Supplementary material

This appendix contains supplementary methods and results for the paper: Filtering artefacts in bacterial community composition can affect the outcome of dissolved organic matter biolability assays.

Cell counts with flow cytometry

At each sampling occasion 10 mL subsamples were taken from each replicate for determination of cell counts using flow cytometry. These subsamples were filtered with 20 μ m filters and stored at 4 °C as 2 mL aliqots for maximum 2 hours prior to measurement.

Measurements were made using a BD Accuri C6 Flow Cytometer on samples stained with SYTO-13 stain (Invitrogen, Carlsbad, CA, USA) at 2.5 μ M final concentration following the protocol of del Giorgio et al. (1996) and using the following software settings: flowrate 14 μ L / min, volume limit 20 μ L, threshold, 20000 on FSC-H.

Figure S1 shows the cell count results, separated by sampling date and filtering treatment.

Time series plots of bacterial taxa

Figure S2 and S3 show time series plots of relative abundances of the 12 most abundant bacterial (sub-) phyla, based on 16S rRNA gene amplicon sequencing. These are the same data used to produce Figure 3 in the main text. See main text methods for details on bioinformatic procedures.

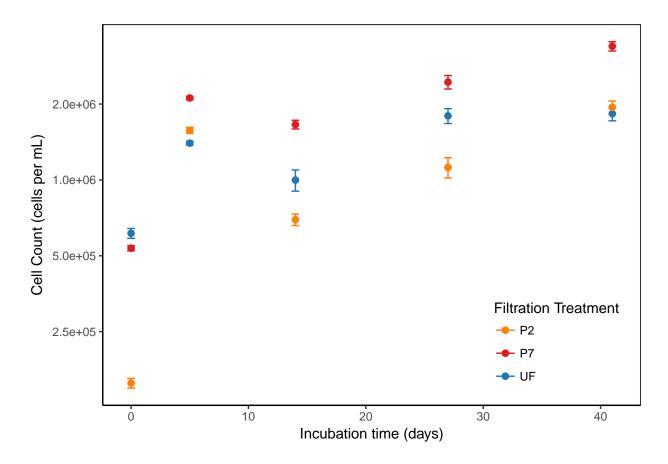


Figure S1: Estimates (mean \pm s.e., n=4) of cell abundance by flow cytometry using samples incubated with nucleic acid stain SYTO-13, note log10 scale on y-axis.

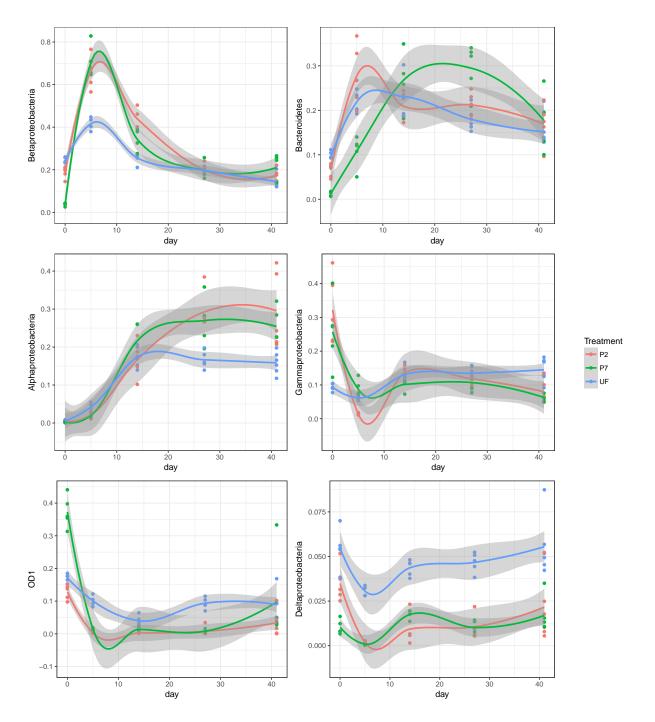


Figure S2: Time series of relative abundances of selected bacterial (sub) phyla measured by 16S amplicon sequencing. Points represent individual replicates. Lines and grey shaded areas are loss smoothers and associated 95% confidence intervals.

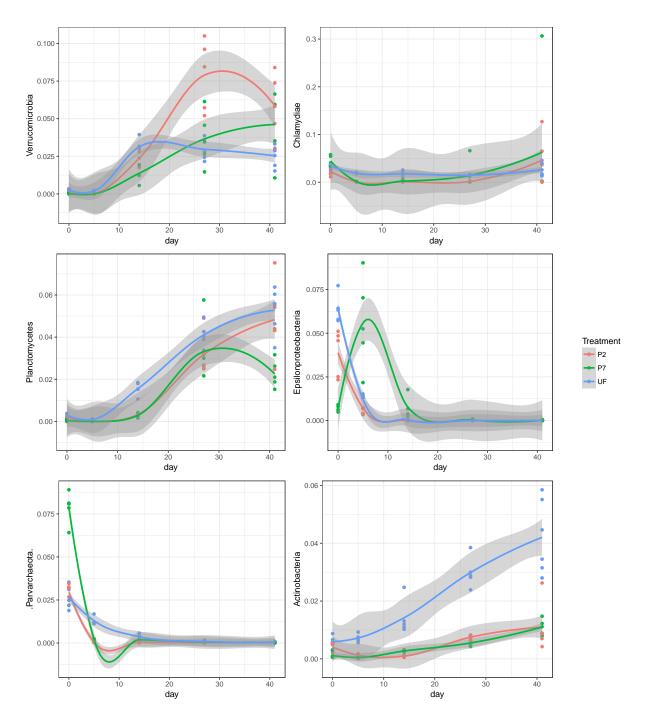


Figure S3: Time series of relative abundances of selected bacterial (sub) phyla measured by 16S amplicon sequencing. Points represent individual replicates. Lines and grey shaded areas are loss smoothers and associated 95% confidence intervals.

References

del Giorgio, P., D. F. Bird, Y. T. Prairie, and D. Planas. 1996. Flow cytometric determination of bacterial abundance in lake plankton with the green nucleic acid stain SYTO 13. *Limnology and Oceanography* 41:783-789.