Atmospheric Plasma Jet for Spacecraft Surface Sterilization for Planetary Protection

J.R. Prager (EHT)*; T. M. Ziemba (EHT); J. A. Miller (EdCC);
*prager@eagleharbortech.com

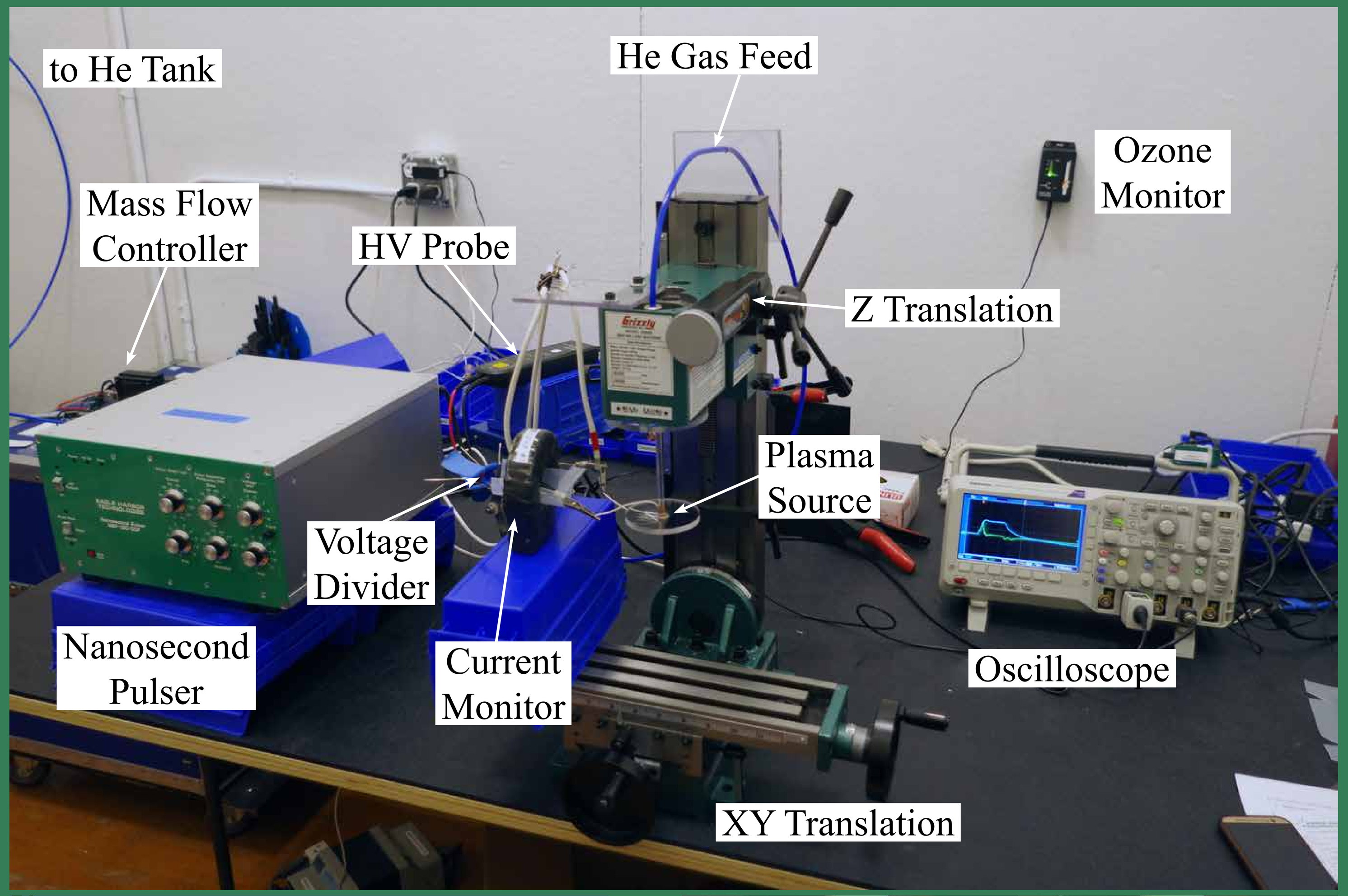
EAGLE HARBOR TECHNOLOGIES

Introduction

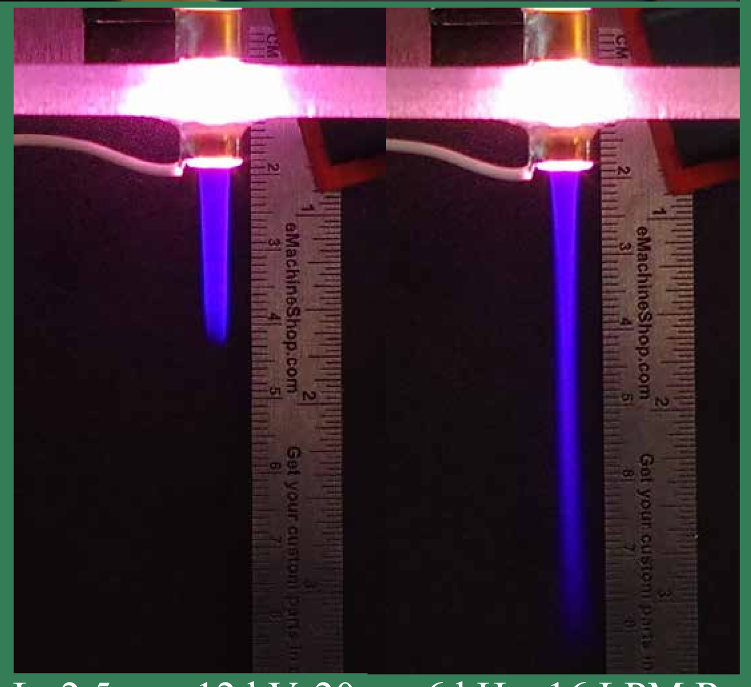
New technology is required for Contamination Control for Planetary Protection as the planetary science community explores solar system bodies with the potential for detecting life. Currently, dry heat microbial reduction (DHMR) and vapor hydrogen peroxide (VHP) are the only microbial reduction methods approved for planetary protection. DHMR is extremely time consuming and chamber time is difficult to schedule, while the use of VHP is not usable for some state-of-the-art electronics and materials.

Eagle Harbor Technologies, Inc. (EHT) has developed an atmospheric pressure plasma jet (APPJ) array that utilizes the EHT Nanosecond Pulser to kill endospores on surfaces. A microbiology lab at Edmonds Community College conducted a wide parameter sweep to determine what pulse and gas parameters are most effective. During the Phase I, they demonstrated a six-log reduction of endospores in petri dishes and provided preliminary evidence of real-world sterilization. Additionally, an FPGA and SDRAM continued to function after plasma exposures showing that this treatment will not impact sensitive electronics.

Experimental Setup



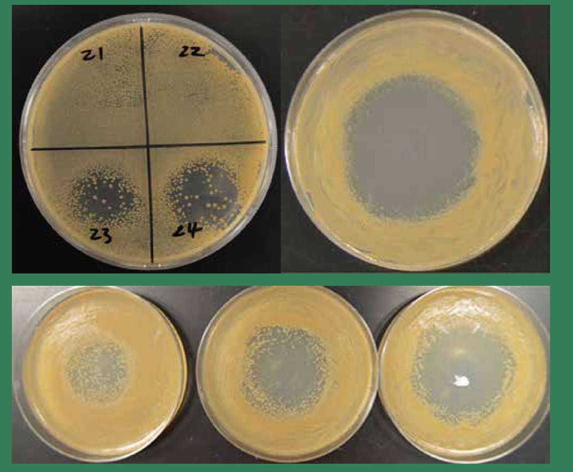
Top: Current (green) and voltage (blue) waveforms. Bottom: Energy per pulse.



L: 2.5 cm, 12 kV, 20 ns, 6 kHz, 16 LPM R: 6.2 cm, 12 kV, 220 ns, 4 kHz, and 4 LPM

Determination of *Bacillus atrophaeus* Sterilization Parameters

EHT and EdCC conducted a broad parameter survey, quickly looking at over 900 combinations. Table below highlights some of the best parameters.



Dist. (cm)	He (slpm)	PW (ns)	PRF (kHz)	Voltage (kV)	Clearing Zone (cm)	% killing
1	4	50	4	20	2.3	99.7
2	8	350	7	10	3.1	99.6
1	4	50	7	20	1	95.5
1	4	50	10	15	1	96.0
1	8	50	7	20	1.7	98.6
1	8	50	10	15	1	95.0
1	8	250	4	15	1	95.0
1	8	550	1	20	0.9	95.1
1	8	50	4	20	1.3	97.9

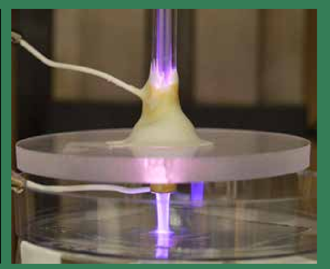
Top: Killing effect of first condition for different exposure times and endospore concentration (20 s and 10 min). Bottom: Killing efficacy of second condition for (left to right) 2, 5, 10 min with 0.1 ml of stock concentration of 2.9 x 10^7endospores/ml.

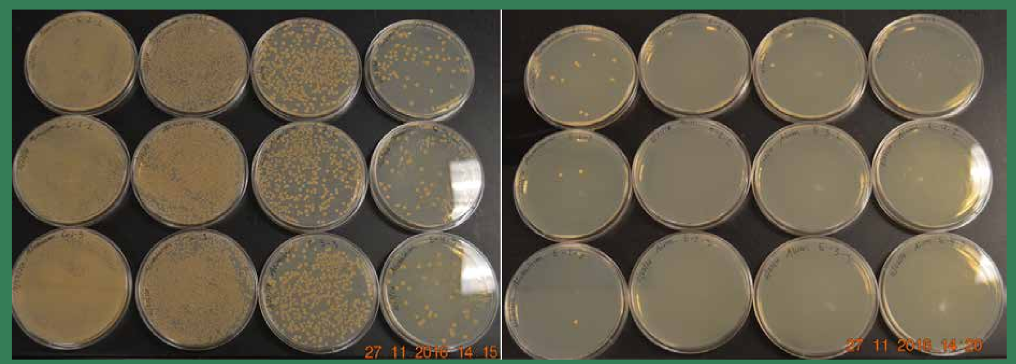
D-Value Determination on Al Surface with *Bacillus atrophaeus*

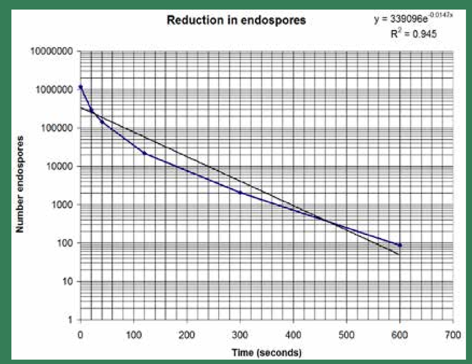
0.1 ml drop of stock *B. atrophaeus* endospore solution containing 2.9 x 10^7 endospores/ml was placed onto Al tape.

Exposed to the APPJ (4 slpm, 50 ns, 4 kHz, 20 kV) for 20 - 600 s. Untreated Al tape with endospores served as a control.

Recovery by dilution series, and calculate killing percentage.



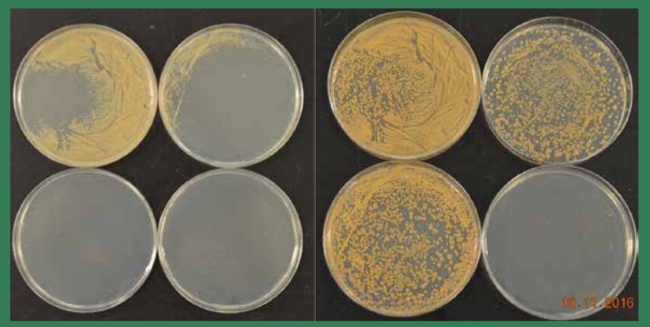




Left: Results of dilution series for control plates (left) and 10-min exposure (right). Columns represent 0.5 ml plated from dilutions of 1:10, 1:100, 1:1,000, and 1:10,000 (left to right). Rows represent triplicate plating of dilution series onto TSA. Plates were incubated for 24 hrs at 35 °C, with no more growth observed after 48-hour incubation.

Right: Data yields an average D value of 160 s under these conditions on an Al surface. D values are typically considered to be linear, the killing pattern here was faster initially, with a D value of <60 seconds for the first log reduction, ~120 seconds for the second log reduction, and closer to 180 seconds for the third and fourth log reductions.

Large-Area with Single Jet



Jet passed over surface of Petri dishes containing 2.9 x 10^7 endospores/ml for 1, 2, (top row), 5, and 10 min (bottom row). Results following 24-hr (left) and 48-hr (right) incubation at 35 °C. Only three colonies formed on the 10-min exposure, meaning jet achieved a 6 log reduction in endospores.

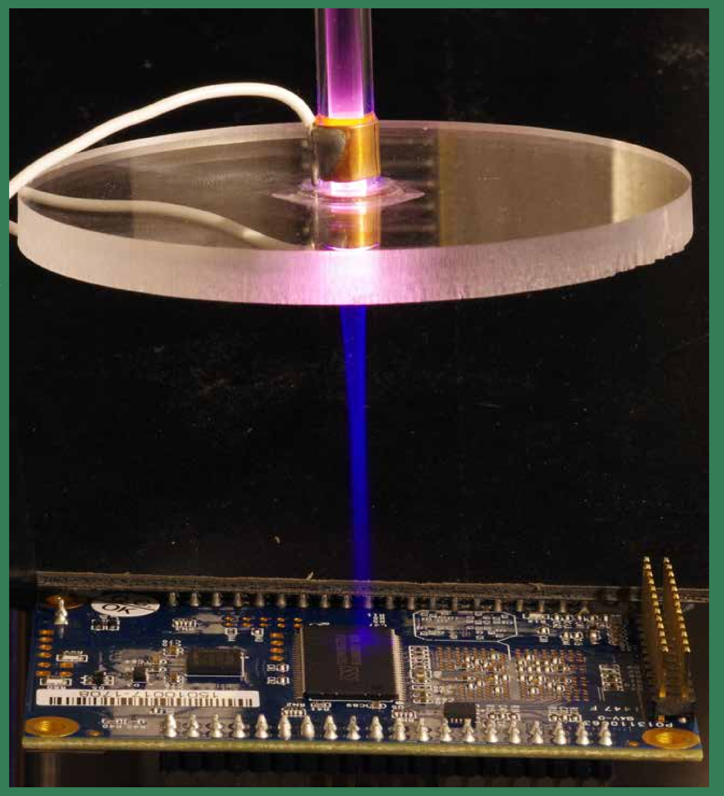
Humidity and O_2 Effect

Treatment	# of Endo-spores (CFU)	% Relative Reduction
0. Untreated Control	1.34 x 10^6	0%
1. He	3.29 x 10^5	75.448%
2. He + 0.5% O_2	4.79 x 10^4	96.425%
3. He + 0.5% O_2 + 0.2 ml dH_20	4.27 x 10^2	99.968%

Conditions were 10 minutes, 1.5 cm, 4 slpm, 50 ns, 4 kHz, 20 kV ring-ring. Treatment 3 yielded greater than 3.5 log reduction in endospores.

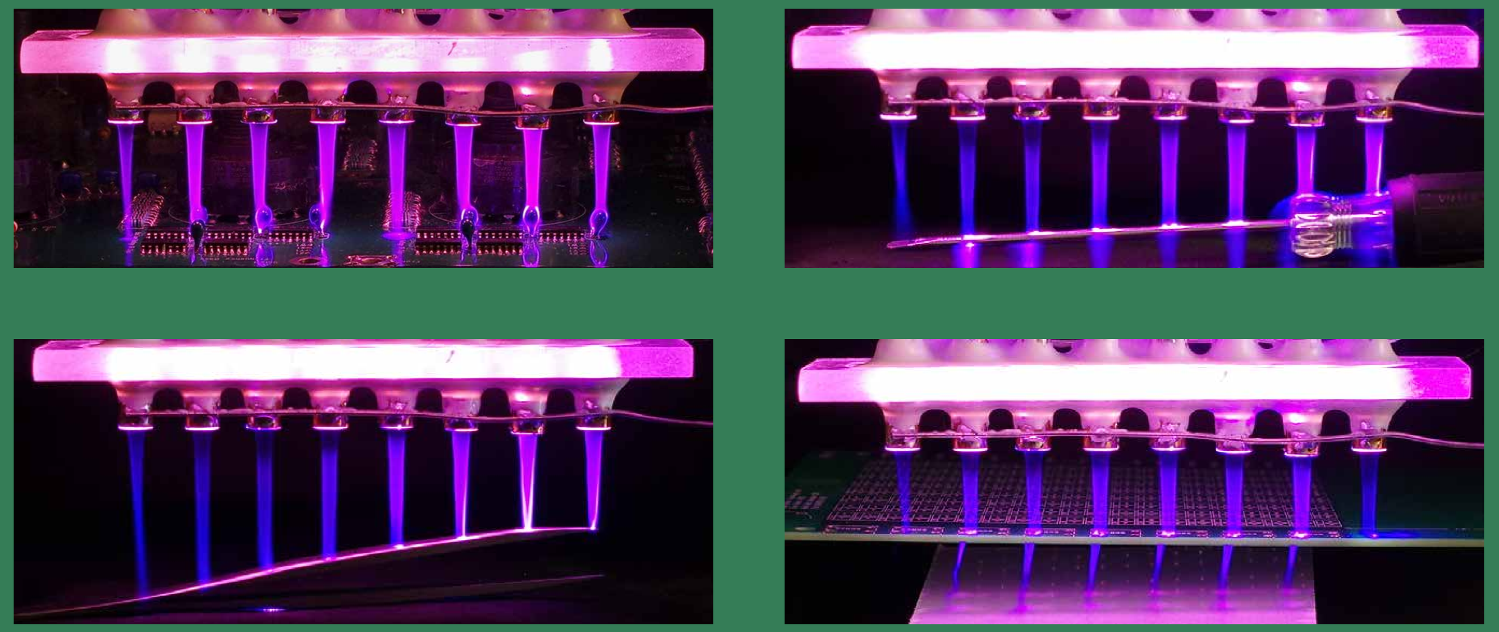
Plasma-Material Interaction

Al, steel, polyimide, and Teflon were exposed to plasma for several minutes with no signs of material damage.



Sensitive electronics, including Altera Cyclone IV (EP4CE22F17C6N) FPGA, 2Kb I2C EEPROM, FPGA Serial Configuration Device (EPCS), and 32MB SDRAM (ISSI IS42S16160G),, were exposed to APPJ. These components continued to function after plasma exposure.

Plasma jet array was used to treat a variety of surfaces to qualitatively look at jet-to-jet interaction.



Conclusion

Using an EHT Nanosecond Pulser to drive an APPJ, researchers at EHT and EdCC accomplished the following:

- Characterized efficacy of over 900 APPJ parameters using lawns of *B. atrophaeus* endospores on Petri dishes and identified conditions yielding greater than two log reduction in 20 s
- Calculated D value for *B. atrophaeus* endospores: 160 s on Al surface
- Demonstrated six log endospores reduction in large-area test of a single jet showing potential efficacy of multi-jet array.
- Identified humidity, water, and low concentrations of oxygen as enhancing killing efficacy of plasma jet.
- Demonstrated APPJ is safe for sensitive electronics including FPGAs SDRAM.

For more information: http://www.eagleharbortech.com/

Acknowledgment

This work was supported by a NASA SBIR Phase I (NNX16CP48P).