# Introduction

Microbial materials such as cells and extracted genomic DNA from a presumably “pure” culture should ideally be free of organismal contaminants. However, high sensitivity detection methods including polymerase chain reaction (PCR) and next generation sequencing (NGS) can detect organismal contaminants previously undetectable by traditional microbiology methods such as culturing, biochemical tests, and microscopy. Characterizing and reducing the level of these contaminants is critical to ensuring high quality microbial materials are used to populate sequence databases, for mock communities used to validate metagenomic methods, to validate biodetection assays, and for basic research using model systems. General contaminant assessment is also needed for the characterization of microbial reference materials, where contaminant profiles allow users to properly determine whether the material is suitable for their application. In addition to organismal contaminants in the material itself, PCR and NGS can also detect reagent impurities, highlighting the need to differentiate material and reagent contaminants. Issues related to reagent contaminants have been well documented and addressed with negative controls, improved methods for removing contaminants, and post-processing of sequence data. However, contaminants in microbial materials, as found in non-axenic cellular materials or genomic materials with foreign DNA, have only been addressed in data processing.

Current approaches for detecting contaminants in microbial materials typically rely on methods such as culture, microscopy, or PCR. Culture and microscopy-based methods lack the required sensitivity for microbial materials being used in NGS and PCR applications, are not appropriate for genomic DNA materials, and assume the contaminants are phenotypically distinct from the material they contaminate. While PCR-based methods can detect contaminants in genomic DNA, the methods are limited as they can only detect specifically targeted contaminants and are not amenable to high-throughput applications. In contrast to these methods, shotgun metagenomic methods can be used to detect contaminants in both cell cultures and genomic DNA materials while only requiring the contaminant has sequencing reads that differentiate it from the material strain.