



## **Microbial structures in an Alpine Thermal Spring – Microscopic techniques for the examination of Biofilms in a Subsurface Environment**

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The research into extreme environments has important implications for biology and other sciences. Many of the organisms found there provide insights into the history of Earth. Life exists in all niches where water is present in liquid form. Isolated environments such as caves and other subsurface locations are of interest for geomicrobiological studies. And because of their “extra-terrestrial” conditions such as darkness and mostly extreme physicochemical state they are also of astrobiological interest.

The slightly radioactive thermal spring at Bad Gastein (Austria) was therefore examined for the occurrence of subsurface microbial communities. The surfaces of the submerged rocks in this warm spring were overgrown by microbial mats. Scanning electron microscopy (SEM) performed by the late Dr. Wolfgang Heinen revealed an interesting morphological diversity in biofilms found in this environment (1, 2). Molecular analysis of the community structure of the radioactive subsurface thermal spring was performed by Weidler et al. (3).

The growth of these mats was simulated using sterile glass slides which were exposed to the water stream of the spring. Those mats were analysed microscopically. Staining, using fluorescent dyes such as 4',6-Diamidino-2-phenylindol (DAPI), gave an overview of the microbial diversity of these biofilms. Additional SEM samples were prepared using different fixation protocols. Scanning confocal laser microscopy (SCLM) allowed a three dimensional view of the analysed biofilms.

This work presents some electron micrographs of Dr. Heinen and additionally new microscopic studies of the biofilms formed on the glass slides. The appearances of the new SEM micrographs were compared to those of Dr. Heinen that were done several years ago. The morphology and small-scale distribution in the microbial mat was analyzed by fluorescence microscopy. The examination of natural biomats and biofilms grown on glass slides using several microscopical techniques suggest that the thermal springs in the Central Alps near Bad Gastein represent a novel and unique habitat for microbial life. Results obtained during these studies revealed reproducibility of Dr. Heinen's micrographs. Hollow reticulated filaments and flat ribbons with parallel hexagonal chambers (web-structures) were found repeatedly. Given the chance that subsurface environments represent a potent opportunity to detect life on planetary bodies it is of big interest to search for representative biosignatures found on earth today.

### References:

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2. Heinen W. & Lauwers A. M. (1985) *Mikroskopie (Wien)* 42, 124-134.
3. Weidler G. W., Dornmayr-Pfaffenhuemer M., Gerbl F. W., Heinen W., Stan-Lotter H. (2007) *AEM* 73, 259-270.