

Table S1. List of oligonucleotides designed and used in the study.

Gene ID	Primer sequence (5' – 3')	Encoding protein	Product size
Csa_5G643380	GGGCGGAGTCAAGAGAGCGT TGCCACCACATCTGACCTGCT	endo-1,3(4)-beta-glucanase 3	146
Csa_2G010370	GCCCTTGAATTGGCGAAACAC TTCGACCTGATGGGCCGACG	pathogenesis-related protein PR-4	68
Csa_2G406640	AGACGCCGAGAAGGACGCTC TCGGCGCAAGAGACTCGACC	peroxidase	117
Csa_3G743950	TTAACGAGCGGTCCAGGCCA ACCGCAATCGCCAGTCATGC	thaumatin-like	165
Csa_2G011420	AGAGCTTTGCCCTCCGACGA GGCCACCCCACTTGTGC	auxin-responsive protein IAA4	119
Csa_7G007930	ACAACATCCGCCGCATCGTG CGCGGTCACTCGAGCCTTCT	indole-3-acetic acid-induced protein ARG7	71
Csa_3G866530	ATCCCTTCCGACGTACCCGC GACTCGTCGCAAGGGATTGCC	auxin-responsive 6B	176
Csa_3G035310	ACCGGGTTCGCTCTACACCG CGCCTGCAGCAATTTCCGGA	auxin-responsive protein SAUR32	143

Table S2. Gene Ontology (GO) enrichment results of up-regulated differentially expressed genes (DEGs) in *Bacillus subtilis* MBI600 (*Bs* MBI600) - treated cucumber roots. DEGs are categorized according to biological process and molecular function.

Biological process								Molecular function			
24 hours				48 hours				24 hours			
Enrichment FDR	Genes in list	Total genes	Functional Category	Enrichment FDR	Genes in list	Total genes	Functional Category	Enrichment FDR	Genes in list	Total genes	Functional Category
8,90E-03	6	20	Regulation of transmembrane transport	2,20E-05	71	1247	Regulation of macromolecule biosynthetic process	5,20E-04	69	1295	DNA binding
1,60E-02	57	1247	Regulation of macromolecule biosynthetic process	2,20E-05	72	1234	Regulation of nucleobase-containing compound metabolic process	5,20E-04	35	496	Transcription regulator activity
1,60E-02	3	7	Amine transport	2,20E-05	110	2223	Regulation of biological process	6,80E-04	32	451	DNA-binding transcription factor activity
1,60E-02	59	1234	Regulation of nucleobase-containing compound metabolic process	2,20E-05	77	1389	Regulation of nitrogen compound metabolic process	3,10E-03	100	2230	Nucleic acid binding
1,60E-02	58	1268	Regulation of cellular biosynthetic process	2,20E-05	70	1200	Regulation of RNA metabolic process				
1,60E-02	3	7	Regulation of organic acid transport	2,20E-05	69	1179	Regulation of transcription, DNA-templated				

1,60E-02	3	7	Amino acid export across plasma membrane	2,20E-05	69	1186	Regulation of nucleic acid-templated transcription
1,60E-02	5	19	Regulation of ion transmembrane transport	2,20E-05	71	1240	Regulation of cellular macromolecule biosynthetic process
1,60E-02	5	22	Regulation of ion transport	2,20E-05	69	1186	Regulation of RNA biosynthetic process
1,60E-02	4	10	Regulation of anion transport	2,90E-05	77	1407	Regulation of primary metabolic process
1,60E-02	8	62	Negative regulation of transcription, DNA-templated	3,30E-05	71	1268	Regulation of cellular biosynthetic process
1,60E-02	6	37	Regulation of transport	3,30E-05	71	1271	Regulation of biosynthetic process
1,60E-02	57	1200	Regulation of RNA metabolic process	3,70E-05	71	1279	Transcription, DNA-templated
1,60E-02	3	7	Regulation of amine transport	3,70E-05	34	442	Ion transport
1,60E-02	3	7	Regulation of amino acid transport	3,80E-05	72	1312	Regulation of gene expression
1,60E-02	55	1179	Regulation of transcription, DNA-templated	3,80E-05	77	1445	Regulation of cellular metabolic process

1,60E-02	3	7	Regulation of amino acid export	3,80E-05	71	1295	RNA biosynthetic process
1,60E-02	4	14	Erythrose 4-phosphate/phosphoenolpyruvate family amino acid catabolic process	3,80E-05	78	1470	Regulation of macromolecule metabolic process
1,60E-02	8	68	Negative regulation of RNA biosynthetic process	3,80E-05	117	2478	Biological regulation
1,60E-02	56	1186	Regulation of nucleic acid-templated transcription	3,80E-05	71	1291	Nucleic acid-templated transcription
1,60E-02	8	68	Negative regulation of nucleic acid-templated transcription	3,80E-05	26	293	Cation transport
1,60E-02	3	7	Regulation of amino acid transmembrane transport	6,10E-05	19	178	Metal ion transport
1,60E-02	4	9	Regulation of anion transmembrane transport	1,00E-04	77	1488	Nucleobase-containing compound biosynthetic process
1,60E-02	56	1240	Regulation of cellular macromolecule biosynthetic process	1,10E-04	82	1622	Heterocycle biosynthetic process
1,60E-02	56	1186	Regulation of RNA biosynthetic process	1,10E-04	97	2013	Regulation of cellular process
1,60E-02	4	14	L-phenylalanine catabolic process	1,50E-04	84	1689	Organic cyclic compound biosynthetic process

1,60E-02	58	1271	Regulation of biosynthetic process	1,70E-04	78	1543	Regulation of metabolic process
1,60E-02	90	2223	Regulation of biological process	2,80E-04	80	1617	Aromatic compound biosynthetic process
1,70E-02	61	1389	Regulation of nitrogen compound metabolic process	4,60E-03	91	2063	Cellular nitrogen compound biosynthetic process
1,70E-02	8	70	Negative regulation of RNA metabolic process	7,30E-03	19	258	Ion transmembrane transport

Table S3. Gene Ontology (GO) enrichment results of down-regulated differentially expressed genes (DEGs) in *Bacillus subtilis* MBI600 (*Bs* MBI600) - treated cucumber roots. DEGs are categorized according to biological process and molecular function.

Biological process							
24 hours				48 hours			
Enrichment FDR	Genes in list	Total genes	Functional Category	Enrichment FDR	Genes in list	Total genes	Functional Category
1,20E-07	115	1247	Regulation of macromolecule biosynthetic process	9,30E-06	7	14	Erythrose 4-phosphate/phosphoenolpyruvate family amino acid catabolic process
1,20E-07	114	1234	Regulation of nucleobase-containing compound metabolic process	9,30E-06	7	14	L-phenylalanine catabolic process
1,20E-07	116	1268	Regulation of cellular biosynthetic process	3,30E-05	7	17	Aromatic amino acid family catabolic process
1,20E-07	112	1200	Regulation of RNA metabolic process	9,90E-05	8	30	Alpha-amino acid catabolic process
1,20E-07	111	1179	Regulation of transcription, DNA-templated	9,90E-05	7	21	Erythrose 4-phosphate/phosphoenolpyruvate family amino acid metabolic process
1,20E-07	112	1186	Regulation of nucleic acid-templated transcription	9,90E-05	7	21	L-phenylalanine metabolic process
1,20E-07	114	1240	Regulation of cellular macromolecule biosynthetic process	1,30E-04	5	9	Cinnamic acid biosynthetic process
1,20E-07	112	1186	Regulation of RNA biosynthetic process	1,30E-04	5	9	Cinnamic acid metabolic process
1,20E-07	116	1271	Regulation of biosynthetic process	1,50E-04	8	33	Cellular amino acid catabolic process
5,50E-07	269	3771	Cellular macromolecule metabolic process	1,70E-04	29	408	Small molecule biosynthetic process

8,40E-07	114	1291	Nucleic acid-templated transcription	3,60E-04	116	2876	Biosynthetic process
8,50E-07	114	1295	RNA biosynthetic process	3,60E-04	10	63	Aromatic amino acid family metabolic process
8,50E-07	113	1279	Transcription, DNA-templated	3,60E-04	48	904	Small molecule metabolic process
1,60E-06	114	1312	Regulation of gene expression	3,60E-04	8	39	Secondary metabolite biosynthetic process
4,20E-06	121	1445	Regulation of cellular metabolic process	6,60E-04	10	69	Small molecule catabolic process
4,80E-06	117	1389	Regulation of nitrogen compound metabolic process	8,50E-04	5	14	Benzene-containing compound metabolic process
5,00E-06	118	1407	Regulation of primary metabolic process	8,50E-04	49	968	Organonitrogen compound biosynthetic process
1,10E-05	155	2013	Regulation of cellular process	1,00E-03	109	2752	Cellular biosynthetic process
2,40E-05	119	1470	Regulation of macromolecule metabolic process	1,20E-03	110	2796	Organic substance biosynthetic process
2,40E-05	156	2060	Macromolecule biosynthetic process	1,20E-03	29	471	Peptide metabolic process
2,70E-05	128	1617	Aromatic compound biosynthetic process	1,30E-03	8	49	Organic acid catabolic process
3,10E-05	123	1543	Regulation of metabolic process	1,30E-03	8	49	Carboxylic acid catabolic process
3,40E-05	153	2028	Cellular macromolecule biosynthetic process	1,30E-03	6	25	Phenylpropanoid biosynthetic process
4,60E-05	164	2223	Regulation of biological process	1,40E-03	31	529	Cellular amide metabolic process
4,60E-05	131	1689	Organic cyclic compound biosynthetic process	1,50E-03	9	65	Secondary metabolic process

4,60E-05	37	307	Response to oxygen-containing compound	1,50E-03	15	169	Alpha-amino acid metabolic process
4,60E-05	77	856	Protein phosphorylation	2,20E-03	116	3055	Organonitrogen compound metabolic process
4,70E-05	15	69	Response to fungus	2,30E-03	27	448	Translation
4,90E-05	12	45	Response to wounding	2,70E-03	6	30	Xyloglucan metabolic process
5,70E-05	7	14	Benzene-containing compound metabolic process	2,70E-03	27	454	Peptide biosynthetic process
Molecular function							
24 hours				48 hours			
Enrichment FDR	Genes in list	Total genes	Functional Category	Enrichment FDR	Genes in list	Total genes	Functional Category
3,80E-17	75	451	DNA-binding transcription factor activity	3,20E-06	8	19	Ammonia-lyase activity
3,80E-17	79	496	Transcription regulator activity	2,80E-05	9	35	Carbon-nitrogen lyase activity
5,80E-09	122	1295	DNA binding	3,60E-05	28	356	Structural molecule activity
1,80E-06	31	197	Calcium ion binding	4,00E-05	25	301	Structural constituent of ribosome
8,50E-06	37	281	Sequence-specific DNA binding	6,50E-05	5	9	Phenylalanine ammonia-lyase activity
6,90E-05	75	828	Protein kinase activity	3,60E-04	20	242	Lyase activity
7,60E-05	8	19	Ammonia-lyase activity	9,60E-04	6	24	Cysteine-type endopeptidase inhibitor activity
7,80E-05	12	46	Calmodulin binding	2,10E-03	6	28	Xyloglucan:xyloglucosyl transferase activity
8,80E-05	85	992	Kinase activity	2,80E-02	9	97	Magnesium ion binding
2,60E-04	79	932	Phosphotransferase activity, alcohol group as acceptor	2,80E-02	6	46	Calmodulin binding
4,00E-04	297	4633	Organic cyclic compound binding	3,00E-02	6	49	Endopeptidase inhibitor activity

4,00E-04	297	4632	Heterocyclic compound binding	3,00E-02	6	49	Peptidase inhibitor activity
4,50E-04	158	2230	Nucleic acid binding	3,00E-02	6	49	Endopeptidase regulator activity
5,80E-04	120	1614	Catalytic activity, acting on a protein	3,10E-02	6	50	Peptidase regulator activity
6,10E-04	5	9	Phenylalanine ammonia-lyase activity				
3,20E-03	87	1147	Transferase activity, transferring phosphorus-containing groups				
4,70E-03	8	35	Carbon-nitrogen lyase activity				
5,80E-03	7	28	Xyloglucan:xyloglucosyl transferase activity				
6,30E-03	157	2356	Transferase activity				
1,30E-02	13	94	Glucosyltransferase activity				
2,10E-02	27	285	Protein serine/threonine kinase activity				
2,60E-02	10	68	Aspartic-type endopeptidase activity				
2,60E-02	10	68	Aspartic-type peptidase activity				
3,60E-02	14	120	Ubiquitin-protein transferase activity				
3,70E-02	9	61	Protein serine/threonine phosphatase activity				
4,50E-02	14	124	Ubiquitin-like protein transferase activity				

Table S4. Metabolite amounts (means \pm SE) expressed as relative abundance based on the relative response compared to internal standard adonitol, 24 h post application of *Bacillus subtilis* MBI600 (*Bs* MBI600).

Substance	Control	<i>Bs</i> MBI600
Water-soluble sugars		
4-Ketoglucose	0.078 \pm 0.015	0.095 \pm 0.0001
Ribofuranose	0.159 \pm 0.039	0.102 \pm 0.0001
Fructose	0.626 \pm 0.014	0.723 \pm 0.062
Sorbose	0.560 \pm 0.045	0.662 \pm 0.049
Glucose	2.922 \pm 0.470	3.538 \pm 0.347
Mannose	0.456 \pm 0.053	0.600 \pm 0.053
Talopyranose	0.126 \pm 0.016	0.163 \pm 0.0001
Mannopyranoside	0.096 \pm 0.008	0.127 \pm 0.0002
Galactopyranoside	0.227 \pm 0.077	0.233 \pm 0.055
D-Allose	nd ^a	0.190 \pm 0.027
Sucrose	0.194 \pm 0.067	0.172 \pm 0.013
Sugar alcohols		
	0.125 \pm	
Mannitol	0.0003	0.114 \pm 0.009
Glycerol	1.236 \pm 0.086	0.997 \pm 0.108
Myo inositol	1.889 \pm 0.063	2.229 \pm 0.347
Other organic substances		
Aucubin	1.735 \pm 0.258	1.739 \pm 0.519
Gluconic acid, γ -lactone	nd	0.171 \pm 0.044
Phosphoric acid	2.160 \pm 0.103	1.826 \pm 0.175

Urea	0.460 ± 0.155	nd
Organic acids		
Malic acid	0.923 ± 0.128 0.080 ±	4.591 ± 0.189* ^b
Threonic acid	0.0002	0.097 ± 0.006
Erythronic acid	0.249 ± 0.018	0.350 ± 0.022*
Glyceric acid	2.068 ± 0.569	5.721 ± 0.529*
Octadecatrienoic acid	0.088 ± 0.015 0.088 ±	0.112 ± 0.002
Arabinonic acid	0.0007	0.103 ± 0.0001*
Butanoic acid	0.170 ± 0.006	0.229 ± 0.005*
Ribonic acid	0.239 ± 0.097	0.088 ± 0.010
Gluconic acid	0.252 ± 0.177	0.109 ± 0.005
Citric acid	0.302 ± 0.040	0.479 ± 0.096
Hexadecanoic acid	0.098 ± 0.023	nd
Linoleic acid	0.116 ± 0.018	0.117 ± 0.020
Amino acids		
Alanine	0.653 ± 0.133	1.134 ± 0.120*
Valine	0.247 ± 0.069	0.208 ± 0.018
Citrulline	0.131 ± 0.024	nd
Norleucine	0.221 ± 0.027	0.166 ± 0.023
Glycine	0.140 ± 0.029	0.125 ± 0.015
Oxoproline	0.126 ± 0.023	0.174 ± 0.025
Serine	0.195 ± 0.052	0.133 ± 0.0001
Proline	0.989 ± 0.215	0.586 ± 0.139
Threonine	0.407 ± 0.103	0.379 ± 0.048
Glutamic acid	1.425 ± 0.336	1.204 ± 0.121
Aspartic acid	0.468 ± 0.088	0.573 ± 0.051

Gamma-aminobutyric acid (GABA)	1.018 ± 0.146	0.853 ± 0.065
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^and stands for not detected

^bAsterisks indicate values that differ significantly at each time point (pairwise t-test, $P \leq 0.05$)

Table S5. Metabolite amounts (means \pm SE) expressed as relative abundance based on the relative response compared to internal standard adonitol 48 h post application of *Bacillus subtilis* MBI600 (*Bs* MBI600).

Substance	Control	<i>Bs</i> MBI600
Water-soluble sugars		
4-Ketoglucose	0.087 \pm 0.013	0.084 \pm 0.012
Ribofuranose	0.117 \pm 0.034	0.065 \pm 0.011
Fructose	0.629 \pm 0.121	0.326 \pm 0.020* ^b
Sorbose	0.698 \pm 0.050	0.389 \pm 0.079*
Galactose	nd ^a	0.036 \pm 0.010
Glucose	2.973 \pm 0.569	1.610 \pm 0.104
Mannose	0.543 \pm 0.066	0.310 \pm 0.023*
Talopyranose	0.188 \pm 0.024	0.097 \pm 0.009*
Xylopyranose	nd	0.150 \pm 0.066
Mannopyranoside	0.146 \pm 0.027	0.313 \pm 0.205
Galactopyranoside	0.302 \pm 0.026	0.142 \pm 0.019
Ribose	0.117 \pm 0.020	0.075 \pm 0.011
D-Allose	0.115 \pm 0.082	0.107 \pm 0.0001
Allopyranose	0.081 \pm 0.010	0.045 \pm 0.008*
Sucrose	0.433 \pm 0.090	0.147 \pm 0.042
Sugar alcohols		
Mannitol	0.228 \pm 0.017	0.110 \pm 0.019*
Glycerol	1.143 \pm 0.111	1.172 \pm 0.103
Myo inositol	2.740 \pm 0.349	1.270 \pm 0.309*
Other organic substances		

Aucubin	0.529 ± 0.122	$0.154 \pm 0.083^*$
Gluconic acid, γ -lactone	nd	0.085 ± 0.036
PhosphoricAcid	1.908 ± 0.199	1.959 ± 0.184
Urea	nd	0.171 ± 0.031

Organic acid

Malic acid	2.627 ± 0.531	5.011 ± 0.518
Threonic acid	0.190 ± 0.072	$0.074 \pm 0.003^*$
Erythronic acid	0.276 ± 0.024	$0.368 \pm 0.018^*$
Glyceric acid	nd	0.036 ± 0.005
Octadecatrienoic acid	0.095 ± 0.006	0.082 ± 0.010
Citric acid	0.302 ± 0.040	0.479 ± 0.096
Arabinonic acid	0.066 ± 0.013	0.073 ± 0.011
	$0.160 \pm$	
Butanoic acid	0.0001	$0.130 \pm 0.007^*$
Ribonic acid	0.118 ± 0.032	0.116 ± 0.010
DeoxyErythroPentonic acid	0.187 ± 0.040	0.092 ± 0.021
ButanedioicAcid	0.078 ± 0.009	0.109 ± 0.009
Glyceric acid	2.070 ± 0.583	3.132 ± 0.474
Butanoic acid	nd	0.067 ± 0.023
Gluconic acid	0.093 ± 0.012	0.048 ± 0.005
Linoleic acid	0.227 ± 0.070	0.113 ± 0.014

Amino acids

Alanine	0.568 ± 0.074	$0.896 \pm 0.119^*$
Isoleucine	nd	0.078 ± 0.013
Valine	0.232 ± 0.032	$0.435 \pm 0.078^*$
Citrulline	nd	0.043 ± 0.007
Norleucine	0.203 ± 0.037	0.357 ± 0.063

Glycine	0.098 ± 0.016	0.149 ± 0.026
Oxoproline	0.334 ± 0.084	0.249 ± 0.041
Serine	0.161 ± 0.019	0.093 ± 0.019*
Proline	0.236 ± 0.045	0.330 ± 0.063
Threonine	nd	0.037 ± 0.005
Asparagine	0.113 ± 0.023	0.100 ± 0.012
Leucine	0.204 ± 0.029	0.272 ± 0.042
Glutamic acid	0.500 ± 0.055	0.652 ± 0.100
Aspartic acid	1.177 ± 0.250	1.289 ± 0.208
Gamma-aminobutyric acid (GABA)	0.522 ± 0.042	1.169 ± 0.091*

^and stands for not detected

^bAsterisks indicate values that differ significantly at each time point (pairwise t-test, $P \leq 0.05$)