

AnnotIEM: a novel tool for microbiome species-level annotation of 16S gene based microbial sequencing [Abstract]

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In order to generate meaningful results from microbiome analysis, identification of microbial species that are relevant to various clinical conditions (eg, *Staphylococcus aureus* in atopic dermatitis) is needed. However, annotation of microbial taxonomy down to the species level presents a unique challenge because the main databases available (eg, Silva, RDP and Greengenes) are not curated and have many errors and missing information. Here we present AnnotIEM - a new tool for species level annotation of sequences (OTUs or ASVs) derived from 16S rDNA sequencing. A novel hit selection algorithm is used to combine the annotation output from multiple databases to ensure accurate identification of a species and reducing the potential for errors from each of them alone. When tested using mock community control datasets, the precision of annotation with AnnotIEM is greater than 80% on the species level and greater than 90% on the genus level, better than any currently available annotation tool. Benchmarking using a number of real case studies shows that annotation by AnnotIEM gave rise to a higher fraction of sequences annotated on the species level (60-80%) as compared to any other currently available annotation tool (eg, IMNGS, SILVA, RDP Classifier and q2classifier from QIIME2). In conclusion, the AnnotIEM tool shows that using a number of databases combined with a novel hit selection algorithm results in a significantly better

annotation on the species level, which can greatly enhance the results of any microbiome analysis study.