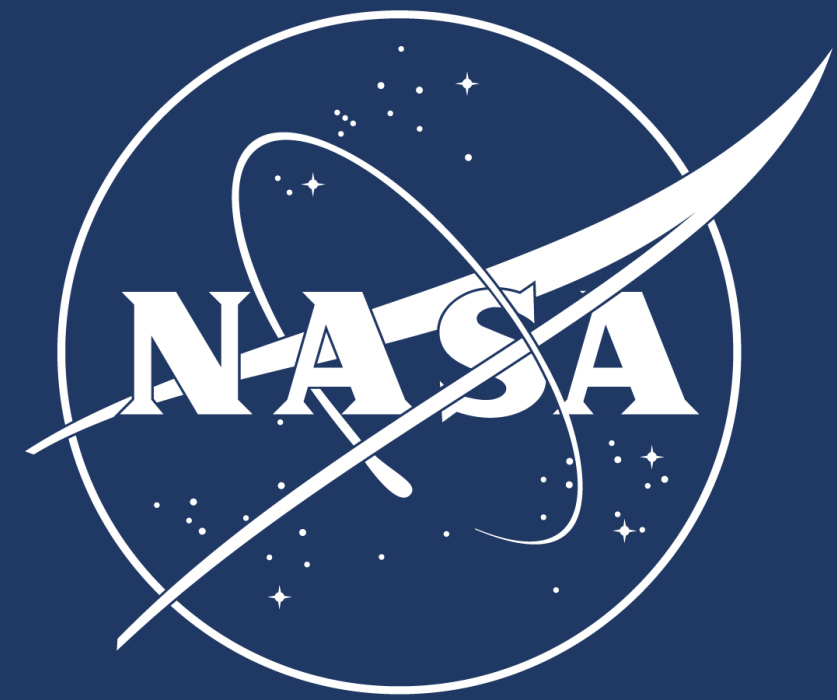


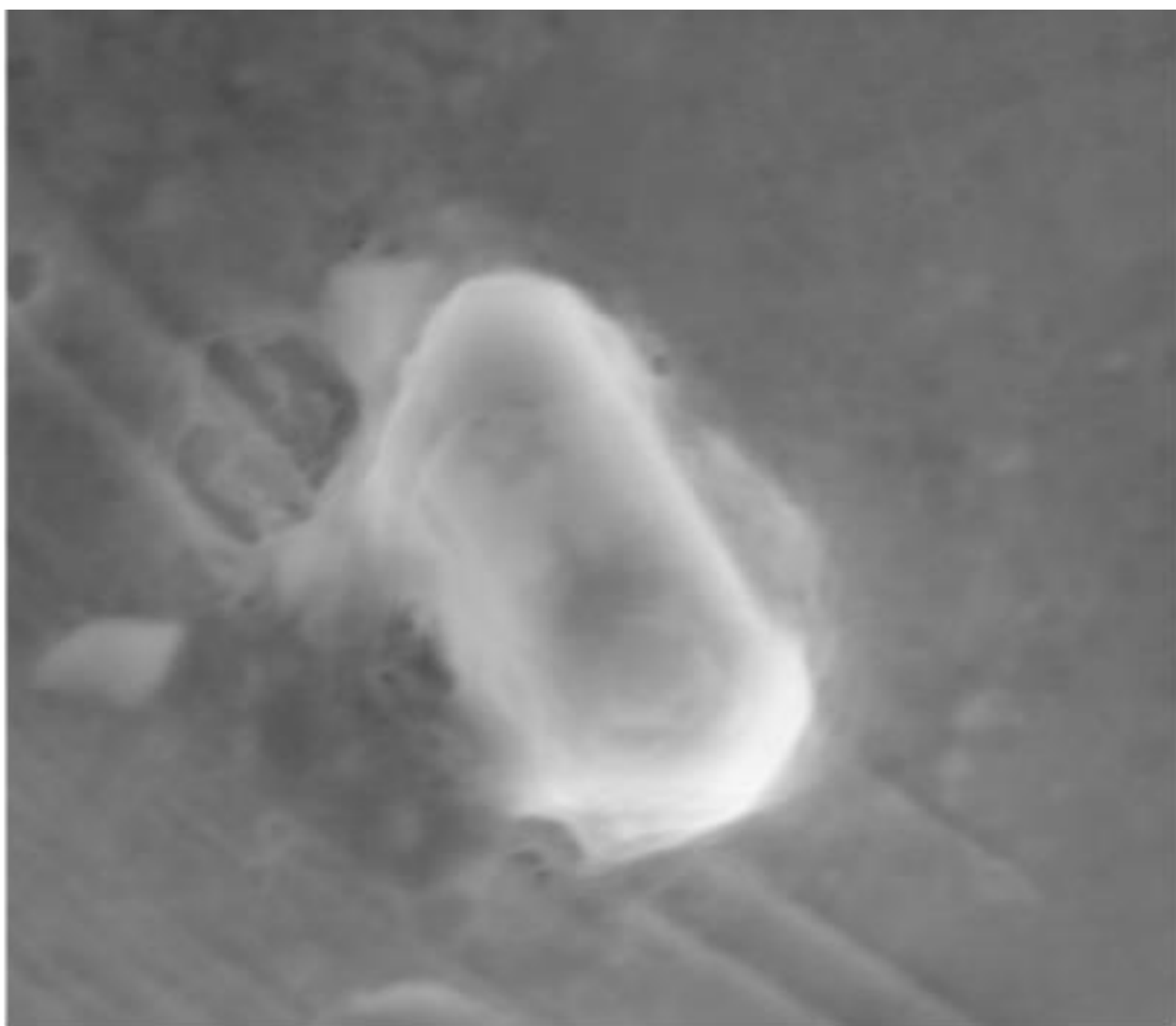
Electron Microscopy of Microbial Biomass



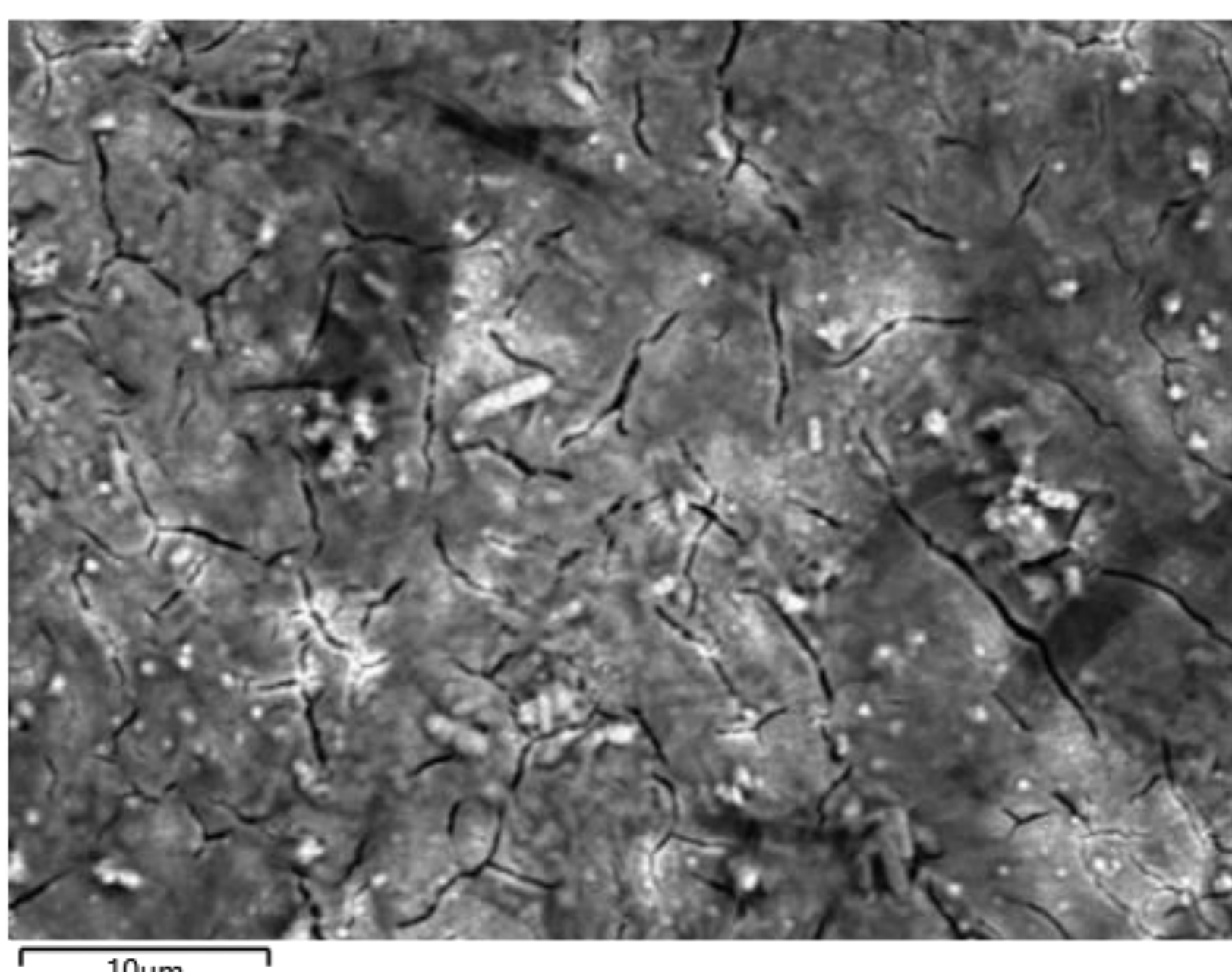
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ABSTRACT

As humans move to long term stays in space their microbes move with them. This poses problems as the microbes can colonize various life support systems, creating biofilms that can grow and spread, which in turn causes obstructions and ultimately degrades function of these systems. MSFC currently has no protocol in place for the Scanning Electron Microscope (SEM) imaging of microbes. As NASA looks to expand human presence in space, it is imperative to understand the impact of microbes on critical life support systems. With an Innovation Grant from Jacobs, we are working to establish a protocol for microbial/biofilm imaging on MSFC’s SEMs.



Microbe on Additively
Manufactured stainless Steel,
imaged on FEI ESEM, 25kx



Microbes on machined Cu
sample imaged using VP-SEM
with BSE, 2500x magnification

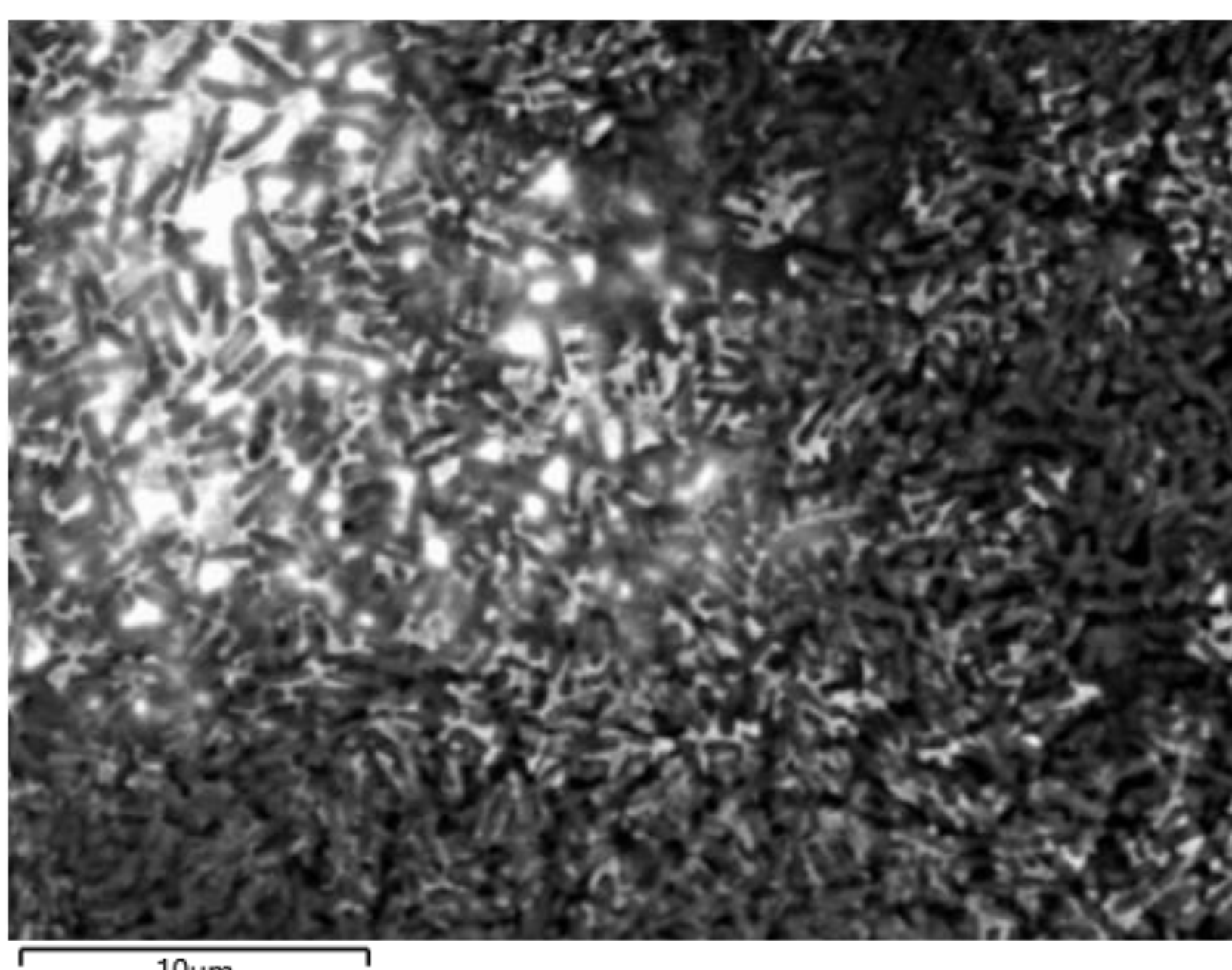
- The use of SEM for detailed visualization of these biofilms:
- Provides insight into the morphology and location contained in the biofilms
 - Is beneficial to understanding biofilm formation, elimination, and prevention
 - Elucidates the interactions of microbes with various alloys used in space applications

This application may also be useful to visualize samples returned from other planetary bodies.

METHOD

Imaging of microbes with SEMs is widely and routinely used today. Frequently, these samples are first dried then chemically fixed and coated with a thin film of various alloys. This process allows for imaging under high vacuum but is time consuming and adds restraints to sample imaging. MSFC has the capability for samples to be imaged under Variable Pressure SEM (VP-SEM), which works at low vacuum and low voltage or Environmental SEM (E-SEM) which allows wet samples to be imaged, with no extra fixation or coating steps, under a low vacuum with water vapor added to the vacuum chamber.

With funding from a Jacobs’ Innovation Grant, we are partnering with an ongoing CIF, the Additive Manufacturing Enabled Biofilm Prevention (AMEBoP) project, to image portions of additively manufactured manifolds that will be inoculated with microbes and undergo various periods of dormancy.



Microbes on machined SS
imaged using VPSEM with
BSE, 3500x magnification



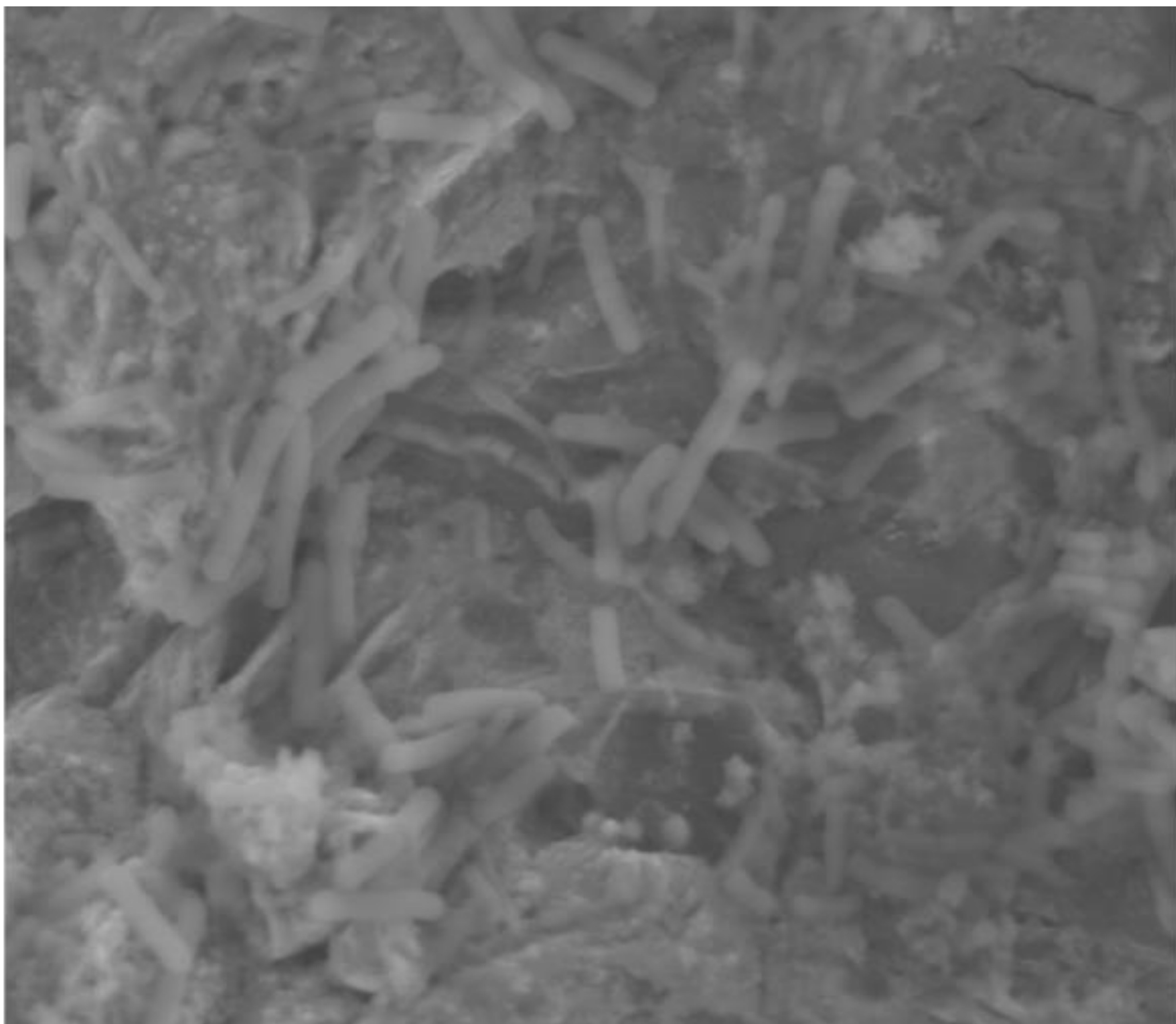
Microbes on Additively
Manufactured Copper,
Imaged on FEI ESEM, 7kx

For trial runs we inoculated 4 separate samples, machined stainless steel and copper and additively manufactured stainless steel and copper, with two strains of microbes that can be found in the life support systems on the ISS- *Pseudomonas aeruginosa* and *Burkholderia cepacia*. These microbes were allowed to grow approximately 3 days and then were imaged using a Variable Pressure SEM (VPSEM) and the ESEM for comparison.

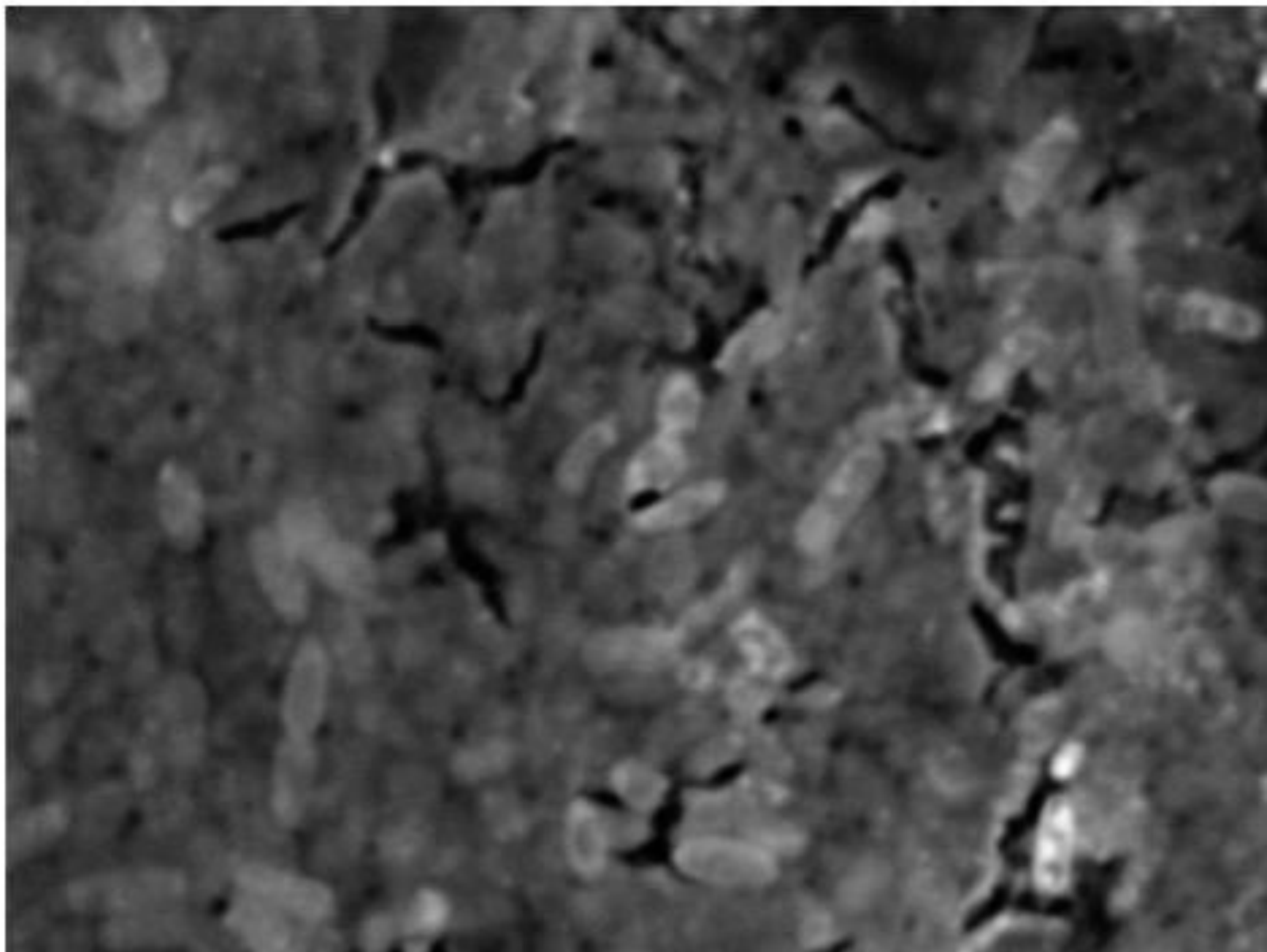
RESULTS

Microbes are imaged better on additively manufactured alloys, but the growth seems to be comparable on both additive manufactured and machined samples. The use of the ESEM appears to allow for more clear and detailed images as compared to VP-SEM.

Images such as these can be useful for many varying microbial applications and can provide insight into the world of microscopic life.



Microbes on AM Cu imaged using
ESEM, 5kx magnification



Microbes on a machined Cu sample imaged on VP-
SEM using BSE, 5kx magnification