

Colloidal graphenes as heterogeneous additive to enhance protein crystal yield

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Supplementary Material

Table 2

The results of Miniaturized 96 well sparse matrix vapour diffusion trials were undertaken with Hampton Research crystal screen 1&2 in ratios of 200:200:50nL of protein solution:crystallisation condition:additive solution (at 20 $\mu\text{g.mL}^{-1}$). Monitoring for crystalline material forming conditions (CCMs). CCMs are inclusive of definite clear micro-crystals, needles or needle clusters, rods and single crystals. A scale commonly used in crystallization to grade the drops as clear, amorphous precipitate, crystalline precipitate, quasi crystalline/phase separation and a condition containing crystalline material was implemented, for examples see http://hamptonresearch.com/documents/growth_101/9.pdf.

	ADH		catalase		lysozyme		trypsin		PSPCI/NONO	
	clr	amo pre quasi cry total	clr	amo pre quasi cry total	clr	amo pre quasi cry total	clr	amo pre quasi cry total	clr	amo pre quasi cry total
control 1	6.0	47.0 40.0 2.0 1.0 96.0	78.0	10.0 4.0 2.0 2.0 96.0	63.0	14.0 6.0 1.0 12.0 96.0	50.0	16.0 23.0 5.0 2.0 96.0	4.0	12.0 79.0 0.0 1.0 96.0
control 2	6.0	47.0 39.0 2.0 2.0 96.0	79.0	9.0 4.0 2.0 2.0 96.0	62.0	14.0 6.0 1.0 13.0 96.0	50.0	16.0 22.0 6.0 2.0 96.0	4.0	13.0 78.0 0.0 1.0 96.0
control 3	6.0	47.0 40.0 2.0 1.0 96.0	79.0	9.0 4.0 2.0 2.0 96.0	65.0	13.0 7.0 2.0 9.0 96.0	51.0	16.0 22.0 5.0 2.0 96.0	4.0	11.0 79.0 0.0 2.0 96.0
control mean	6.0	47.0 39.7 2.0 1.3 96.0	78.7	9.3 4.0 2.0 2.0 96.0	63.3	13.7 6.3 1.3 11.3 96.0	50.3	16.0 22.3 5.3 2.0 96.0	4.0	12.0 78.7 0.0 1.3 96.0
SE		0.3		0.0		1.2		0.0		0.3
graphene 1	2.0	30.0 59.0 1.0 4.0 96.0	78.0	5.0 6.0 2.0 5.0 96.0	59.0	8.0 11.0 2.0 16.0 96.0	44.0	13.0 28.0 8.0 3.0 96.0	2.0	6.0 86.0 0.0 2.0 96.0
graphene 2	2.0	30.0 59.0 1.0 4.0 96.0	79.0	6.0 7.0 1.0 3.0 96.0	58.0	8.0 12.0 2.0 16.0 96.0	44.0	13.0 29.0 7.0 3.0 96.0	2.0	6.0 84.0 0.0 4.0 96.0
graphene 3	2.0	30.0 60.0 1.0 3.0 96.0	79.0	6.0 7.0 1.0 3.0 96.0	62.0	7.0 12.0 2.0 13.0 96.0	44.0	14.0 29.0 7.0 2.0 96.0	2.0	5.0 87.0 0.0 2.0 96.0
graphene mean	2.0	30.0 59.3 1.0 3.7 96.0	78.7	5.7 6.7 1.3 3.7 96.0	59.7	7.7 11.7 2.0 15.0 96.0	44.0	13.3 28.7 7.3 2.7 96.0	2.0	5.7 85.7 0.0 2.7 96.0
SE		0.3		0.7		1.0		0.3		0.7
graphene oxide 1	2.0	39.0 52.0 1.0 2.0 96.0	79.0	7.0 4.0 2.0 4.0 96.0	51.0	8.0 21.0 4.0 12.0 96.0	42.0	16.0 28.0 6.0 4.0 96.0	3.0	14.0 74.0 0.0 5.0 96.0
graphene oxide 2	2.0	39.0 52.0 1.0 2.0 96.0	78.0	6.0 5.0 2.0 5.0 96.0	50.0	7.0 20.0 4.0 15.0 96.0	43.0	15.0 28.0 6.0 4.0 96.0	3.0	13.0 75.0 0.0 5.0 96.0
graphene oxide 3	2.0	39.0 52.0 1.0 2.0 96.0	79.0	7.0 5.0 2.0 3.0 96.0	53.0	7.0 20.0 4.0 12.0 96.0	42.0	16.0 28.0 6.0 4.0 96.0	3.0	11.0 79.0 0.0 3.0 96.0
graphene oxide mean	2.0	39.0 52.0 1.0 2.0 96.0	78.7	6.7 4.7 2.0 4.0 96.0	51.3	7.3 20.3 4.0 13.0 96.0	42.3	15.7 28.0 6.0 4.0 96.0	3.0	12.7 76.0 0.0 4.3 96.0
SE		0.0		0.6		1.0		0.0		0.7

Table 3

Analysis of variance table used to determine p-values using transformed data. The data was transformed by dividing the CCM by 96 (the number of trials), taking the square root and then the arcsine. DF denotes the degrees of freedom associated with the source of variance. Sum sq. denotes the sum of squares associated with the sources of variance, Total, Model and Error. Mean sq denotes the mean square, (Sum sq./DF). F value is the F-statistic (Mean Square Model divided by the Mean Square Error). The Pr (> F) denotes the p-value associated with the F-statistic. It is used in testing the null hypothesis that all means are equal.

	DF	Sum sq.	Mean sq.	F value	Pr(>F)
Protein	4	0.32033	0.0800082	148.6923	<2.2e-16
Treatment	2	0.02801	0.014004	26.0013	2.815e-07
Protein:treatment	8	0.01233	0.001541	2.8606	0.01724
Residuals	30	0.01616	0.000539		

Table 4

Details of diffraction data collected from three lysozyme crystals grown with addition of either graphene or GO at a final concentration of 33 $\mu\text{g.mL}^{-1}$, or a water-only control.

		lysozyme control	lysozyme and graphene	lysozyme and graphene oxide
Space group		$P 4_3 2_1 2$	$P 4_3 2_1 2$	$P 4_3 2_1 2$
Cell Dimensions	Å	$a=78.99, c=36.96$	$a=78.68, c=37.35$	$a=78.83, 36.96$
Resolution limits (highest resolution bin)	Å	78.99-1.36 (1.43-1.36)	78.68-1.67 (1.76-1.67)	78.83-1.53 (1.61-1.53)
R_{pim}^*		0.02 (0.11)	0.02 (0.20)	0.02 (0.11)
Mean ((I)/sd(I))		23.4(6.6)	33.5(3.7)	28.3(5.8)
Completeness	%	99.8(98.7)	99.6 (97.6)	99.8 (99.0)
Multiplicity		13.4 (13.5)	13.0 (9.9)	11.7 (9.9)

* Precision-indicating R-factor