Supplementary Materials

Biotransformation of Benzoate to 2,4,6-Trihydroxybenzophenone by Engineered *Escherichia coli*

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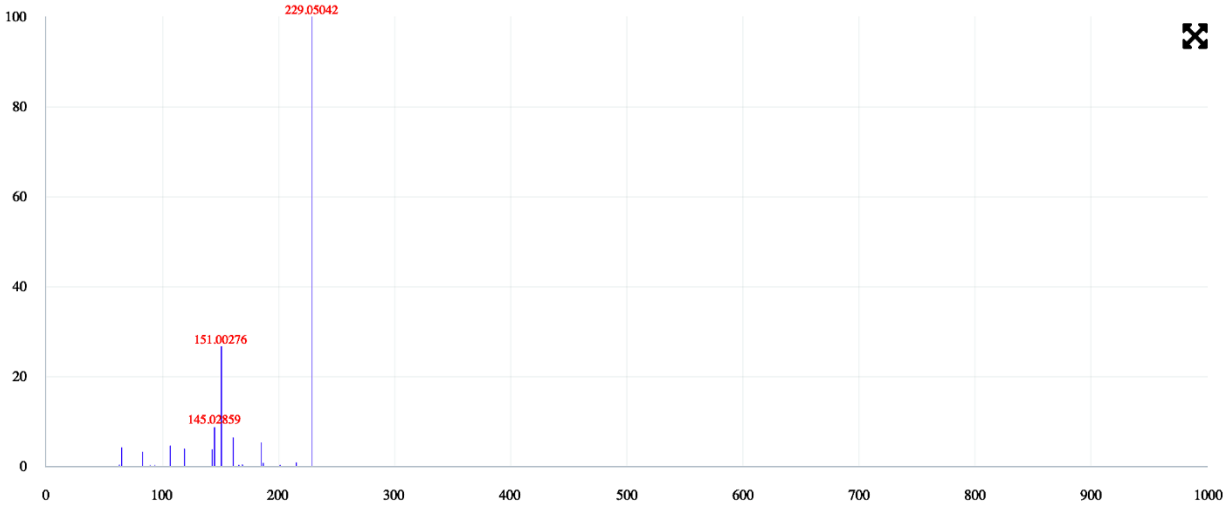
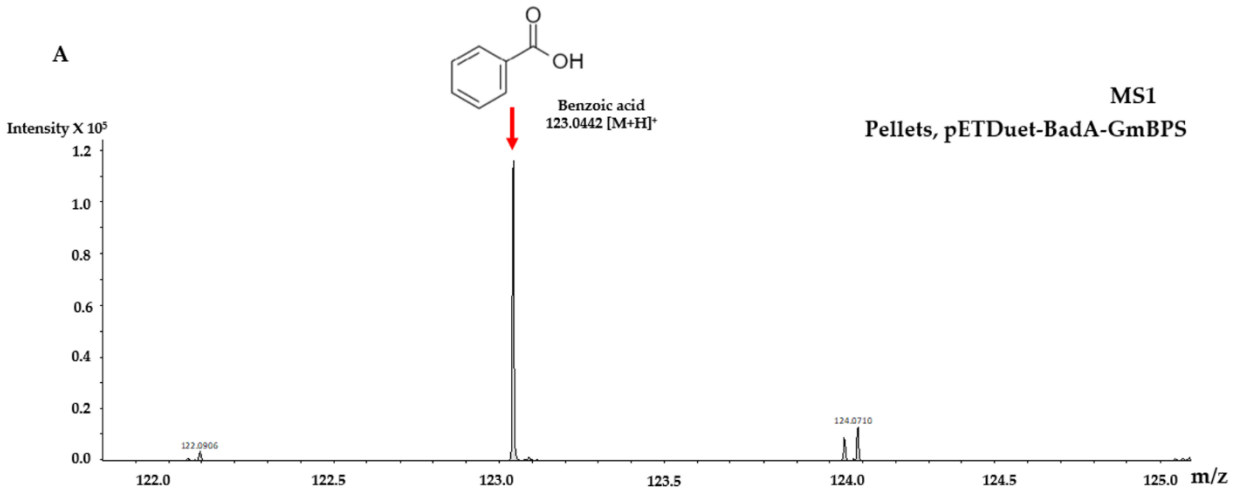


Figure S1. The MS/MS spectra of 2,4,6-TriHB(−) reported in PubChem database.



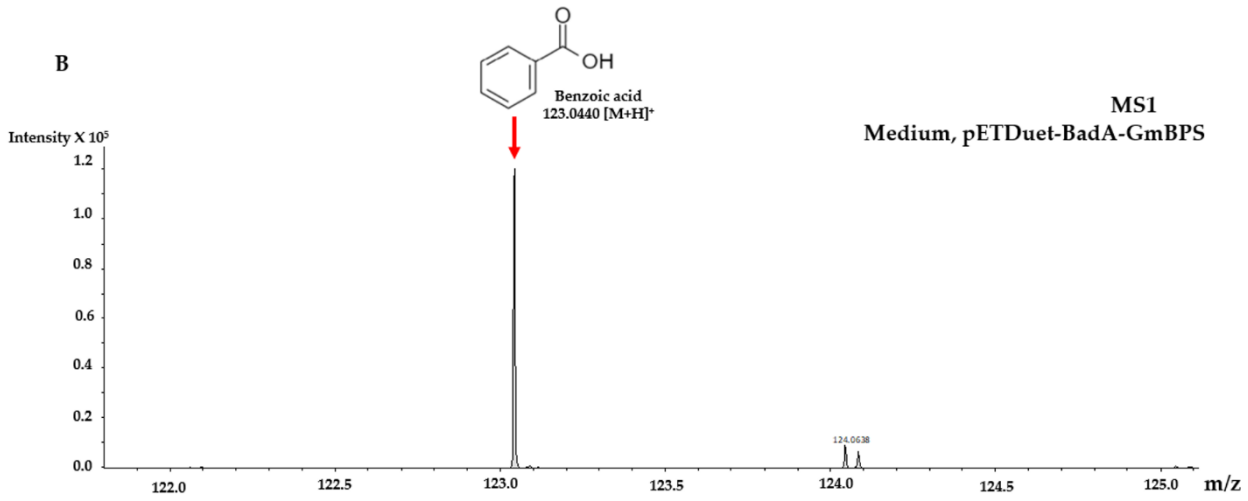


Figure S2. The EICs for the putative benzoic acid (calc. 123.044605 $[M+H]^+$) detected both from pellets (**A**) and medium (**B**) of clones harbored pETDuet-BadA-GmBPS.

>L42322.1:19-1584 Rhodopseudomonas palustris benzoate-coenzyme A ligase (badA) gene, complete cds

ATGAATGCAGCCGCGGTCACGCCGCCACCCGAGAAGTTTAATTTTGCCGAGCACCTGCTGCAGACCAATC

GCGTGCGGCCGGACAAGACGGCGTTCGTCGACGACATCTCGTCGCTGAGCTTCGCGCAACTCGAAGCTCA

GACGCGTCAGCTCGCCGCCGCCTTACGCGCGATCGGGGGTGAAACGCGAAGAGCGCGTGCTGCTGCTGATG

CTCGACGGCACGGATTGGCCGGTGGCGTTTCTCGGCGCAATCTACGCCGGCATCGTGCCGGTCGCGGTCA

ATACGCTGCTGACGGCGGACGACTACGCCTACATG***XhoI***CTCGAGCATTCGCGGGCTCAGGCCGTGCTGGTCAG

CGGCGCGCTGCACCCGGTGCTCAAGGCAGCGCTGACCAAGAGCGATCACGAGGTGCAGCGAGTGATCGTT

TCGCGCCCAGCGCTCCGCTGGAGCCGGGCGAGGTCGACTTCGCTGAGTCGGCGCACATCGCTTGAGAAGC

CTGCCGCTACGCAAGCGGACGATCCGGCGTTCTGGCTGTATTCGTCGGGTTCTACCGGGCGGCCGAAGGG

CGTGGTGCACACTCACGCCAATCCGTACTGGACCTCGGAGCTGTACGGCCGCAACACGCTGCATCTGCGC

GAAGACGACGTCTGCTTTTCGGCGGCCAAACTGTTTTTCGCTTACGGCCTCGGCAACGCGCTGACGTTTC

CGATGACGGTCGGCGCGACCACGCTGCTGATGGGCGAGCGACCGACGCCGGACGCGGTGTTCAAGCGCTG

GCTCGGCGGCGTCGGCGGTGTGAAACCGACCGTGTTCTACGGCGCGCCCACCGGCTACGCCGGCATGTTG

GCCGCGCCGAACCTGCCGTCGCGCGACCAGGTGGCGTTGCGGCTCGCGTCGTCGGCGGGCGAAGCACTGC

CGGCGGAGATTGGGCAGCGCTTCCAGCGCCATTTCGGCCTCGACATCGTCGATGGCATCGGCTCGACCGA

GATGCTGCACATCTTTCTGTCGAACCTGCCAGACCGGGTGCGCTACGGCACCACCGGATGGCCGGTGCCG

GGCTATCAGATCGAGCTGCGCGGCGACGGCGGCGGACCGGTCGCCGACGGAGAGCCGGGCGATCTCTACA

TTCACGGCCCGTCATCGGCGACGATGTATTGGGGCAACCGGGCCAAGAGCCGCGACACCTTCCAGGGCGG

CTGGACCAAGAGCGGCGACAAATACGTCCGCAACGACGACGGCTCCTACACCTATGCGGGCCGCACCGAC

GACATGCTGAAGGTCAGCGGCATCTATGTCAGCCCGTTCGAGATCGAAGCGACGCTGGTGCAGCATCCCG

GTGTGCTCGAAGCCGCAGTGGTCGGGGTCGCCGACGAACACGGCCTGACCAAACCGAAGGCCTATGTGGT

GCCGCGGCCCGGCCAGACCCTGTCGGAGACCGAGCTGAAGACCTTCATCAAGGATCGACTGGCGCCGTAC

AAATATCCGCGCAGCACGGTGTTCGTCGCCGAATTGCCGAAGACGGCGACCGGCAAGATTCAGCGCTTCA

AGCTGCGCGAGGGTGTGTTGGGCTGA

Figure S3. The *XhoI* recognition site* detected in the BadA coding sequence. * The restriction site was analysed by NEBcutter V2.0 (http://nc2.neb.com/NEBcutter2/).

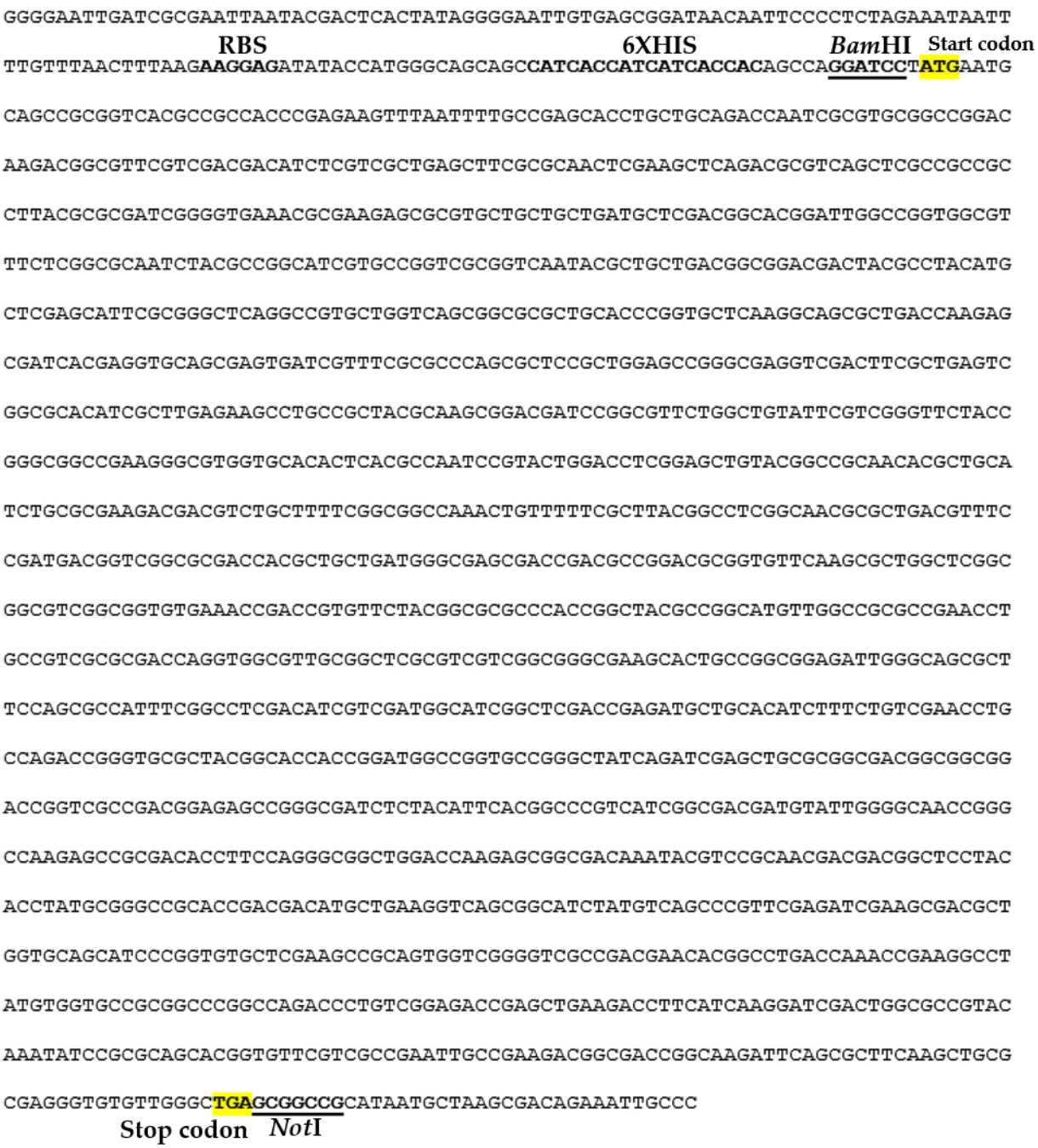


Figure S4. The insertion of BadA in MCS-1 region of pETDuet-1 vector.

RBS

AGGGAAACTACTATAGGGGATTGTGAGCGGATAACAATTCCCCATCTTAGTATATTAGTTAAGTATAAG**AAGGAG**AT

Start codon

ATA**CATATG**GCACCTGCAATGGATTCTGCCCAAAATGGCCACCAAAGCCGAGGTTCTGCCAATGTTCTAGCAATTGG

*Nde*I

CACTGCAAATCCACCAAACGTCATTCTGCAAGAAGACTATCCTGATTTTTACTTTAAAGTCACAAATAGCGAGCATT

TAACTGACTTGAAAGAAAAATTTAAGCGTATATGTGTTAAATCAAAAACCAGGAAGAGGCATTTTTATCTAACAGAG

CAAATCCTTAAAGAGAACCCAGGCATTGCAACCTATGGAGCAGGATCTCTCGACTCTCGTCAAAAGATTCTCGAAAC

GGAGATTCCTAAGCTCGGAAAAGAGGCTGCAATGGTAGCCATCCAAGAATGGGGACAACCAGTGTCGAAAATCACCC

ACGTTGTATTCGCCACCACTTCAGGCTTTATGATGCCCGGAGCTGACTACTCAATCACCAGGCTTCTCGGCCTAAAT

CCCAATGTAAGGCGCGTAATGATCTATAACCAAGGATGCTTTGCTGGTGGCACGGCCCTTCGTGTTGCCAAGGACCT

TGCAGAGAACAACAAGGGTGCTCGTGTCCTTGTAGTTTGCGCTGAGAACACAGCCATGACCTTCCACGGACCTAATG

AGAATCATTTAGATGTGTTGGTTGGTCAGGCTATGTTCTCTGATGGCGCGGCTGCTTTGATCATTGGAGCCAACCCC

AATTTACCAGAGGAACGTCCAGTTTATGAGATGGTAGCAGCCCATCAAACCATTGTCCCTGAGTCGGATGGGGCGAT

AGTTGCGCATTTTTATGAGATGGGAATGAGTTACTTCCTCAAAGAGAATGTTATTCCTCTTTTTGGCAACAACATTG

AGGCTTGCATGGAGGCCGCATTTAAGGAATATGGAATTAGTGACTGGAACTCCTTGTTCTATTCTGTGCATCCTGGT

GGCCGGGCTATCGTTGATGGAATCGCAGAGAAGTTGGGTCTTGATGAAGAAAACCTCAAGGCAACAAGGCACGTTTT

GAGTGAATATGGGAATATGGGTTCTGCTTGTGTCATCTTTATCCTCGATGAGCTGAGGAAGAAATCCAAAGAGGAGA

AGAAGCTCACCACTGGAGATGGAAAGGAGTGGGGTTGCCTTATTGGATTAGGCCCGGGACTCACCGTGGAGACCGTC

GTACTTCGTAGTGTGCCAATAGCA**CTCGAG**TCTGGT**AAAGAAACCGCTGCTGCGAAATTTGAACGCCAGCACATGGA**

*Xho*I

S-Tag

CTCGTCTACTAGCGCAGCTTTATTTCCTTTCCCCCT

Figure S5. The insertion of GmBPS in MCS-2 region of pETDuet-1 vector.

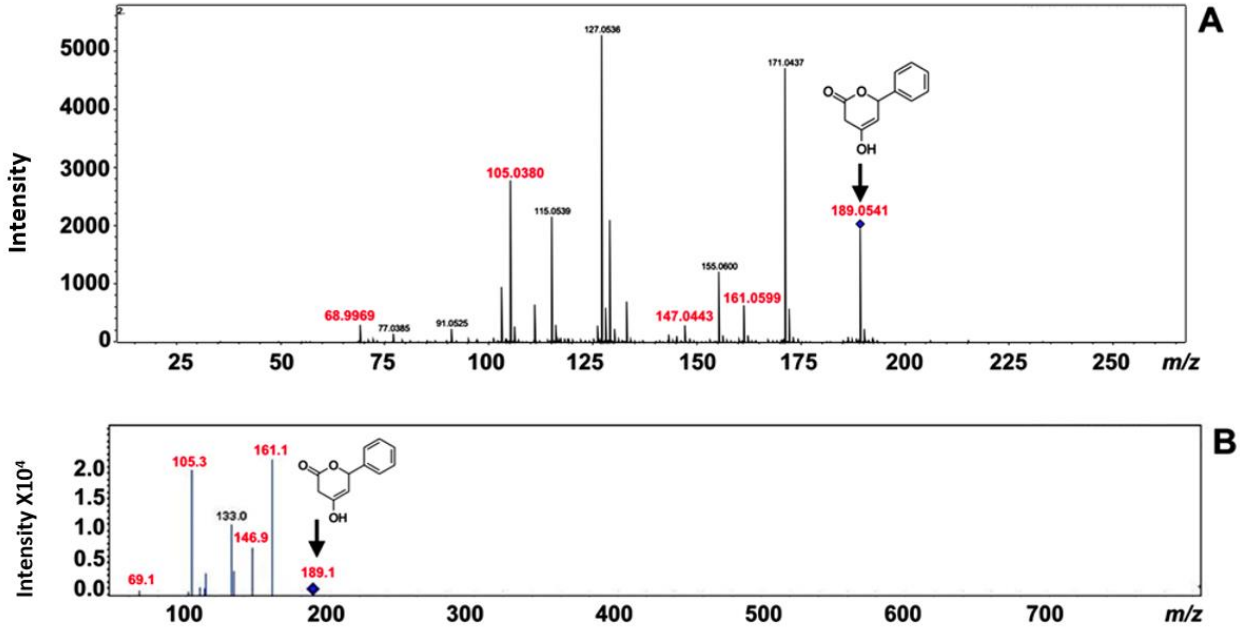


Figure S6. The MS/MS spectra of triketide lactone. (**A**) The MS/MS spectra of triketide lactone obtained from the enzymatic reaction of GmBPS reported in this study; (**B**) The MS/MS spectra of triketide lactone reported by Nualkaew et al. 2012 [12].

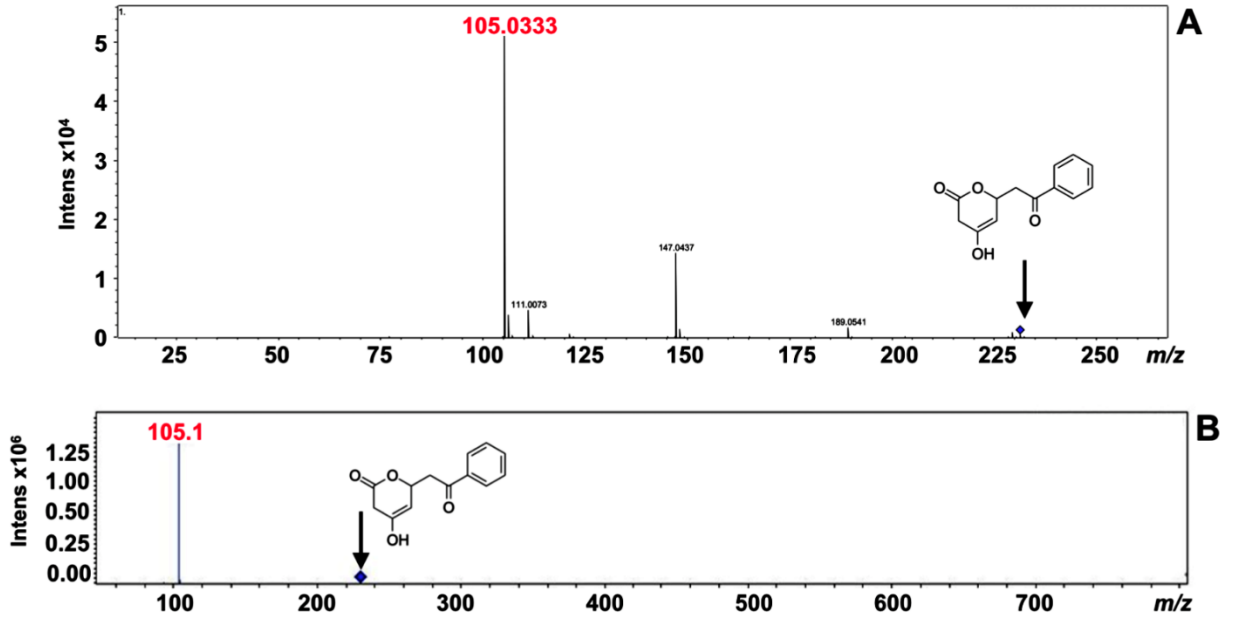


Figure S7. The MS/MS spectra of tetraketide lactone. (**A**) The MS/MS spectra of tetraketide lactone obtained from the enzymatic reaction of GmBPS reported in this study; (**B**) The MS/MS spectra of tetraketide lactone reported by Nualkaew et al. 2012 [12].

m/z	Intensity
105.0335	1809
153.0179	12899
231.0646	3009

Figure S8. The raw mass data of 2,4,6-TriHB measured by positive ion mode LC-MS/MS.

m/z	Intensity
145.0288	17212
151.0033	7548
161.0604	1480
185.0597	788
229.0496	788

Figure S9. The raw mass data of 2,4,6-TriHB measured by negative ion mode LC-MS/MS.