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Establishing a PMA-based, molecular protocol for microbial life-dead distinction in specialized indoor environments

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Microbial contamination and quality control usually relies on cultivation and quantification of colony forming units. This process, however, is laborious and gives only information on the cultivable microbial diversity.

One field, which requires quick decisions on relevant microbial contamination, is Planetary Protection. Herein, mission goals, such as other planets, need to be protected from terrestrial microbial contamination. This includes a stringent quality control of spacecraft. To avoid delays in construction and testing, decision on spacecraft cleanliness have to be taken in timely manner.

In collaboration with the German Aerospace Center and the European Space Agency, we are establishing a novel protocol for the quick analysis of cleanroom and spacecraft contamination, based on the application of propidium monoazide (PMA), which blocks the signal from non-viable cells. Classical cleanroom contaminants were selected to form a representative mock community for all experiments. We simulated cleanroom sampling and sample concentration on filters. After exposure to radiation, desiccation and freezing, stresses which are expected during a space flight, the filters are treated with PMA, subjected to DNA extraction, microbiome analysis and quantitative PCR. In parallel to confirm our results, cultivation-based tests are performed.

Our results indicate that the protocol developed is well suited for a quick (about 14 h) quality assessment of the viability of possible contaminants. We also could show, based on the stress-exposure that the viability of the mock community is largely reduced. This protocol might also be useful for applications in the field of hygiene, food contamination control or pharmaceutical industry.