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Biocidal Activity of Fast Pyrolysis Biochar against *E. coli* O157:H7 in Soil Varies Based on Production Temperature or Age of Biochar

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SUPPLEMENTAL MATERIAL

TABLE S1. *pH readings of varying concentrations of fast pyrolysis biochar in soil*

Soil Amendment	Biochar Treatment Temperatures		
	450°C	500°C	600°C
Soil negative control (0%)	7.70	7.70	7.70
1.0% Biochar	7.90	7.72	7.95
1.5% Biochar	7.90	7.77	7.99
2.0% Biochar	7.91	7.77	7.91
2.5% Biochar	7.89	7.73	7.95
3.0% Biochar	7.95	7.78	7.89
3.5% Biochar	7.90	7.81	7.96

TABLE S2. Survival of *E. coli* O157:H7 (log CFU/g) in soil with varying concentrations of 450°C fast pyrolysis biochar stored at 22°C^a

Soil Amendment	Storage time (weeks)							
	0	1	2	3	4	5	6	7
Soil positive control (0%)	7.42 ± 0.05 ^b	6.70 ± 0.03	6.66 ± 0.03	6.48 ± 0.04	6.64 ± 0.14	6.41 ± 0.08	6.54 ± 0.11	6.40 ± 0.16
1.0% Biochar	7.42 ± 0.05	6.49 ± 0.10	6.68 ± 0.13	6.53 ± 0.11	6.34 ± 0.10	6.44 ± 0.09	6.49 ± 0.24	6.39 ± 0.14
1.5% Biochar	7.42 ± 0.05	6.81 ± 0.19	6.57 ± 0.21	6.55 ± 0.09	6.47 ± 0.17	6.42 ± 0.19	6.56 ± 0.18	6.37 ± 0.16
2.0% Biochar	7.42 ± 0.05	6.78 ± 0.17	6.64 ± 0.12	6.65 ± 0.12	6.55 ± 0.18	6.56 ± 0.12	6.55 ± 0.07	6.40 ± 0.13
2.5% Biochar	7.42 ± 0.05	6.84 ± 0.18	6.83 ± 0.15	6.62 ± 0.25	6.58 ± 0.09	6.61 ± 0.14	6.65 ± 0.15	6.48 ± 0.11
3.0% Biochar	7.42 ± 0.05	6.96 ± 0.06	6.84 ± 0.12	6.67 ± 0.17	6.64 ± 0.11	6.60 ± 0.17	6.80 ± 0.14	6.65 ± 0.12
3.5% Biochar	7.42 ± 0.05	7.03 ± 0.16	6.78 ± 0.19	6.67 ± 0.02	6.56 ± 0.19	6.53 ± 0.18	6.66 ± 0.13	6.52 ± 0.10

^aData points are means of observations ± SEM.

^bDay 0 counts consist of original population of inoculum added to soil/biochar mixture.

TABLE S3. Survival of *E. coli* O157:H7 (log CFU/g) in soil with varying concentrations of 500°C fast pyrolysis biochar stored at 22°C^a

Soil amendment	Storage time (weeks)							
	0	1	2	3	4	5	6	7
Soil positive control (0%)	7.74 ± 0.04	6.69 ± 0.14	6.53 ± 0.18	6.70 ± 0.15	6.60 ± 0.12	6.60 ± 0.14	6.58 ± 0.11	6.53 ± 0.13
1.0% Biochar	7.74 ± 0.04	6.83 ± 0.14	6.70 ± 0.19	7.00 ± 0.15	6.79 ± 0.24	6.78 ± 0.20	6.79 ± 0.20	6.52 ± 0.22
1.5% Biochar	7.74 ± 0.04	6.77 ± 0.11	6.35 ± 0.46	6.88 ± 0.16	6.57 ± 0.34	6.57 ± 0.22	6.62 ± 0.15	6.48 ± 0.15
2.0% Biochar	7.74 ± 0.04	6.78 ± 0.03	6.88 ± 0.12	6.72 ± 0.12	6.70 ± 0.22	6.78 ± 0.11	6.81 ± 0.09	6.59 ± 0.17
2.5% Biochar	7.74 ± 0.04	6.83 ± 0.11	6.90 ± 0.14	6.80 ± 0.13	6.49 ± 0.36	6.71 ± 0.11	6.62 ± 0.15	6.54 ± 0.10
3.0% Biochar	7.74 ± 0.04	7.02 ± 0.05	6.95 ± 0.13	6.92 ± 0.14	6.63 ± 0.29	6.80 ± 0.20	6.68 ± 0.17	6.52 ± 0.27
3.5% Biochar	7.74 ± 0.04	6.88 ± 0.12	6.78 ± 0.26	6.89 ± 0.13	6.89 ± 0.14	6.96 ± 0.16	6.88 ± 0.16	6.58 ± 0.22

^aData points are means of observations ± SEM.

^bDay 0 counts consist of original population of inoculum added to soil/biochar mixture.

Table S4. Analysis of variance for 450 and 500°C pyrolyzed FPBC^a

Source	DF	450°C FPBC		500°C FPBC	
		F-value	p-value	F-value	p-value
Concentration	6	3.82	0.0078	0.42	0.8497
Week	6	9.65	<.0001	7.48	<.0001
Conc × Week	36	0.65	0.9234	0.81	0.7439

^aFast pyrolysis biochar.

Table S5. Log CFU/g means and mean comparison for 450 and 500°C pyrolyzed FPBC^a

Concentration	450°C	Week	450°C	500°C
Control	6.55ab ^b	1	6.80a ^c	6.83a
1.0%	6.48b	2	6.71ab	6.73ab
1.5%	6.54ab	3	6.60bcd	6.84a
2.0%	6.59ab	4	6.54cd	6.67ab
2.5%	6.66ab	5	6.51cd	6.76a
3.0%	6.74a	6	6.61bc	6.71a
3.5%	6.68ab	7	6.46d	6.54b

^a Fast pyrolysis biochar.

^b Concentration means with different letters are different at the 0.05 significance level.

^c Week means with different letters are different at the 0.05 significance level.

TABLE S6. *Log CFU/g means and mean comparison for 600°C pyrolyzed FPBC^a*

<u>Concentration</u>	<u>600C</u>
Control	6.70a ^b
1.0%	4.68b
1.5%	2.75c
2.0%	2.92c
2.5%	2.05cd
3.0%	1.36d
3.5%	0.86d

^aFast-pyrolyzed biochar

^bConcentration means with different letters are different at the 0.05 significance level.