

SUPPORTING ONLINE MATERIAL

Infrared and Raman Spectroscopic Studies of the Antimicrobial Effects of Garlic Concentrates and Diallyl Constituents on Foodborne Pathogens

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CONTENTS:

Table S1. Effect of garlic concentrate (*Allium sativum*) on growth of *Escherichia coli* O157:H7 in sterilized tryptic soy broth at different temperature treatments for 0, 1, 3, 5 and 7 days (N=3).

Temperature treatments (°C)	Concentration (µl/ml)	Viable cell counts (log CFU/ml)				
		0 day	1 day	3 days	5 days	7 days
4	0	5.03±0.21	5.08±0.41	5.01±0.25	5.06±0.09	4.95±0.15
	25	5.14±0.31	5.09±0.28	5.02±0.31	5.00±0.18	4.93±0.08
	50	5.12±0.24	5.04±0.17	4.93±0.28	4.90±0.21	4.89±0.32
	75	5.10±0.09	5.09±0.31	4.93±0.37	4.81±0.08	4.83±0.17
	100	5.01±0.12	5.02±0.26	4.91±0.24	4.99±0.06	4.93±0.34
	200	5.33±0.23	5.00±0.13	5.01±0.31	4.85±0.14	4.20±0.26
22	0	4.92±0.35	9.31±0.73	9.25±0.24	9.23±0.81	9.17±0.47
	25	4.98±0.12	4.85±0.21	4.01±0.41	2.93±0.16	0
	50	4.86±0.09	4.71±0.54	2.76±0.26	0	0
	75	4.81±0.41	4.65±0.31	0	0	0
	100	4.86±0.12	4.39±0.44	0	0	0
	200	4.87±0.23	4.16±0.18	0	0	0
35	0	5.00±0.13	9.24±0.49	9.26±0.25	9.12±0.79	9.18±0.31
	25	5.02±0.24	2.28±0.34	0	0	0
	50	5.01±0.17	0	0	0	0
	75	5.03±0.41	0	0	0	0
	100	4.87±0.07	0	0	0	0
	200	5.05±0.21	0	0	0	0

Table S2. Effect of garlic concentrate (*Allium sativum*) on growth of *Listeria monocytogenes* in sterilized tryptic soy broth at different temperature treatments for 0, 1, 3, 5 and 7 days (N=3).

Temperature treatments (°C)	Concentration (µl/ml)	Viable cell counts (log CFU/ml)				
		0 day	1 day	3 days	5 days	7 days
4	0	4.99±0.26	5.13±0.29	5.39±0.19	6.07±0.40	6.22±0.23
	25	5.03±0.38	4.86±0.37	4.77±0.12	4.43±0.53	4.30±0.26
	50	4.99±0.39	4.75±0.24	4.66±0.09	4.61±0.12	4.45±0.31
	75	4.94±0.13	4.91±0.26	4.69±0.19	4.71±0.07	4.36±0.36
	100	4.88±0.08	4.86±0.27	4.76±0.20	4.53±0.32	4.68±0.36
	200	4.99±0.27	4.82±0.14	4.80±0.39	4.34±0.26	4.34±0.23

22	0	4.96±0.31	8.21±0.37	8.34±0.28	7.49±0.40	7.28±0.19
	25	5.03±0.21	4.98±0.24	4.92±0.31	4.64±0.43	3.54±0.29
	50	5.03±0.17	4.96±0.31	4.77±0.24	4.42±0.31	3.43±0.42
	75	4.99±0.09	4.80±0.54	4.59±0.08	4.23±0.36	2.11±0.26
	100	4.97±0.09	4.62±0.51	4.27±0.18	4.13±0.22	1.85±0.34
	200	4.97±0.27	4.68±0.49	4.24±0.34	2.97±0.24	1.48±0.24
35	0	4.94±0.23	8.89±0.14	9.01±0.29	8.72±0.37	8.36±0.63
	25	4.99±0.31	3.63±0.30	1.48±0.39	0	0
	50	4.79±0.32	2.92±0.21	0	0	0
	75	5.03±0.43	2.88±0.32	0	0	0
	100	5.00±0.25	2.45±0.12	0	0	0
	200	4.95±0.42	0	0	0	0

Table S3. Effect of diallyl sulfide on growth of *E. coli* O157:H7 in sterilized tryptic soy broth at different temperature treatments for 0, 1, 2, 3, 4, 5, 6 and 7 days (N=3).

Temperature treatments	Concentration	<i>E. coli</i> O157:H7 viable cell counts (log CFU/ml)							
		0 day	1 day	2 days	3 days	4 days	5 days	6 days	7 days
4°C	control	5.08±0.06	5.18±0.12	5.20±0.18	5.17±0.21	5.16±0.19	4.93±0.19	4.96±0.20	4.89±0.23
	5 μM	5.03±0.11	5.09±0.21	5.12±0.12	4.81±0.16	4.71±0.15	4.30±0.16	4.23±0.18	4.22±0.18
	10 μM	5.00±0.11	4.62±0.22	4.29±0.31	4.01±0.41	3.85±0.37	2.01±0.34	0	0
	20 μM	5.10±0.09	4.43±0.19	4.26±0.21	4.14±0.23	0	0	0	0
22°C	control	4.96±0.11	9.18±0.15	9.14±0.11	9.03±0.18	8.96±0.15	8.98±0.14	9.17±0.12	9.10±0.20
	5 μM	4.92±0.10	8.17±0.22	8.48±0.25	8.54±0.15	8.56±0.34	8.43±0.29	8.26±0.12	7.98±0.17
	10 μM	4.99±0.09	7.09±0.21	5.46±0.14	3.79±0.43	2.48±0.21	1.31±0.12	0	0
	20 μM	4.99±0.09	7.03±0.14	4.38±0.33	0	0	0	0	0
35°C	control	4.98±0.12	9.02±0.19	9.26±0.22	9.04±0.17	9.18±0.19	9.02±0.20	9.00±0.12	9.15±0.18
	5 μM	4.98±0.12	8.88±0.24	8.79±0.15	8.99±0.29	9.10±0.26	9.02±0.31	8.98±0.12	9.06±0.23
	10 μM	5.00±0.15	5.79±0.22	0	0	0	0	0	0
	20 μM	5.02±0.08	4.40±0.17	0	0	0	0	0	0

Table S4. Effect of diallyl sulfide on growth of *Listeria monocytogenes* in sterilized tryptic soy broth at different temperature treatments for 0, 1, 2, 3, 4, 5, 6 and 7 days (N=3).

Temperature treatments	Concentration	<i>L. monocytogenes</i> viable cell counts (log CFU/ml)							
		0 day	1 day	2 days	3 days	4 days	5 days	6 days	7 days
4°C	control	4.93±0.11	5.11±0.22	5.17±0.17	5.24±0.18	5.33±0.19	5.85±0.13	5.99±0.11	6.15±0.12
	5 μM	5.00±0.09	5.08±0.11	4.98±0.19	5.03±0.18	5.08±0.21	5.00±0.12	5.14±0.31	5.08±0.16
	10 μM	4.99±0.09	5.16±0.13	5.16±0.18	5.29±0.09	4.91±0.15	4.96±0.16	4.91±0.20	4.73±0.18
	20 μM	4.97±0.08	5.25±0.18	5.25±0.13	5.05±0.14	4.94±0.16	4.93±0.17	4.86±0.11	4.83±0.19
22°C	control	4.98±0.12	8.01±0.18	8.81±0.15	8.57±0.21	8.57±0.19	8.31±0.31	8.37±0.22	7.84±0.24
	5 μM	5.01±0.18	7.79±0.19	8.61±0.22	7.54±0.20	7.03±0.18	6.42±0.36	6.03±0.28	5.19±0.27
	10 μM	5.02±0.09	8.00±0.17	8.64±0.19	6.64±0.21	4.57±0.12	3.40±0.11	2.90±0.09	0

	20 μ M	5.08 \pm 0.10	7.72 \pm 0.16	8.64 \pm 0.15	5.92 \pm 0.10	0	0	0	0
35°C	control	4.98 \pm 0.09	9.27 \pm 0.17	8.32 \pm 0.18	9.04 \pm 0.17	8.96 \pm 0.12	8.76 \pm 0.21	8.68 \pm 0.23	8.62 \pm 0.17
	5 μ M	5.00 \pm 0.14	9.13 \pm 0.11	6.92 \pm 0.31	5.48 \pm 0.29	0	0	0	0
	10 μ M	5.00 \pm 0.13	9.08 \pm 0.09	4.27 \pm 0.27	0	0	0	0	0
	20 μ M	5.02 \pm 0.12	8.97 \pm 0.18	3.45 \pm 0.29	0	0	0	0	0

Table S5 (a). Band assignments of bacterial Raman spectra

<i>Escherichia coli</i> O157:H7		<i>Listeria monocytogenes</i>	
Wavenumber (cm ⁻¹)	Assignment*	Wavenumber (cm ⁻¹)	Assignment
754	symmetric breathing of tryptophan	666	ring breathing modes in DNA base
788	O-P-O stretching DNA	788	O-P-O stretching DNA
823	out-of-plane ring breathing of tyrosine	1008	phenylalanine
879	tryptophan	1090	sym phosphate str vibr
968	lipids	1112	saccharide
1008	phenylalanine	1134	lipids
1043	carbohydrate	1168	lipids
1093	sym PO ₂ ⁻ str vibr of DNA backbone vibration	1243	amide III
1130	phospholipid	1261	amide III
1174	tyrosine, phenylalanine and C-H bend of protein	1297	fatty acids
1243	amide III	1321	amide III
1309	CH ₃ /CH ₂ twisting or bending mode of lipid	1335	CH ₃ CH ₂ wagging of nucleic acid
1343	CH ₃ and CH ₂ wagging	1367	vs (CH ₃) of phospholipids
1401	bending modes of methyl group	1415	ring structure of nucleic acid
1454	phospholipids	1453	phospholipids
1586	phenylalanine	1531	amide structure
1676	amide I (β -sheet)	1577	bound and free NADH
2935	chain end CH ₃ sym band	1669	carbonyl stretch
3059	unsaturated =CH stretch	1689	amide I

Table S5 (b). Band assignments of bacterial FT-IR spectra

Wavenumber (cm ⁻¹)	Assignment
918	phosphodiester
970	phosphodiester
1060	polysaccharides
1080	sym P=O of nucleic acid
1236	asym P=O of nucleic acid
1400	sym str of C-O
1455	CH ₂ bending of lipids
1545	amide II
1647	amide I
2854	methylene groups from lipids
2929	methylene groups from lipids
2966	methylene groups from lipids
3074	C-H ring
3290	N-H str of proteins and O-H str of polysaccharide and water

* sym: symmetric, asym: asymmetric, str: stretching, vibr: vibration

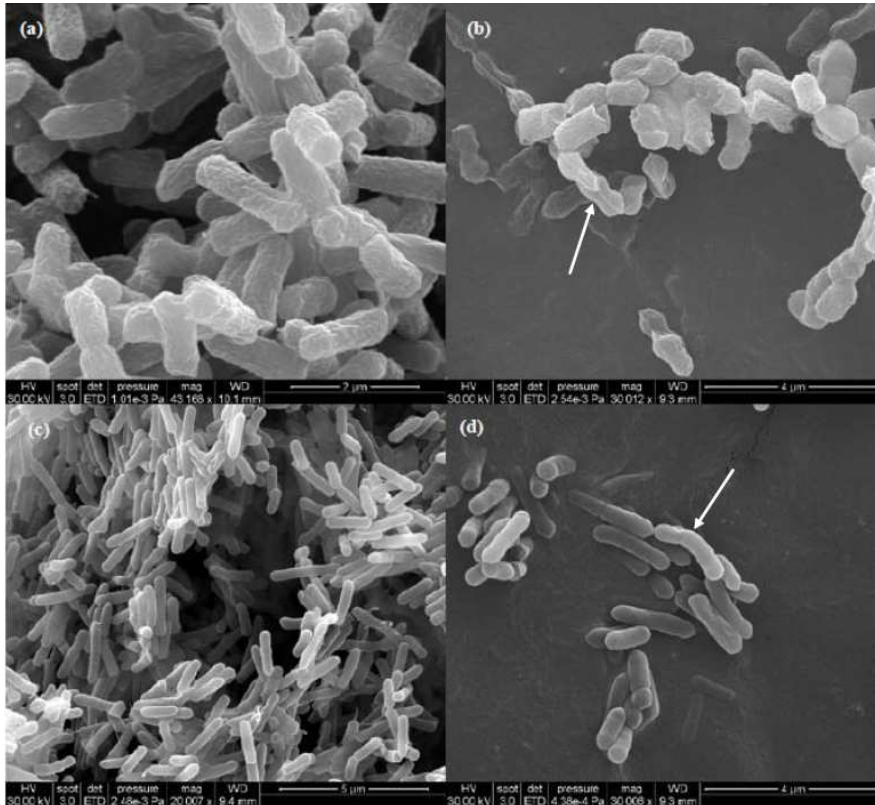


Figure S1. Scanning electron microscope images of *Escherichia coli* O157:H7 without (a) and with (b) the treatment of organosulfur compounds derived from garlic (*Allium sativum*) and *Listeria monocytogenes* without (c) and with (d) the treatment of organosulfur compounds derived from garlic (*Allium sativum*).

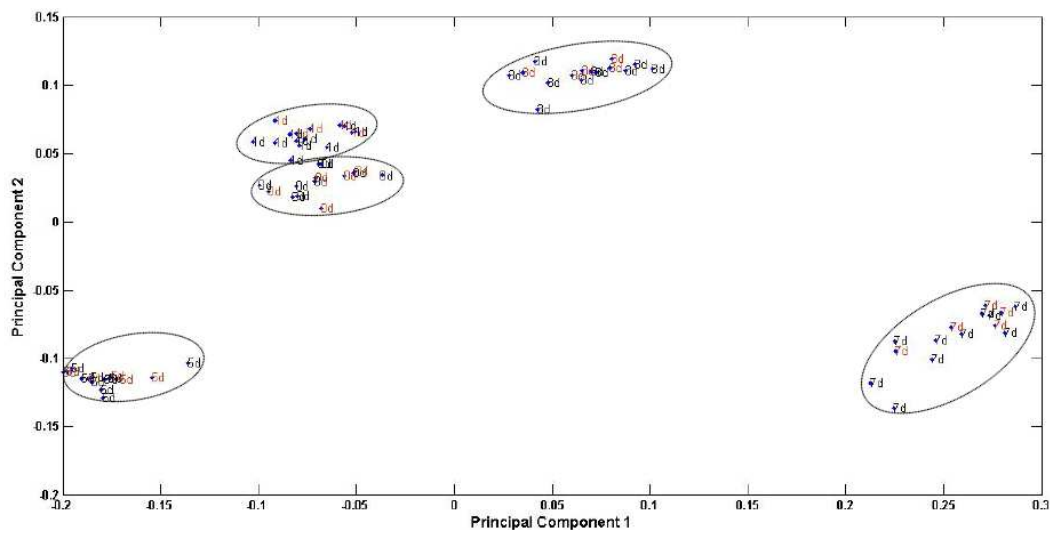


Figure S2 (a). Representative two-dimensional principal component analysis of *E. coli* O157:H7 cocktail in sterilized broth under 10 μ M diallyl sulfide treatment for 0 (control), 1, 3, 5, and 7 days at room temperature (22°C) using infrared spectra.

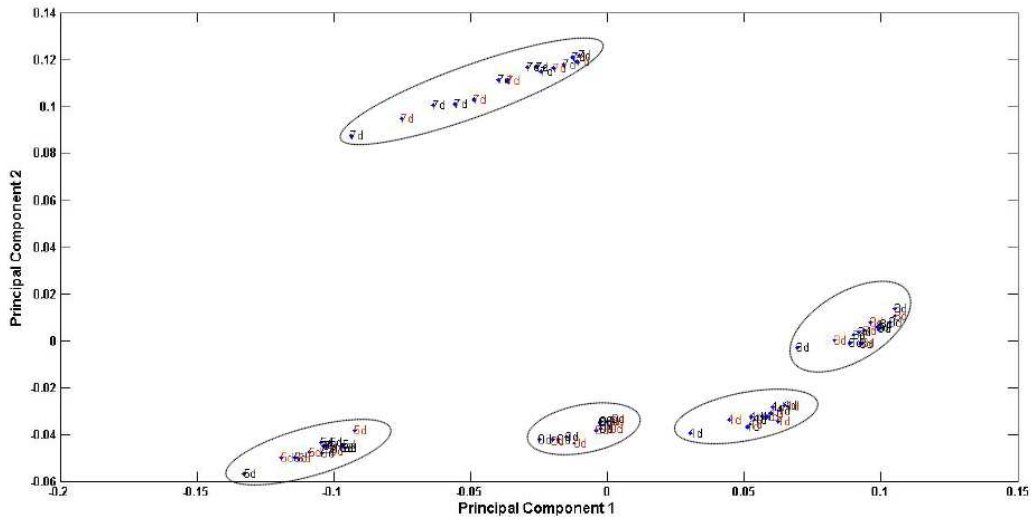


Figure S2 (b). Representative two-dimensional principal component analysis of *E. coli* O157:H7 cocktail in sterilized broth under 25 μ l/ml garlic concentrate treatment for 0 (control), 1, 3, 5, and 7 days at room temperature (22°C) using infrared spectra.

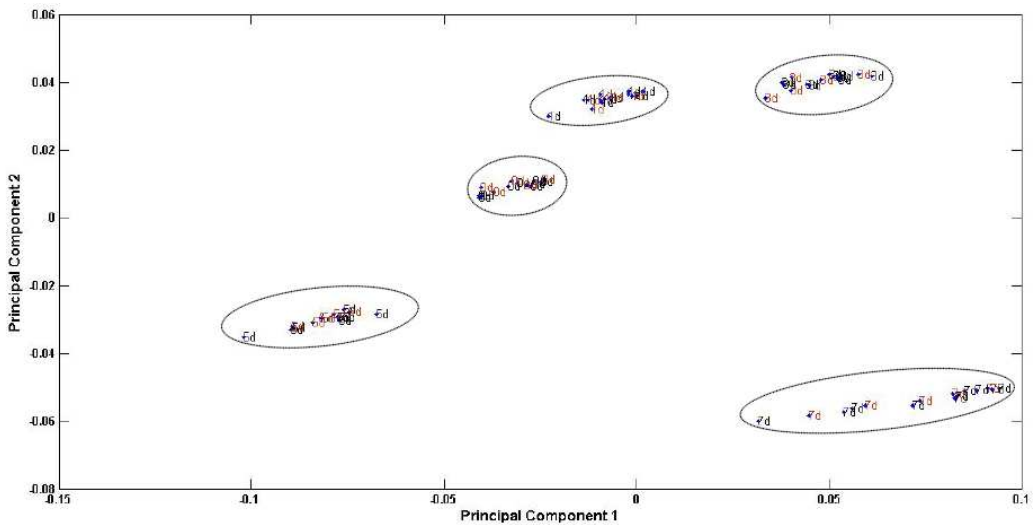


Figure S2 (c). Representative two-dimensional principal component analysis of *L. monocytogenes* cocktail in sterilized broth under 10 μ M diallyl sulfide treatment for 0 (control), 1, 3, 5, and 7 days at room temperature (22°C) using infrared spectra.

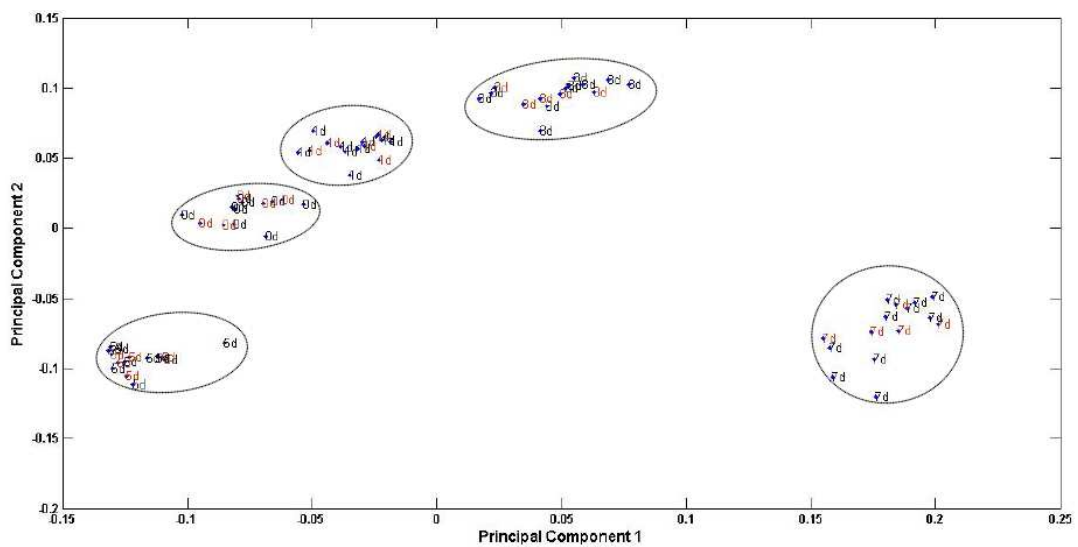


Figure S2 (d). Representative two-dimensional principal component analysis of *L. monocytogenes*

cocktail in sterilized broth under 25 $\mu\text{l/ml}$ garlic concentrate treatment for 0 (control), 1, 3, 5, and 7 days at room temperature (22°C) using infrared spectra.

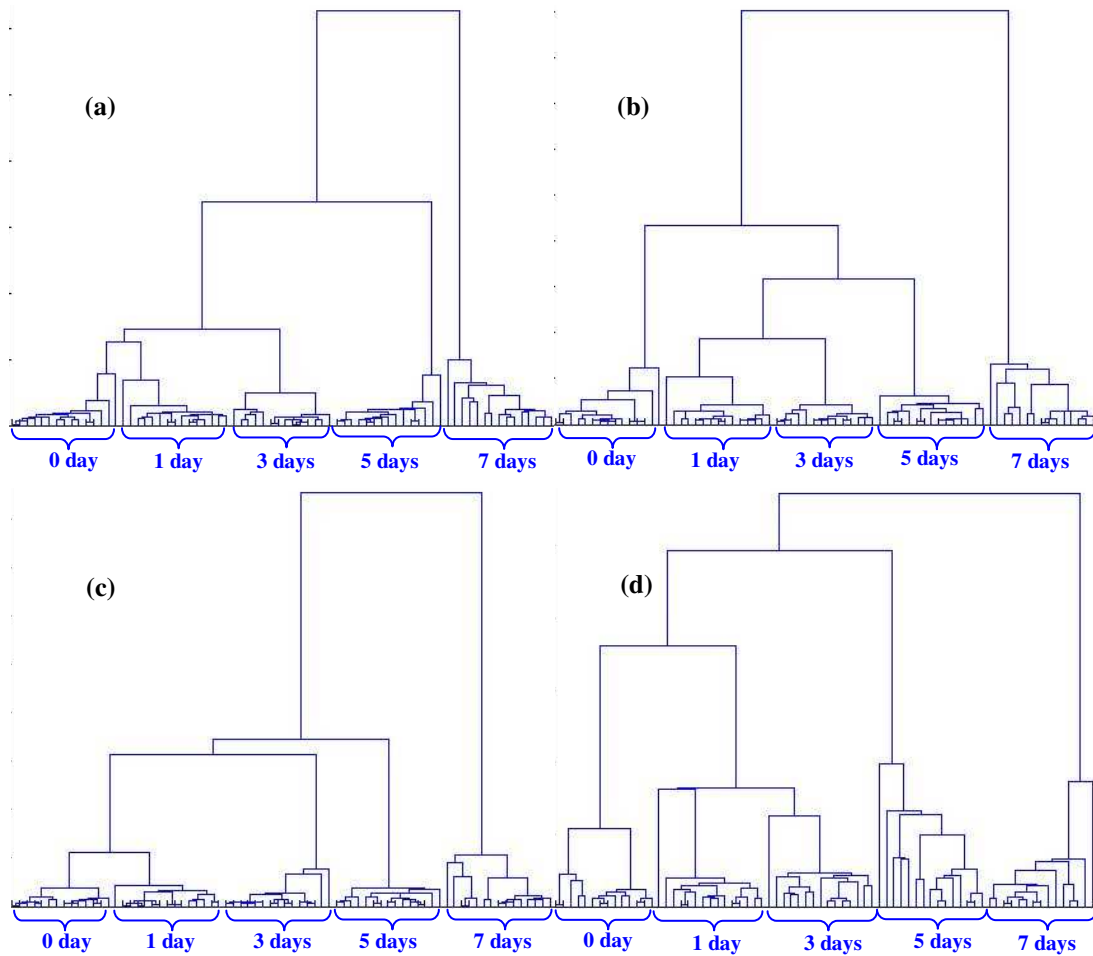


Figure S3. Representative dendrogram analysis of *E. coli* O157:H7 cocktail in sterilized broth under (a) 10 μM diallyl sulfide treatment (b) 25 $\mu\text{l/ml}$ garlic concentrate treatment and *L. monocytogenes* cocktail in sterilized broth under (c) 10 μM diallyl sulfide treatment (d) 25 $\mu\text{l/ml}$ garlic concentrate treatment at room temperature (22°C) using Raman spectra.