

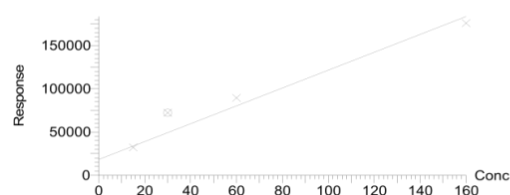
Article

Multifunctional Properties of a *Bacillus thuringiensis* Strain (BST-122): Beyond the Paraspore Crystal

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A

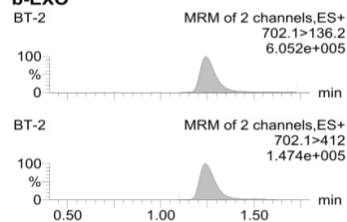
Compound name: b-EXO
Correlation coefficient: $r = 0.993281$, $r^2 = 0.986608$
Calibration curve: $1032.88 * x + 18238.3$
Response type: External Std, Area
Curve type: Linear, Origin: Exclude, Weighting: $1/x$, Axis trans: None



B

Name: 20181121A9, Date: 21-Nov-2018, Time: 11:06:37, ID: , Description: BT-2

b-EXO

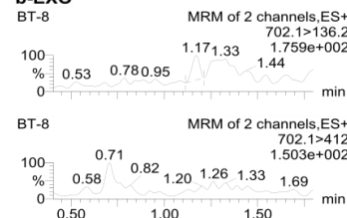


#	Name	Sample Text	Trace	RT	Area	Response	Conc.
1	1 b-EXO	BT-2	702.1>136.2	1.24	61285.598	61285.598	41.6769

C

Name: 20181121A15, Date: 21-Nov-2018, Time: 12:13:48, ID: , Description: BT-8

b-EXO



#	Name	Sample Text	Trace	RT	Area	Response	Conc.
1	1 b-EXO	BT-8	702.1>136.2				

Figure S1. Results of HPLC analysis for detection of production of type I β -exotoxin (thuringiensin). (A): Calibration curve of HPLC analysis, * multiplication symbol; (B): HD-2 strain standard results (positive control); (C): BST-122 results.

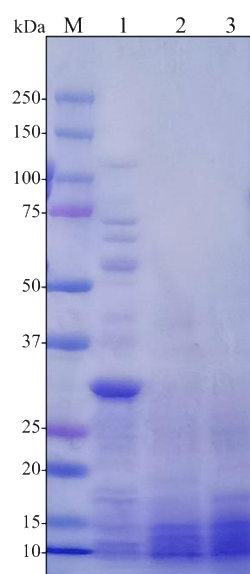


Figure S2. SDS-PAGE from BST-122 strain. M: protein weight marker (kDa); 1: spores and crystal mixture; 2: solubilized in carbonate buffer (pH 11.3); 3: solubilized protein dialyzed in HEPES (pH 8.0).

Table S1. Mean lethal concentration (LC₅₀) value of the BST-122 solubilized protein crystals for J2 *M. incognita* individuals.

LC	Concentration (µg/ml)	Lower limits	Upper limits	χ^2	df	Slope	SE slope	Intercept
LC ₅₀	458	276	11711	24.0	6	1.37	0.224	-3.64

LC: lethal concentration; χ^2 : chi-square; df: degree of freedom; SE: standard error.

Table S2. Mortality percentages of J2 *M. incognita* individuals treated with Cry5_orf65 solubilized protein crystals.

Treatment	Concentration (µg/ml)	Mortality (%) Mean ± SE
Control (HEPES)	0	14.5 ± 5.2 a
BMB171-pSTAB-empty	50	37.7 ± 2.9 a
	150	97.1 ± 2.9 b
BMB171-Cry5_orf65	50	50.7 ± 20.4 ab
	150	86.9 ± 2.5 b

SE: Standard Error. Different letters were used to denote statistical significance between values. Kruskal-Wallis ($\chi^2 = 11.552$, df = 4, $p = 0.02101$), Bonferroni test (p -value < 0.05).

Table S3. Insecticidal protein content of *Bacillus thuringiensis* BMB171.

Target Database ^a	Identity (%) ^b	MW (kDa)	Length (No. Residues)
CalY	55	22.1	197
Endochitinase	99	75.8	688
Bmp1	35	60.9	566
InhA2	99	87.9	799
Sphingomyelin phosphodiesterase	99	33	294
DNRLRE domain-containing protein	100	251.7	2253
CalY	62	21.8	197
CalY	99	20.7	189

InhA1	96	86.7	796
InhA2	29	72.9	656
Bmp1	97	89.4	817
InhA3	99	76.2	692
InhA1	78	86.9	795
Bmp1	61	62.4	567
Hemolytic enterotoxin HBL	99	52.3	466
Bmp1	61	62.5	565
Enhancin_Bel	100	85.6	742
Exochitinase	99	39.4	360
ColB	78	48.3	426
ColB	34	35.6	309
CalY	50	22.3	201
Bmp1	36	65.3	591
Spp1Aa1	82	40.2	362
Bmp1	32	64.6	583

^a**Bold**, described toxicity to nematodes. ^bLocal alignment using BLASTP.

Table S4. Comparative of strains BST-122 and BMB171 secreted factors.

Target Database ^a	BST-122		BMB171	
	% Identity ^b	MW (kDa)	% Identity ^b	MW (kDa)
Bmp1	36	65.3	97	89.4
ColB	78	48.3	78	48.3
Endochitinase	-	-	99	75.8
Exochitinase	99	39.4	99	39.4
CalY	56	22	99	20.7
InhA1	96	86.7	96	86.7
InhA2	99	87.9	99	87.9
InhA3	-	-	99	76.2
Enhancin_Bel	99	85.5	100	85.6
Sphingomyelin phosphodiesterase	100	37.0	99	33.0

^a**Bold**, described toxicity to nematodes. ^bLocal alignment using BLASTP.

Table S5. Primers used in this study.

Primers	Sequences (5'-3') ^a
For gene cloning	
Cry5Ad-like_NcoI_FW	CCCATGGCCAAATGATTGCATATCTTGATT
Orf2_Cry65_Eco52I_RV	CCCGGCCGCTCCGCGTGTATTCTTATA
For gene sequencing	
Cry5As-like-seq-FW	CATTACAGCAATTGCACACC
Cry5As-like-seq-RV	CCAGTAACAACAAAGCTGC

^a*Italic*, restriction enzyme site included in the primer sequence.