

Online supplementary material to

High-throughput analysis of microbiomes in a meat processing facility: Are food processing facilities an establishment niche for persisting bacterial communities?

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Table S1. Strains used for phylogenetic analysis and multi-species biofilm reconstitution in this study (provided as excel file).

Table S2. Permutational multivariate analysis of variance on Bray-Curtis distance matrix of samples in March 2023 by both culture-dependent and culture-independent approaches.

Table S3. Pairwise single nucleotide polymorphisms (SNPs) matrix for isolates of *C. divergens*, *R. rivi*, *R. inusitata* and *S. proteamaculans*. Only isolates with more than 99% ANI value were included for SNP variant analysis (provided as excel file).

Table S4. Bacterial relative abundance in biofilm samples determined by 16S rRNA sequencing. Only taxa with a relative abundance of 1% or more are presented (provided as excel file).

Figure S1. Heatmap of bacterial isolates collected from meat samples, environmental surface samples during the first time sampling, September 2022.

Figure S2. Relative abundance of uncultured genera that were only detected by 16S sequencing, among samples collected in the first sampling, September 2022.

Figure S3. Principal coordinate analysis, using Bray-Curtis distance with 605 isolates classified at species level for 40 sampling sites, collected at the first sampling time, September 2022.

Figure S4. Bacterial coexistence network based on the microbial communities characterized by 16S rRNA gene sequencing.

Figure S5. Phylogenetic tree of strains of *C. divergens* (A), *Rahnella* (B) and *S. proteamaculans* (C) isolated from a meat processing facility.

Table S2. Permutational multivariate analysis of variance on Bray-Curtis distance matrix of samples from March 2023 by culture-dependent and culture-independent approaches to test the association of community variance.

Method	Comparison	P. adjusted value
Culture-independent	FCS vs NFCS	0.516
	FCS vs Meat	0.096
	FCS vs Water	0.396
	NFCS vs Meat	0.072
	NFCS vs Water	0.252
	Meat vs Water	1.000
	zone1 vs zone3	0.405
	zone1 vs zone2	0.750
	zone1 vs zone4	0.525
	zone1 vs Meat	0.465
	zone1 vs water	0.915
	zone3 vs zone2	0.015
	zone3 vs zone4	1.000
	zone3 vs Meat	0.855
	zone3 vs water	1.000
	zone2 vs zone4	0.105
	zone2 vs Meat	0.165
	zone2 vs water	0.840
	zone4 vs Meat	1.000
	zone4 vs water	1.000
	Cutting vs Packaging	1.000
	Cutting vs Cooling	0.450
	Cutting vs Killing	1.000
	Cutting vs Shipping	1.000
	Cutting vs Meat	0.090
	Cutting vs Water	0.468
	Packaging vs Cooling	1.000
	Packaging vs Killing	1.000
	Packaging vs Shipping	1.000
	Packaging vs Meat	1.000
	Packaging vs Water	1.000
	Cooling vs Killing	1.000
	Cooling vs Shipping	1.000
	Cooling vs Meat	1.000
	Cooling vs Water	1.000
	Killing vs Shipping	NA
	Killing vs Meat	1.000
	Killing vs Water	NA
	Shipping vs Meat	1.000
	Shipping vs Water	NA
	Prior vs Post	0.066
	Prior vs Meat	0.336
	Prior vs Water	1.000
	Post vs Meat	0.060

	Post vs Water	0.102
Culture-dependent	FCS vs NFCS	0.660
	FCS vs Meat	0.042
	FCS vs Water	0.150
	NFCS vs Meat	0.048
	NFCS vs Water	0.156
	Meat vs Water	1.000
	zone1 vs zone3	0.105
	zone1 vs zone2	1.000
	zone1 vs zone4	1.000
	zone1 vs Meat	0.135
	zone1 vs water	0.480
	zone3 vs zone2	0.555
	zone3 vs zone4	0.435
	zone3 vs Meat	0.060
	zone3 vs water	1.000
	zone2 vs zone4	0.615
	zone2 vs Meat	0.180
	zone2 vs water	0.900
	zone4 vs Meat	0.855
	zone4 vs water	1.000
	Cutting vs Packaging	0.198
	Cutting vs Cooling	0.720
	Cutting vs Killing	1.000
	Cutting vs Shipping	1.000
	Cutting vs Meat	0.108
	Cutting vs Water	0.324
	Packaging vs Cooling	1.000
	Packaging vs Killing	1.000
	Packaging vs Shipping	1.000
	Packaging vs Meat	0.900
	Packaging vs Water	1.000
	Cooling vs Killing	1.000
	Cooling vs Shipping	1.000
	Cooling vs Meat	0.540
	Cooling vs Water	1.000
	Killing vs Shipping	NA
	Killing vs Meat	1.000
	Killing vs Water	NA
	Shipping vs Meat	1.000
	Shipping vs Water	NA
	Prior vs Post	0.006
	Prior vs Meat	0.036
	Prior vs Water	1.000
	Post vs Meat	0.012
	Post vs Water	0.102

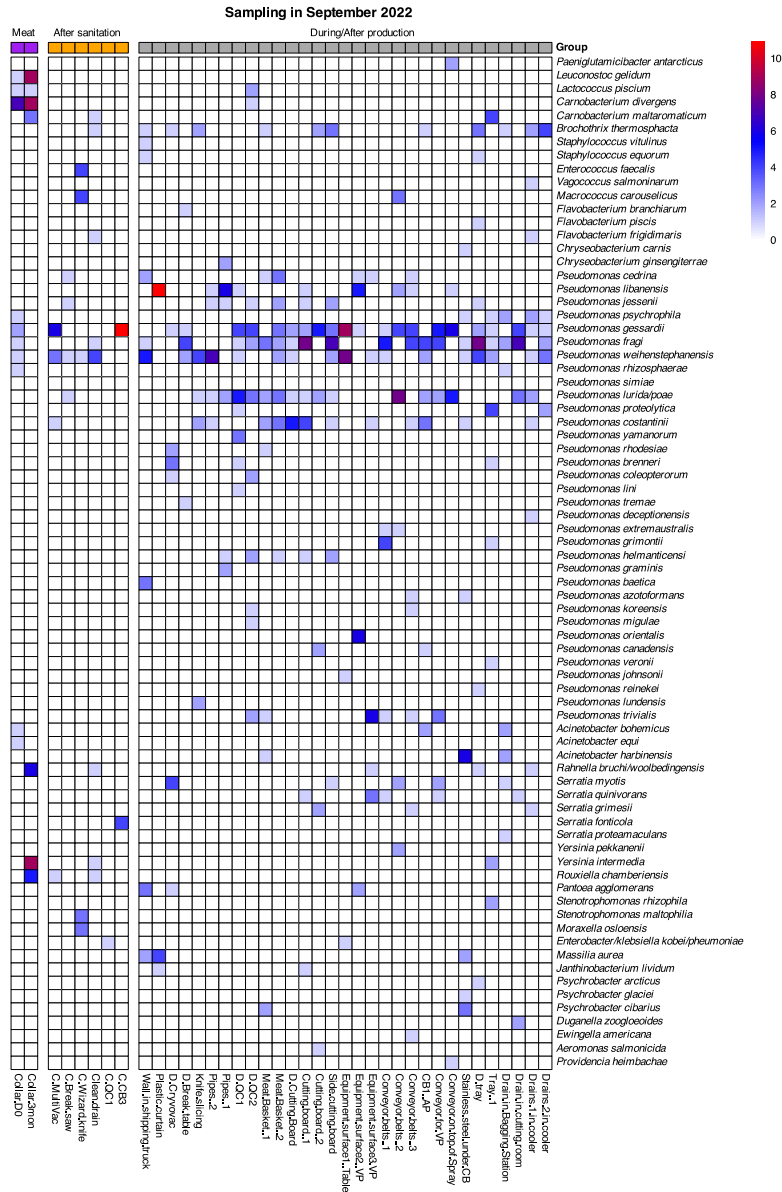


Figure S1. Heatmap of bacterial isolates collected from meat samples, environmental surface samples during the first time sampling, September 2022. Taxonomy classification was determined based on 16S rRNA gene and blasted against NCBI database.

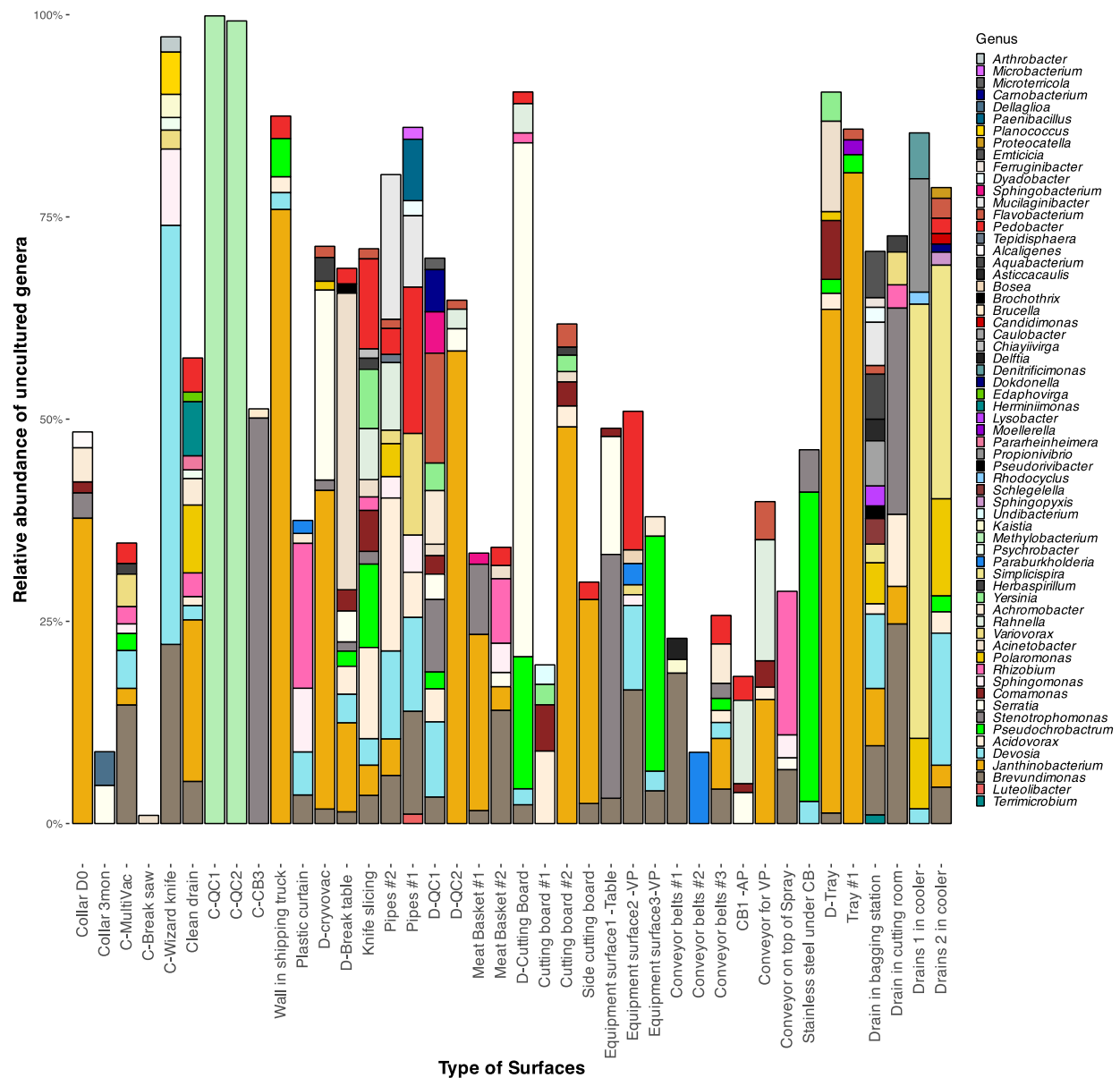


Figure S2. Relative abundance of uncultured genera that were only detected by 16S sequencing, among samples collected in the first sampling, September 2022. Only genera with no less than 1% relative abundance were plotted.

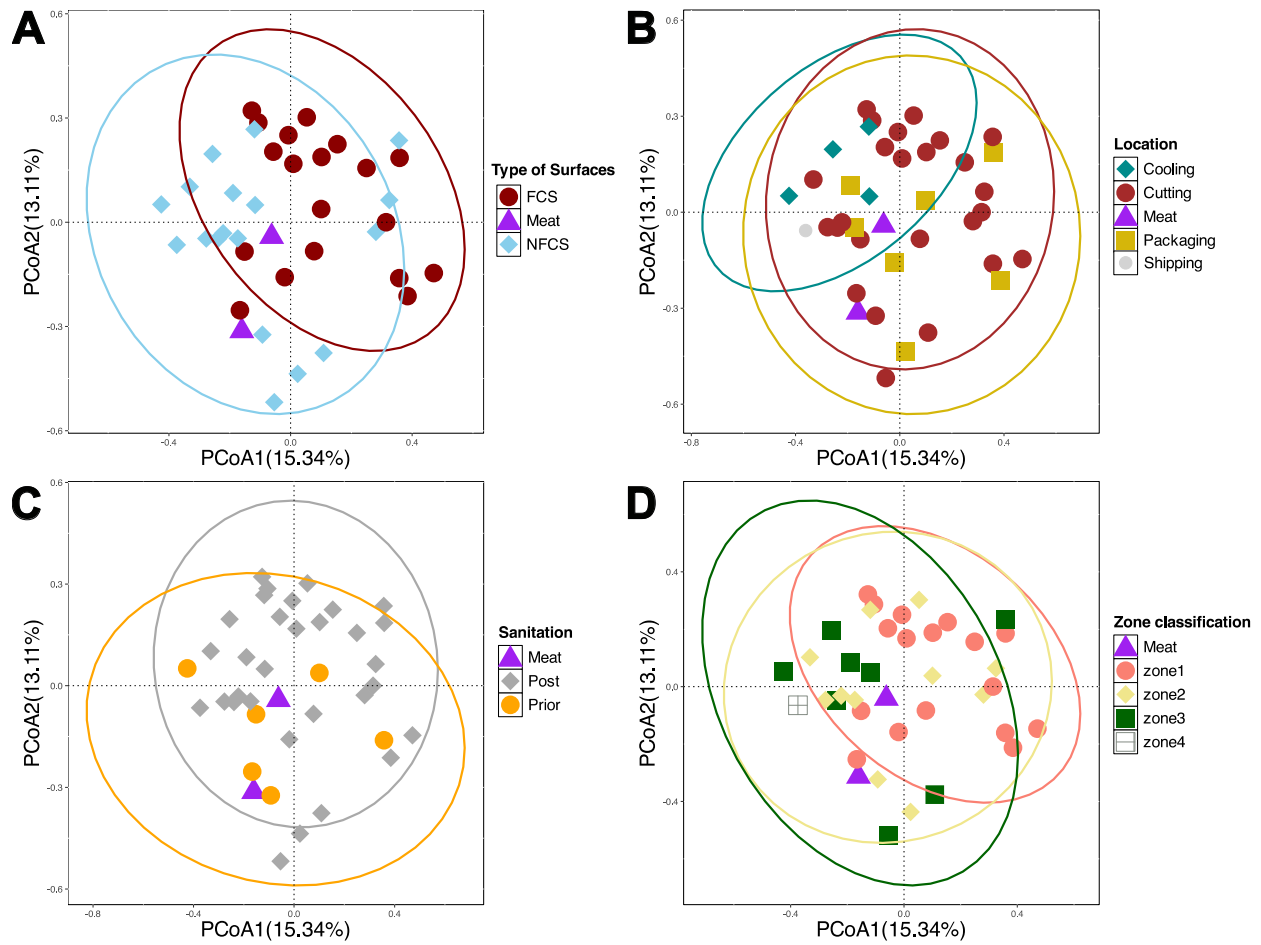


Figure S3. Principal coordinate analysis, using Bray-Curtis distance with 605 isolates classified at species level for 40 sampling sites, collected at the first sampling time, September 2022.

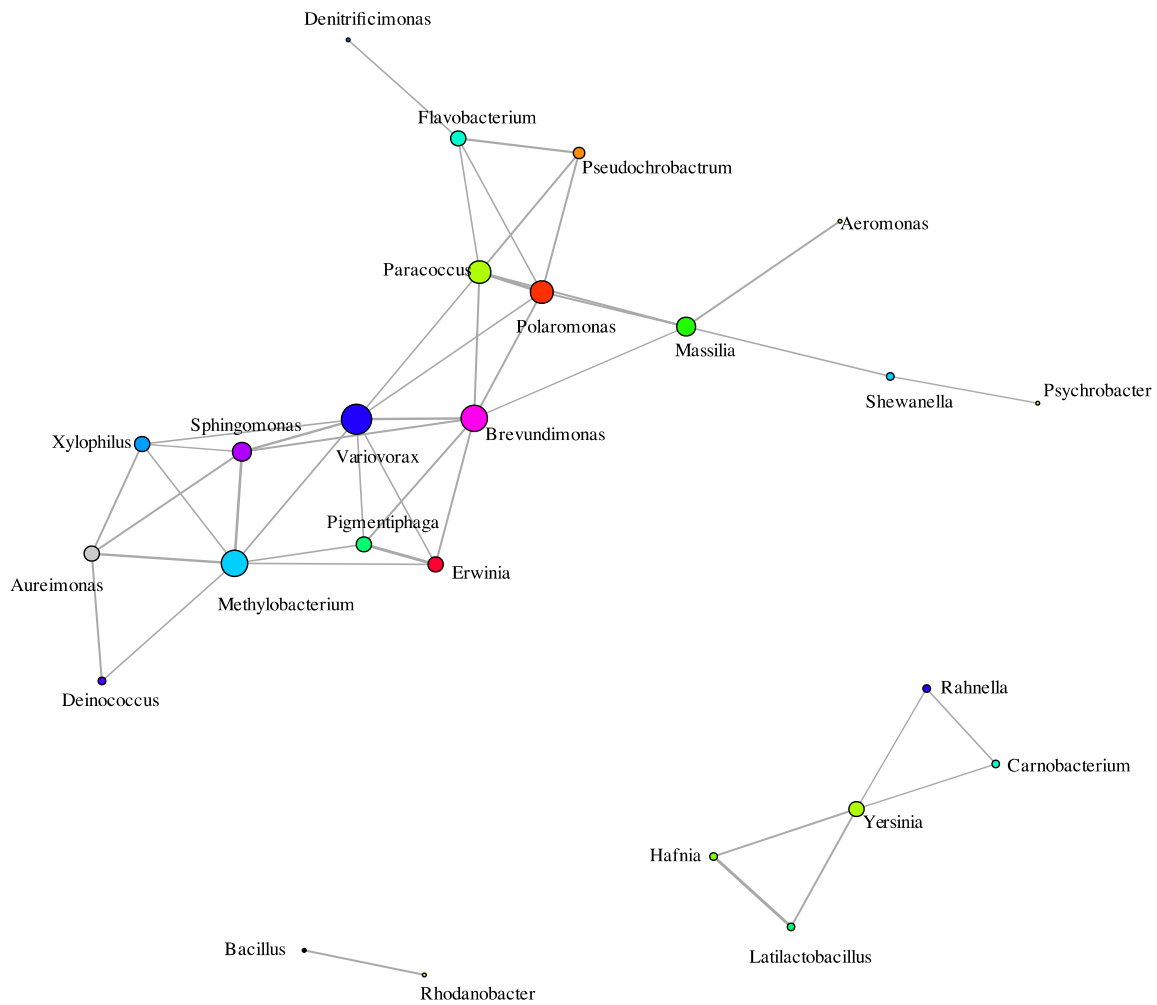


Figure S4. Bacterial coexistence network based on the microbial communities characterized by 16S rRNA gene sequencing. Nodes are colored at genus level. The network connections are determined using Spearman correlation test. Only correlations with a significance level of $P < 0.0001$ and a coefficient of ≥ 0.5 are included.

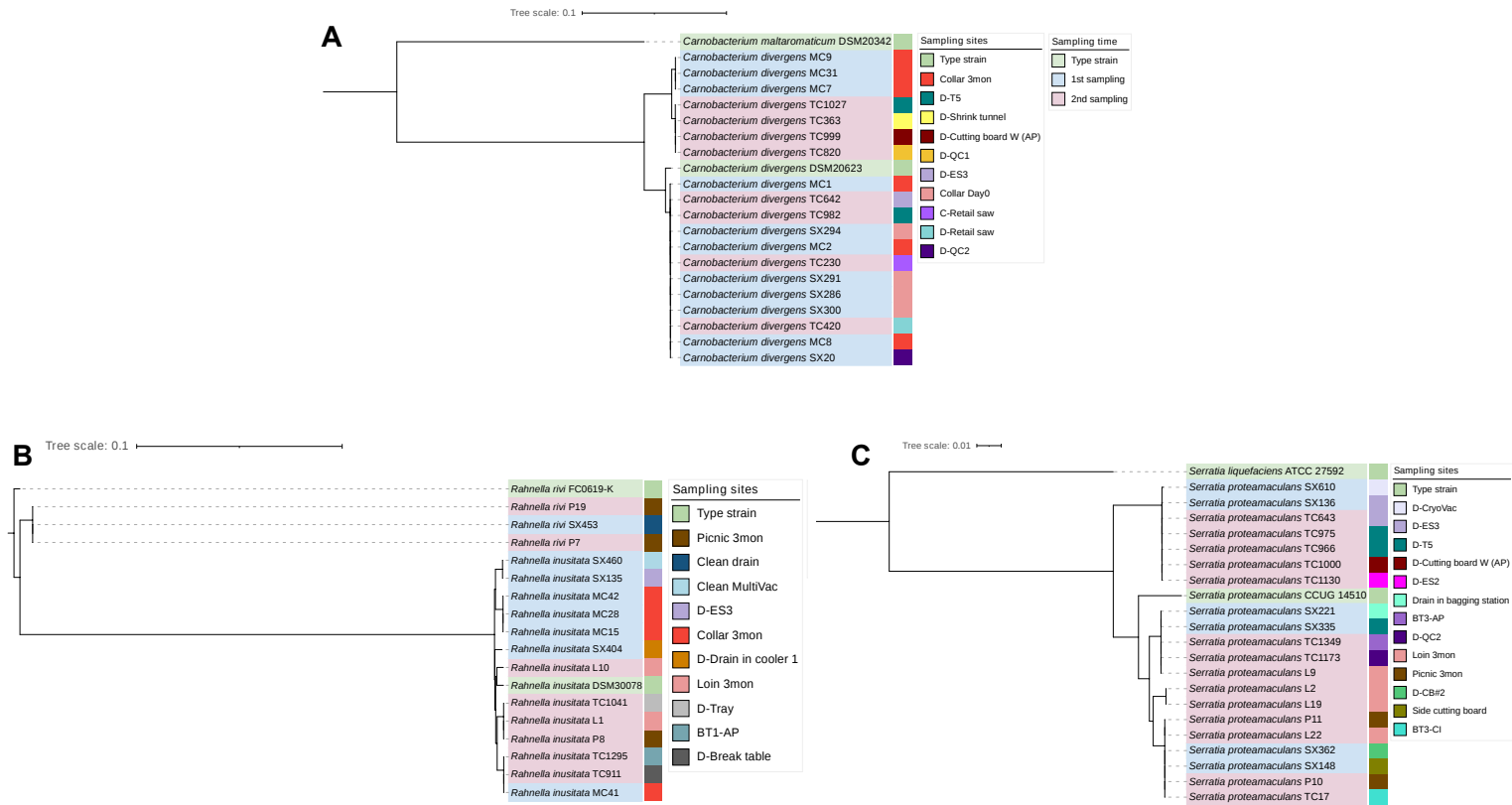


Figure S5. Phylogenetic tree of strains of *C. divergens* (A), *Rahnella* (B) and *S. proteamaculans* (C) isolated from a meat processing facility. The phylogeny was inferred based on core genome alignment, using the GTR+I+G4 model with 1000 bootstrap replicates. Strains are color coded based on sampling time (clades) and sampling sites (legend). Type strains of the target species and its closely related species as outgroups were included for tree visualization.