), and synchrotron-based X-ray tomographic microscopy (SRXTM) to visualise and characterise maxillofacial biomaterials.[1,2] We report a method of 3-dimensional scanning electron microscopy (3D-SEM) to visualise maxillofacial biomaterials.

We used one dental implant (Straumann SLActive®; Straumann, Basel, Switzerland), soft-tissue matrices (Mucoderm®, Botiss, Berlin, Germany; Mucograft®, Geistlich Pharma AG, Wolhusen, Switzerland), and one bony substitute (Maxgraft® bonering, Botiss Biomaterials GmbH, Berlin, Germany) for 3D-SEM imaging.

All samples were mounted on specimen holders with side-adhesive tape and thin copper wires, and were fixed to the holders as recording electrodes to ensure a high image resolution. They were then sputtered with gold in an argon atmosphere. We used a Philips ESEM XL-30 scanning electron microscope (Philips, Eindhoven, Netherlands), and obtained the stereo-pair SEM images by using tilt angles of 6°.[3] These were then digitally overlapped and the resulting 3-dimensional images were examined using bi-coloured (red/green) glasses.

3D-SEM combines the advantages of the high-resolution of SEM, the features of 3-dimensional reconstruction, and the ability to examine μCT and SRXTM scans 3-dimensionally.

Figs. 1–4 show detailed information about the specific characteristics of the maxillofacial biomaterials, such as the surface of the implant (Fig. 1), the typical architecture of the collagen fibre of the soft-tissue matrices (Figs. 2 and 3), and the osseoconductive geometry of the bony substitute (Fig. 4). All four can be viewed using red/green 3-dimensional glasses. 3D-SEM gives a better assessment of the surface, sample proportions, and spatial intersections within the visualised samples than conventional SEM.

The 3-dimensional effect of 3D-SEM images depends on two factors: the structure and geometry of the samples, and the angle between the electron beam and the surface. Samples with a 3-dimensional architecture (for example, spheres and cubes), and large differences in the levels of the surface (in the form of rises and grooves on implants or bony substitutes), allow more of a 3-dimensional effect than ones that are flat with a smooth surface (such as discs) (Figs. 1 and 4). This effect can be guided by the angle between the electron beam and the surface of the sample, when angles greater than, or

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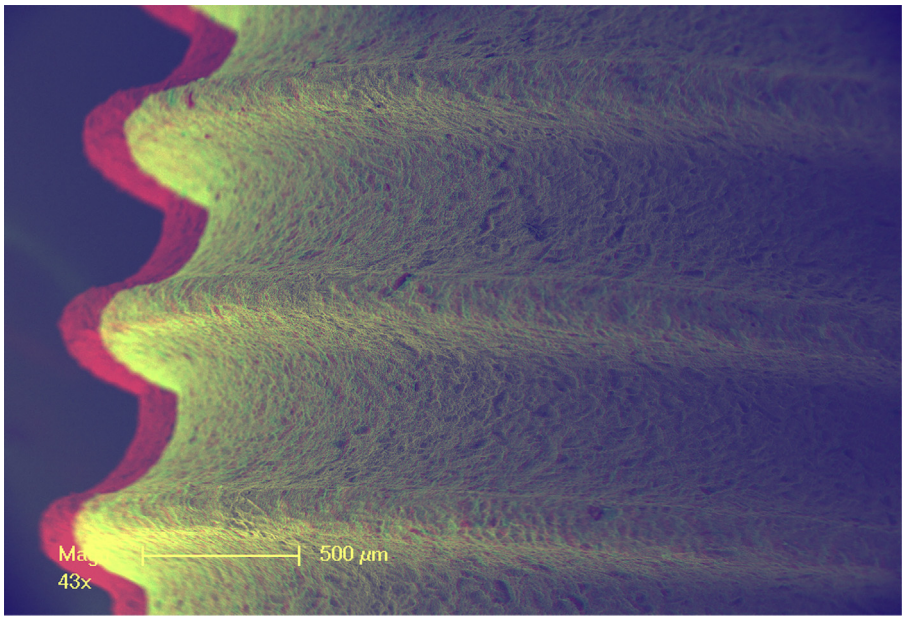

Fig. 1. Three-dimensional scanning electron microscopy visualisation of the surface of an implant (SLActive®, Straumann) shows its characteristic structure and roughness. The visualisation allows an in-depth 3-dimensional analysis, because the implant has a pronounced 3-dimensional architecture, specifically at the thread of the screw.

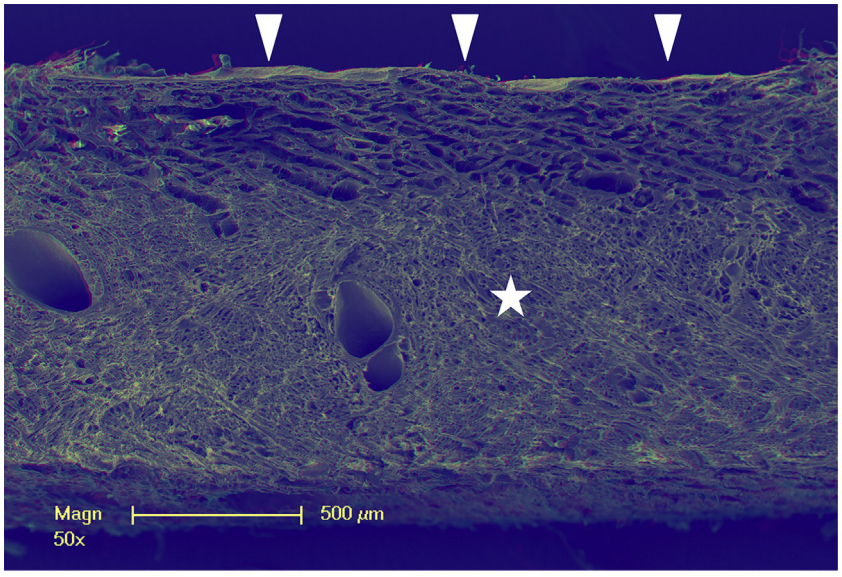

Fig. 2. Three-dimensional scanning electron microscopy visualisation of the collagen matrix (Mucoderm®, Botiss) shows the mono-layered design of the scaffold with the surface layer on the top side of the matrix (white arrows) and the spongious internal structure with a parallel arrangement of densely-packed collagen bundles (white asterisks). The 3-dimensional effect of the image was relatively small because the visualised cross-section of the sample was rectangular and aligned to the electron beam.

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