

Supplemental Information:

Table S1: Bacterial species identified in the smokeless tobacco products

<u>Product</u>	<u>AR1</u>	<u>DC1</u>	<u>AR2</u>	<u>DC2</u>	<u>AR3</u>	<u>DC3</u>
Brand A Straight	<i>Bacillus species</i> <i>B. licheniformis</i> <i>B. pumilus</i> <i>B. safensis</i>	<i>B. pumilus</i> <i>B. subtilis</i> <i>B. species</i> <i>B. safensis</i> <i>B. licheniformis</i>	<i>B. pumilus</i> <i>B. safensis</i> <i>B. species</i> <i>B. licheniformis</i> <i>B. subtilis</i>	<i>B. species</i> <i>B. licheniformis</i> <i>B. safensis</i> <i>B. pumilus</i> <i>B. altitudinis</i>	<i>B. pumilus</i> <i>B. safensis</i> <i>B. species</i> <i>B. subtilis</i> <i>B. licheniformis</i>	<i>B. species</i> <i>B. subtilis</i> <i>B. licheniformis</i> <i>B. sonorensis</i> <i>B. safensis</i>
Brand A Wintergreen	<i>B. licheniformis</i> <i>B. subtilis</i> <i>B. amyloliquefaciens</i> <i>B. species</i> <i>B. pumilus</i> <i>B. safensis</i> <i>Paenibacillus species</i> <i>unclassified Bacillales</i>	<i>B. pumilus</i> <i>B. subtilis</i> <i>B. licheniformis</i> <i>B. clausii</i> <i>B. safensis</i> <i>B. species</i> <i>Thiomargarita namibiensis</i>	<i>B. pumilus</i> <i>B. licheniformis</i> <i>B. species</i> <i>B. safensis</i> <i>B. aerophilus</i>	<i>B. pumilus</i> <i>B. species</i> <i>B. safensis</i> <i>B. subtilis</i> <i>B. tequilensis</i> <i>B. licheniformis</i>	<i>B. species</i> <i>B. licheniformis</i> <i>B. pumilus</i> <i>B. safensis</i>	<i>B. species</i> <i>B. licheniformis</i> <i>B. sonorensis</i> <i>B. pumilus</i>
Brand B Straight	<i>B. species</i> <i>B. licheniformis</i> <i>B. mojavensis</i> <i>B. amyloliquefaciens</i> <i>B. safensis</i> <i>B. subtilis</i> <i>B. pumilus</i> <i>Tetragenococcus halophilus</i>	<i>B. pumilus</i> <i>B. safensis</i> <i>B. subtilis</i> <i>B. species</i> <i>B. licheniformis</i>	<i>B. pumilus</i> <i>B. species</i> <i>B. licheniformis</i> <i>B. safensis</i> <i>B. aerophilus</i> <i>B. aerophilus</i> <i>B. altitudinis</i>	<i>B. subtilis</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. species</i> <i>B. licheniformis</i>	<i>B. pumilus</i> <i>B. species</i> <i>B. licheniformis</i> <i>B. safensis</i>	<i>B. species</i> <i>B. licheniformis</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. amyloliquefaciens</i> <i>B. subtilis</i>

	<i>Tetragenococcus species</i> <i>unclassified Bacillales</i>					
Brand B Wintergreen	<i>B. pumilus</i> <i>B. species</i> <i>B. safensis</i> <i>B. licheniformis</i> <i>Oceanobacillus oncorhynchi</i> <i>Oceanobacillus species</i> <i>unclassified Bacillales</i>	<i>B. pumilus</i> <i>B. species</i> <i>B. licheniformis</i> <i>B. subtilis</i> <i>B. safensis</i>	<i>B. licheniformis</i> <i>B. pumilus</i> <i>B. species</i> <i>B. subtilis</i> <i>B. sonorensis</i> <i>B. aerius</i> <i>B. safensis</i> <i>B. aerophilus</i>	<i>B. species</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. licheniformis</i>	<i>B. species</i> <i>B. licheniformis</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. cereus</i>	<i>B. species</i> <i>B. licheniformis</i> <i>B. pumilus</i> <i>B. safensis</i>
Brand C Wintergreen	<i>B. species</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. licheniformis</i> <i>B. subtilis</i> <i>B. aerophilus</i> <i>B. sonorensis</i> <i>unclassified Bacillales</i>	<i>B. species</i> <i>B. pumilus</i> <i>B. subtilis</i> <i>B. safensis</i> <i>B. licheniformis</i> <i>B. megaterium</i> <i>Oceanobacillus species</i>	<i>B. licheniformis</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. sonorensis</i> <i>B. species</i> <i>Brevibacillus species</i> <i>Terribacillus species</i>	<i>B. species</i> <i>B. pumilus</i> <i>B. megaterium</i> <i>B. licheniformis</i> <i>B. sonorensis</i> <i>B. safensis</i>	<i>B. species</i> <i>B. pumilus</i> <i>B. licheniformis</i> <i>B. safensis</i>	<i>B. species</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. licheniformis</i> <i>Paenibacillus species</i>
Brand D Straight	<i>Staphylococcus species</i> <i>B. pumilus</i> <i>B. species</i> <i>B. licheniformis</i> <i>B. subtilis</i> <i>B. sonorensis</i> <i>B. species</i>	<i>B. licheniformis</i> <i>B. species</i> <i>B. sonorensis</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. subtilis</i>	<i>B. species</i> <i>B. pumilus</i> <i>B. species</i> <i>B. licheniformis</i> <i>B. sonorensis</i> <i>B. safensis</i>	<i>B. pumilus</i> <i>B. species</i> <i>B. sonorensis</i> <i>B. licheniformis</i> <i>B. subtilis</i> <i>Uncultured bacterium</i>	<i>B. species</i> <i>B. altitudinis</i> <i>B. pumilus</i> <i>B. licheniformis</i> <i>B. subtilis</i>	<i>B. species</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. licheniformis</i>

	<i>unclassified Bacillales</i>					
Brand B Pouches	<i>B. licheniformis</i> <i>B. species</i> <i>B. safensis</i> <i>B. pumilus</i> <i>B. subtilis</i> <i>unclassified Bacillales</i>	<i>B. pumilus</i> <i>B. species</i> <i>B. safensis</i> <i>B. clausii</i> <i>B. licheniformis</i>	<i>B. pumilus</i> <i>B. safensis</i> <i>B. species</i> <i>B. subtilis</i> <i>B. licheniformis</i> <i>B. aerophilus</i> <i>Paenibacillus species</i>	<i>B. species</i> <i>B. pumilus</i> <i>B. licheniformis</i> <i>B. altitudinis</i> <i>B. tequilensis</i>	<i>B. licheniformis</i> <i>B. species</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. sonorensis</i> <i>B. licheniformis</i>	<i>B. species</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. sonorensis</i> <i>B. licheniformis</i>
Brand A Pouches	<i>B. sonorensis</i> <i>B. species</i> <i>B. licheniformis</i> <i>B. safensis</i> <i>B. pumilus</i> <i>B. amylolyquefaciens</i> <i>Tetragenococcus halophilus</i> <i>unclassified Bacillales</i>	<i>B. aerius</i> <i>B. licheniformis</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. species</i> <i>B. subtilis</i>	<i>B. pumilus</i> <i>B. species</i> <i>B. licheniformis</i> <i>B. safensis</i>	<i>B. species</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. subtilis</i> <i>B. tequilensis</i> <i>B. licheniformis</i>	<i>B. species</i> <i>B. pumilus</i> <i>B. safensis</i>	<i>B. species</i> <i>B. pumilus</i> <i>B. sonorensis</i> <i>B. safensis</i>
Brand B Mint Pouches	<i>B. licheniformis</i> <i>B. species</i> <i>B. safensis</i> <i>B. pumilus</i> <i>Tetragenococcus species</i>	<i>B. safensis</i> <i>B. pumilus</i> <i>B. licheniformis</i> <i>B. species</i> <i>B. circulans</i> <i>Terribacillus species</i> <i>Tetragenococcus species</i>	<i>B. aerophilus</i> <i>B. clausii</i> <i>B. licheniformis</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. species</i> <i>Geobacillus species</i> <i>Ornithinibacillus species</i>	<i>B. species</i> <i>B. safensis</i> <i>B. pumilus</i> <i>B. aerophilus</i> <i>Thermus species</i>	<i>B. pumilus</i> <i>B. safensis</i> <i>B. species</i> <i>B. licheniformis</i>	<i>B. pumilus</i> <i>B. safensis</i> <i>B. sonorensis</i> <i>B. licheniformis</i> <i>B. species</i>

Brand E Snus Mellow	<i>B. species</i>	<i>B. infantis</i>	<i>B. pumilus</i>	<i>B. pumilus</i>	<i>None Identified</i>	<i>None Identified</i>
	<i>B. pumilus</i>	<i>B. licheniformis</i>	<i>B. safensis</i>			
	<i>B. licheniformis</i>	<i>Uncultured bacterium</i>	<i>Staphylococcus hominis</i>			
		<i>Staphylococcus epidermidis</i>	<i>Staphylococcus epidermidis</i>			
Brand F Snus Mint	<i>B. species</i>	<i>Staphylococcus capitis</i>	<i>Staphylococcus species</i>	<i>None Identified</i>	<i>None Identified</i>	<i>None Identified</i>
		<i>Staphylococcus hominis</i>				
Brand B Snus Mint	<i>B. species</i>	<i>B. niabensis</i>	<i>Oceanobacillus species</i>	<i>None Identified</i>	<i>B. species</i>	<i>None Identified</i>
	<i>Staphylococcus epidermidis</i>				<i>B. safensis</i>	
					<i>B. pumilus</i>	
Brand G Chewing Tobacco	<i>B. pumilus</i>	<i>B. species</i>	<i>B. pumilus</i>	<i>B. pumilus</i>	<i>B. pumilus</i>	<i>B. safensis</i>
	<i>B. species</i>	<i>B. pumilus</i>	<i>B. safensis</i>	<i>B. species</i>		<i>B. pumilus</i>
	<i>B. safensis</i>	<i>B. safensis</i>	<i>B. species</i>	<i>B. safensis</i>		<i>B. licheniformis</i>
	<i>B. licheniformis</i>	<i>B. licheniformis</i>	<i>B. licheniformis</i>	<i>B. megaterium</i>		<i>B. species</i>
	<i>B. megaterium</i>	<i>B. subtilis</i>	<i>B. subtilis</i>	<i>B. licheniformis</i>		
	<i>Gracilibacillus species</i>	<i>B. clausii</i>	<i>B. aerophilus</i>	<i>Oceanobacillus oncorhynchi</i>		
	<i>Oceanobacillus species</i>	<i>Paenibacillus species</i>	<i>Oceanobacillus species</i>			
		<i>Geobacillus species</i>				
		<i>Oceanobacillus oncorhynchi</i>				
		<i>Oceanobacillus iheyensis</i>				
Brand H Chewing Tobacco	<i>B. licheniformis</i>	<i>B. clausii</i>	<i>B. pumilus</i>	<i>B. licheniformis</i>	<i>None Identified</i>	<i>B. cereus</i>
	<i>B. sonorensis</i>	<i>B. licheniformis</i>	<i>Oceanobacillus species</i>	<i>B. pumilus</i>		
	<i>B. species</i>	<i>Oceanobacillus species</i>	<i>B. clausii</i>	<i>B. species</i>		
	<i>B. pumilus</i>		<i>B. aerophilus</i>	<i>Micrococcus luteus</i>		
	<i>Oceanobacillus species</i>			<i>Staphylococcus cohnii</i>		

				<i>Staphylococcus hominis</i>		
Brand G Golden Blend Chewing Tobacco	<i>B. species</i> <i>B. licheniformis</i> <i>B. subtilis</i> <i>B. pumilus</i> <i>B. safensis</i> <i>Oceanobacillus species</i> unclassified <i>Bacillales</i>	<i>B. clausii</i> <i>B. species</i> <i>B. licheniformis</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. sonorensis</i> <i>Lysinibacillus fusiformis</i> <i>Paenibacillus species</i> <i>Oceanobacillus species</i> <i>Oceanobacillus iheyensis</i> <i>Staphylococcus epidermidis</i>	<i>B. pumilus</i> <i>B. safensis</i> <i>B. subtilis</i> <i>B. species</i> <i>B. licheniformis</i> <i>B. clausii</i>	<i>B. aerius</i> <i>B. clausii</i> <i>B. licheniformis</i> <i>B. pumilus</i> <i>B. safensis</i> <i>B. species</i> <i>Oceanobacillus oncorynchi</i>	<i>B. safensis</i> <i>B. pumilus</i> <i>B. licheniformis</i>	<i>B. safensis</i> <i>B. pumilus</i> <i>B. licheniformis</i> <i>B. species</i>

Note: Colors are shown to highlight major taxonomic groups. The light purple highlighted isolates are those that did not fall into another major group.

Table S2: MG-RAST Accession Information and Sequence Counts

Sample	Sample ID	Metagenome Name	MG-RAST Accession #: V1/V2	Sequence Count: V1/V2	Shannon Diversity Index: V2	MG-RAST Accession #: V6	Sequence Count: V6	Shannon Diversity Index: V6
Brand A Straight AR1	AR1_1	NCTR1	4573045.3	6,765	4.73	4582872.3	13,717	4.55
Brand A Straight AR2	AR2_1	NCTR17	4573052.3	56,203	4.26	4582879.3	30,407	12.38
Brand A Straight AR3	AR3_1	NCTR32	4573069.3	17,700	3.80	4582896.3	53,568	10.95
Brand A Straight DC1	DC1_1	CTP1	4573000.3	16,262	4.78	4582827.3	9,955	1.74
Brand A Straight DC2	DC2_1	CTP25	4573017.3	17,858	4.83	4582844.3	38,942	10.89
Brand A Straight DC3	DC3_1	CTP31	4573024.3	18,627	4.17	4582851.3	45	7.48
Brand A Wintergreen AR1	AR1_2	NCTR2	4573055.3	11,809	2.83	4582882.3	11,333	11.14
Brand A Wintergreen AR2	AR2_2	NCTR18	4573053.3	19,374	5.93	4582880.3	17,982	11.53
Brand A Wintergreen AR3	AR3_2	NCTR33	4573070.3	2,942	12.78	4582897.3	20,652	12.36
Brand A Wintergreen DC1	DC1_2	CTP2	4573011.3	12,267	2.40	4582838.3	18,193	5.44
Brand A Wintergreen DC2	DC2_2	CTP17	4573008.3	45,400	4.03	4582835.3	27,024	9.06
Brand A Wintergreen DC3	DC3_2	CTP33	4573026.3	24,327	2.27	4582853.3	34,251	11.60
Brand B Straight AR1	AR1_3	NCTR3	4573066.3	6,945	7.52	4582893.3	12,476	6.03
Brand B Straight AR2	AR2_3	NCTR19	4573054.3	60,792	6.46	4582881.3	21,146	9.22
Brand B Straight AR3	AR3_3	NCTR34	4573071.3	8,485	16.78	4582898.3	11,971	10.44

Brand B Straight								
DC1	DC1_3	CTP3	4573022.3	15,791	3.32	4582849.3	16,745	1.37
Brand B Straight								
DC2	DC2_3	CTP18	4573009.3	23,108	4.35	4582836.3	26,501	7.83
Brand B Straight								
DC3	DC3_3	CTP32	4573025.3	19,785	3.04	4582852.3	39,607	6.82
Brand B								
Wintergreen AR1	AR1_4	NCTR4	4573077.3	7,190	2.65	4582904.3	9,934	5.61
Brand B								
Wintergreen AR2	AR2_4	NCTR20	4573056.3	15,127	6.61	4582883.3	20,874	13.27
Brand B								
Wintergreen AR3	AR3_4	NCTR35	4573072.3	28,242	6.32	4582899.3	20,501	13.29
Brand B								
Wintergreen DC1	DC1_4	CTP4	4573033.3	14,598	2.33	4582860.3	12,256	2.17
Brand B								
Wintergreen DC2	DC2_4	CTP19	4573010.3	6,375	12.70	4582837.3	28,175	9.58
Brand B								
Wintergreen DC3	DC3_4	CTP34	4573027.3	17,390	3.34	4582854.3	11,262	11.19
Brand C								
Wintergreen AR1	AR1_5	NCTR6	4573086.3	9,713	11.12	4582913.3	11,448	10.86
Brand C								
Wintergreen AR2	AR2_5	NCTR21	4573057.3	11,049	6.56	4582884.3	12,425	10.59
Brand C								
Wintergreen AR3	AR3_5	NCTR36	4573073.3	34,846	11.14	4582900.3	20,957	11.74
Brand C								
Wintergreen DC1	DC1_5	CTP9	4573044.3	26	1.00	4582871.3	9,099	12.43
Brand C								
Wintergreen DC2	DC2_5	CTP24	4573016.3	14,259	9.76	4582843.3	17,227	11.77
Brand C								
Wintergreen DC3	DC3_5	CTP35	4573028.3	24,232	5.76	4582855.3	64,404	13.10
Brand D Straight								
AR1	AR1_6	NCTR7	4573087.3	14,041	5.19	4582914.3	11,273	12.75
Brand D Straight								
AR2	AR2_6	NCTR22	4573058.3	2,519	6.99	4582885.3	15,634	7.51
Brand D Straight								
AR3	AR3_6	NCTR37	4573074.3	16,226	12.56	4582901.3	46,772	9.11

AR3

Brand D Straight								
DC1	DC1_6	CTP5	4573040.3	9,543	3.27	4582867.3	14,137	2.96
Brand D Straight								
DC2	DC2_6	CTP20	4573012.3	15,308	7.23	4582839.3	24,896	10.14
Brand D Straight								
DC3	DC3_6	CTP36	4573029.3	21,234	7.24	4582856.3	8,241	9.41
Brand B Pouches								
AR1	AR1_7	NCTR8	4573088.3	39,161	10.12	4582915.3	10,870	5.30
Brand B Pouches								
AR2	AR2_7	NCTR23	4573059.3	14,699	11.58	4582886.3	26,794	10.32
Brand B Pouches								
AR3	AR3_7	NCTR38	4573075.3	26,886	10.50	4582902.3	27,548	13.73
Brand B Pouches								
DC1	DC1_7	CTP6	4573041.3	13,037	2.45	4582868.3	13,020	3.08
Brand B Pouches								
DC2	DC2_7	CTP21	4573013.3	27,726	3.72	4582840.3	22,190	4.32
Brand B Pouches								
DC3	DC3_7	CTP38	4573031.3	26,322	9.35	4582858.3	12,384	9.63
Brand A Pouches								
AR1	AR1_8	NCTR9	4573089.3	9,258	3.27	4582916.3	13,538	6.99
Brand A Pouches								
AR2	AR2_8	NCTR24	4573060.3	98,786	15.06	4582887.3	36,552	11.21
Brand A Pouches								
AR3	AR3_8	NCTR39	4573076.3	26,562	8.79	4582903.3	23,577	10.71
Brand A Pouches								
DC1	DC1_8	CTP10	4573001.3	14,740	2.65	4582828.3	11,097	9.31
Brand A Pouches								
DC2	DC2_8	CTP16	4573007.3	47,752	4.90	4582834.3	20	7.00
Brand A Pouches								
DC3	DC3_8	CTP37	4573030.3	25,639	12.36	4582857.3	37,162	13.54
Brand B Mint								
Pouches AR1	AR1_9	NCTR41	4574738.3	1,087	4.80	4582911.3	12,007	12.69
Brand B Mint								
Pouches AR2	AR2_9	NCTR25	4573061.3	64,245	8.87	4582888.3	30,770	9.37

Brand B Mint								
Pouches AR3	AR3_9	NCTR40	4573078.3	25,706	8.03	4582905.3	13,644	14.51
Brand B Mint								
Pouches DC1	DC1_9	CTP11	4573002.3	12,238	5.12	4582829.3	10,097	3.83
Brand B Mint								
Pouches DC2	DC2_9	CTP26	4573018.3	29,321	4.21	4582845.3	48,986	11.93
Brand B Mint								
Pouches DC3	DC3_9	CTP39	4573032.3	26,684	2.99	4582859.3	16,517	14.27
Brand E Snus								
Mellow AR1	AR1_10	NCTR10	4573046.3	10,256	1.80	4582873.3	13,222	12.83
Brand E Snus								
Mellow AR2	AR2_10	NCTR26	4573062.3	7,508	6.10	4582889.3	23,182	9.47
Brand E Snus								
Mellow AR3	AR3_10	NCTR42	4573080.3	18,799	8.70	4582906.3	14,370	8.86
Brand E Snus								
Mellow DC1	DC1_10	CTP7	4573042.3	10,502	8.52	4582869.3	9,354	10.38
Brand E Snus								
Mellow DC2	DC2_10	CTP22	4573014.3	27,993	9.69	4582841.3	22,568	10.18
Brand E Snus								
Mellow DC3	DC3_10	CTP40	4573034.3	18,002	2.50	4582861.3	12,672	10.13
Brand F Snus Mint								
AR1	AR1_11	NCTR11	4573047.3	5,907	5.95	4582874.3	14,783	13.90
Brand F Snus Mint								
AR2	AR2_11	NCTR27	4573063.3	7,287	3.97	4582890.3	15,594	8.54
Brand F Snus Mint								
AR3	AR3_11	NCTR43	4573081.3	21,958	10.78	4582907.3	14,760	7.12
Brand F Snus Mint								
DC1	DC1_11	CTP15	4573006.3	32,965	11.44	4582833.3	22,321	4.81
Brand F Snus Mint								
DC2	DC2_11	CTP30	4573023.3	14,899	7.31	4582850.3	20,100	9.14
Brand F Snus Mint								
DC3	DC3_11	CTP41	4573035.3	20,333	14.82	4582862.3	27,479	11.33
Brand B Snus Mint								
AR1	AR1_12	NCTR12	4573048.3	9,001	2.95	4582875.3	14,980	12.21
Brand B Snus Mint								
AR2	AR2_12	NCTR28	4573064.3	11,190	5.53	4582891.3	26,663	8.83

AR2

Brand B Snus Mint								
AR3	AR3_12	NCTR44	4573082.3	30,919	11.14	4582908.3	15,525	7.89
Brand B Snus Mint								
DC1	DC1_12	CTP12	4573003.3	3,612	7.75	4582830.3	13,619	13.00
Brand B Snus Mint								
DC2	DC2_12	CTP27	4573019.3	119,437	3.86	4582846.3	13,630	9.47
Brand B Snus Mint								
DC3	DC3_12	CTP42	4573036.3	22,727	9.25	4582863.3	23,238	9.60
Brand G Chewing								
Tobacco AR1	AR1_13	NCTR13	4573049.3	42,733	7.68	4582876.3	13,269	18.73
Brand G Chewing								
Tobacco AR2	AR2_13	NCTR29	4573065.3	3,801	8.35	4582892.3	19,775	10.31
Brand G Chewing								
Tobacco AR3	AR3_13	NCTR45	4573083.3	22,288	11.29	4582909.3	12,246	10.59
Brand G Chewing								
Tobacco DC1	DC1_13	CTP8	4573043.3	18,130	10.23	4582870.3	9,959	15.61
Brand G Chewing								
Tobacco DC2	DC2_13	CTP23	4573015.3	8,179	11.74	4582842.3	23,284	6.57
Brand G Chewing								
Tobacco DC3	DC3_13	CTP43	4573037.3	913	4.75	4582864.3	12,659	7.91
Brand H Chewing								
Tobacco AR1	AR1_14	NCTR14	4573050.3	13,388	7.61	4582877.3	11,227	15.81
Brand H Chewing								
Tobacco AR2	AR2_14	NCTR30	4573067.3	2,043	11.93	4582894.3	17,036	8.20
Brand H Chewing								
Tobacco AR3	AR3_14	NCTR46	4573084.3	29,054	5.61	4582910.3	13,063	9.09
Brand H Chewing								
Tobacco DC1	DC1_14	CTP13	4573004.3	9,669	3.61	4582831.3	26,104	7.34
Brand H Chewing								
Tobacco DC2	DC2_14	CTP28	4573020.3	815	1.89	4582847.3	19,415	8.32
Brand H Chewing								
Tobacco DC3	DC3_14	CTP44	4573038.3	18,450	8.71	4582865.3	19,414	11.29
Brand G Golden								
Blend Chewing	AR1_15	NCTR16	4573051.3	52,779	6.74	4582878.3	12,894	13.26

Tobacco AR1								
Brand G Golden Blend Chewing								11.04
Tobacco AR2	AR2_15	NCTR31	4573068.3	33,419	7.93	4582895.3	36,023	
Brand G Golden Blend Chewing								12.36
Tobacco AR3	AR3_15	NCTR5	4573085.3	10,897	4.01	4582912.3	6,062	
Brand G Golden Blend Chewing								21.27
Tobacco DC1	DC1_15	CTP14	4573005.3	13,742	3.69	4582832.3	30,097	
Brand G Golden Blend Chewing								9.85
Tobacco DC2	DC2_15	CTP29	4573021.3	15,259	11.09	4582848.3	15,755	
Brand G Golden Blend Chewing								11.07
Tobacco DC3	DC3_15	CTP45	4573039.3	21,852	19.07	4582866.3	12,555	

Table S3: Results of the DGGE analysis of microbial population changes

Product	Visual Changes in DGGE Profiles During 15-Day Period?					
	AR1	DC1	AR2	DC2	AR3	DC3
Brand A Straight	Yes	No	No	No	No	No
Brand A Wintergreen	Yes	No	No	No	Yes	No
Brand B Straight	No	No	No	No	Yes	No
Brand B Wintergreen	No	No	No	No	Yes	No
Brand C Wintergreen	No	No	No	Yes	No	No
Brand D Straight	No	No	No	No	Yes	Yes
Brand B Pouches	No	No	Yes	Yes	Yes	No
Brand A Pouches	Yes	Yes	Yes	No	Yes	No
Brand B Mint Pouches	Yes	Yes	No	No	No	Yes
Brand E Snus Mellow	No	No	No	No	No	No
Brand F Snus Mint	No	No	No	No	No	No
Brand B Snus Mint	No	No	No	No	No	No
Brand G Chewing Tobacco	Yes	No	Yes	No	No	No
Brand H Chewing Tobacco	Yes	No	No	No	No	No
Brand G Golden Blend Chewing Tobacco	Yes	No	No	No	No	No

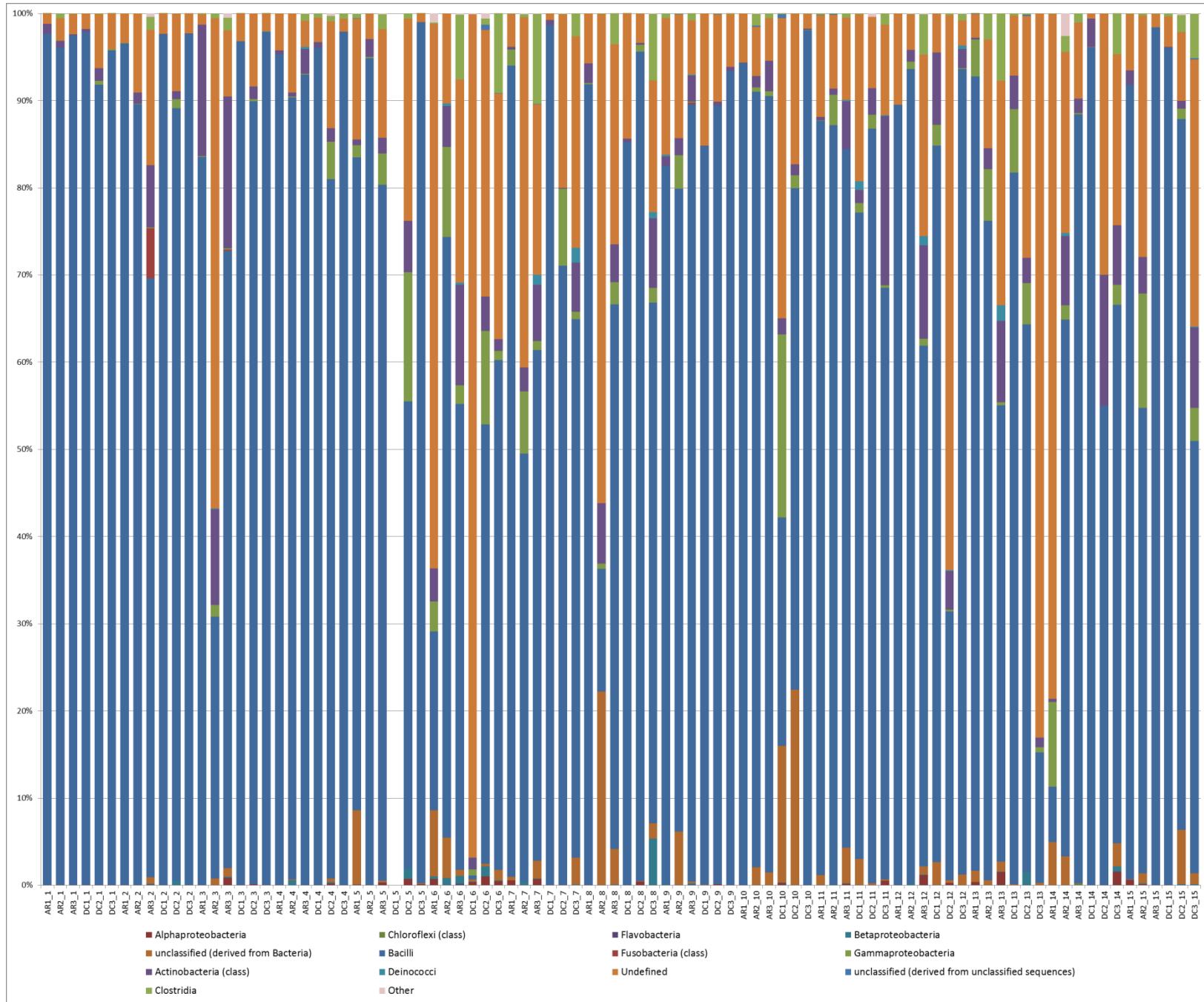


Figure S1: Results of pyrosequencing for the V1/V2 region of the 16S rRNA for each of the different types of smokeless tobacco samples analyzed in the study. The sample names are presented on the bottom with the sampling period (AR1, DC1, etc.) and sample type (1, 2,...14, 15), which correspond to the sample type (i.e., 1 = Brand A Straight, etc; See Supplementary Table 3 for a key). The bar graph indicated the percent of sequences detected that belonged to a particular bacterial class. The classes of organisms and color schemes are presented on the bottom of the figure. The DNA sequences identified as plant material have been removed due to their likely non-microbial origins.

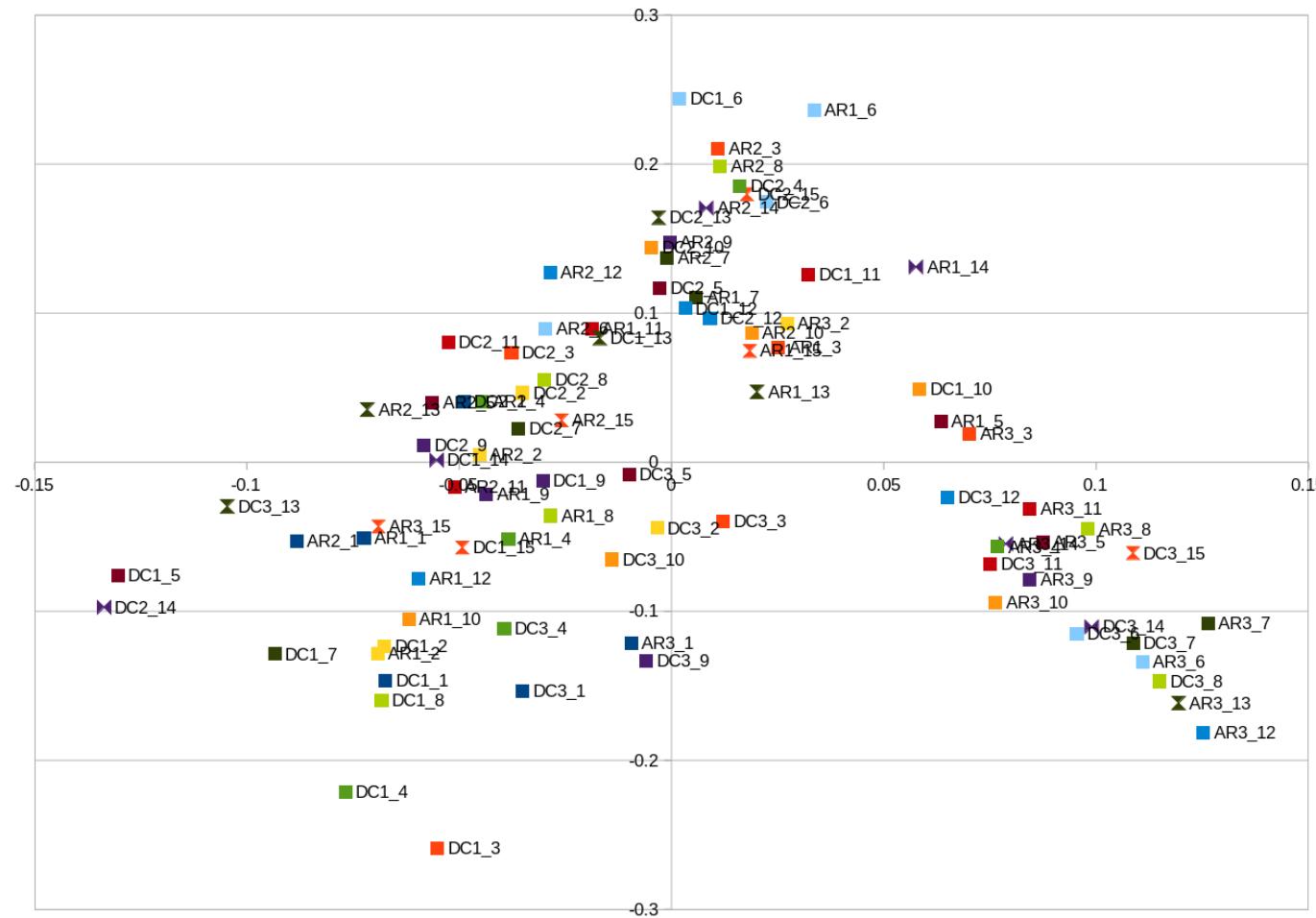
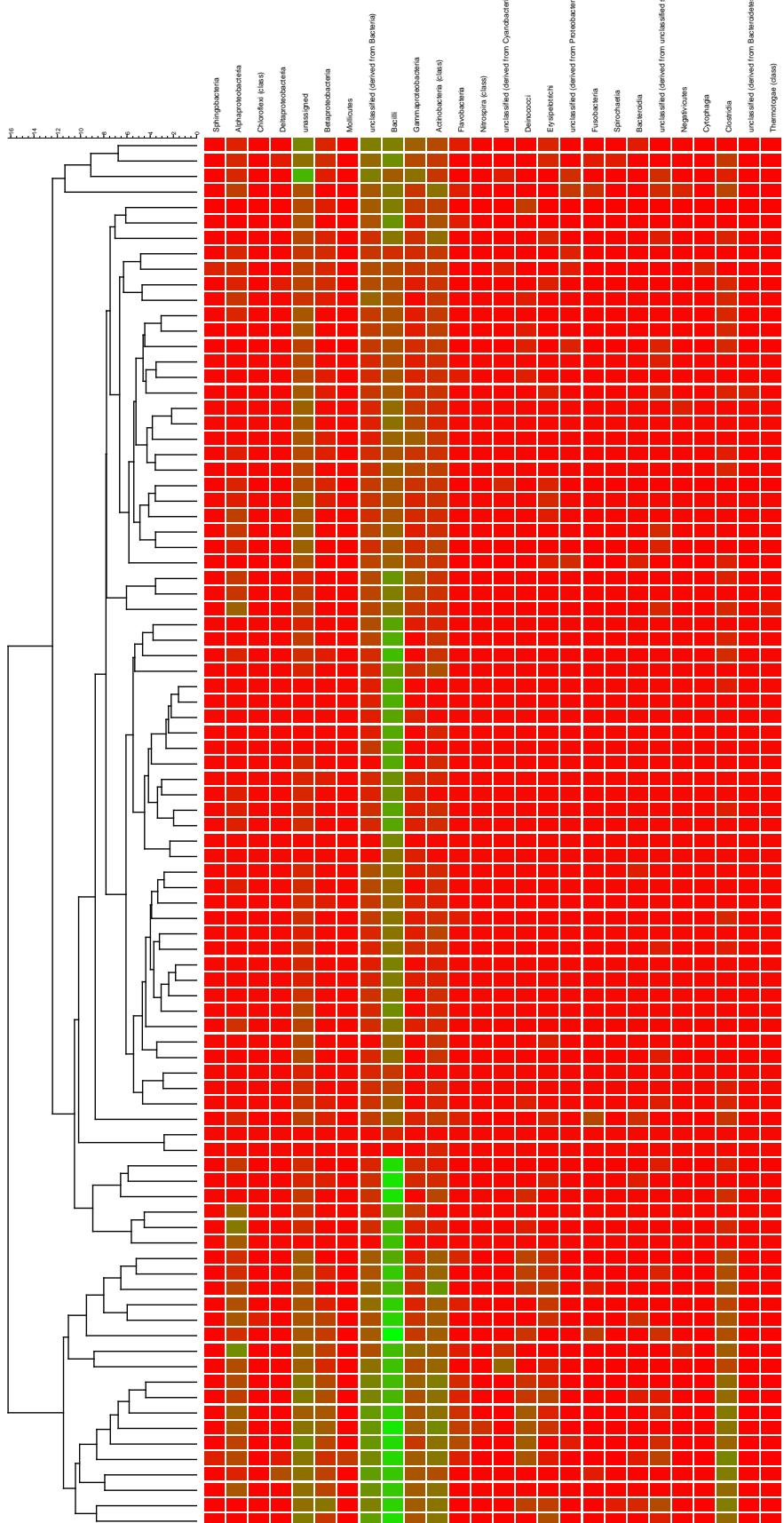


Figure S2: PCoA plot generated based on the Class-level analysis of the V2 region of the 16S rRNA for each of the different types of smokeless tobacco samples analyzed in the study. The sample names are presented on the bottom with the sampling period (AR1, DC1, etc.) and sample type (1, 2,...14, 15), which correspond to the sample type (i.e., 1 = Brand A Straight, etc; See Supplementary Table 3 for a key).

Greengenes Class

Greengenes Class



Sample ID	Product (2)	MG RAST ID
DC1_10	Snus 1 DC1	4573042.3
AR1_6	Loose Moist Snuff 6 AR1	4573086.3
AR1_14	Chewing Tobacco 2 AR1	4573050.3
AR3_3	Loose Moist Snuff 3 AR3	4573071.3
DC1_11	Snus 2 DC1	4573006.3
AR1_15	Chewing Tobacco 3 AR1	4573051.3
AR1_3	Loose Moist Snuff 3 AR1	4573066.3
DC1_6	Loose Moist Snuff 6 DC1	4573040.3
AR1_7	Moist Snuff Pouch 1 AR1	4573087.3
DC2_6	Loose Moist Snuff 6 DC2	4573012.3
DC2_12	Snus 3 DC2	4573019.3
DC2_5	Loose Moist Snuff 5 DC2	4573016.3
AR2_3	Loose Moist Snuff 3 AR2	4573054.3
AR2_14	Chewing Tobacco 2 AR2	4573067.3
DC2_11	Snus 2 DC2	4573023.3
AR2_6	Loose Moist Snuff 6 AR2	4573058.3
AR2_7	Moist Snuff Pouch 1 AR2	4573059.3
DC2_2	Loose Moist Snuff 2 DC2	4573008.3
DC2_7	Moist Snuff Pouch 1 DC2	4573013.3
AR2_9	Moist Snuff Pouch 3 AR2	4573061.3
DC2_13	Chewing Tobacco 1 DC2	4573015.3
DC1_13	Chewing Tobacco 1 DC1	4573043.3
DC2_4	Loose Moist Snuff 4 DC2	4573010.3
AR2_15	Chewing Tobacco 3 AR2	4573068.3
DC2_10	Snus 3 DC2	4573014.3
DC1_12	Snus 1 DC1	4573003.3
AR2_8	Moist Snuff Pouch 2 AR2	4573060.3
DC2_15	Chewing Tobacco 3 DC2	4573021.3
AR1_13	Chewing Tobacco 1 AR1	4573049.3
AR1_8	Moist Snuff Pouch 2 AR1	4573088.3
AR2_10	Snus 1 AR2	4573062.3
DC1_9	Moist Snuff Pouch 3 DC1	4573002.3
DC3_5	Loose Moist Snuff 5 DC3	4573028.3
DC3_10	Snus 1 DC3	4573034.3
AR1_9	Moist Snuff Pouch 3 AR1	4573089.3
DC1_8	Moist Snuff Pouch 2 DC1	4573001.3
DC1_4	Loose Moist Snuff 4 DC1	4573033.3
AR1_2	Loose Moist Snuff 2 AR1	4573055.3
DC1_2	Loose Moist Snuff 2 DC1	4573011.3
DC1_1	Loose Moist Snuff 1 DC1	4573000.3
AR1_10	Snus 1 AR1	4573046.3
DC1_15	Chewing Tobacco 3 DC1	4573005.3
AR1_12	Snus 3 AR1	4573048.3
DC3_4	Loose Moist Snuff 4 DC3	4573027.3
AR1_4	Loose Moist Snuff 4 AR1	4573007.3
DC1_7	Moist Snuff Pouch 1 DC1	4573041.3
AR2_1	Loose Moist Snuff 1 AR2	4573052.3
AR2_2	Loose Moist Snuff 2 AR2	4573053.3
AR2_4	Loose Moist Snuff 4 AR2	4573056.3
DC2_1	Loose Moist Snuff 1 DC2	4573017.3
AR1_11	Snus 2 AR1	4573047.3
DC1_14	Chewing Tobacco 2 DC1	4573004.3
DC2_3	Loose Moist Snuff 3 DC2	4573009.3
AR1_1	Loose Moist Snuff 1 AR1	4573045.3
AR1_5	Loose Moist Snuff 5 AR1	4573085.3
DC2_9	Moist Snuff Pouch 3 DC2	4573018.3
AR2_11	Snus 2 AR2	4573063.3
DC2_8	Moist Snuff Pouch 2 DC2	4573007.3
AR2_5	Loose Moist Snuff 5 AR2	4573057.3
AR2_12	Snus 3 AR2	4573064.3
DC3_13	Chewing Tobacco 1 DC3	4573037.3
AR2_13	Chewing Tobacco 1 AR2	4573065.3
AR3_10	Snus 1 AR3	4574738.3
AR3_2	Loose Moist Snuff 2 AR3	4573070.3
DC2_14	Chewing Tobacco 2 DC2	4573020.3
DC1_5	Loose Moist Snuff 5 DC1	4573044.3
DC3_3	Loose Moist Snuff 3 DC3	4573025.3
DC3_2	Loose Moist Snuff 2 DC3	4573026.3
DC3_9	Moist Snuff Pouch 3 DC3	4573032.3
DC3_1	Loose Moist Snuff 1 DC3	4573024.3
AR3_1	Loose Moist Snuff 1 AR3	4573069.3
DC1_3	Loose Moist Snuff 3 DC1	4573022.3
DC3_12	Snus 3 DC3	4573036.3
AR3_4	Loose Moist Snuff 4 AR3	4573072.3
DC3_11	Snus 2 DC3	4573035.3
AR3_11	Snus 2 AR3	4573080.3
AR3_15	Chewing Tobacco 3 AR3	4573084.3
AR3_9	Moist Snuff Pouch 3 AR3	4573078.3
AR3_5	Loose Moist Snuff 5 AR3	4573073.3
AR3_12	Snus 3 AR3	4573081.3
DC3_15	Chewing Tobacco 3 DC3	4573039.3
AR3_6	Loose Moist Snuff 6 AR3	4573074.3
AR3_14	Chewing Tobacco 2 AR3	4573083.3
AR3_13	Chewing Tobacco 1 AR3	4573082.3
DC3_7	Moist Snuff Pouch 1 DC3	4573031.3
AR3_7	Moist Snuff Pouch 1 AR3	4573075.3
DC3_6	Loose Moist Snuff 6 DC3	4573029.3
DC3_14	Chewing Tobacco 2 DC3	4573038.3
DC3_8	Moist Snuff Pouch 2 DC3	4573030.3
AR3_8	Moist Snuff Pouch 2 AR3	4573076.3

Figure S3: V1/V2-based Unweighted Pair Group Method with Arithmetic Mean (UPGMA) dendrogram calculated based on the Euclidean distance of the normalized sequence abundance at the Class taxonomic unit. The red to green continuum ranges from 0.00 to 1.00 normalized abundance. The color blocks to the right of the normalized abundance are representative of the 15 different products analyzed during the study.

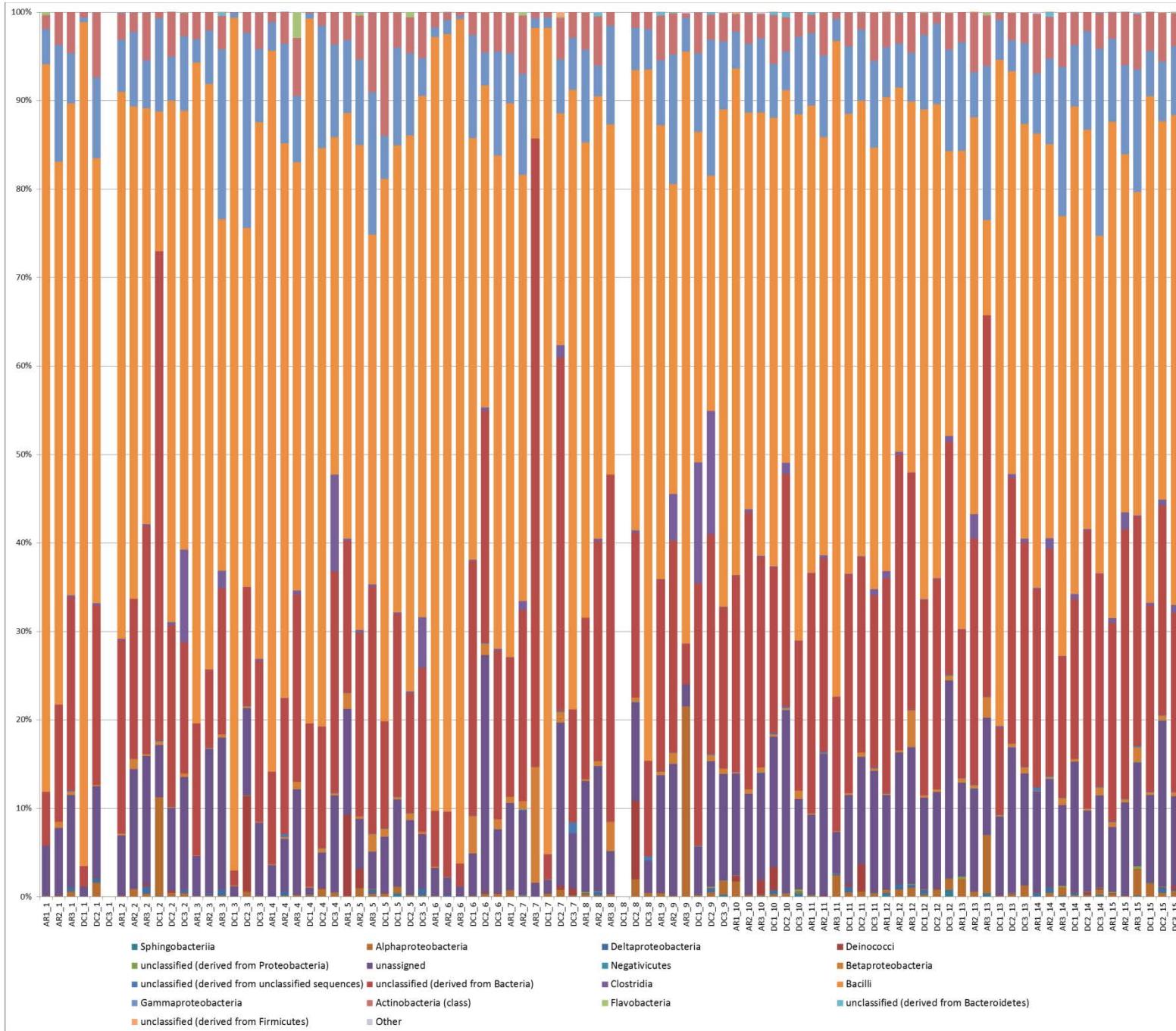


Figure S4: Results of pyrosequencing for the V6 region of the 16S rRNA for each of the different types of smokeless tobacco samples analyzed in the study. The sample names are presented on the bottom with the sampling period (AR1, DC1, etc.) and sample type (1, 2,...14, 15), which correspond to the sample type (i.e., 1 = Brand A Straight, etc; See Supplementary Table 3 for a key). The bar graph indicated the percent of sequences detected that belonged to a particular bacterial class. The classes of organisms and color schemes are presented on the bottom of the figure. The DNA sequences identified as plant material have been removed due to their likely non-microbial origins.

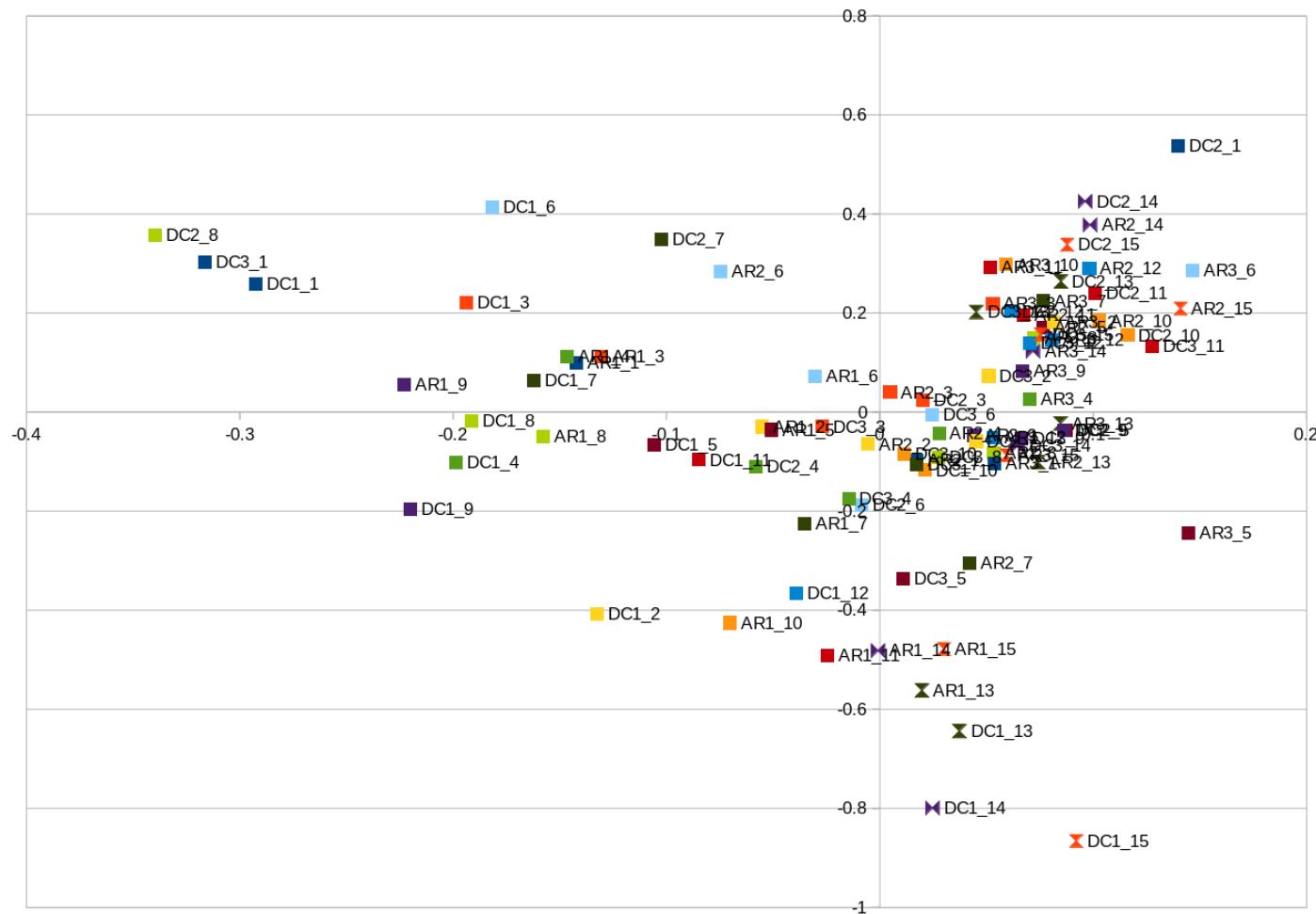


Figure S5: PCoA plot generated based on the Class-level analysis of the V6 region of the 16S rRNA for each of the different types of smokeless tobacco samples analyzed in the study. The sample names are presented on the bottom with the sampling period (AR1, DC1, etc.) and sample type (1, 2,...14, 15), which correspond to the sample type (i.e., 1 = Brand A Straight, etc; See Supplementary Table 3 for a key).

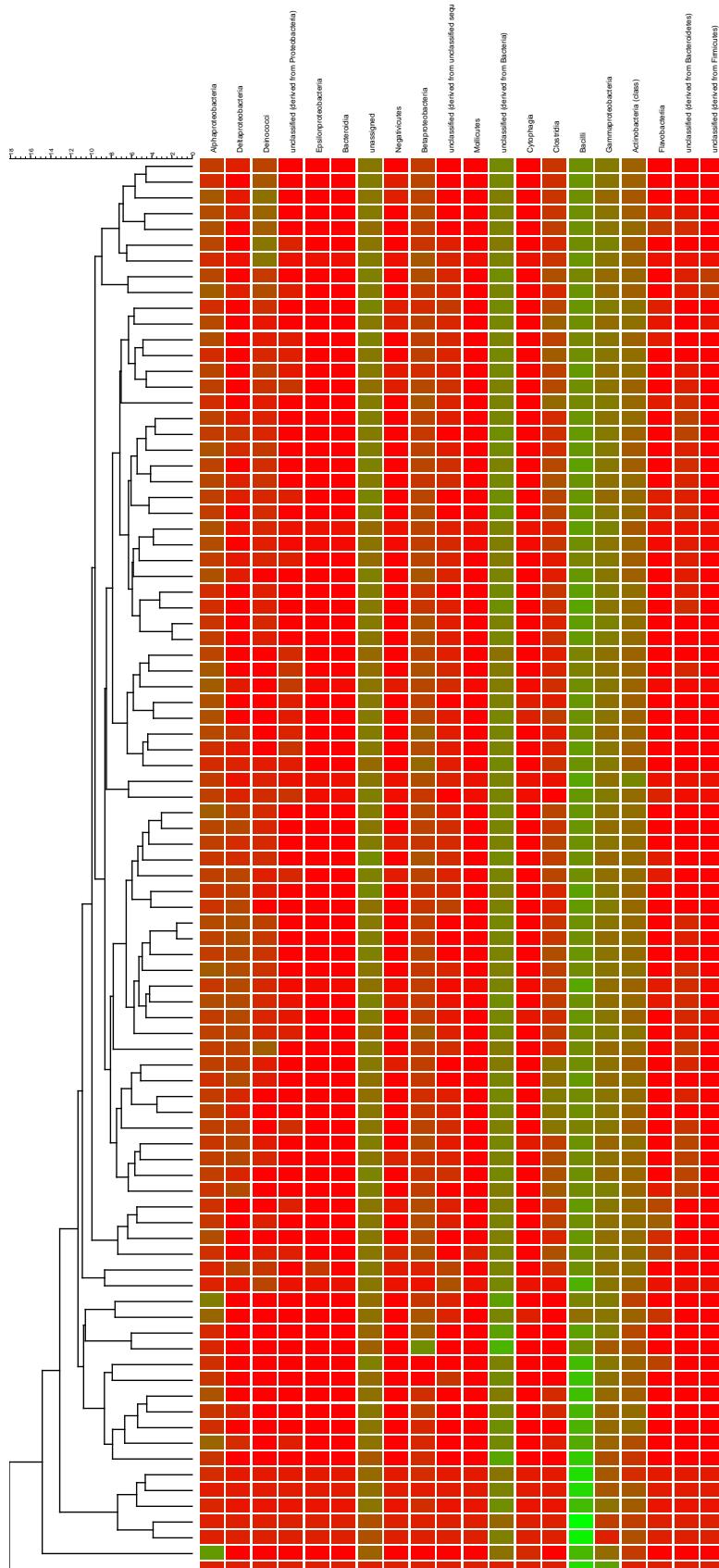
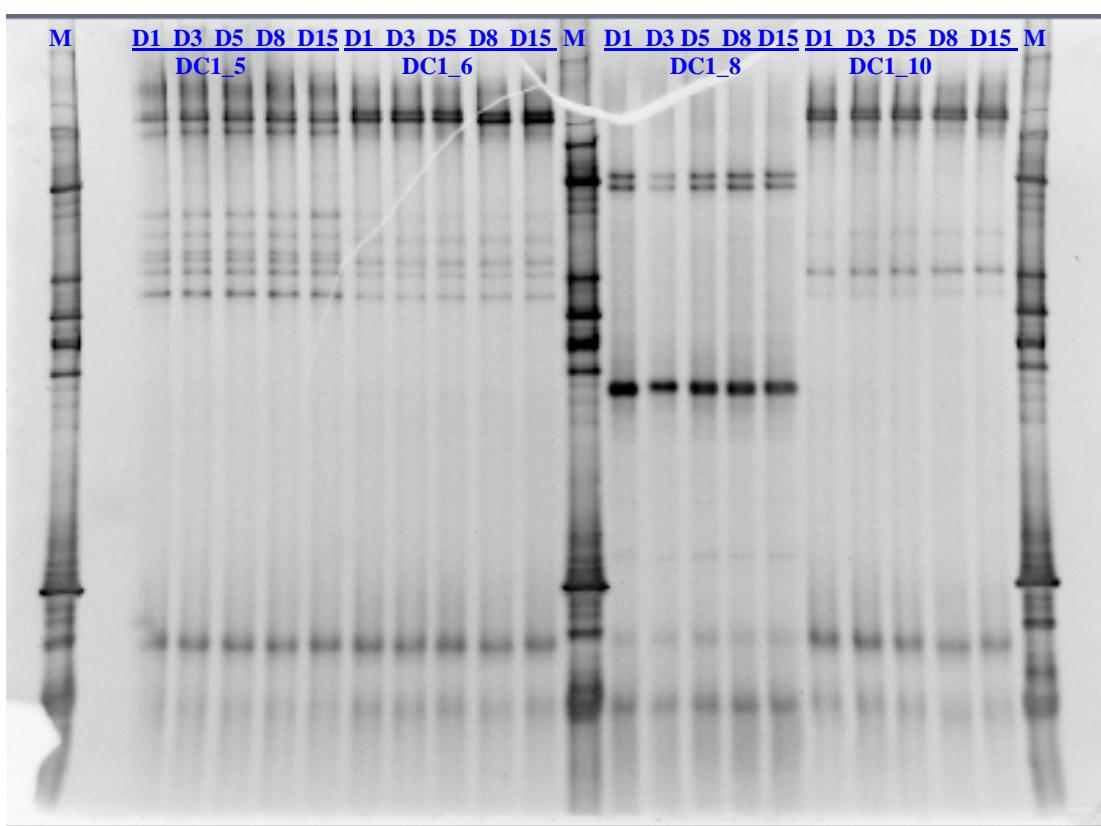
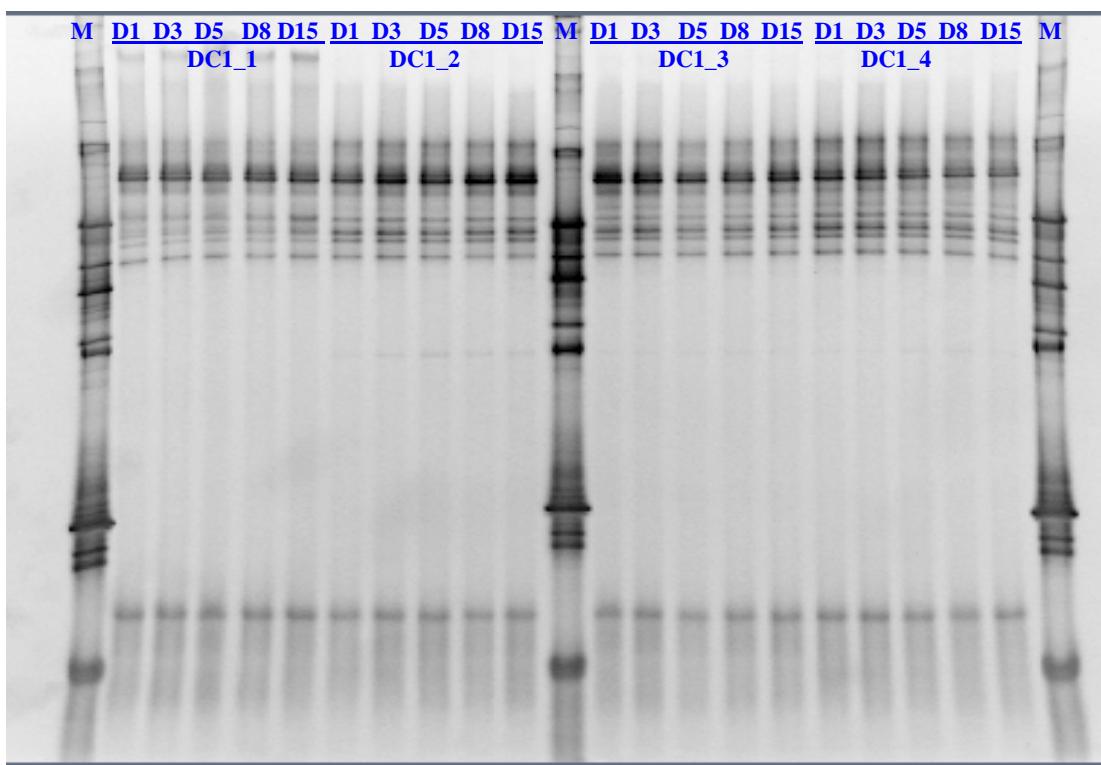
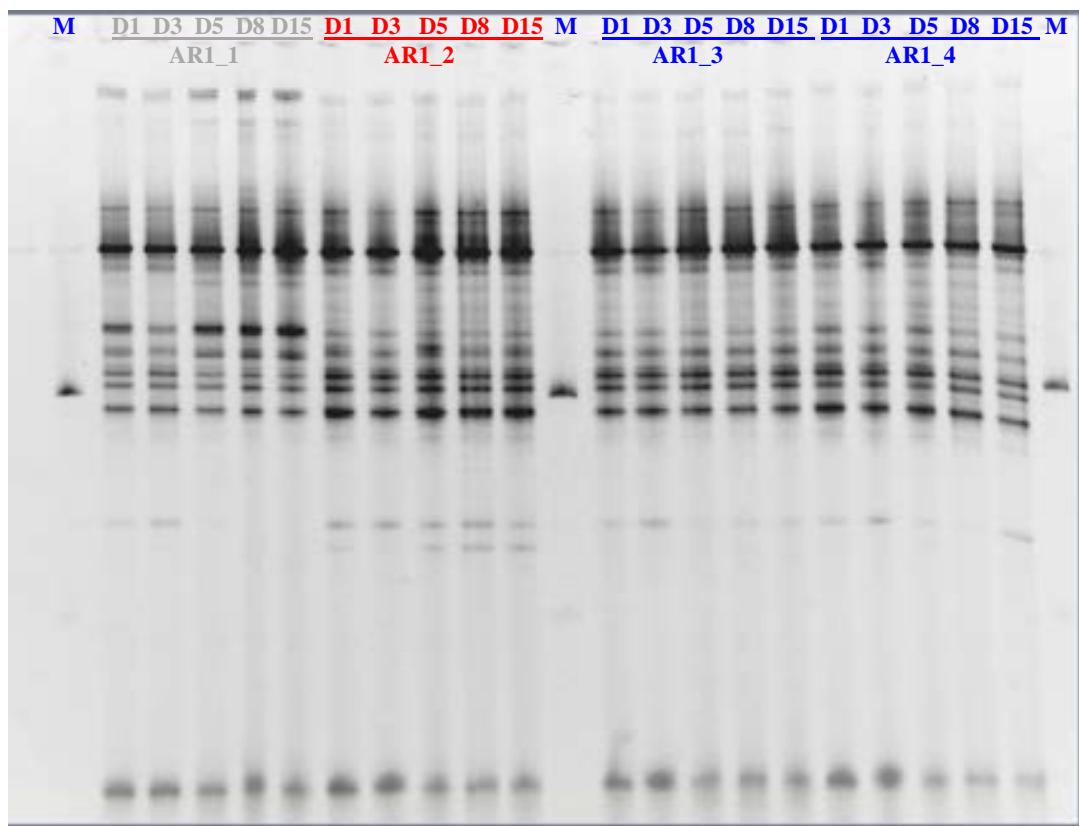
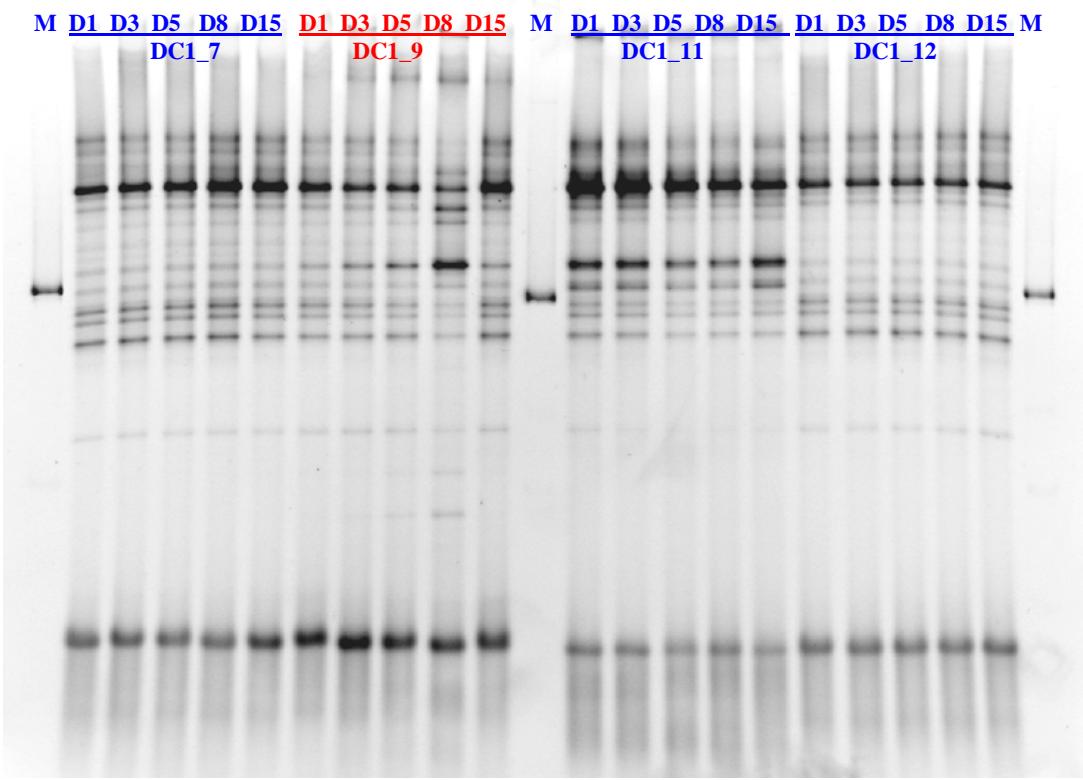
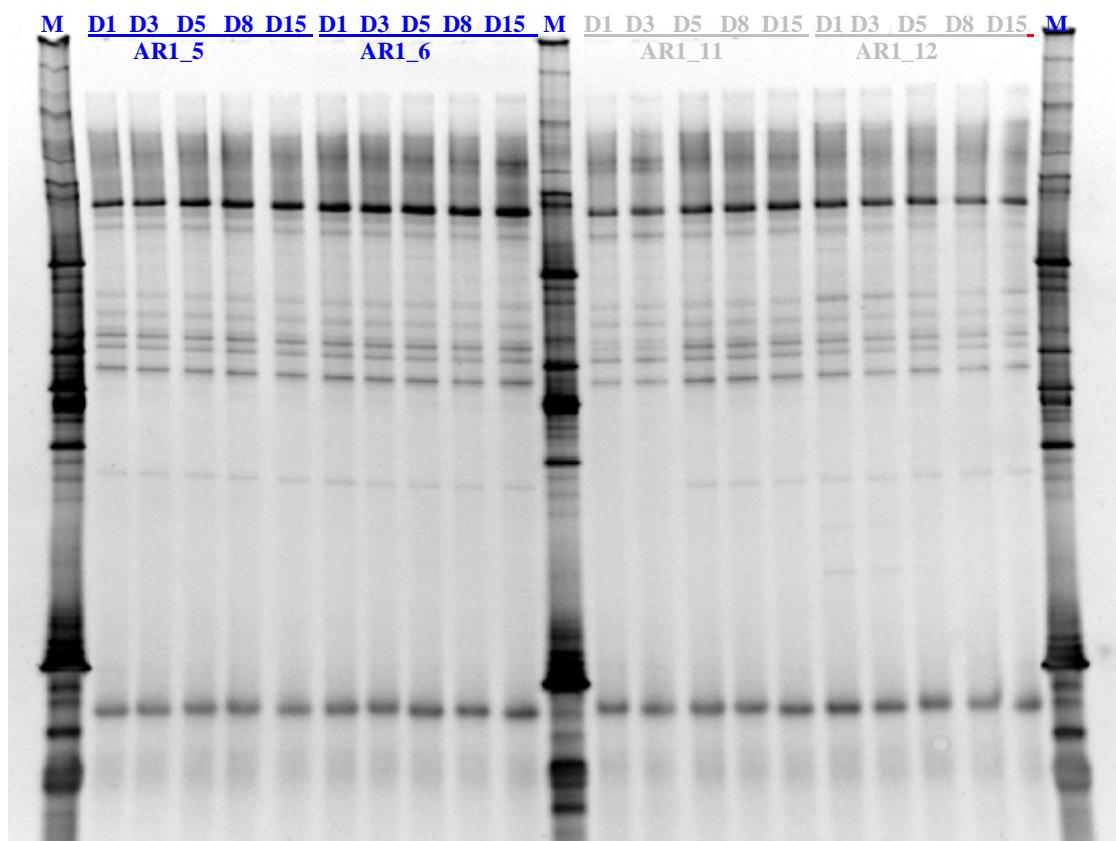
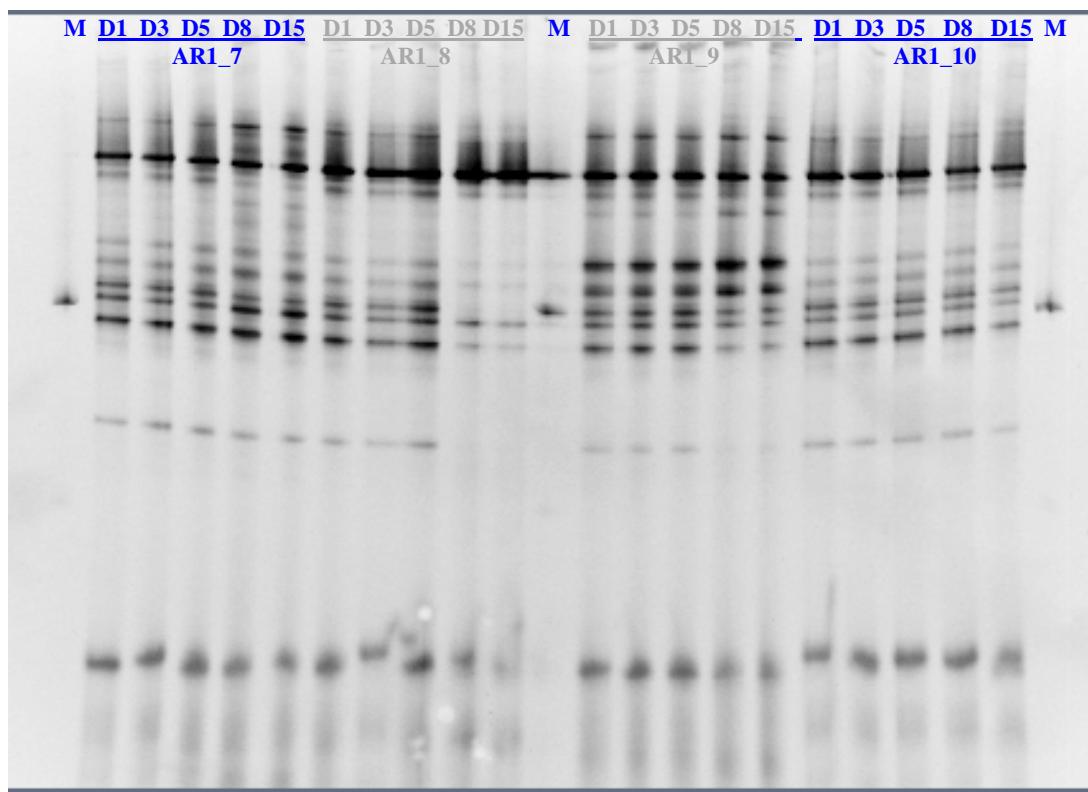


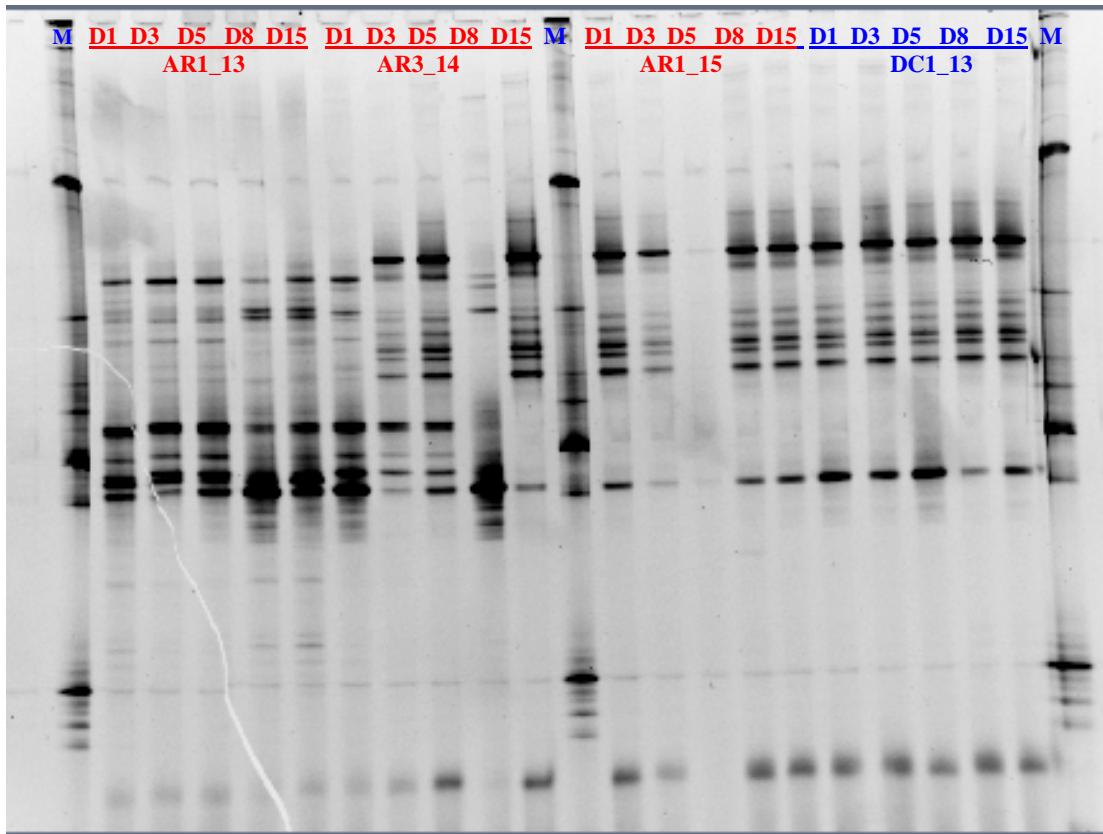
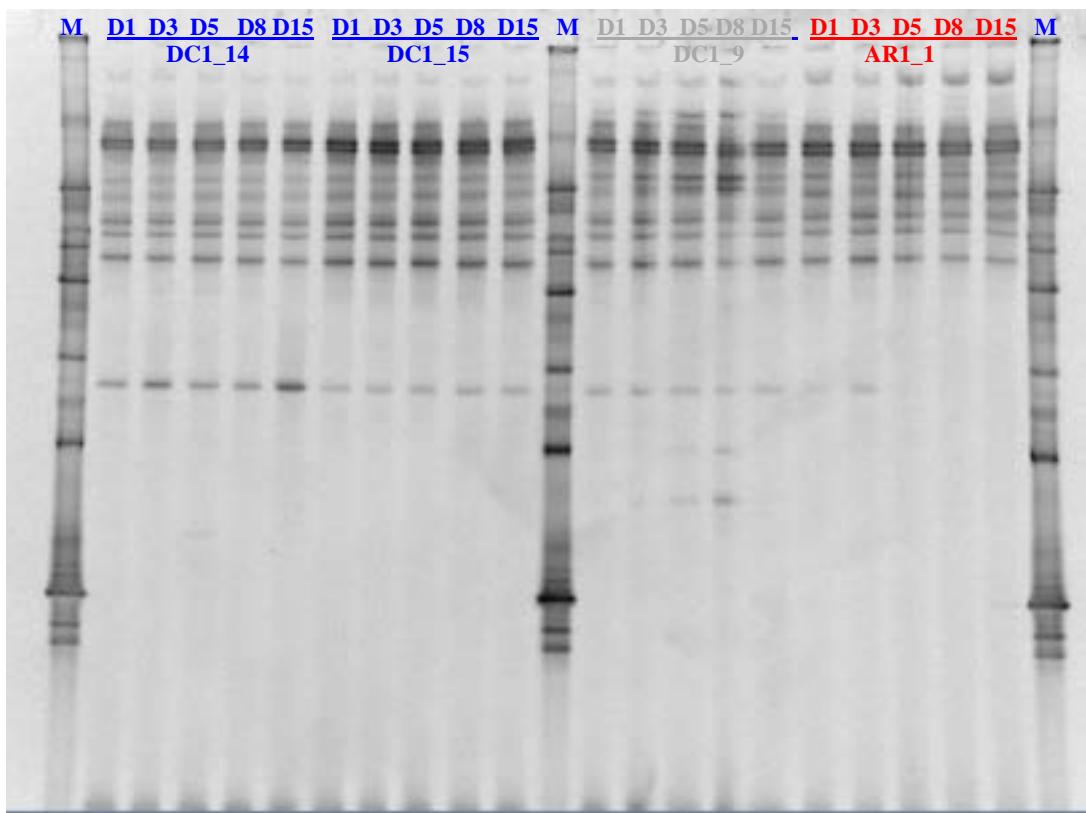
Figure S6: V6-based Unweighted Pair Group Method with Arithmetic Mean (UPGMA) dendrogram calculated based on the Euclidean distance of the normalized sequence abundance at the Class taxonomic unit. The red to green continuum ranges from 0.00 to 1.00 normalized abundance. The color blocks to the right of the normalized abundance are representative of the 15 different products analyzed during the study.

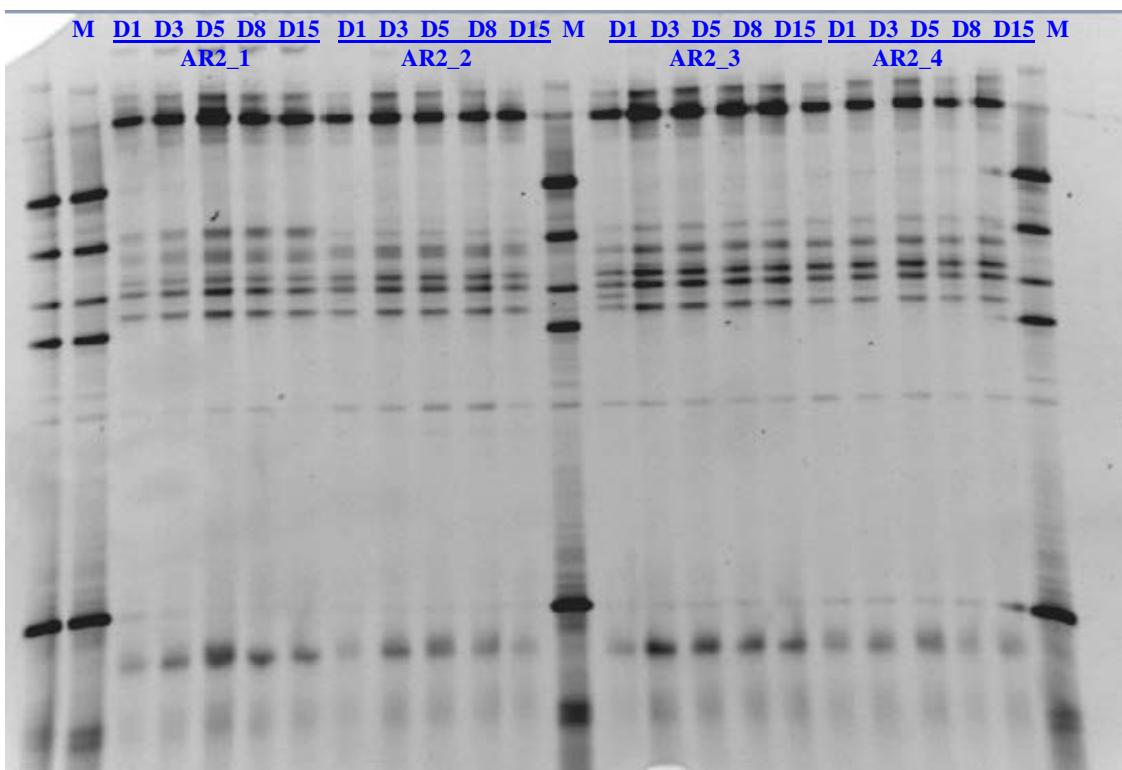
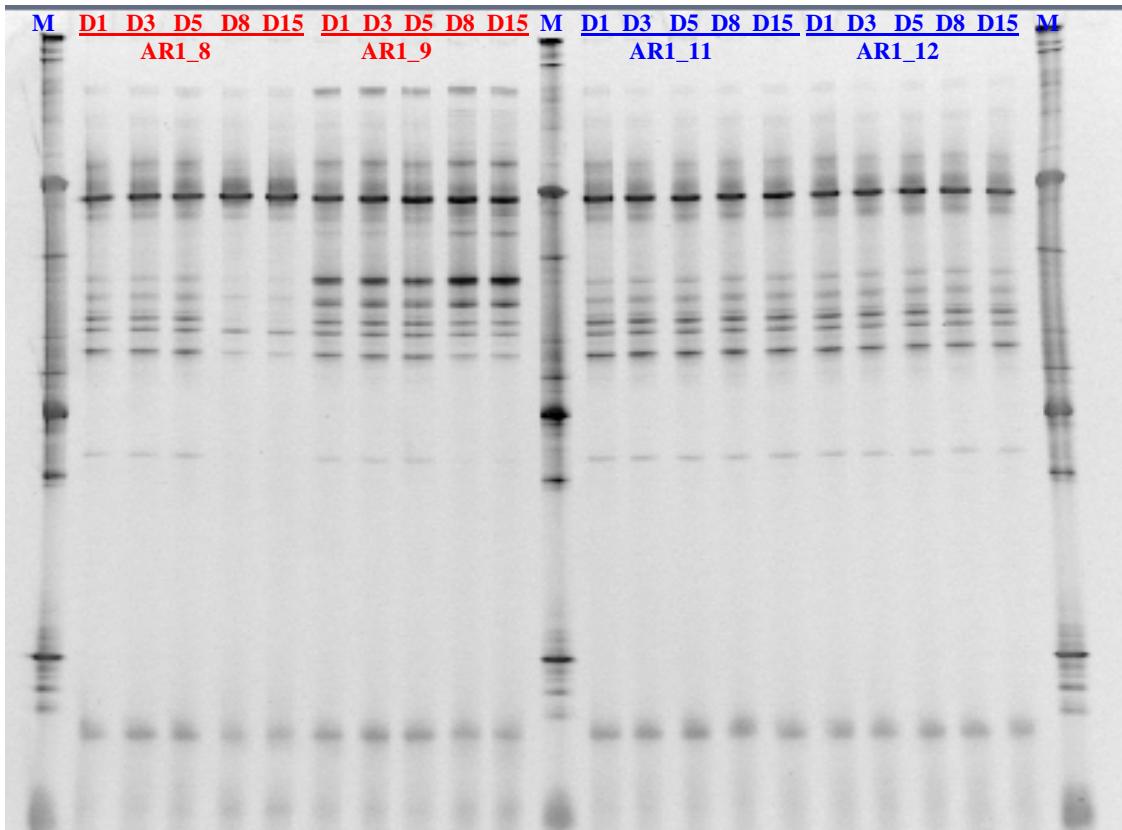
Figure S7 (below): Denaturing Gradient Gel Electrophoresis (DGGE) gels used to analyze the 16S rRNA gene profiles of total bacteria isolated from open STP samples during a 15-day sampling period. Changes in the DGGE profiles would be indicative of changes in the bacterial populations over the 15-day period. The 26 gels below contain the data from each of the samples at day 1 (D1; when the product was opened initially) and subsequently days 3 (D3), 5 (D5), 8 (D8) and 15 (D15). DNA samples isolated each day from a particular product are shown adjacent to one another to evaluate profile changes over the sampling period. Products/samples identified in blue labels did not appear to have any profile changes, those in red likely had changes in the banding profiles over the study period and those in grey were repeated on another gel due to poor initial banding resolution. The sample IDs are defined in Supplementary Table S3 and an overview of the DGGE results is shown in Supplemental Table S4.









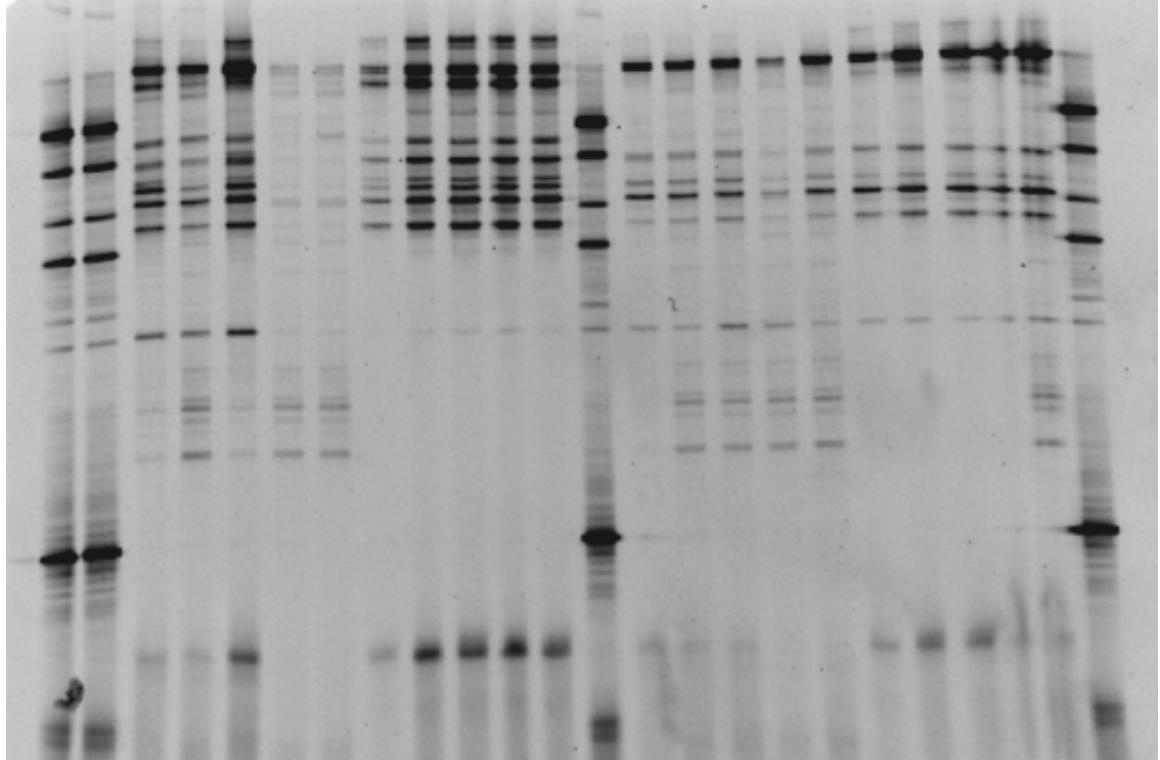


M D1 D3 D5 D8 D15 D1 D3 D5 D8 D15 M D1 D3 D5 D8 D15 D1 D3 D5 D8 D15 M

AR2_5 AR2_6

AR2_7

AR2_8



M D1 D3 D5 D8 D15 D1 D3 D5 D8 D15 M D1 D3 D5 D8 D15 D1 D3 D5 D8 D15 M

NCTR_9

NCTR_10

NCTR_11

NCTR_12

