communications

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The City of Escondido
Attn: Jerry Van Leeuewen
Director, Community Services
475 N. Spruce Street
Escondido, CA 92025

Subject: Mayflower Dog Park Spore Analysis

Dear Mr. Van Leeuewen,

Introduction:
The L-3 communications team employed their BIT Plasma technology to decontaminate the Mayflower Dog Park. The potential contamination of the park with infectious Parvovirus was suspected following the presence of an animal that was positive for this virus. Our treatment is expected to eradicate a virus such as Parvovirus quickly and efficiently. There is no direct test for environmental presence of Parvovirus so we have used a surrogate pathogen on site during the decontamination to evaluate the effectiveness of our treatment. We chose a spore form of bacteria that is significantly more resistant to a variety of decontaminants when compared to Parvovirus. We used commercially prepared paper strips that contain a high density of spores. When the strips are cultured, the degree of killing is proportional to the amount of growth that occurs over time when compared to controls. As an example, a strip containing 10,000 spores can be expected to show growth when cultured as early as 12 hours. If a treated strip shows growth at the same time and to the same extent as controls, the treatment was not effective. If the treated strip shows delayed and/or diminished growth, then killing has occurred. All of the sample strips placed on site during treatment had growth that was significantly delayed when compared to controls and many showed no growth at all. Based upon our analysis of the amount of growth over time, we conclude that more than 99% of the surrogate pathogen was killed. While the degree of actual killing is likely much higher than 99%, the qualitative nature of the test strips used does not permit a more precise measurement. Additionally, since the surrogate is expected to be more resistant to our treatment than Parvovirus, we expect that the rate of killing of the Parvovirus was even greater.

Method:
15 samples were placed at random throughout the area treated. Each sample consisted of two spore strips containing a known quantity of *Geobacillus stearothermophilus*; one strip at 10,000 spores and one strip at 1,000,000 spores. The area was sprayed with cold plasma activated BIT

solution applied at approximately 25 gallons/acre. Following treatment, the strips were collected and placed into sterile culture media, and incubated at 55 deg C.

Results:
All samples examined at 12 hr were negative for growth. Control samples (untreated spore strips) were all positive for growth at this time. At 18 hr 71% of the samples remained negative for growth while 29% showed slight growth. At 36 hr 46% of the samples remained negative for growth while 54% of the samples showed light, moderate or heavy growth.

Analysis:
Based upon the large number of negative samples at 36 hr and the diminished growth in the remaining samples, the treatment appears to have been effective. When the number of starting spores is considered along with the level of growth in any positive samples we can conclude that we achieved a significant reduction of viable spores. Since the strips are not quantitative, we can only express the diminished growth against time and a visual assessment of the density of the resulting culture. Based upon this type of analysis it is reasonable to conclude that we achieved at least a 99% kill rate of the spores.

Conclusion:
Geobacillus stearothermophilus is a very hardy organism. It is used in the health care industry as the surrogate organism to establish the effectiveness of sterilization of various procedures. The treatment of the samples in the park showed a significant diminished growth from the spore strips when cultured for 36 hours. The positive controls all produced significant growth. The effect of this treatment on a significantly more labile pathogen such as Parvovirus can be expected to eliminate the infectious virus from the treated surfaces. In my opinion, the park is safe for the return to use by the public.

Sincerely,

Kurt R. Klimpel, Ph.D.
Senior Program Manager
Binary Ionization Technology (BIT)
L-3 Communications