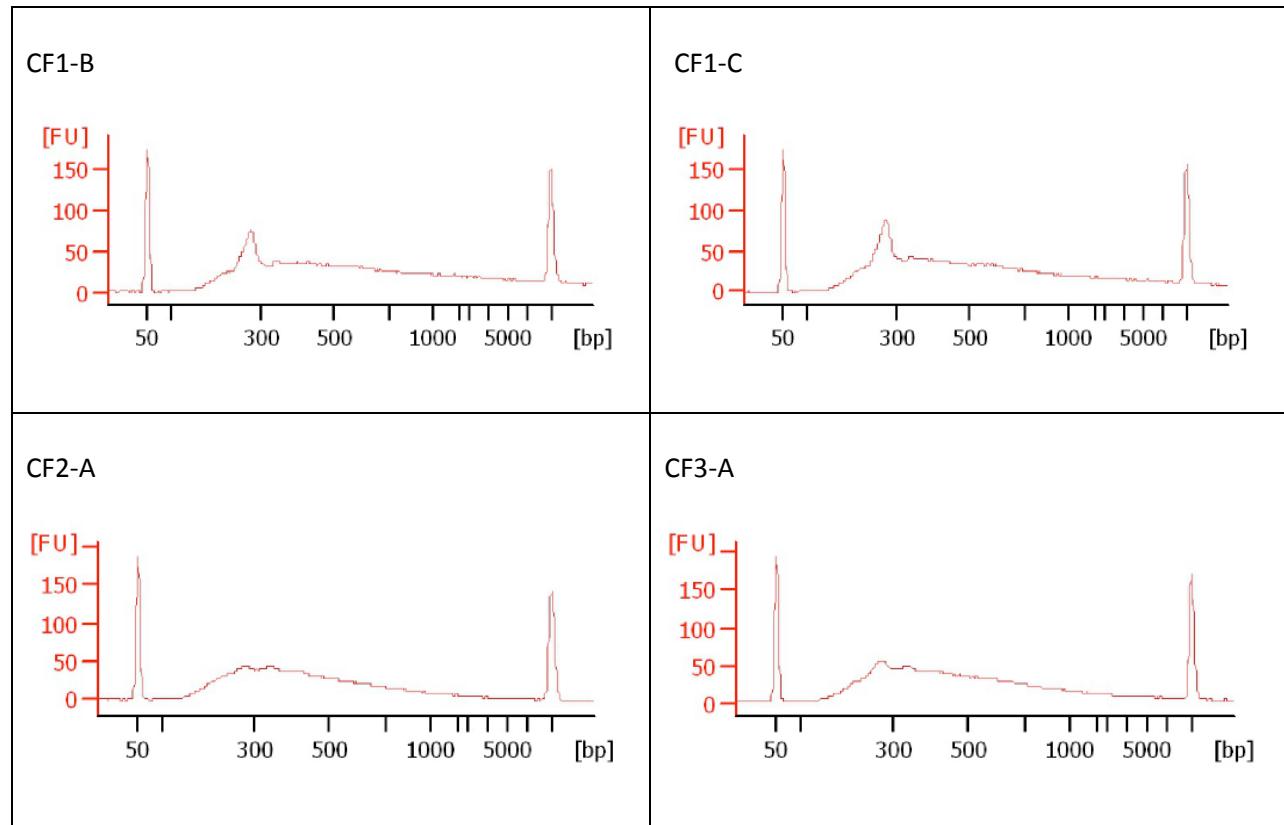
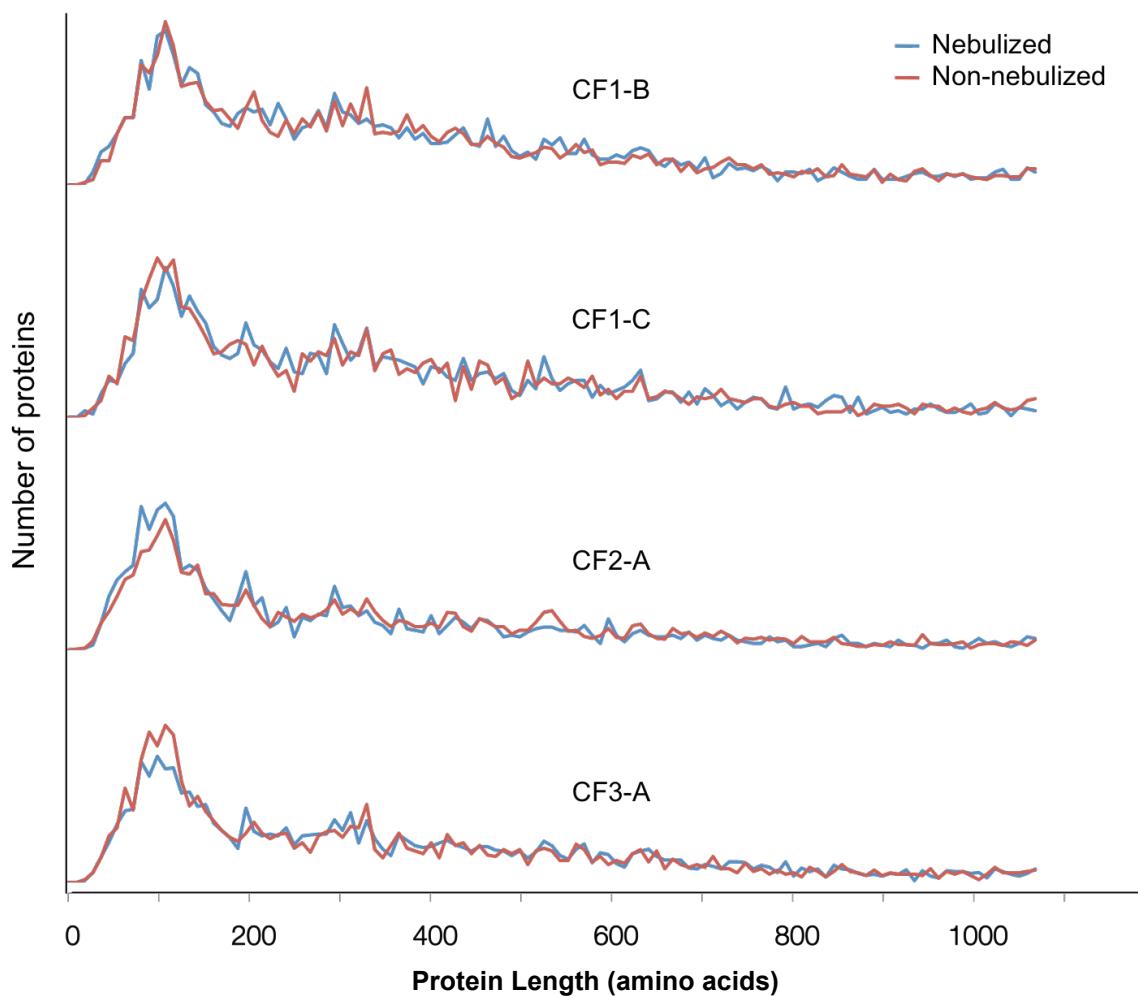


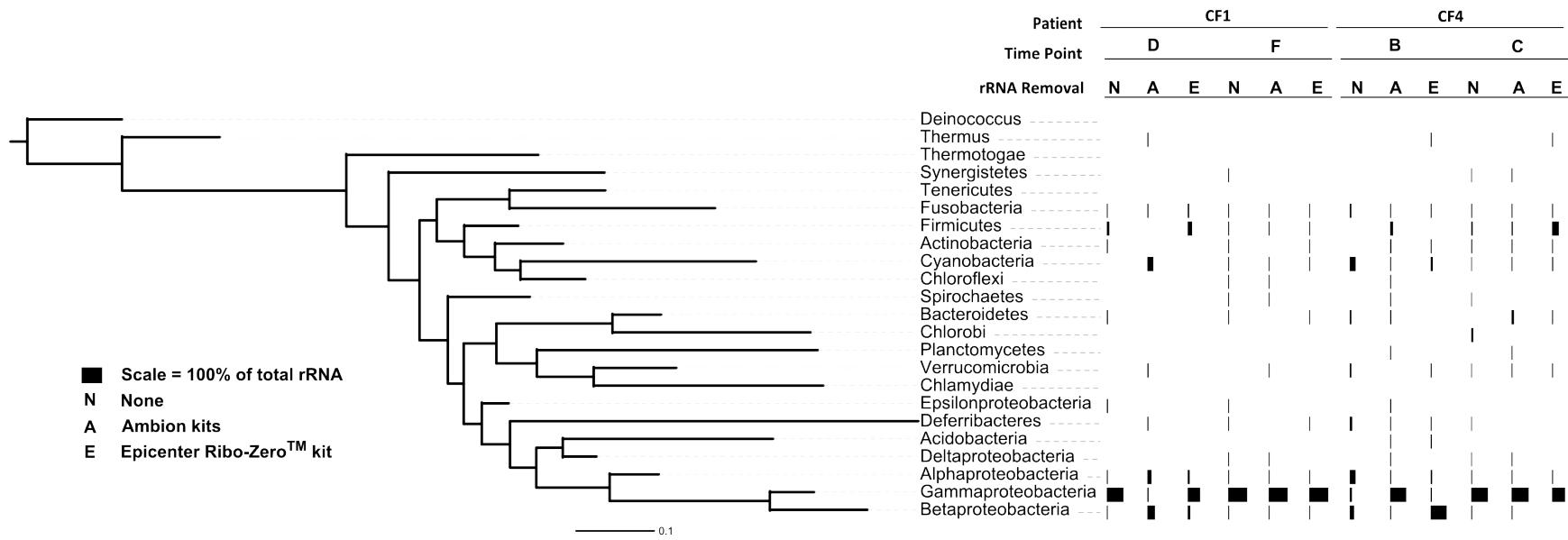
SUPPLEMENTARY FIGURE 1: The relative proportion of 16S and 18S rRNA gene based on quantitative PCR following the modified Breitenstein *et al.* method used to deplete host-associated (human) DNA in sample CF5-A and CF5-B. (B) Total DNA, (P) Pellet following hypotonic lysis and DNase treatment, (S) supernatant of the resuspended pellet. The ratio of 16S:18S increased following the hypotonic lysis and DNase treatment, indicated depletion of the 18S (human DNA) and the enrichment of 16S (microbial DNA).



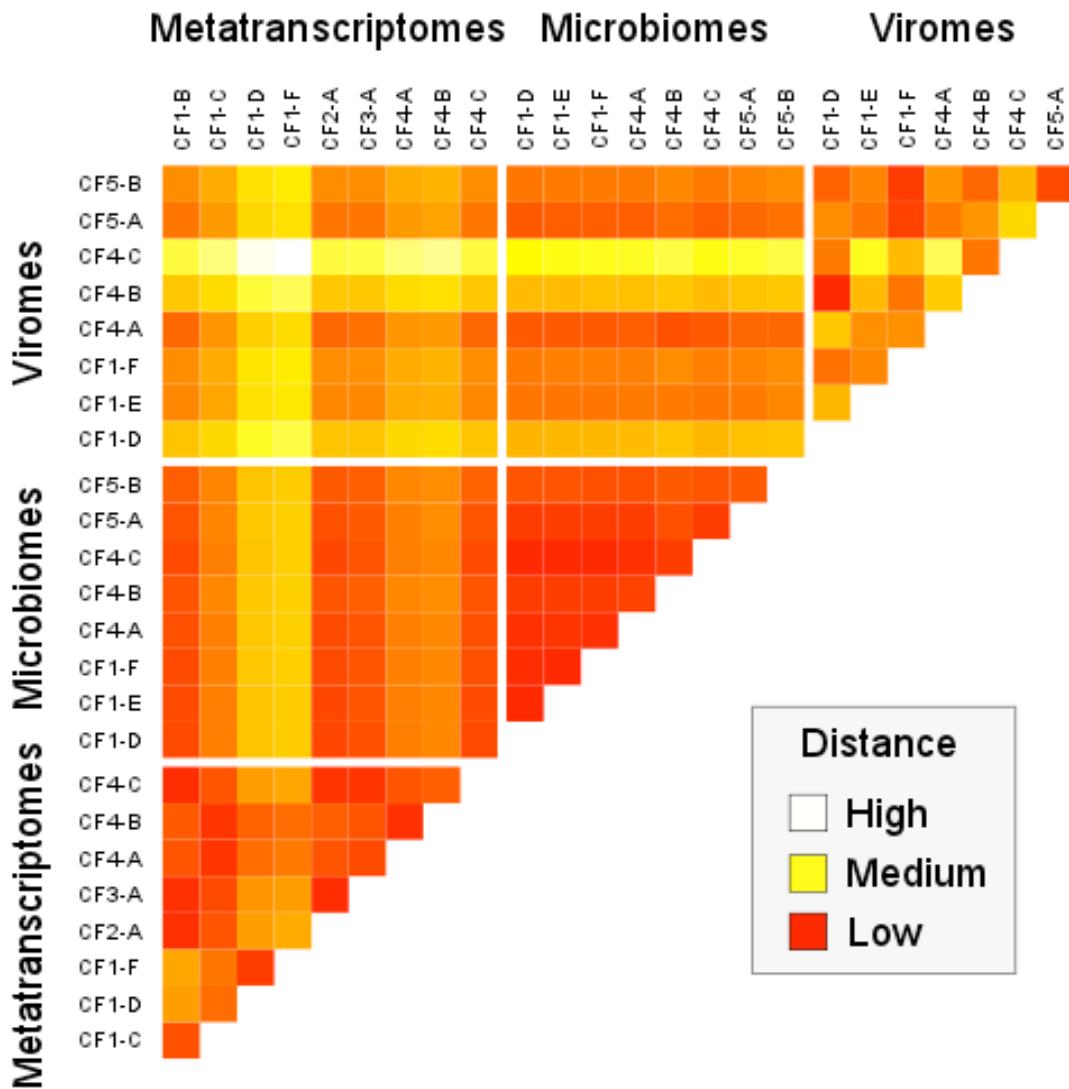
SUPPLEMENTARY FIGURE 2: cDNA electropherograms for the metatranscriptomes generated to assess the effects of nebulization. The cDNA length profiles shown here were generated by the Agilent 2100 Bioanalyzer before nebulization.



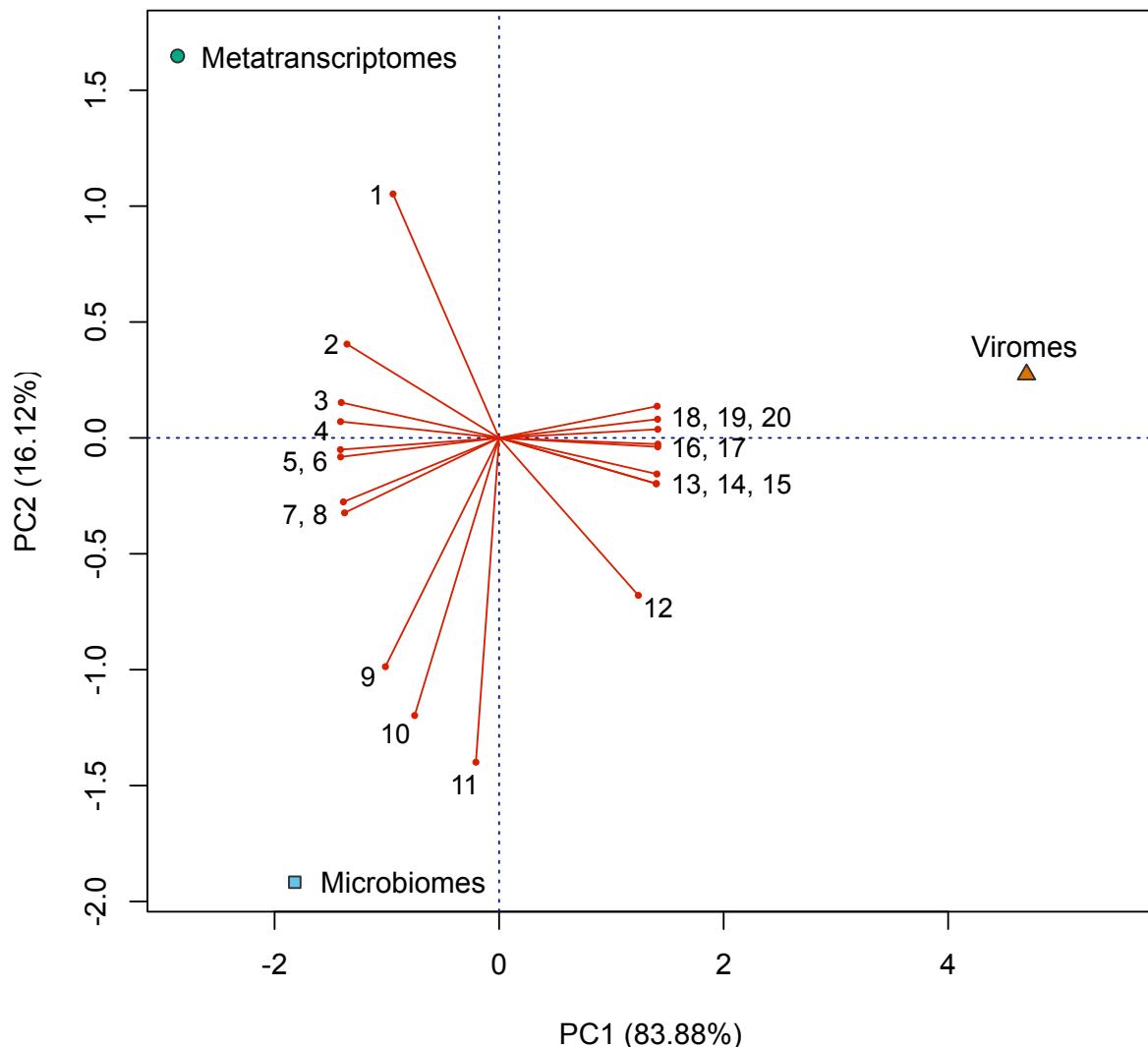
SUPPLEMENTARY FIGURE 3: Effect of nebulization on translated protein length. Protein lengths were predicted for cDNA transcripts by BLASTx against the NCBI non-redundant protein database. Protein length profiles for non-Chordata proteins were plotted with a cutoff at 1,200 amino acids.



SUPPLEMENTARY FIGURE 4: Relative abundance of microbial phyla/classes identified from rRNA reads. The 12 metatranscriptomes compare Epicentre rRNA depletion, Ambion rRNA depletion, and no RNA depletion in each of four samples



SUPPLEMENTARY FIGURE 5: Similarities between the predicted metabolic profiles of the virome, microbiome, and metatranscriptome datasets. The heatmap represents the Euclidian distance between datasets ranging from white (least similar) to red (most similar).



SUPPLEMENTARY FIGURE 6: Principle component analysis (PCA) showing the top 20 metabolic pathways that displayed the greatest variance between the three dataset groups. CF1-A was omitted in both analysis due to insufficient data.

¹Ribosome (ko03010); ²Amino sugar and nucleotide sugar metabolism (ko00520); ³Fructose and mannose metabolism (ko00051); ⁴Biotin metabolism (ko00780); ⁵Lipoic acid metabolism (ko00785); ⁶Aminoacyl-tRNA biosynthesis (ko03015); ⁷Peptidoglycan biosynthesis (ko00550); ⁸Protein export (ko03060); ⁹Pantothenate and CoA biosynthesis (ko00770); ¹⁰One carbon pool by folate (ko00670); ¹¹D-glutamine and D-glutamate metabolism (ko00471); ¹²Valine, leucine and isoleucine biosynthesis (ko00290); ¹³Pyrimidine metabolism (ko00240); ¹⁴Riboflavin metabolism (ko00740); ¹⁵Folate biosynthesis (ko00790); ¹⁶Homologous recombination (ko03440); ¹⁷Mismatch repair (ko03430); ¹⁸Alanine, aspartate, and glutamate metabolism (ko00250); ¹⁹DNA replication (ko03030); ²⁰D-Alanine metabolism (ko00473)