



PREVENTING BIOLOGICAL ACTIVITY OF *ULOCLADIUM SP* SPORES IN ARTIFACTS USING 157nm LASER

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Abstract

High resolution AFM images of immobilized *ulocladium sp* cultures on silicon wafers reveal cease of biological activity after laser illumination at 157 nm.

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

The population of a monolayer culture was successfully destroyed following illumination with 150 laser pulses at the fluence of 1mJ/cm² per pulse. A thin layer of 0.3 nm was removed on the average from the external membrane per pulse, and a thin layer of 45 nm had to be removed from the external membrane before cell explosion.

Thus the use of 157 nm laser is an effective and controllable method for stopping biological activity of *ulocladium sp* spores in artifacts.

Experimental

Ulocladium sp spores were collected from mycelia cultures grown in agar, the aggregation containing 1.2x10⁵ spores/ml with 20 % /hour rate. They were placed after 10-12 hours on silica wafer and sticky tape substrates,



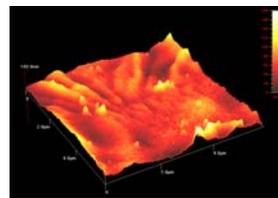
Monolayer aggregation of *ulocladium sp* spores grown on Si substrate. The average length of the spores was 10 μm.

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



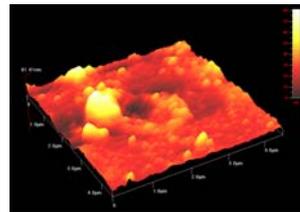
Results

Before 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 nodes or rod let patterns 10-20 nm long.



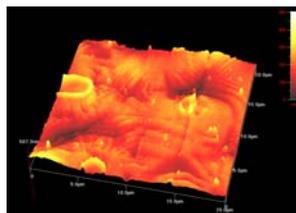
- AFM image of a hole on the surface of *ulocladium sp*.
- Around the hole there are spaced nodes and rodlets.
- The spore wall consisted of two zones and the holes are discontinuities which connect the two layers

After irradiation

- ❖ The external membrane of the cell exploded following sample's illumination with 150 pulses at the fluence of 1 mJ/cm².
- ❖ It is estimated that the cell was exploded when an average layer of 45 nm was removed from the membrane.

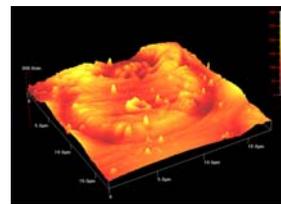


Exposed at 157nm 1mJ/cm² per pulse



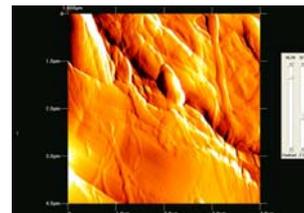
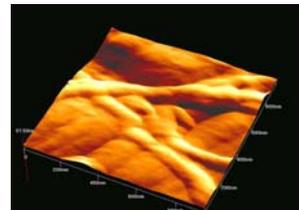
AFM image of one spore consisted of two cells following illumination at 157 nm.

- The spore was exploded after illumination indicating that the nucleus material is under high pressure.



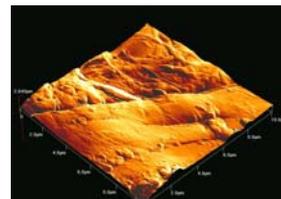
AFM image of two connected spores. They seem to be empty from the nucleus material.

There are strong arguments to believe that almost all foxing in paper is produced by the proliferation of various forms of fungi, provided that the metabolic requirements of fungi are fulfilled.



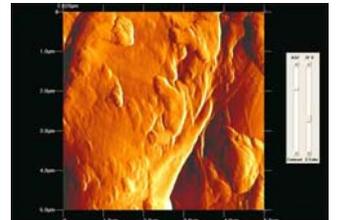
The AFM image of non infected part of historic paper is shown in The integrity of the cellulose fibres is retained.

The boundary between infected and non infected areas is clearly seen. The integrity of the cellulose fibres in the infected areas is not any more retained and the paper fibres appear brittle

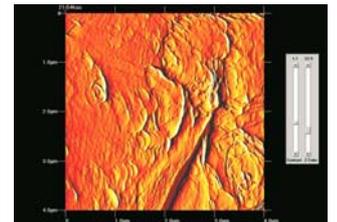


AFM image of infected by foxing and non- infected areas are shown.

The remains of the biological activity, possibly secretion of mucilage, have dimensions 0.5-1 μm



AFM image of infected by foxing areas is shown



AFM face mode image of infected by foxing areas

Conclusion

- ✓ Laser light dissociates the external multi layered proteinaceous membrane of the spores.
- ✓ The high internal pressure of the nucleus explodes them.
- ✓ Thin layers of 0.3 nm thick were removed from the external protective membrane when it was illuminated with 1mJ/cm² per pulse.
- ✓ The spores were exploded when 45 nm were removed from the external membrane.
- ✓ The use of 157 nm lasers is an effective and controllable method for stopping biological activity of *ulocladium sp* spores in artefacts.
- ✓ Rodlets are involved in attachment of spores on surfaces of the artefacts as a step of biological colonization and in spores aggregation.
- ✓ Silicon wafers used in micro lithography are possibly the best substrate to grow *ulocladium sp* cultures.

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