New approaches to unveil biodeterioration processes applied to petroglyphs sites in Israel and Austria

Laura RABBACHIN, Institute of Natural Sciences and Technology in the Arts (INTK), Academy of Fine Arts Vienna, Austria
Guadalupe PIÑAR, Institute of Natural Sciences and Technology in the Arts (INTK), Academy of Fine Arts Vienna, Austria
Irit NIR, Department of Biotechnology Engineering, Ben-Gurion University of the Negev, Be'er Sheva, Israel
Ariel KUSHMARO, Department of Biotechnology Engineering, Ben-Gurion University of the Negev, Be'er Sheva, Israel
Mariela J. PAVAN, Ilse Katz Institute for Nanoscale Science and Technology, Ben-Gurion University of the Negev, Beer Sheva, Israel
Elisabeth EITENBERGER, Institute of Chemical Technology and Analytics, TU Wien, Austria
Monika WALDHERR, Applied Life Sciences/Bioengineering/Bioinformatics, FH Campus, Vienna, Austria
Alexandra GRAF, Applied Life Sciences/Bioengineering/Bioinformatics, FH Campus, Vienna, Austria
Katja STERFLINGER, Institute of Natural Sciences and Technology in the Arts (INTK), Academy of Fine Arts Vienna, Austria

Keywords: *petroglyphs – extreme environments – biodeterioration – metagenomics – analytical techniques*

CHNT Reference: Rabbachin, L., *et al* (2022). 'New approaches to unveil biodeterioration processes applied to petroglyphs sites in Israel and Austria', in CHNT Editorial board. *Proceedings of the 27th International Conference on Cultural Heritage and New Technologies, November 2022.* Heidelberg: Propylaeum.

Introduction and aims of the project

Rock art, in the form of petroglyphs and pictograms, is found worldwide and has undoubtedly an immense value, as it is considered one of the first forms of expression of ancient societies, and the prehistoric precursor to art. Due to their outstanding universal value, a number of rock art sites were added to the World Heritage List (WHL) established by UNESCO (https://whc.unesco.org/en/list/).
As part of the natural landscape, petroglyphs are constantly exposed to anthropogenic and natural weathering processes, but despite this, the knowledge regarding the preservation and conservation practices related to this valuable cultural heritage is very limited. In order to protect and preserve them, a better knowledge of the degradation patterns is needed with special attention to biodeterioration. Indeed, the research focusing on the role of biological agents in the deterioration processes specific to rock art is still minimal. Nevertheless, physical and chemical weathering processes initiated by stone-dwelling microorganism (biodeterioration) can play a significant role in the degradation patterns of stone (Gorbushina and Krumbein, 2005). Biodeterioration includes surface alterations by crusts and pigments that can change the colour of the surface, physico-chemical disintegration of the stone material as well as the dissolution of the lithic substrate by inorganic and organic acids (Steiger, Charola and Sterflinger, 2014). Therefore, the aims of the project are to characterize the microbial communities, understand the biochemical interaction between microbes and rock and the

mechanisms of biodeterioration and to learn about the environmental factors that determine the microbial communities.

In a joint project of the Academy of Fine Arts Vienna (Austria) and the Ben Gurion University (Israel) two different petroglyph sites are considered: one in the Negev desert of Israel (Fig. 1a) and one in the East Alpine region (Dachstein area) of Austria (Fig. 1b), two very different environments, but sharing extreme conditions.

a b

Fig. 1. A petroglyph site in the Negev desert, Israel (a) and the Notgasse petroglyph site in the Dachstein region, Austria.

In this contribution, some preliminary results obtained from samples collected in the Negev desert are presented. In the Negev desert, most of the petroglyph sites are found in the west/central highlands, which is considered climatically as an arid area. These petroglyphs are carved on white limestone rocks covered by a dark crust, the so-called desert varnish, and include figurative images, geometric shapes, symbols, and inscriptions, reflecting the life of desert people throughout history (Nir *et al.*, 2019). They are considered to be from 3000 BCE, or older.

The sampling was carried out during winter (February and March) 2021. The stone samples were collected in proximity of the petroglyphs from similar rock types (limestone covered with desert varnish) using a hammer and chisel previously sterilized with 70% ethanol.

Interdisciplinary approach

To achieve the goals of this study, an interdisciplinary approach was used combining microscopy and physico-chemical techniques (X-Ray Fluorescence, scanning electron microscopy coupled with EDX microanalysis (SEM-EDX), 3D microscopy, Raman spectroscopy) as well as culture-independent microbiological methods (metagenomics).

The metagenomic analysis were performed with the Oxford Nanopore technology (MinION platform) following a Whole Genome Amplification (WGA) protocol, which allows to obtain simultaneously the real proportions of different groups of microorganisms (fungi, bacteria and archaea) as a whole in a given sample. The results showed that the microbiomes of the samples are dominated by bacteria, revealing a high biodiversity dominated by Actinobacteria and Cyanobacteria. These microorganisms are believed to be closely connected with the formation of the dark crust, and, on the other hand, might contribute to the rock deterioration.

Through physico-chemical techniques such as XRF and Raman spectroscopy the composition of the stone and the dark crust was studied, revealing a bedrock composed of silicified limestone (Si and Ca) and a dark crust composed of clay minerals, iron (Fe) and (Mn) oxides, as previously reported in literature (Goldsmith, Stein and Enzel, 2014).

With microscopy, applied both on raw sample material and on petrographic thin-sections of the samples, the morphology of the samples was studied. The observation of the petrographic thin sections unveiled from moderately to heavily weathered areas underneath the black varnish (Fig.2 a and b). In these areas, grains of the bedrock have been leached, leaving behind a very porous zone. In some samples, biological colonization (cyanobacterial aggregates) was clearly visible. In the areas colonized by cyanobacteria the black varnish appears completely fragmented and detached from the bedrock, or is no longer present (Fig. 2c). Interestingly, in the weathered areas present underneath the black crust, the elemental mappings performed with SEM-EDX revealed that the calcium matrix of the limestone is completely missing, and only the silicate component is left, leaving a very porous zone. This phenomenon was attributed to the ability of some microorganisms detected through the metagenomics analysis to dissolve the carbonates in limestone rocks to reach optimal living conditions inside the stone such as temperature, humidity and nutrient supply (Krumbein and Jens, 1981).

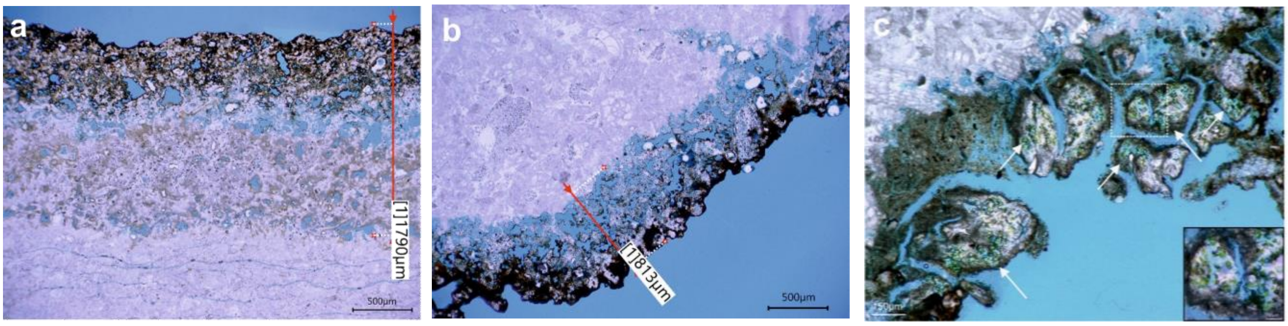

Fig. 2. Microscope images of the petrographic thin sections showing heavily weathered areas underneath the black crust (a and b) and an area colonized by cyanobacteria aggregates where the black crust is detaching from the bedrock (c).

Conclusions

The study pointed out that rocks harbouring petroglyphs in the Negev desert are colonized by a complex microbial community and patterns of deterioration in the bedrock and in the black crust were unveiled, feasibly connected to microbial action. Hitherto, it was never discussed if any measures – as e.g. the protection by a canopy or regular cleaning - could prevent or at least slow down the process of biodeterioration in valuable petroglyph sites. However, before being able to give recommendations, it is essential to understand the mechanisms of biodeterioration. The interdisciplinary approach used in this study allowed a better understating of the mineral-microbial interaction, thus a first step towards a better knowledge of the biodeterioration processes of petroglyph sites, which might be useful for a more effective conservation of this valuable cultural heritage.

Funding

This research was funded by the Austrian Science Fund (FWF – Der Wissenschaftsfonds) project I 4748-B “Stone cultural heritage in Austria and Israel” and by the Ministry of Science and Technology (MOST), Israel.

Conflict of Interests Disclosure

The authors declare no conflict of interest.

Author Contributions

Conceptualization: K.S., G. P., I.N., A.K.
Data curation: L.R., G.P., K.S.
Funding acquisition: K.S., A.K.
Investigation: L.R., G.P., I.N., E. E., M.J.P.
Methodology: L.R., G.P., I.N., K.S., A.K.
Project Administration: K.S., A.K.
Software: M.W., A.G.
Supervision: K.S., A.K.
Writing – original draft: L.R., K.S., G.P.
Writing – review & editing: L.R, G.P., K.S., I.N., A.K.

References

Goldsmith, Y., Stein, M. and Enzel, Y. (2014) ‘From dust to varnish: Geochemical constraints on rock varnish formation in the Negev Desert, Israel’, *Geochimica et Cosmochimica Acta*, 126, pp. 97–111. doi: 10.1016/j.gca.2013.10.040.

Gorbushina, A. A. and Krumbein, W. E. (2005) ‘Role of Microorganisms in Wear Down of Rocks and Minerals’, *Microorganisms in Soils: Roles in Genesis and Functions*, 3, pp. 59–84. doi: 10.1007/3-540-26609-7_3.

Krumbein, W. E. and Jens, K. (1981) ‘Biogenic rock varnishes of the negev desert (Israel) an ecological study of iron and manganese transformation by cyanobacteria and fungi’, *Oecologia*, 50(1), pp. 25–38. doi: 10.1007/BF00378791.

Nir, I. *et al.* (2019) ‘Seasonal diversity of the bacterial communities associated with petroglyphs sites from the Negev Desert, Israel’, *Annals of Microbiology*, 69(10), pp. 1079–1086. doi: 10.1007/s13213-019-01509-z.

Steiger, M., Charola, E. and Sterflinger, K. (2014) ‘Weathering and Deterioration’, in *Stone in Architecture: Properties, Durability: Fifth Edition*. Springer, pp. 225–316. doi: 10.1007/978-3-642-45155-3.