Text S3. Estimation of the proportion of archaea obtained with the non-specific ArBa primer pair using qPCR and pyrosequencing libraries. The ratio of bacteria to archaea obtained in the pyrosequencing library generated with the ArBa primer pair was validated by qPCR of 16S rRNA genes from bacteria and archaea in the same samples (Figure S7). The pyrosequencing libraries showed a significantly higher average abundance of archaea (4.9% ± 2.1%; bacteria: 95.1% ± 2.1%) than qPCR (1.3% ± 0.7%; bacteria: 98.7% ± 0.7%). A similarly high abundance of the archaeal component was also reported by Brulc et al. using a primer-independent metagenomics approach [1]. The overall trend of bacterial, and thus archaeal abundances in the 12 pyrosequencing libraries was correlated with the results obtained from qPCR (Pearson correlation coefficient = 0.83, \( P < 0.001 \); Figure S7), indicating that generalized between-sample comparisons can be validly made from data generated using the ArBa primer pair.

REFERENCES