**Library construction and sequencing**

For amplification of the V3-V4 domain of bacterial 16S rRNA, we used primers 341F (5′-CCTACGGGNGGCWGCAG-3′) and 806R (5′-GGACTACHVGGGTWTCTAAT-3′) ([Vasileiadis et al., 2012](#_ENREF_5); [Klindworth et al., 2013](#_ENREF_4)). These primers include overhang adapter sequences for compatibility with Illumina index and sequencing adapters. Illumina sequencing adapters and dual-index barcodes were added to purified amplicons using the Nextera XT index kit (Illumina). Purified libraries were quantified using the Quant-iT™ PicoGreen® dsDNA assay kit (ThermoFisher) and KAPA qPCR Library quantification kit for Illumina (Kapa Biosystems), and validated using a BioAnalyzer (Agilent Technologies). The libraries were pooled in equimolar amounts and contained 20% of a phiX control library. Sequencing was performed on an Illumina MiSeq platform at the Tokyo University of Agriculture and Technology using a 250-bp paired-end kit from Illumina.

**Microbial community analysis**

After paired-end reads were merged using fastq-join ([Aronesty, 2011](#_ENREF_1)), phiX sequences were removed using BWA (version 0.7.10; http://bio-bwa.sourceforge.net/), SAMtools (version 0.1.19; http://samtools.sourceforge.net/) and a custom Perl script. Quality filtering of merged sequences and OTU clustering was performed using the Quantitative Insights into Microbial Ecology (QIIME) v.1.8.0 pipeline ([Caporaso et al., 2010](#_ENREF_2)). Sequences were assigned to OTUs using a closed-reference OTU picking protocol at 97% sequence identity using UCLUST ([Edgar, 2010](#_ENREF_3)) implemented in QIIME against the non-redundant SILVA Reference database (release SSU Ref119 NR), obtaining OTU relative abundance data at each taxonomic level.

**References**

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