Supplementary Material

One-Pot Biocatalytic Upgrading Lignin-Derived Phenol and Catechol to Hydroxytyrosol

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1. Chemicals

Catechol, phenol and L-DOPA were from Titan Scientifc (Shanghai, China). Hydroxytyrosol, 3,4-dihydroxyphenylpyruvic acid and dopamine were from Bide Pharmatech Ltd (Shanghai, China). Pyridoxal-5'-phosphate (PLP) were from Energy Chemical (Shanghai, China). Tryptone, yeast extract, isopropyl β-D-thiogalactoside (IPTG), kanamycin, ampicillin, streptomycin and Taq plus DNA polymerase were from Sangon Biotech (Shanghai, China). Molecular biological reagents, such as T4 DNA ligase and restriction endonuclease were from New England Biolabs (Beijing, China). Plasmid isolation kit was from Tiangen (Shanghai, China). All other chemicals were of analytical grade and were commercially available.

2. Strains, Plasmids, and Culture Conditions

E. coli T7 super-competent cells was obtained from New England Biolabs (Beijing, China). The plasmids pET28a, pETduet-1, pCDFduet-1 and pRSFduet-1 for the heterogeneous expression studies were obtained from Novagen (Shanghai, China). The *E. coli* strains were grown at 37°C in Luria–Bertani (LB) medium for 7 h, and then a 250 mL Erlenmeyer flask containing 50 mL of TB medium was inoculated with 1 mL of seed liquid and incubated at 37 °C and 200 rpm. When the optical density (OD) at 600 nm reached 0.6-0.8, IPTG was added to a final concentration of 0.5 mM and cultured overnight (12-14 h) at 20 °C and 200 rpm.

3. Construction of the recombinant *E. coli* strains

All the primers used in this study were synthesized by Tsingke (Beijing, China) and list in Table S1.

For *E. coli* (pET28a-FnTPL), the gene of tyrosine phenol-lyase (FnTPL) from *Fusobacterium nucleatum* [1] was synthesized by Tsingke (Beijing, China), and the codon of the FnTPL gene sequence was optimized. PCR program settings: initial denaturation at 95°C for 5 min followed by 30 cycles of denaturation at 95°C for 40 s, annealing at 68°C for 40 s, extension at 72°C for 1 min and followed by a final extension at 72°C for 10 min. The PCR product of FnTPL genes were isolated and double-digested with corresponding restriction endonucleases (*BamH* I, *Not* I), and ligated into the pET28a at the *BamH* I/*Not* I sites to form the recombinant plasmids pET28a-FnTPL. The recombinant plasmids pET28a-FnTPL was transformed into the competent *E. coli* T7 to form *E. coli* (pET28a-FnTPL).

For *E. coli* (pET28a-sfTDC), the gene of tyrosine decarboxylase (sfTDC) from *Streptococcus faecalis* [2] was synthesized by Tsingke (Beijing, China), and the codon of the sfTDC gene sequence was optimized. PCR program settings: initial denaturation at 95°C for 5 min followed by 30 cycles of denaturation at 95°C for 40 s, annealing at 68°C for 40 s, extension at 72°C for 1 min and followed by a final extension at 72°C for 10 min. The PCR product of sfTDC genes were isolated and digested with corresponding restriction endonucleases (*EcoR* I, *Hind*III), ligated into the pET28a vector to form the recombinant plasmid pET28a-sfTDC. The recombinant plasmid pET28a-sfTDC was transformed into the competent *E. coli* T7 to form *E. coli* (pET28a-sfTDC).

For *E. coli* (pET28a-CepTA), the gene of amine transaminase (CepTA) (EXJ89053.1) from *Capronia epimyces* was synthesized by Tsingke (Beijing, China). PCR program settings: initial denaturation at 95°C for 5 min followed by 30 cycles of denaturation at 95°C for 40 s, annealing at 68°C for 40 s, extension at 72°C for 1 min and followed by a final extension at 72°C for 10 min. The PCR product of CepTA genes were isolated and double-digested with corresponding restriction endonucleases (*Nde I*, *Xho* I), and ligated into the pET28a at the *Nde* I/*Xho* I sites to form the recombinant plasmids pET28a-CepTA. In addition, the recombinant plasmid pET28a-CepTA was transformed into the competent *E. coli* T7 to form *E. coli* (pET28a-CepTA).

For *E. coli* (pET28a-ARO10), the gene of α-keto acid decarboxylase (ARO10) from *Saccharomyces cerevisiae* [3] was synthesized by Tsingke (Beijing, China), and the codon of the ARO10 gene sequence was optimized. PCR program settings: initial denaturation at 95°C for 5 min followed by 30 cycles of denaturation at 95°C for 40 s, annealing at 68°C for 40 s, extension at 72°C for 1 min and followed by a final extension at 72°C for 10 min. The PCR product of ARO10 genes were isolated and double-digested with corresponding restriction endonucleases (*BamH* I, *Hind* III), and ligated into the pET28a at the *BamH* I/*Hind* III sites to form the recombinant plasmids pET28a-ARO10. In addition, the recombinant plasmid pET28a-ARO10 was transformed into the competent *E. coli* T7 to form *E. coli* (pET28a-ARO10).

For *E. coli* (pET28a-LAAD), the gene of L-amino acid deaminase (LAAD) from *Proteus mirabilis* [4] was synthesized by Tsingke (Beijing, China), and the codon of the LAAD gene sequence was optimized. PCR program settings: initial denaturation at 95°C for 5 min followed by 30 cycles of denaturation at 95°C for 40 s, annealing at 68°C for 40 s, extension at 72°C for 1 min and followed by a final extension at 72°C for 10 min. The PCR product of LAAD genes were isolated and digested with

corresponding restriction endonucleases (*Nde* I, *Xho* I), ligated into the pET28a vector to form the recombinant plasmid pET28a-LAAD. The recombinant plasmid pET28a-LAAD was transformed into the competent *E. coli* T7 to form *E. coli* (pET28a-LAAD).

For *E. coli* (pET28a-yahK), the gene of aldehyde reductase (yahK) from *Escherichia coli* [5] was synthesized by Tsingke (Beijing, China), and the codon of the yahK gene sequence was optimized. PCR program settings: initial denaturation at 95°C for 5 min followed by 30 cycles of denaturation at 95°C for 40 s, annealing at 68°C for 40 s, extension at 72°C for 1 min and followed by a final extension at 72°C for 10 min. The PCR product of yahK genes were isolated and digested with corresponding restriction endonucleases (*BgI* II, *Xho* I), ligated into the pET28a vector to form the recombinant plasmid pET28a-yahK. The recombinant plasmid pET28a-yahK was transformed into the competent *E. coli* T7 to form *E. coli* (pET28a-yahK).

For *E. coli* (pET28a-HpaB), the gene of 4-hydroxyphenylacetate 3-monooxygenase (HpaB) from *Escherichia coli* [6] was synthesized by Tsingke (Beijing, China), and the codon of the HpaB gene sequence was optimized. PCR program settings: initial denaturation at 95°C for 5 min followed by 30 cycles of denaturation at 95°C for 40 s, annealing at 68°C for 40 s, extension at 72°C for 1 min and followed by a final extension at 72°C for 10 min. The PCR product of HpaB genes were isolated and double-digested with corresponding restriction endonucleases (*BamH* I, *Hind* III), and ligated into the pET28a at the *BamH* I/*Hind* III sites to form the recombinant plasmids pET28a-HpaB. In addition, the recombinant plasmid pET28a-HpaB was transformed into the competent *E. coli* T7 to form *E. coli* (pET28a-HpaB).

For *E. coli* (pET28a-HpaC), the gene of 4-hydroxyphenylacetate 3-monooxygenase reductase (HpaC) from *Escherichia coli* [6] was synthesized by Tsingke (Beijing, China), and the codon of the HpaC gene sequence was optimized. PCR program settings: initial denaturation at 95°C for 5 min followed by 30 cycles of denaturation at 95°C for 40 s, annealing at 68°C for 40 s, extension at 72°C for 1 min and followed by a final extension at 72°C for 10 min. The PCR product of HpaC genes were isolated and digested with corresponding restriction endonucleases (*Nde* I, *Xho* I), ligated into the pET28a vector to form the recombinant plasmid pET28a-HpaC. The recombinant plasmid pET28a-HpaC was transformed into the competent *E. coli* T7 to form *E. coli* (pET28a-HpaC).

For *E. coli* (pRSFduet-yahK), the plasmid pET28a-yahK was used as template for gene amplification of yahK by PCR. The PCR product of yahK genes were isolated and digested with corresponding

restriction endonucleases (*BgI* II, *Xho* I), ligated into the pRSFduet-1 vector to form the recombinant plasmid pRSFduet-yahK. The recombinant plasmid pRSFduet-yahK was transformed into the competent *E. coli* T7 to form *E. coli* (pRSFduet-yahK). The recombinant *E. coli* (pCDFduet-yahK) and *E. coli* (pETduet-yahK) were constructed as described above.

For *E. coli* (pRSFduet-FnTPL-yahK), the plasmid pET28a-FnTPL was used as template for gene amplification of FnTPL by PCR. The PCR product of FnTPL genes were isolated and double-digested with corresponding restriction endonucleases (*BamH* I, *Not* I), and ligated into the pRSFduet-yahK at the *BamH* I/*Not* I sites to form the recombinant plasmids pRSFduet-FnTPL-yahK (known as RFK). In addition, the recombinant plasmid pRSFduet-FnTPL-yahK was transformed into the competent *E. coli* T7 to form *E. coli* (pRSFduet-FnTPL-yahK) which was named *E. coli* (RFK). The recombinant *E. coli* (pCDFduet-FnTPL-yahK) and *E. coli* (pETduet-FnTPL-yahK) were also constructed as described above.

For *E. coli* (pETDuet-LAAD), the plasmid pET28a-LAAD was used as template for gene amplification of LAAD by PCR. The PCR product of LAAD genes were isolated and digested with corresponding restriction endonucleases (*Nde* I, *Xho* I), ligated into the pETDuet-1 vector to form the recombinant plasmid pETDuet-LAAD. The recombinant plasmid pETDuet-LAAD was transformed into the competent *E. coli* T7 to form *E. coli* (pETDuet-LAAD). The recombinant *E. coli* (pRSFduet-LAAD) was also constructed as described above.

For *E. coli* (pETDuet-ARO10-LAAD), the plasmid pET28a-ARO10 was used as template for gene amplification of ARO10 by PCR. The PCR product of ARO10 genes were isolated and double-digested with corresponding restriction endonucleases (*BamH* I, *Hind* III), and ligated into the pETDuet-LAAD at the *BamH* I/*Hind* III sites to form the recombinant plasmids pETDuet-ARO10-LAAD (known as EAL). In addition, the recombinant plasmid pETDuet-ARO10-LAAD was transformed into the competent *E. coli* T7 to form *E. coli* (pETDuet-ARO10-LAAD) which was named *E. coli* (EAL). The recombinant *E. coli* (pRSFduet-ARO10-LAAD) was also constructed as described above.

For *E. coli* (pETDuet-HpaC), the plasmid pET28a-HpaC was used as template for gene amplification of HpaC by PCR. The PCR product of HpaC genes were isolated and digested with corresponding restriction endonucleases (*Nde* I, *Xho* I), ligated into the pETDuet-1 vector to form the recombinant plasmid pETDuet-HpaC. The recombinant plasmid pETDuet-HpaC was transformed into the competent *E. coli* T7 to form *E. coli* (pETDuet-HpaC). The recombinant *E. coli* (pCDFduet-HpaC) was also constructed as described above.

For *E. coli* (pETDuet-HpaB-HpaC), the plasmid pET28a-HpaB was used as template for gene amplification of HpaB by PCR. The PCR product of HpaB genes were isolated and double-digested with corresponding restriction endonucleases (*BamH* I, *Hind* III), and ligated into the pETDuet-HpaC at the *BamH* I/*Hind* III sites to form the recombinant plasmids pETDuet-HpaB-HpaC (known as EBC). In addition, the recombinant plasmid pETDuet-HpaB-HpaC was transformed into the competent *E. coli* T7 to form *E. coli* (pETDuet-HpaB-HpaC) which was named *E. coli* (EBC). The recombinant *E. coli* (pCDFduet-HpaB-HpaC) was also constructed as described above.

For *E. coli* (pRSFDuet-sfTDC), the plasmid pET28a-sfTDC was used as template for gene amplification of sfTDC by PCR. The PCR product of sfTDC genes were isolated and digested with corresponding restriction endonucleases (*EcoR* I, *Hind*III), ligated into the pRSFDuet-1 vector to form the recombinant plasmid pRSFDuet-sfTDC. The recombinant plasmid pRSFDuet-sfTDC was transformed into the competent *E. coli* T7 to form *E. coli* (pRSFDuet-sfTDC).

For *E. coli* (pRSFduet-sfTDC-CepTA), the plasmid pET28a-CepTA was used as template for gene amplification of CepTA by PCR. The PCR product of CepTA genes were isolated and double-digested with corresponding restriction endonucleases (*Nde I*, *Xho* I), and ligated into the pRSFDuet-sfTDC at the *Nde* I/*Xho* I sites to form the recombinant plasmids pRSFduet-sfTDC-CepTA (known as RTA). In addition, the recombinant plasmid pRSFduet-sfTDC-CepTA was transformed into the competent *E. coli* T7 to form *E. coli* (pRSFduet-sfTDC-CepTA) which was named *E. coli* (RTA).

For *E. coli* (RFK-EAL), the constructed recombinant plasmids pETDuet-ARO10-LAAD and pRSFduet-FnTPL-yahK were simultaneously transformed into the competent *E. coli* T7 to form the recombinant *E. coli* (pRSFduet-FnTPL-yahK/pETDuet-ARO10-LAAD), designated as *E. coli* (RFK-EAL). The recombinant *E. coli* (CFK-EAL), *E. coli* (CFK-RAL) and *E. coli* (EFK-RAL) were also constructed as described above.

For *E. coli* (CFK-RTA), the constructed recombinant plasmids pRSFDuet-sfTDC-CepTA and pCDFduet-FnTPL-yahK were simultaneously transformed into the competent *E. coli* T7 to form the recombinant *E. coli* (pCDFduet-FnTPL-yahK/pRSFDuet-sfTDC-CepTA), designated as *E. coli* (CFK-RTA). The recombinant *E. coli* (EFK-RTA) was also constructed as described above.

For *E. coli* (CFK-EBC-RAL), the constructed recombinant plasmids pETDuet-HpaB-HpaC, pRSFduet-ARO10-LAAD and pCDFduet-FnTPL-yahK were simultaneously transformed into the competent *E. coli* T7 to form the recombinant *E. coli* (pCDFduet-FnTPL-yahK/pETDuet-HpaB-

HpaC/pRSFDuet-ARO10-LAAD), designated as *E. coli* (CFK-EBC-RAL). The recombinant *E. coli* (EFK-CBC-RAL) and *E. coli* (RFK-CBC-EAL) were also constructed as described above.

All the constructed recombinant *E. coli* strains were listed in Table S2.

4. Gene Sequences

Gene sequence of FnTPL with codon optimized

ATGCGTTTTGAAGATTATCCGGCCGAACCGTTTCGTATTAAATCAGTTGAAACCGTTAAAAT
GATCGATAAAGCCGCCCGTGAAGAAGTTATTAAGAAGGCCGGTTATAATACCTTTCTGATTA
ATAGCGAAGATGTTTATATCGATCTGCTGACCGATAGCGGTACCAATGCAATGAGCGATAAA
CAGTGGGGTGGTCTGATGCAGGGTGATGAAGCATATGCCGGTAGCCGTAATTTCTTCCATCT
GGAAGAAACCGTGAAAGAAATTTTCGGTTTTAAACATATCGTTCCGACACATCAGGGTCGT
GGTGCAGAAAACATTCTGAGCCAGATTGCGATTAAACCGGGTCAGTATGTCCCGGGCAATA
TGTATTTTACCACCACCCGTTATCATCAGGAACGCAATGGTGGCATCTTTAAAGATATTATTC
GTGATGAAGCCCATGATGCCACCCTGAACGTTCCTTTTAAAGGTGATATTGATCTGAACAAA
CTGCAGAAACTGATTGATGAAGTGGGTGCAGAAAATATTGCATATGTTTGTCTGGCAGTTAC
CGTTAACCTGGCAGGCGGTCAGCCGGTTAGCATGAAAAATATGAAAGCAGTCCGTGAACTG
ACAAAGAAGCATGGTATTAAAGTATTTTATGATGCCACCCGTTGTGTTGAAAATGCCTATTTT
ATTAAAGAACAGGAAGAAGGTTATCAGGATAAAACCATTAAAGAAATCGTGCATGAAATGT
TTTCCTATGCAGATGGTTGTACCATGAGTGGCAAGAAGGATTGTCTGGTTAACATTGGCGGT
TTTCTGTGTATGAATGATGAAGATCTGTTTCTGGCAGCGAAAGAAATTGTTGTTGTTTATGA
AGGTATGCCGAGCTATGGTGGTCTGGCAGGTCGTGATATGGAAGCGATGGCAATTGGTCTG
CGTGAAAGCCTGCAGTATGAATATATTCGTCATCGTATTCTGCAGGTTCGTTATCTGGGTGAA
AAGCTGAAAGAAGCAGGTGTTCCGATTCTGGAACCGGTAGGCGGTCATGCAGTTTTCCTGG
ATGCCCGTCGTTTTTGTCCGCATATTCCGCAGGAAGAATTTCCAGCCCAGGCACTGGCCGCC
GCAATTTATGTTGAATGTGGTGTTCGTACTATGGAACGTGGTATTATTAGTGCTGGTCGTGAT
GTTAAAACCGGTGAAAACCATAAACCGAAACTGGAAACCGTTCGTGTTACCATTCCGCGCC
GTGTTTATACGTATAAACATATGGATGTTGTGGCAGAAGGTATTATTAAACTGTATAAACATA
AAGAAGATATCAAACCGCTGGAATTTGTTTATGAACCTAAACAGCTGCGTTTCTTCACCGCA
CGCTTTGGTATTAAGAAGTAA

Gene sequence of LAAD with codon optimized

ATGAACATTTCACGTCGCAAGCTGCTGCTGGGTGTTGGTGCTGCTGGTGTGTTGGCGG
GTGGTGCAGCTCTGGTTCCAATGGTGCGTCGTGATGGTAAATTTGTTGAAGCAAAGAG
CCGTGCGAGCTTCGTGGAAGGTACCCAAGGTGCGCTGCCGAAAGAAGCTGACGTTGT
GATTATCGGTGCTGGTATTCAGGGTATCATGACCGCTATTAATCTGGCAGAACGTGGTAT
GAGCGTTACCATTCTGGAAAAGGGTCAAATCGCAGGTGAACAGAGCGGTCGTGCGTA
CAGCCAAATTATCAGCTATCAGACCAGCCCGGAAATTTTTCCGCTGCATCACTACGGTA
AAATCCTGTGGCGTGGTATGAACGAAAAGATTGGTGCGGATACCAGCTATCGTACCCA
AGGTCGTGTTGAAGCTCTGGCAGATGAAAAAGCACTGGACAAGGCGCAGGCTTGGAT
CAAAACCGCGAAGGAAGCTGCAGGTTTTGACACCCCGCTGAATACCCGTATTATCAAA
GGTGAAGAACTGAGCAACCGTCTGGTTGGTGCTCAAACCCCGTGGACCGTGGCTGCT
TTCGAAGAAGATAGCGGTAGCGTTGACCCGGAAACCGGTACCCCGGCACTGGCTCGTT
ACGCTAAACAGATTGGTGTTAAGATCTATACCAACTGCGCTGTGCGTGGTATTGAAACC
GCGGGTGGTAAAATCAGCGATGTTGTGAGCGAAAAAGGTGCGATCAAGACCAGCCAA

GTGGTGCTGGCGGGTGGTATTTGGAGCCGTCTGTTTATGGGTAATATGGGTATTGACATC
CCGACCCTGAACGTTTACCTGAGCCAACAACGTGTTAGCGGTGTGCCAGGTGCGCCGC
GTGGTAATGTGCATCTGCCGAACGGTATCCACTTTCGTGAACAAGCTGATGGTACCTAT
GCTGTTGCACCGCGTATTTTCACCAGCAGCATCGTGAAAGACAGCTTTCTGCTGGGTC
CGAAGTTCATGCATCTGCTGGGTGGTGGTGAACTGCCGCTGGAATTTTCTATCGGTGAA
GACCTGTTTAATAGCTTCAAAATGCCGACCAGCTGGAACCTGGACGAAAAGACCCCGT
TTGAACAATTCCGTGTTGCGACCGCTACCCAAAATACCCAGCACCTGGATGCAGTTTTT
CAGCGTATGAAAACCGAATTTCCGGTGTTCGAAAAGAGCGAAGTTGTGGAACGTTGG
GGTGCTGTTGTGAGCCCGACCTTCGACGAACTGCCGATTATCAGCGAAGTTAAGGAAT
ACCCGGGTCTGGTTATTAACACCGCTACCGTGTGGGGTATGACCGAAGGTCCGGCAGC
GGGTGAAGTTACCGCAGATATTGTGATGGGTAAAAAGCCGGTTATTGATCCGACCCCGT
TTAGTTTGGATCGTTTTAAGAAGTAA

Gene sequence of ARO10 with codon optimized

ATGGCACCTGTTACGATTGAAAAGTTTGTTAATCAGGAAGAACGCCATCTGGTTAGCAACC
GTAGCGCCACCATTCCGTTTGGTGAATATATTTTCAAACGTCTGCTGAGCATTGATACCAAAT
CCGTGTTTGGTGTGCCGGGTGATTTTAATCTGAGTCTGCTGGAATATCTGTATAGCCCGAGT
GTTGAAAGCGCCGGTCTGCGTTGGGTTGGTACCTGTAATGAACTGAATGCGGCATATGCTG
CAGATGGTTATAGTCGTTATAGCAATAAAATCGGTTGTCTGATTACCACCTATGGTGTTGGTG
AACTGTCTGCACTGAATGGTATTGCAGGTAGCTTTGCAGAAAATGTTAAAGTTCTGCATATC
GTGGGTGTTGCCAAAAGCATTGATAGCCGTTCTTCTAATTTTAGTGATCGTAATCTGCATCAT
CTGGTTCCGCAGCTGCATGATTCAAATTTTAAAGGCCCGAATCATAAAGTTTATCATGATATG
GTTAAAGATCGCGTGGCATGTTCTGTTGCATATCTGGAAGATATCGAAACCGCCTGTGATCA
GGTTGATAACGTTATTCGTGATATCTATAAATATAGTAAACCTGGTTATATCTTTGTTCCGGCA
GATTTTGCAGATATGAGTGTGACGTGTGATAATCTGGTGAATGTTCCGCGTATTAGTCAGCA
GGATTGTATTGTTTATCCGAGCGAAAATCAGCTGAGTGATATTATTAATAAAATCACCTCCTG
GATCTATAGCAGCAAAACACCGGCCATTCTGGGCGATGTCCTGACCGATCGTTATGGTGTTA
GCAATTTTCTGAATAAACTGATTTGCAAAACAGGCATTTGGAATTTTTCAACCGTTATGGGT
AAAAGCGTCATTGATGAAAGCAATCCGACCTATATGGGCCAGTATAACGGTAAAGAAGGTC
TGAAACAGGTGTATGAACATTTTGAACTGTGTGATCTGGTTCTGCATTTTGGTGTTGATATCA
ATGAAATTAACAACGGCCATTATACCTTTACATATAAACCAAATGCCAAAATCATCCAGTTTC
ATCCGAATTATATCCGTCTGGTTGATACTCGTCAGGGTAATGAACAGATGTTTAAAGGTATTA
ACTTTGCACCGATTCTGAAAGAACTGTATAAACGCATTGATGTTAGCAAACTGTCCCTGCAG
TATGATAGTAATGTTACGCAGTATACCAATGAAACAATGCGTCTGGAAGATCCGACCAATGG
TCAGAGCAGCATTATTACCCAGGTGCATCTGCAGAAAACCATGCCGAAATTTCTGAATCCGG
GTGATGTGGTTGTTTGTGAAACGGGTAGCTTTCAGTTTAGCGTGCGTGATTTTGCATTTCCG
AGCCAGCTGAAATATATTAGCCAGGGTTTCTTCCTGAGTATCGGTATGGCGCTGCCGGCAGC
ACTGGGTGTTGGTATTGCGATGCAGGATCATTCAAATGCTCATATTAATGGTGGTAATGTTAA
AGAAGATTATAAACCACGTCTGATTCTGTTTGAAGGTGATGGTGCCGCACAGATGACTATTC
AGGAACTGAGTACCATTCTGAAATGTAATATTCCTCTGGAAGTTATTATCTGGAATAATAATG
GTTATACAATCGAACGTGCAATTATGGGTCCGACACGTAGTTATAATGATGTTATGAGCTGGA
AATGGACCAAACTGTTTGAAGCATTTGGTGATTTTGATGGTAAATATACAAACAGTACCCTG
ATTCAGTGTCCAAGCAAACTGGCACTGAAACTGGAAGAACTGAAAAATAGCAACAAACGT
AGCGGCATTGAACTGCTGGAAGTGAAACTGGGTGAACTGGATTTTCCAGAACAGCTGAAA

TGTATGGTTGAAGCGGCTGCACTGAAACGTAATAAGAAGTAA

Gene sequence of yahK with codon optimized

ATGAAAATCAAAGCCGTCGGTGCATATAGCGCAAAACAGCCGCTGGAACCGATGGATATCA
CCCGTCGTGAACCGGGTCCGAACGATGTCAAAATTGAAATTGCCTATTGTGGTGTGTGTCAT
AGCGATCTGCATCAGGTTCGTAGCGAATGGGCAGGTACTGTGTATCCGTGTGTTCCGGGTCA
TGAAATTGTTGGTCGCGTTGTTGCTGTTGGTGATCAGGTTGAAAAGTATGCCCCTGGTGATC
TGGTTGGTGTTGGTTGTATTGTTGATAGTTGTAAACATTGTGAAGAATGTGAAGATGGTCTG
GAAAATTATTGTGATCATATGACGGGTACATATAATAGTCCGACGCCGGATGAACCTGGTCAT
ACCCTGGGTGGTTATAGCCAGCAGATTGTTGTTCATGAACGTTATGTGCTGCGTATTCGCCAT
CCGCAGGAACAGCTGGCAGCAGTTGCACCACTGCTGTGTGCAGGTATTACAACCTATAGCC
CGCTGCGTCATTGGCAGGCCGGTCCTGGTAAGAAGGTTGGTGTTGTTGGCATTGGTGGTCT
GGGTCATATGGGTATTAAACTGGCACATGCCATGGGTGCGCATGTTGTTGCGTTTACCACTT
CAGAAGCGAAACGTGAAGCTGCAAAAGCGCTGGGTGCGGATGAAGTGGTGAATAGCCGTA
ACGCGGATGAAATGGCTGCACATCTGAAAAGCTTTGATTTTATTCTGAACACCGTTGCAGC
ACCGCATAATCTGGATGATTTTACAACCCTGCTGAAACGTGATGGTACCATGACCCTGGTTG
GTGCACCGGCAACCCCTCATAAATCACCGGAAGTTTTCAATCTGATTATGAAACGTCGTGCA
ATCGCCGGTAGTATGATTGGCGGCATTCCGGAAACCCAGGAAATGCTGGATTTTTGTGCAGA
ACATGGCATTGTTGCGGATATTGAAATGATCCGTGCCGATCAGATCAATGAAGCATATGAAC
GTATGCTGCGTGGTGATGTTAAATATCGCTTTGTTATTGATAACCGTACCCTGACCGATTAA

Gene sequence of sfTDC with codon optimized

ATGAAAAATGAAAAGCTGGCAAAAGGTGAAATGAACCTGAATGCGCTGTTTATCGGCGATA
AAGCAGAAAATGGTCAGCTGTATAAAGATCTGCTGATTGATCTGGTTGATGAACATCTGGGC
TGGCGTCAGAATTATATGCCGCAGGATATGCCTGTTATTTCAAGTCAGGAACGTACCAGCGA
AAGCTATGAAAAGACCGTGAATCATATGAAAGATGTGCTGAATGAAATCAGCAGTCGTATG
CGTACCCATTCCGTTCCATGGCATACCGCAGGCCGTTATTGGGGTCATATGAATAGCGAAAC
CCTGATGCCAAGCCTGCTGGCGTATAATTTTGCAATGCTGTGGAATGGCAATAACGTTGCAT
ATGAAAGTAGCCCGGCCACCAGTCAGATGGAAGAAGAAGTCGGTCATGAATTTGCACATCT
GATGTCATATAAAAACGGCTGGGGTCATATTGTTGCGGATGGTAGCCTGGCGAATCTGGAAG
GTCTGTGGTATGCACGCAACATTAAAAGCCTGCCTTTTGCAATGAAAGAAGTTAAACCGGA
ACTGGTTGCAGGTAAAAGCGATTGGGAACTGCTGAATATGCCTACCAAAGAAATTATGGAT
CTGCTGGAAAGCGCAGAAGATGAAATCGATGAAATTAAAGCACATAGCGCACGTAGCGGTA
AACATCTGCAGGCAATTGGTAAATGGCTGGTGCCGCAGACCAAACATTATAGCTGGCTGAA
AGCAGCAGATATTATTGGTATTGGTCTGGATCAGGTTATTCCGGTTCCTGTTGATCATAATTAT
CGTATGGATATTAACGAACTGGAAAAGATTGTTCGTGGTCTGGCAGAAGAACAGATTCCGG
TTCTGGGTGTTGTGGGTGTTGTTGGTAGTACCGAAGAAGGTGCGGTTGATAGTATTGATAAA
ATTATTGCACTGCGTGATGAACTGATGAAAGATGGTATTTATTATTATGTTCATGTTGATGCGG
CGTATGGTGGTTATGGTCGCGCAATTTTCCTGGATGAAGATAATAACTTTATCCCGTATGAAG
ATCTGCAGGATGTTCATGAAGAATATGGTGTTTTCAAAGAAAAGAAGGAACATATTTCCCGT
GAAGTTTATGATGCATATAAAGCAATTGAACTGGCAGAAAGCGTGACCATTGATCCGCATAA
AATGGGTTATATTCCGTATAGCGCAGGTGGCATTGTGATTCAGGATATTCGTATGCGTGATGT
GATTAGTTATTTTGCAACCTATGTTTTCGAAAAGGGTGCCGATATTCCGGCCCTGCTGGGTG
CATATATTCTGGAAGGTAGCAAAGCAGGTGCAACCGCAGCAAGCGTTTGGGCAGCACATCA

TGTTCTGCCTCTGAATGTTGCAGGTTATGGTAAACTGATTGGTGCAAGCATCGAAGGTTCCC
ATCATTTTTATAATTTTCTGAACGATCTGACCTTTAAAGTTGGTGATAAAGAAATTGAAGTTC
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Gene sequence of HpaB with codon optimized

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Gene sequence of HpaC with codon optimized

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Gene sequence of CepTA

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5. Screening of amine transaminases

To identify suitable ATA biocatalysts, multiple ATAs available in our lab were screened using dopamine as a substrate. The standard reaction mixture was Tris-HCl buffer (50 mM, pH 8.0) containing 10 mM dopamine, 0.1 mM PLP, 10 mM sodium pyruvate and 15 g cdw/L *E. coli* (ATA) resting cells. The reactions were incubation for 6 h at 30 °C, and 200 rpm shaking. The reaction solution (100 μL) was mixed well with acetonitrile (900 μL) containing 2 mM benzaldehyde (internal standard) and centrifuged at 12,000 rpm for 5 minutes. The obtained supernatant filtered through the 0.22 μm filters and submitted to HPLC analysis.

6. *In vitro* cascade biocatalysis for conversion of catechol to HT with the cell free extract of *E. coli* (FnTPL), *E. coli* (sfTDC), *E. coli* (CepTA) and *E. coli* (yahK)

In the enzyme cascade reaction of FnTPL-sfTDC-TA-yahK, the cascade combining the four steps in a concurrent one-pot fashion was investigated with the cell free extract of *E. coli* (FnTPL), *E. coli* (sfTDC),

E. coli (CepTA) and *E. coli* (yahK). The standard reaction mixture was Tris-HCl buffer (50 mM, pH 8.0) containing 10 mM catechol, 0.1 mM PLP, 40 mM sodium pyruvate, 200 mM NH_4Cl, 15 mg/mL FnTPL, 15 mg/mL sfTDC, 15 mg/mL CepTA and 15 mg/mL yahK, 0.5 mM NADH, 10 U glucose dehydrogenase (GDH), 50 mM glucose. The reactions were incubation for 24 h at 30 °C, and 200 rpm shaking. The reaction solution (100 μl) was mixed well with acetonitrile (900 μl) containing 2 mM benzaldehyde (internal standard) and centrifuged at 12,000 rpm for 5 minutes. The obtained supernatant filtered through the 0.22 μm filters and submitted to HPLC analysis.

7. *In vitro* cascade biocatalysis for conversion of catechol to HT with the cell free extract of *E. coli* (FnTPL), *E. coli* (LAAD), *E. coli* (ARO10) and *E. coli* (yahK)

In the enzyme cascade reaction of FnTPL-LAAD-ARO10-yahK, the cascade combining the four steps in a concurrent one-pot fashion was investigated with the cell free extract of *E. coli* (FnTPL), *E. coli* (LAAD), *E. coli* (ARO10) and *E. coli* (yahK). The standard reaction mixture was Tris-HCl buffer (50 mM, pH 8.0) containing 10 mM catechol, 0.1 mM PLP, 40 mM sodium pyruvate, 200 mM NH_4Cl, 15 mg/mL FnTPL, 15 mg/mL LAAD, 15 mg/mL ARO10 and 15 mg/mL yahK, 0.5 mM NADH, 10 U GDH, 50 mM glucose. The reactions were incubation for 24 h at 30 °C, and 200 rpm shaking. The reaction solution (100 μL) was mixed well with acetonitrile (900 μL) containing 2 mM benzaldehyde (internal standard) and centrifuged at 12,000 rpm for 5 minutes. The obtained supernatant filtered through the 0.22 μm filters and submitted to HPLC analysis.

8. *In vitro* cascade biocatalysis for conversion of phenol to HT with the cell free extract of *E. coli* (FnTPL), *E. coli* (HpaBC), *E. coli* (LAAD), *E. coli* (ARO10) and *E. coli* (yahK)

In the enzyme cascade reaction of FnTPL-HpaBC-LAAD-ARO10-yahK, the cascade combining the four steps in a concurrent one-pot fashion was investigated with the cell free extract of *E. coli* (FnTPL), *E. coli* (HpaBC), *E. coli* (LAAD), *E. coli* (ARO10) and *E. coli* (yahK). The standard reaction mixture was Tris-HCl buffer (50 mM, pH 8.0) containing 10 mM phenol, 0.1 mM PLP, 40 mM sodium pyruvate, 200 mM NH_4Cl, 15 mg/mL FnTPL, 10 mg/mL HpaB, 10mg/mL HpaC, 15mg/mL LAAD, 15 mg/mL ARO10 and 15 mg/mL yahK, 0.5 mM NADH, 10 U GDH, 50 mM glucose. The reactions were incubation for 24 h at 30 °C, and 200 rpm shaking. The reaction solution (100 μL) was mixed well with acetonitrile (900

μL) containing 2 mM benzaldehyde (internal standard) and centrifuged at 12,000 rpm for 5 minutes. The obtained supernatant filtered through the 0.22 μm filters and submitted to HPLC analysis.

9. Cascade conversion of catechol to HT with the recombinant *E. coli* resting cells co-expression of FnTPL, sfTDC, CepTA and yahK

The recombinant *E. coli* (CFK-RTA) and *E. coli* (EFK-RTA) induced overnight were collected by centrifugation for 5 min at 4 °C and 8000 rpm, then washed twice with Tris-HCl buffer (50 mM, pH 8.0). The obtained cells were suspended with 50 mM Tris-HCl buffer (pH 8.0) to a cell density of 15 g CDW L^{-1}. The standard reaction mixture was containing 10 mM catechol, 0.1 mM PLP, 40 mM sodium pyruvate, 200 mM NH_4Cl. The reaction mixtures were shaken at 200 rpm at 30°C. The samples were collected at regular intervals. The reaction solution (100 μL) was mixed well with acetonitrile (900 μL) containing 2 mM benzaldehyde (internal standard) and centrifuged at 12,000 rpm for 5 minutes. The obtained supernatant filtered through the 0.22 μm filters and submitted to HPLC analysis.

10. Cascade conversion of catechol to HT with the recombinant *E. coli* resting cells co-expression of FnTPL, LAAD, ARO10 and yahK

The recombinant *E. coli* (RFK-EAL), *E. coli* (CFK-EAL), *E. coli* (CFK-RAL) and *E. coli* (EFK-RAL) induced overnight were collected by centrifugation for 5 min at 4 °C and 8000 rpm, then washed twice with Tris-HCl buffer (50 mM, pH 8.0). The obtained cells were suspended with 50 mM Tris-HCl buffer (pH 8.0) to a cell density of 15 g CDW L^{-1}. The standard reaction mixture was containing 10 mM catechol, 0.1 mM PLP, 40 mM sodium pyruvate, 200 mM NH_4Cl. The reaction mixtures were shaken at 200 rpm at 30°C. The samples were collected at regular intervals. The reaction solution (100 μL) was mixed well with acetonitrile (900 μL) containing 2 mM benzaldehyde (internal standard) and centrifuged at 12,000 rpm for 5 minutes. The obtained supernatant filtered through the 0.22 μm filters and submitted to HPLC analysis.

11. Cascade conversion of phenol to HT with the recombinant *E. coli* resting cells co-expression of FnTPL, HpaBC, LAAD, ARO10 and yahK

The recombinant *E. coli* (CFK-EBC-RAL), *E. coli* (EFK-CBC-RAL) and *E. coli* (RFK-CBC-EAL) induced overnight were collected by centrifugation for 5 min at 4 °C and 8000 rpm, then washed twice

with Tris-HCl buffer (50 mM, pH 8.0). The obtained cells were suspended with 50 mM Tris-HCl buffer (pH 8.0) to a cell density of 15 g CDW L^{-1}. The standard reaction mixture was containing 10 mM phenol, 0.1 mM PLP, 40 mM sodium pyruvate, 200 mM NH_4Cl. The reaction mixtures were shaken at 200 rpm at 30°C. The samples were collected at regular intervals. The reaction solution (100 μL) was mixed well with acetonitrile (900 μL) containing 2 mM benzaldehyde (internal standard) and centrifuged at 12,000 rpm for 5 minutes. The obtained supernatant filtered through the 0.22 μm filters and submitted to HPLC analysis.

12. Conversion of phenol and catechol to HT with fed-batch strategy

The standard reaction mixture was containing 10 mM catechol or phenol, 120 mM sodium pyruvate, 600 mM NH_4Cl, 15 g CDW L^{-1} of *E. coli* (RFK-EAL) or 25 g cdw /L *E. coli* (CFK-EBC-RAL) at pH 8.0 and 30 °C. The strategy commences with an initial substrate concentration of 10 mM, followed by the addition of 10 mM substrate when the substrate is almost gone for a total of 2 feedings, resulting in a final substrate concentration of 30 mM. The samples were collected at regular intervals. The reaction solution (100 μL) was mixed well with acetonitrile (900 μL) containing 2 mM benzaldehyde (internal standard) and centrifuged at 12,000 rpm for 5 minutes. The obtained supernatant filtered through the 0.22 μm filters and submitted to HPLC analysis.

13. Preparation experiments

Using the fed-batch method, preparative experiments were conducted on a 100 mL scale with the constructed recombinant *E. coli* (RFK-EAL) and *E. coli* (CFK-EBC-RAL) resting cells. The initial reaction conditions are: 10 mM catechol or phenol, 120 mM sodium pyruvate, 600 mM NH_4Cl ,15 g CDW L^{-1} of *E. coli* (RFK-EAL) or 25 g CDW L^{-1} *E. coli* (CFK-EBC-RAL) at pH 8.0 and 30 °C. 10 mM catechol or phenol was added when the substrate is almost gone for a total of 2 feedings. After the reactions were finished, the reaction mixtures were extracted with ethyl acetate (EtOAc) (50 mL) for three times. The combined organic phases were dried over anhydrous Na_2SO_4. The solvent was removed by evaporation and the crude products were purified by flash chromatography on a silica gel column to give HT as a yellow oily. ^1H NMR (600 MHz, CD_3OD) δ 6.67 (d, J = 8.0 Hz, 1H), 6.65 (d, J = 2.0 Hz, 1H), 6.52 (dd, J = 8.0, 2.1 Hz, 1H), 3.67 (t, J = 7.3 Hz, 2H), 2.66 (t, J = 7.2 Hz, 2H).

14. Analytical Methods

The determination of both standards and products was performed with a Shimadzu HPLC LC-10A system equipped with an ultraviolet absorption detector (SPD) and reversed-phase C18 column (4.6 mm × 250 mm × 5 μm, Phenomenex, Shanghai) at 30°C. The mobile phase consisted of A (0.1% trifluoroacetic acid in acetonitrile) and B (0.1 % trifluoroacetic acid in water) and maintained A: B = 50: 50 for 10 min. The detection wavelength and flow rate were controlled at 210 nm and 1 mL/min. The injection volume of each sample was 20 μL.

Table S1. Primers used in this study.

Entry	Names	Primers (5'-3')	REA[a]
1	FnTPL-F	CGC***GGATCC***GATGCGTTTTGAAGATTATCCGGC	*BamH*I
2	FnTPL-R	AAGGAAAAAA***GCGGCCGC***TTACTTCTTAATACCAAAGCGTGCGG	*Not*I
3	CepTA-F	GGGAATTC***CATATG***GCTTCGATGGACAAGGTGTTTGCCG	*NdeI*
4	CepTA-R	CCG***CTCGAG***TCACTTGTGCACGCCATTGACACC	*Xho*I
5	yahK-F	GGA***AGATCT***CATGAAAATCAAAGCCGTCGGTGCATATAGCGC	*BgI* II
6	yahK-R	CCG***CTCGAG***TTAATCGGTCAGGGTACGGTTATCAATAAC	*Xho*I
7	sfTDC-F	CCG***GAATTC***GATGAAAAATGAAAAGCTGGCAAAAGGTGAAATGAACC	*EcoR*I
8	sfTDC-R	CCC***AAGCTT***TTATTTAACATCATAAATCTGTTCCAGCTTTTCCTGCAGCG	*Hind*III
9	ARO10-F	CGC***GGATCC***GATGGCACCTGTTACGATTGAAAAGTTTG	*BamH*I
10	ARO10-R	CCC***AAGCTT***TTACTTCTTATTACGTTTCAGTGCAGCCGC	*Hind*III
11	LAAD-F	GGGAATTC***CATATG***AACATTTCACGTCGCAAGCTGCTGC	*NdeI*
12	LAAD-R	CCG***CTCGAG***TTACTTCTTAAAACGATCCAAACTAAACGGGG	*Xho*I
13	HpaB-F	CGC***GGATCC***GATGAAACCGGAAGATTTTCGC	*BamH*I
14	HpaB-R	CCC***AAGCTT***TTATTTCAGCAGTTTATCCAGCATATTGATATCATC	*Hind*III
15	HpaC-F	GGGAATTC***CATATG***CAGCTGGATGAACAGCGTCTGC	*NdeI*
16	HpaC-R	CCG***CTCGAG***TTAAATCGCTGCTTCCATTTCCAGCATAACC	*Xho*I

[a] REA: restriction endonuclease

Table S2. Strains and plasmids used in this study.

Recombinant *E. coli*	Recombinant plasmid	Enzyme expressed	Reference
E. coli (FnTPL)	pET28a-FnTPL	FnTPL (*Fusobacterium nucleatum*)	[1]
E. coli (sfTDC)	pET28a-sfTDC	sfTDC (*Streptococcus faecalis*)	[2]
E. coli (CepTA)	pET28a-CepTA	CepTA (*Capronia epimyces*)	this study
E. Coli (ARO10)	pET28a-ARO10	ARO10 (*Saccharomyces cerevisiae*)	[3]
E. coli (LAAD)	pET28a-LAAD	LAAD (*Proteus mirabilis*)	[4]
E. coli (yahK)	pET28a-yahK	yahK (*Escherichia coli*)	[5]
E. coli (HpaC)	pET28a-HpaC	HpaC (*Escherichia coli*)	[6]
E. coli (HpaB)	pET28a-HpaB	HpaB (*Escherichia coli*)	[6]
E. coli (CFK)	pCDFDuet-FnTPL-yahK	FnTPL and yahK	this study
E. coli (RFK)	pRSFDuet-FnTPL-yahK	FnTPL and yahK	this study
E. coli (EFK)	pETDuet-FnTPL-yahK	FnTPL and yahK	this study
E. coli (EAL)	pETDuet-ARO10-LAAD	ARO10 and LAAD	this study
E. coli (RAL)	pRSFDuet-ARO10-LAAD	ARO10 and LAAD	this study
E. coli (RTA)	pRSFDuet-sfTDC-CepTA	sfTDC and CepTA	this study
E. coli (CBC)	pCDFDuet-HpaB-HpaC	HpaB and HpaC	this study
E. coli (EBC)	pETDuet-HpaB-HpaC	HpaB and HpaC	this study
E. coli (CFK-RTA)	pRSF-sfTDC-CepTA pCDF-FnTPL-yahK	FnTPL, sfTDC,CepTA and yahK	this study
E. coli (EFK-RTA)	pRSF-sfTDC-CepTA pET-FnTPL-yahK	FnTPL, sfTDC,CepTA and yahK	this study
E. coli (RAL-EFK)	pRSF-ARO10-LAAD pET-FnTPL-yahK	FnTPL, LAAD, ARO10 and yahK	this study
E. coli (RAL-CFK)	pRSF-ARO10-LAAD pCDF-FnTPL-yahK	FnTPL, LAAD, ARO10 and yahK	this study
E. coli (EAL-CFK)	pET-ARO10-LAAD pCDF-FnTPL-yahK	FnTPL, LAAD, ARO10 and yahK	this study
E. coli (EAL-RFK)	pET-ARO10-LAAD pRSF-FnTPL-yahK	FnTPL, LAAD, ARO10 and yahK	this study
E. coli (CFK-EBC-RAL)	pET-HpaB-HpaC pRSF-ARO10-LAAD pCDF-FnTPL-yahK	FnTPL, HpaBC, LAAD, ARO10 and yahK	this study
E. coli (EFK-CBC-RAL)	pCDF-HpaB-HpaC pRSF-ARO10-LAAD pET-FnTPL-yahK	FnTPL, HpaBC, LAAD, ARO10 and yahK	this study
E. coli (RFK-CBC-EAL)	pCDF-HpaB-HpaC pET-ARO10-LAAD pRSF-FnTPL-yahK	FnTPL, HpaBC, LAAD, ARO10 and yahK	this study

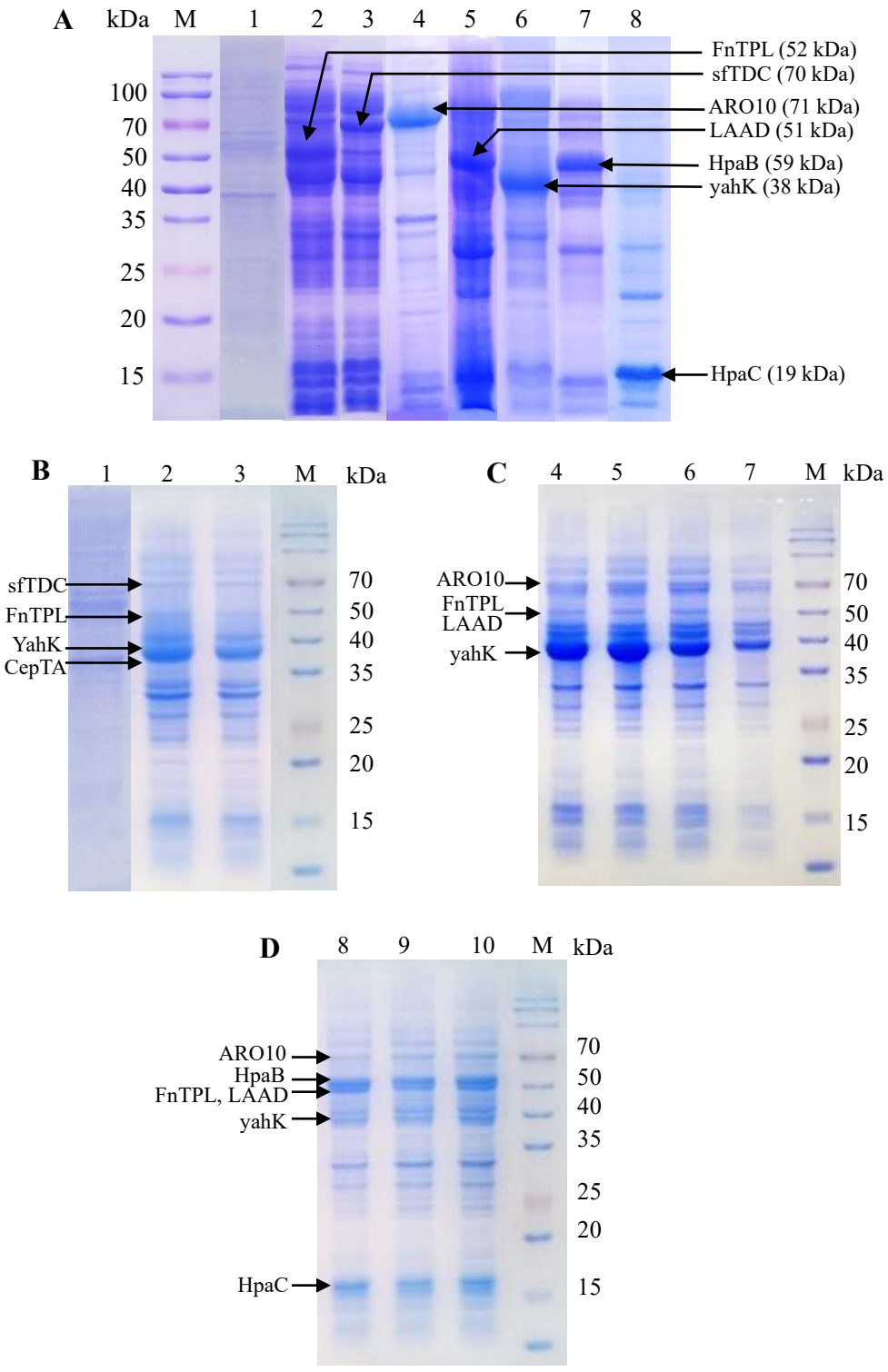

Figure S1. SDS-PAGE of the cell-free extracts of recombinant *E. coli* cells co-expression of multiple enzymes. **A**: Lane M： marker, lane 1: *E. coli* (pET28a), lane 2: *E. coli* (FnTPL), lane 3: *E. coli* (sfTDC), lane 4: *E. coli* (ARO10), lane 5: *E. coli* (LAAD), lane 6: *E. coli* (yahK), lane 7: *E. coli* (HpaB), lane 8: *E. coli* (HpaC); **B**: lane 1: *E. coli* (pET28a), lane 2: *E. coli* (CFK-RTA), lane 3: *E. coli* (EFK-RTA); **C**: lane 4: *E. coli* (CFK-EBC-RAL), lane 5: *E. coli* (RFK-CBC-EAL), lane 6: *E. coli* (EFK-CBC-RAL), lane 7: *E. coli* (RFK-EAL); **D**: lane 8: *E. coli* (EFK-RAL), lane 9: *E. coli* (CFK-EAL), lane 10: *E. coli* (CFK-RAL).

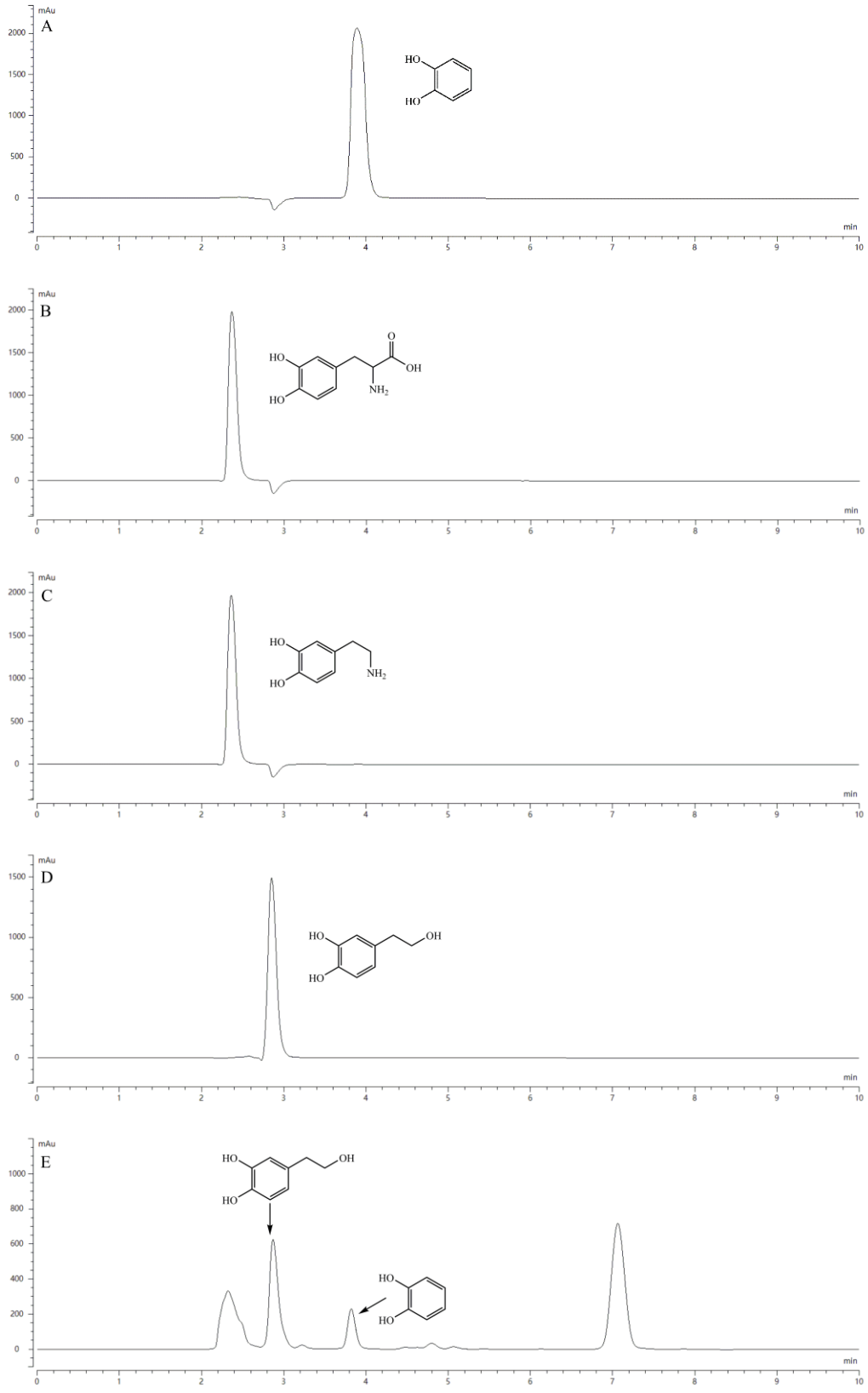

Figure S2. HPLC analysis of HT produced from catechol with the system 1. A: Catechol standard; B: L-3,4-dihydroxyphenylalanine standard; C: Dopamine standard; D: HT standard; E: HT produced by conversion of catechol (10 mM) with the resting cells of *E. coli* (CFK-RTA) (15 g cdw/L) at 24 h.

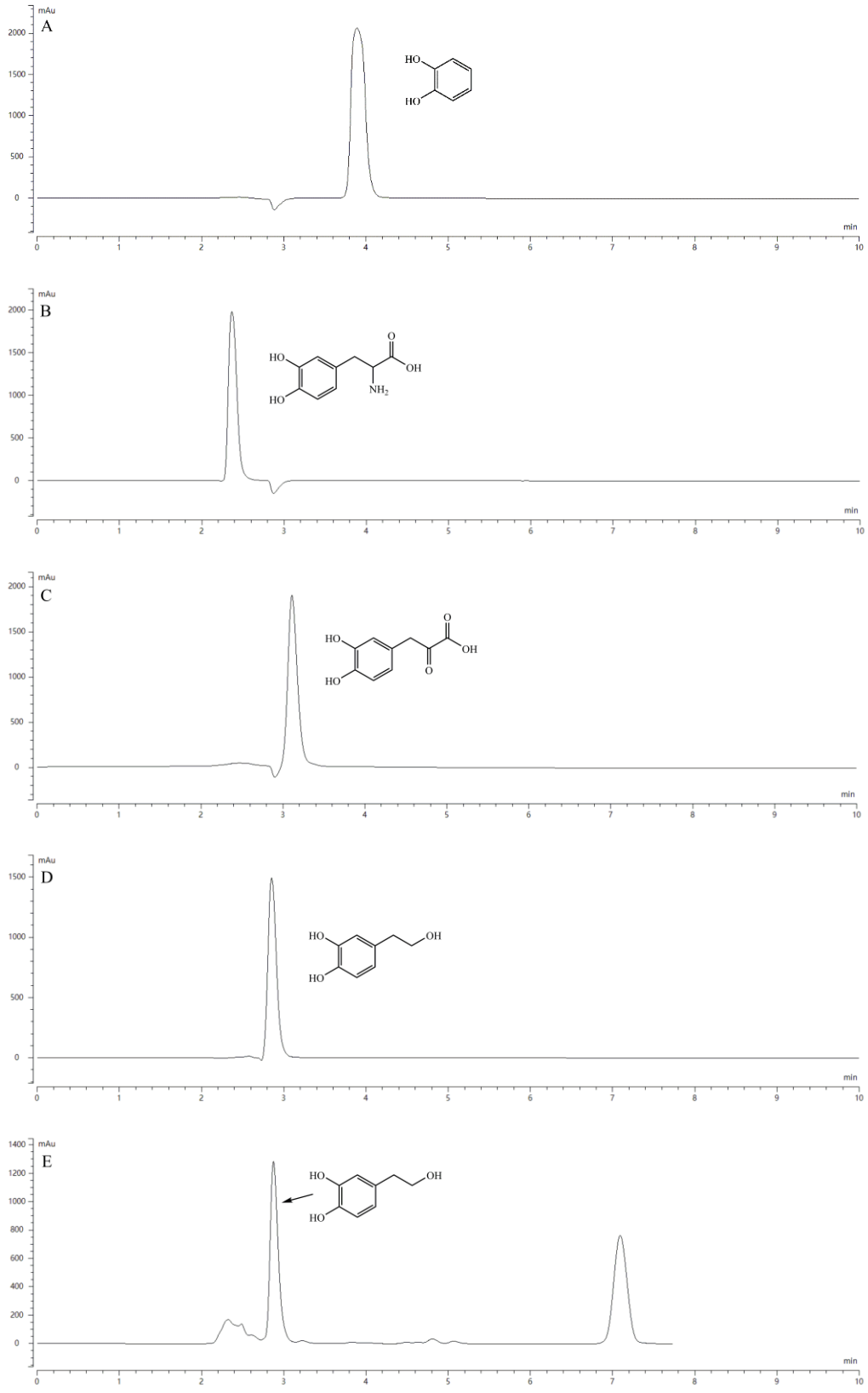

Figure S3. HPLC analysis of HT produced from catechol with the system 2. A: Catechol standard; B: L-3,4-dihydroxyphenylalanine standard; C: 3,4-Dihydroxyphenylpyruvic acid standard; D: HT standard; E: HT produced by conversion of catechol (10 mM) with the resting cells of *E. coli* (RFK-EAL) (15 g cdw/L) at 24 h.

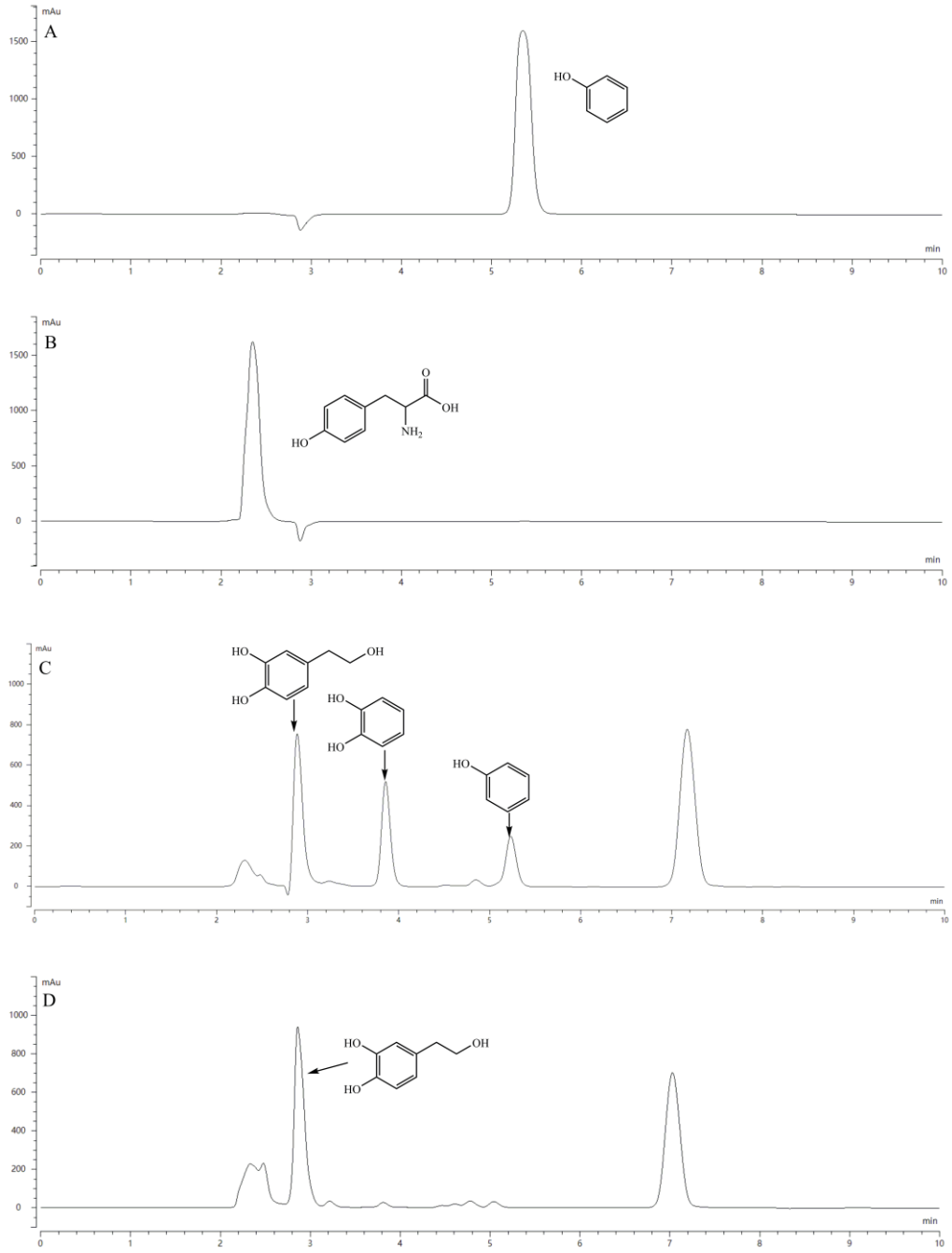

Figure S4. HPLC analysis of HT produced from phenol with the system 3. A: phenol standard; B: L-Tyrosine standard; C: HT produced by conversion of phenol (10 mM) with resting cells of *E. coli* (CFK-EBC-RAL) (25 g cdw/L) at 6 h. D: HT produced by conversion of phenol (10 mM) with resting cells of *E. coli* (CFK-EBC-RAL) (25 g cdw/L) at 24 h.

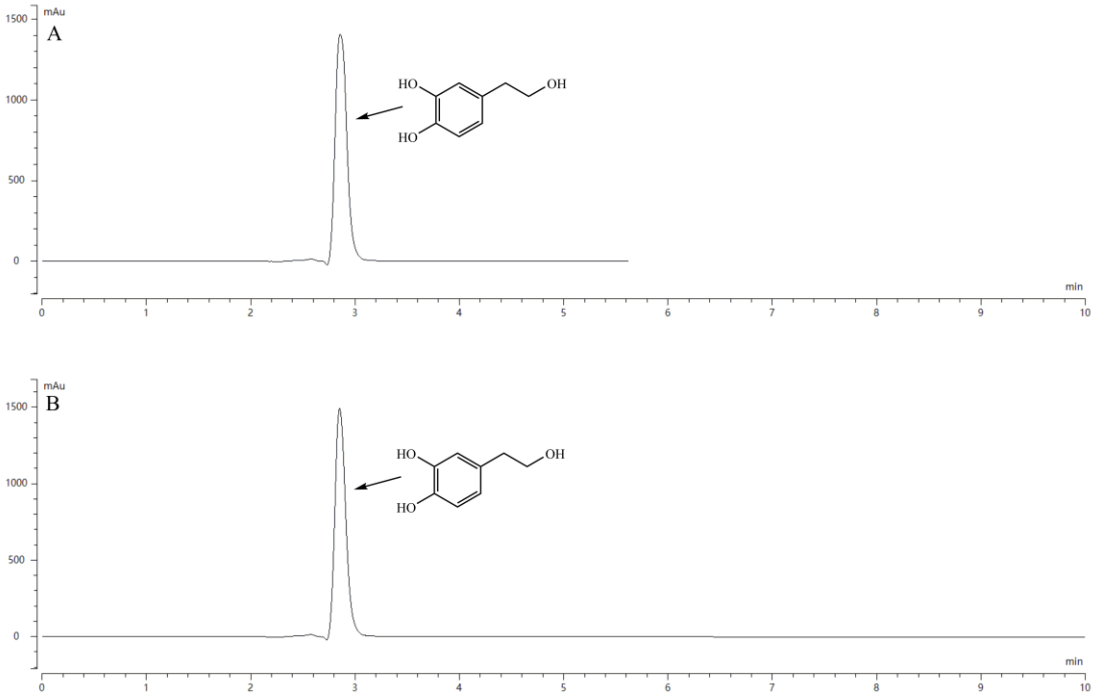

Figure S5. HPLC analysis of HT prepared from catechol (A) and phenol (B). A: HT prepared from catechol with the recombinant *E. coli* (RFK-EAL) resting cells. B: HT prepared from phenol with the recombinant *E. coli* (CFK-EBC-RAL) resting cells.

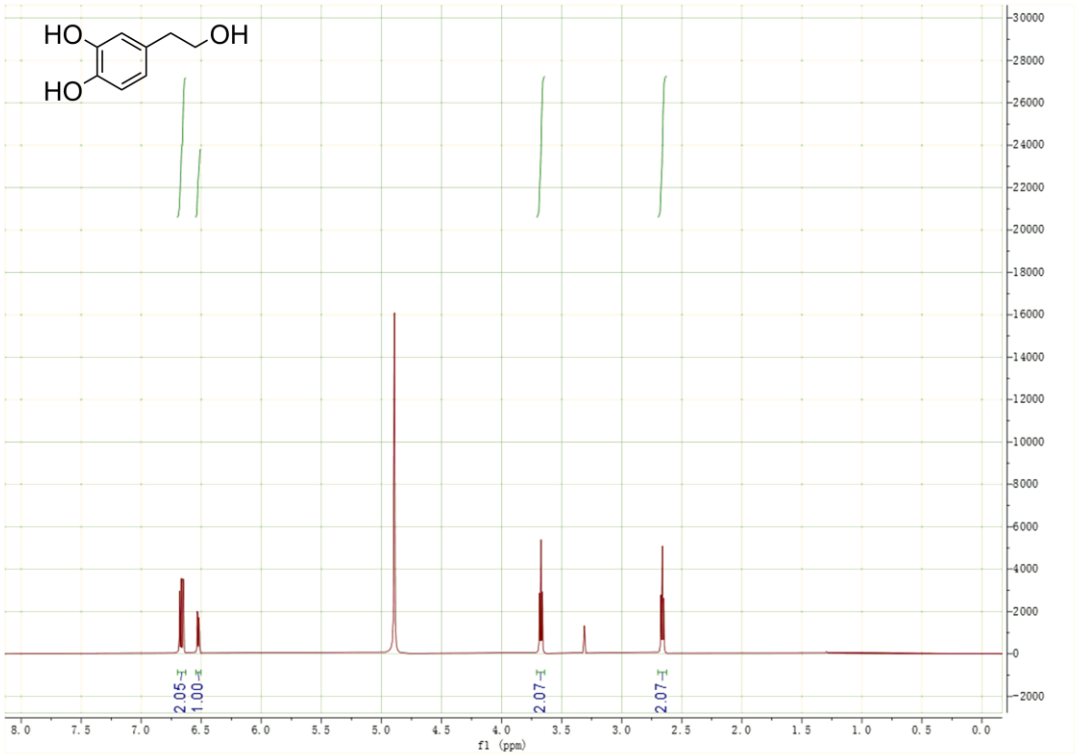

Figure S6. ^{1}H NMR spectra analysis of HT.

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