SUPPLEMENTARY METHODS

Bioinformatics datasets pre-processing and merging

mothur (v.1.42.3) was run in "batch mode" on a linux server. The complete mothur "logfile" is included as supplementary information. Initial steps deviated from the mothur MiSeq standard operation procedure (https://mothur.org/wiki/miseq_sop/; Kozich *et al.*, 2013). Paired sequences from the 2015 and 2017 datasets contained both "forward" and "reverse" reads in both "R1" and "R2" fastq files, preventing the generation of a ".files" or ".oligos" file in mothur and as such barcodes and primer sequences could not be removed following the first steps of the mothur MiSeq SOP. Instead, paired reads were first converted into contigs using the make.contigs() commands and barcode and primer sequences then removed using trim.seqs() with an ".oligos" file. For the 2018 dataset, barcode and primer sequences were removed using pcr.seqs(). All 3 trimmed contig datasets when then merged into a single dataset using the merge.files() command. The rest of the sequence pre-processing process followed the mothur MiSeq SOP as detailed on https://mothur.org/wiki/miseq_sop/. The full list and details of mothur commands can be found in the "mothur.command.batch" file. The batch file used to run initial mothur commands for quality checks and OTU generation is also included as supplementary information.

References

Kozich JJ, Westcott SL, Baxter NT, Highlander SK & Schloss PD (2013) Development of a Dual-Index Sequencing Strategy and Curation Pipeline for Analyzing Amplicon Sequence Data on the MiSeq Illumina Sequencing Platform. *Applied and Environmental Microbiology* **79**: 5112-5120.