

Supplementary Material

# Elucidation of Mechanical, Physical, Chemical and Thermal Properties of Microbial Composite Films by Integrating Sodium Alginate with *Bacillus subtilis* sp.

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**Table S1.** Tensile strength, breaking strain and toughness of microbial composite films.

Mass of <i>Bacillus subtilis</i> (g)	Triplicate	Tensile strength (MPa)	Breaking strain (%)	Toughness (MJ/m <sup>3</sup> )
0	1	0.4	85.644	0.004
	2	0.584	84.384	0.009
	3	0.849	83.088	0.034
Average		0.611	84.372	0.016
Standard deviation		0.226	6.906	0.016
0.1	1	0.581	85.314	0.025
	2	0.626	84.35	0.015
	3	0.653	84.882	0.009
Average		0.62	84.849	0.016
Standard deviation		0.036	0.483	0.008
0.2	1	0.753	84.65	0.02
	2	0.567	84.484	0.016
	3	0.585	86.144	0.019
Average		0.635	85.092	0.0183
Standard deviation		0.102	0.914	0.0021
0.3	1	0.834	86.01	0.034
	2	0.68	85.048	0.022
	3	0.611	84.45	0.015
Average		0.709	85.169	0.024
Standard deviation		0.114	0.787	0.009
0.4	1	0.776	82.39	0.022
	2	0.604	83.354	0.016
	3	0.829	89.832	0.035
Average		0.736	85.192	0.024
Standard deviation		0.118	4.047	0.009
0.5	1	0.947	89.166	0.061
	2	0.756	85.812	0.03
	3	0.871	87.24	0.044
Average		0.858	87.406	0.0450
Standard deviation		0.096	1.683	0.016
0.6	1	0.888	85.646	0.033
	2	0.776	85.878	0.022
	3	0.829	86.94	0.039
Average		0.831	86.155	0.0310
Standard deviation		0.056	0.69	0.009

**Table S2.** Thickness of microbial composite films.

Mass of <i>Bacillus subtilis</i> (g)	Triplicate	Readings of Precision Micrometer (mm)					Average (mm)	Overall Average (mm)
0.0 g	1	0.505	0.604	0.524	0.576	0.513	0.544	0.578
	2	0.583	0.636	0.581	0.617	0.642	0.612	
	3	0.661	0.516	0.528	0.587	0.601	0.578	
0.1 g	1	0.607	0.623	0.643	0.677	0.611	0.632	0.665
	2	0.618	0.703	0.692	0.687	0.717	0.683	
	3	0.762	0.603	0.702	0.656	0.677	0.680	
0.2 g	1	0.766	0.781	0.793	0.79	0.658	0.758	0.761
	2	0.689	0.776	0.782	0.743	0.762	0.750	
	3	0.801	0.812	0.793	0.762	0.699	0.773	
0.3 g	1	0.885	0.909	0.913	0.927	0.816	0.890	0.892
	2	0.805	0.912	0.973	0.873	0.991	0.911	
	3	0.943	0.947	0.868	0.796	0.818	0.874	
0.4 g	1	1.011	1.073	0.934	0.987	0.907	0.982	1.002
	2	0.963	0.998	1.087	1.106	0.977	1.026	
	3	0.936	0.948	1.044	1.087	0.976	0.998	
0.5 g	1	1.082	1.073	1.172	1.007	1.086	1.084	1.057
	2	1.073	1.088	1.074	1.096	0.992	1.065	
	3	0.976	1.067	1.082	1.012	1.006	1.029	
0.6 g	1	1.103	1.057	0.974	1.109	1.008	1.050	1.059
	2	1.063	0.969	1.043	1.101	1.048	1.045	
	3	1.17	1.112	1.051	1.008	1.072	1.826	

**Table S3.** Opacity (white light) of microbial composite films.

Mass of <i>Bacillus subtilis</i> (g)	Triplicate	Opacity (White light, %)	Average	High	Low	Standard deviation
0	0.1	7.46	7.52	7.73	7.39	0.18
	0.2	7.39				
	0.3	7.73				
0.1	1.1	7.83	8.1	8.28	7.83	0.24
	1.2	8.28				
	1.3	8.18				
0.2	2.1	8.3	8.39	8.44	8.3	0.07
	2.2	8.44				
	2.3	8.48				
0.3	3.1	10.31	9.19	10.31	8.49	0.98
	3.2	8.49				
	3.3	8.78				
0.4	6.1	15.7	12.16	15.7	10.02	3.09
	6.2	10.02				
	6.3	10.74				
0.5	5.1	13.43	13.65	14.15	13.38	0.43

	5.2	14.15				
	5.3	13.38				
0.6	4.1	16.76	16.45	16.76	16.21	0.28
	4.2	16.21				
	4.3	16.38				

**Table S4.** Opacity (black light) of microbial composite films.

Mass of <i>Bacillus subtilis</i> (g)	Triplicate	Opacity (Black light, %)	Average	High	Low	Standard deviation
0	0.1	38.88	38.41	39.14	38.09	0.64
	0.2	38.09				
	0.3	39.14				
0.1	1.1	37.9	37.86	38.44	37.34	0.6
	1.2	38.44				
	1.3	37.34				
0.2	2.1	37.92	38.61	38.98	37.92	0.6
	2.2	38.94				
	2.3	38.98				
0.3	3.1	43.03	39.29	43.03	36.92	3.28
	3.2	36.92				
	3.3	37.93				
0.4	6.1	50.56	41.69	50.56	36.13	7.77
	6.2	36.13				
	6.3	38.38				
0.5	5.1	39.99	40.55	50.56	39.99	0.52
	5.2	40.66				
	5.3	41.01				
0.6	4.1	41.12	41.18	42.07	40.36	0.86
	4.2	40.36				
	4.3	42.07				

**Table S5.** Brightness of microbial composite films.

Mass of <i>Bacillus subtilis</i> (g)	Triplicate	Readings			Average	High	Low	Standard deviation
		1	2	3				
0	0.1	4.23	4.14	4.06	4.24	4.76	3.9	0.25
	0.2	4.05	3.9	4.37				
	0.3	4.76	4.35	4.34				
0.1	1.1	4.23	4.14	4.06	4.24	4.76	3.9	0.25
	1.2	4.05	3.9	4.37				
	1.3	4.76	4.35	4.34				
0.2	2.1	7.06	6.52	6.62	6.65	7.06	6.24	0.27
	2.2	6.28	6.24	6.79				
	2.3	6.84	6.62	6.86				
0.3	3.1	6.14	7.2	6.54	6.67	7.3	5.57	0.54

	3.2	5.57	6.88	6.68				
	3.3	7.07	7.3	6.69				
0.4	4.1	7.38	8.99	5.22	7.38	8.99	5.22	1.17
	4.2	7.42	6.47	6.57				
	4.3	8.51	7.51	8.34				
0.5	6.1	7.32	8.23	8.48	7.88	8.48	7.32	0.49
	6.2	7.91	8.23	8.47				
	6.3	7.33	7.38	7.58				
0.6	5.1	10.62	9.26	9.2	8.19	10.62	6.64	1.32
	5.2	6.92	7.39	6.64				
	5.3	7.36	7.7	8.66				

Table S6. Conductivity of microbial composite films.

Mass of <i>Bacillus subtilis</i> (g)	Triplicate	Conductivity (mV)	Average (mV)
0	1	11	11.33
	2	10	
	3	13	
0.1	1	12	12
	2	13	
	3	11	
0.2	1	16	16.33
	2	15	
	3	18	
0.3	1	15	17.33
	2	19	
	3	18	
0.4	1	19	19.67
	2	19	
	3	21	
0.5	1	27	26
	2	26	
	3	25	
0.6	1	25	26.33
	2	28	
	3	26	

Table S7. Water absorption of microbial composite films.

Mass of <i>Bacillus subtilis</i> (g)	Triplicate	Weight of dried films (g)	Weight of wet films (g)	Percentage of water absorption (%)	Average (%)
0	1	0.07	0.26	271.43	264.29
	2	0.07	0.26	271.43	
	3	0.08	0.28	250	
0.1	1	0.07	0.27	285.71	269.05
	2	0.08	0.28	250	

	3	0.07	0.26	271.43	
0.2	1	0.08	0.29	262.5	270.83
	2	0.08	0.31	287.5	
	3	0.08	0.29	262.5	
0.3	1	0.08	0.3	275	279.17
	2	0.08	0.3	275	
	3	0.08	0.31	287.5	
0.4	1	0.08	0.31	287.5	289.35
	2	0.09	0.32	255.56	
	3	0.08	0.34	325	
0.5	1	0.08	0.34	325	300.93
	2	0.09	0.34	277.78	
	3	0.09	0.36	300	
0.6	1	0.09	0.37	311.11	307.78
	2	0.1	0.39	290	
	3	0.09	0.38	322.22	

Table S8. Thermogravimetric Analysis (TGA).

Step	Value	Sodium alginate film (Control)	Step	0.5 g microbial composite film
1	Onset Temperature	208.36 °C	-9.58% (0.52 mg)	211.21 °C
	Residue	70.11% (5.27 mg)		66.12% (3.62 mg)
	Inflection Point	211.99 °C		218.99 °C
	Midpoint	212.25 °C		221.95 °C
2	Onset Temperature	248.02 °C	-25.42% (-1.39 mg)	252.69 °C
	Residue	42.86% (3.22 mg)		40.69% (2.23 mg)
	Inflection Point	267.82 °C		270.44 °C
	Midpoint	267.73 °C		277.19 °C
3	Onset Temperature	417.09 °C	-6.21% (-0.34 mg)	431.08 °C
	Residue	34.05% (2.56 mg)		34.48% (1.89 mg)
	Inflection Point	436.80 °C		445.46 °C
	Midpoint	443.61 °C		466.16 °C
4	Onset Temperature	746.39 °C	-13.30% (0.73 mg)	657.30 °C
	Residue	25.75% (1.94 mg)		21.18% (1.16 mg)
	Inflection Point	777.03 °C		754.14 °C
	Midpoint	744.01 °C		701.79 °C