VUV LASER CLEANING OF FUNGUS and LICHENS FROM HELLENIC ARCHAEOLOGICAL STONES

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Abstract

Cease of biological activity of species, which are growing on various Hellenic archeological stones was observed following laser illumination of the specimens at 157 nm.

Laser light dissociates the external multilayered proteinacious membrane of the spores reducing their thickness to a critical value prior to cell explosion due to the high internal pressure of the nucleus.

High resolution AFM imaging reveals that the population of a monolayer culture was successfully destroyed following illumination with fluence from 1-200mJ/cm^2 pp.

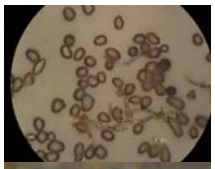
The use of 157nm laser is an effective and controllable method for stopping biological activity of *Fungus and Lichens* from archaeological stones.

Experimental

Fungus were collected from mycelia cultures grown in agar, the aggregation containing 1.2x10^5 spores/ml with 20 % /hour rate. They were placed after 10-12 hours on silica wafer and sticky tape substrates,

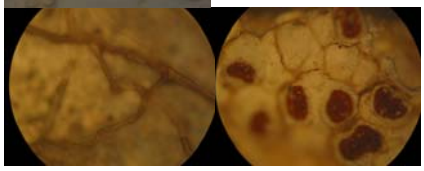
Lichens were collected from Hellenic archaeological stones in Lucius of Peloponnesus.

The species were de-hydrated and then they were illuminated with a number of laser pulses of known fluence at 157 nm.



Monolayer aggregation of ulocladium sp spores grown on Si substrate. The average length of the spores was 10 μm.

Aspergillus sp.4-1 fungi . Conidiophore with vesicle and chains of spores.



Branched hyphae with a thin sheath of polysschrides

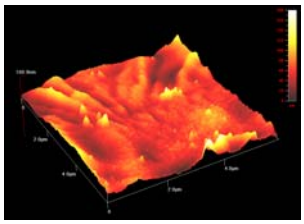
General view of Lichens layer collected from archaeological stones

The experimental set up consists of the laser apparatus at 157nm which is the VUV exposure tool, the focusing optics and the high precision X-Y-Z-Θ micromachining stage where the samples were placed.

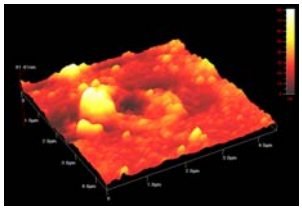
Results and Discussion

Ulocladium

Prior to irradiation



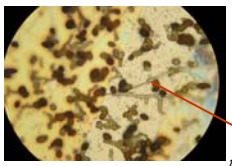
AFM image of ulocladium sp spore.
- *Large conic holes 200-500 nm wide on the top of the surface which become narrower towards the centre.*
- *Nubs 100 nm long.*
- *Regularly spaced nods or rod let patterns 10-20 nm long.*



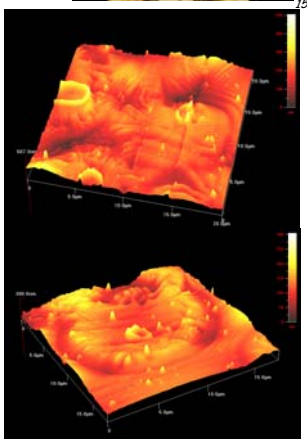
AFM image of a hole on the surface of ulocladium sp.
- *Around the hole there are spaced nods and rodlets.*
- *The spore wall consisted of two zones and the holes are discontinuities which connect the two layers*

After irradiation

❖The external membrane of the cell exploded following sample's illumination with 1mJ/cm^2.
❖It is estimated that the cell was exploded when an average layer of 45 nm was removed from the membrane.



Exposed at 157nm 1mJ/cm^2 per pulse

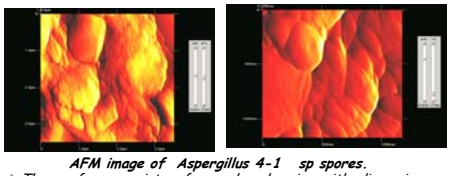


AFM image of one spore consisted of two cells following illumination at 157 nm.
- *The spore was exploded after illumination indicating that the nucleus material is under high pressure.*

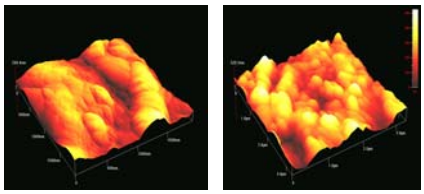
AFM image of two connected spores.
- *The spores seem to be empty from the nucleus material.*

Aspergillus

Prior to irradiation

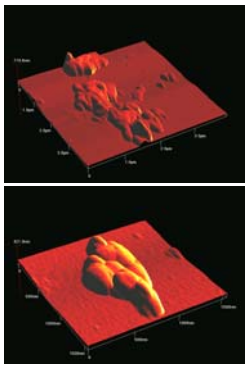


AFM image of Aspergillus 4-1 sp spores.
- *The surface consists of granular domains with dimensions 100-200 nm.*
- *Higher resolution images (phase mode) reveals the presence of rodlet-like structures in the surface of the granular domains. The rodlets are approximately 20nm wide and a few hundred nm long, (left image)*



- *The surface of the Aspergillus 4-1 sp*
- *Part of the conidiophore can be seen on the righ (left image)*

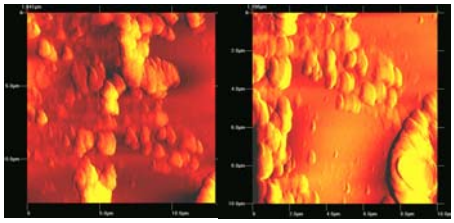
After irradiation



- *AFM images of the exposed Aspergillus 4-1 spores at 157nm.*
- *Parts of spores with dimensions approximately 100-200nm, spread in the area of destroyed spores can be seen.*
- *A destroyed spore can be seen and a part of about 250nm thick has been removed from the center of the spore.*

Lichens

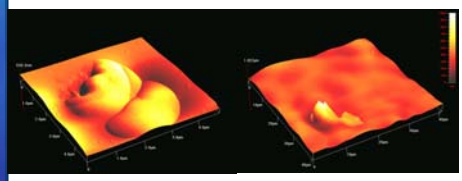
Prior to irradiation



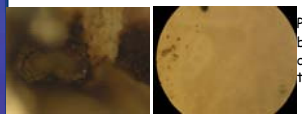
AFM images of the non exposed lichen .

Long hyphae with crossed walls

After irradiation



AFM images of the exposed lichen at 157nm.
Parts of destroyed biological species with dimensions approximately 100-200nm, spread in the area.



Parts of destroyed biological species at different irradiation time