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Figure S1. Nucleotide and predicted amino acid sequence of HDT1 cDNA (Genbank MF115997). Oligonucleotide primer annealing sites are underlined and the corresponding primer names (see Table 1) are indicated above the nucleotide sequence at the 5' primer end. Forward primers (annealing to the antisense strand) are indicated by “>” and reverse primers (annealing to the sense strand) are indicated by “<”.

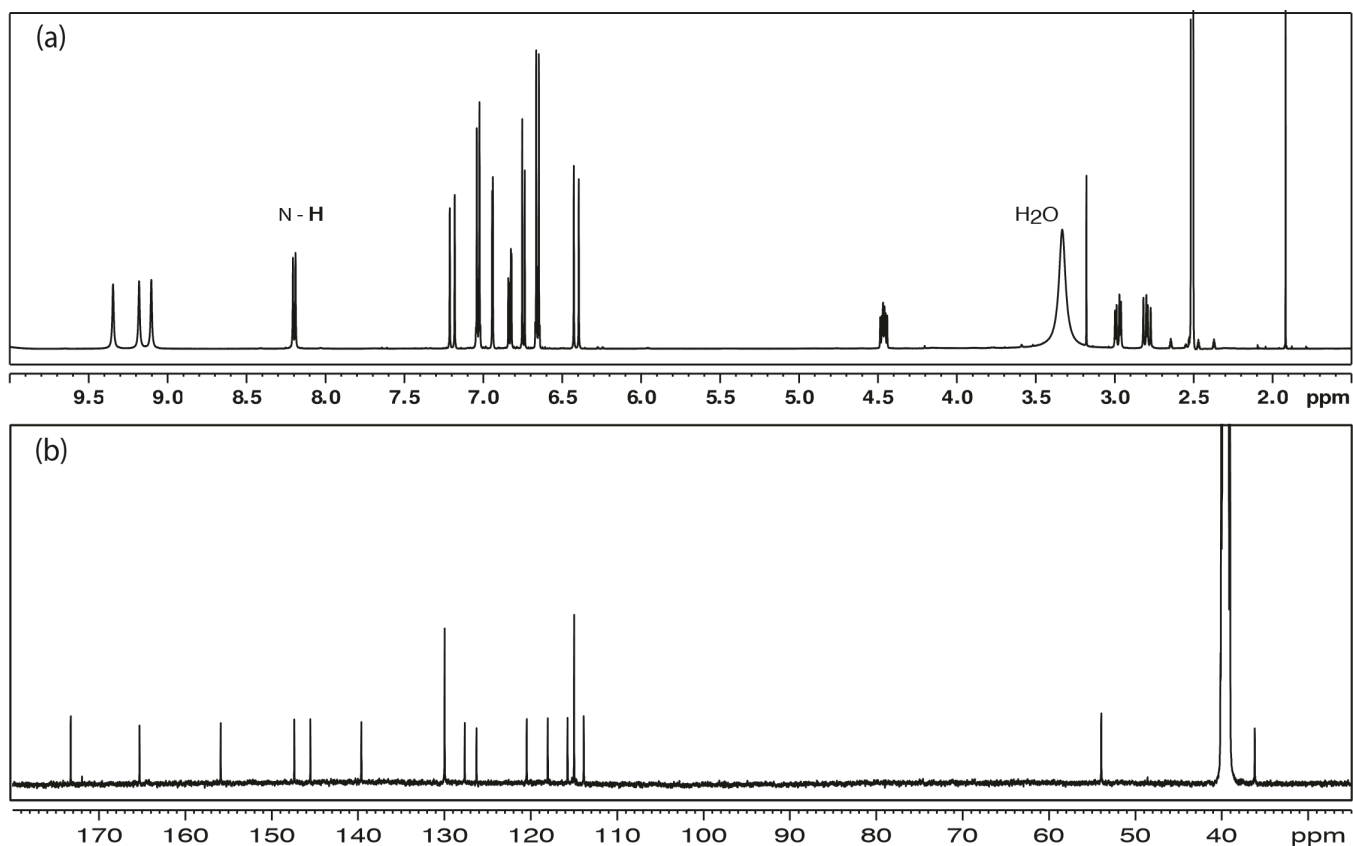


Figure S2. NMR evaluation of caffeoyl-L-Tyr generated by HDT1 *in vitro*. Caffeoyl-L-Tyr was synthesized in a 6 mL reaction by incubating 1 mM caffeoyl-CoA, 3 mM L-tyrosine, and HDT1 (25 $\mu\text{g}/\text{mL}$ expressed and purified from *E. coli* as described in the Materials and methods section) in a buffer of 100 mM sodium phosphate and 25 mM ascorbic acid, pH 8.0. Following a 90 min incubation at 30°C, the reaction was acidified by the addition of 1/10th volume 1 N HCl. The acidified reaction mixture was applied to a 100 mg ENVI-18 solid phase extraction column (Supelco, Bellefonte, PA, USA) preequilibrated with 3 X 1 mL methanol and 3 X 1 mL 1% (v/v) acetic acid in water, the column was washed with 3 X 1 mL 1% (v/v) acetic acid in water and 3 X 1 mL 0.1% (v/v) acetic acid in water, and product was eluted with 1 mL ethanol. The eluate was dried under a stream of nitrogen and the residue dissolved in DMSO-*d*₆. NMR spectra were recorded on a 500MHz Bruker Biospin Avance III HD spectrometer (Bruker, Billerica, MA, USA) equipped with a 5 mm inverse gradient TCI cryoprobe at 298 K. ¹H NMR (500 MHz, DMSO-*d*₆) (a) and ¹³C NMR (125 MHz, DMSO *d*₆) (b) spectra of the caffeoyl-L-Tyr product are shown.

Below, chemical shifts (δ , ppm) for the *in vitro* product are referenced to the residual undeuterated solvent peak, 2.5 for ¹H NMR and 39.5 for ¹³C NMR. The following abbreviations are used to denote the multiplicities: s = singlet, d = doublet, t = triplet, and m = multiplet, and are followed by their coupling constants (*J*, Hz).

¹H NMR (500 MHz, DMSO-*d*₆): δ 2.79 (1H, dd, 9.5, 14.0), 2.98 (1H, dd, 4.5, 13.5), 4.44-4.48 [1H, m), 6.41 (1H, d, 15.8), 6.66 (2H, d, 8.6), 6.75 (1H, d, 8.1), 6.83 (1H, dd, 2.1, 8.3), 6.94 (1H, d, 2.1), 7.02 (2, 2H, 8.5), 7.20 (1H, d, 15.7), 8.2 (1H, d, H-N), 9.10, 9.18, 9.35 (3 \times 1H, 3 s),

12.61 (1H, s); ^{13}C NMR (125 MHz, $\text{DMSO-}d_6$): δ 36.1, 53.9, 113.8, 115.0, 115.7, 118.0, 120.4, 126.3, 127.6, 130.0, 139.6, 145.5, 147.4, 155.9, 165.3, 173.3.

Spectroscopic data is consistent with that reported for (-)-N-[3',4'-Dihydroxy-(E)-cinnamoyl]-L-tyrosine in the literature (Stark and Hofmann, 2005).

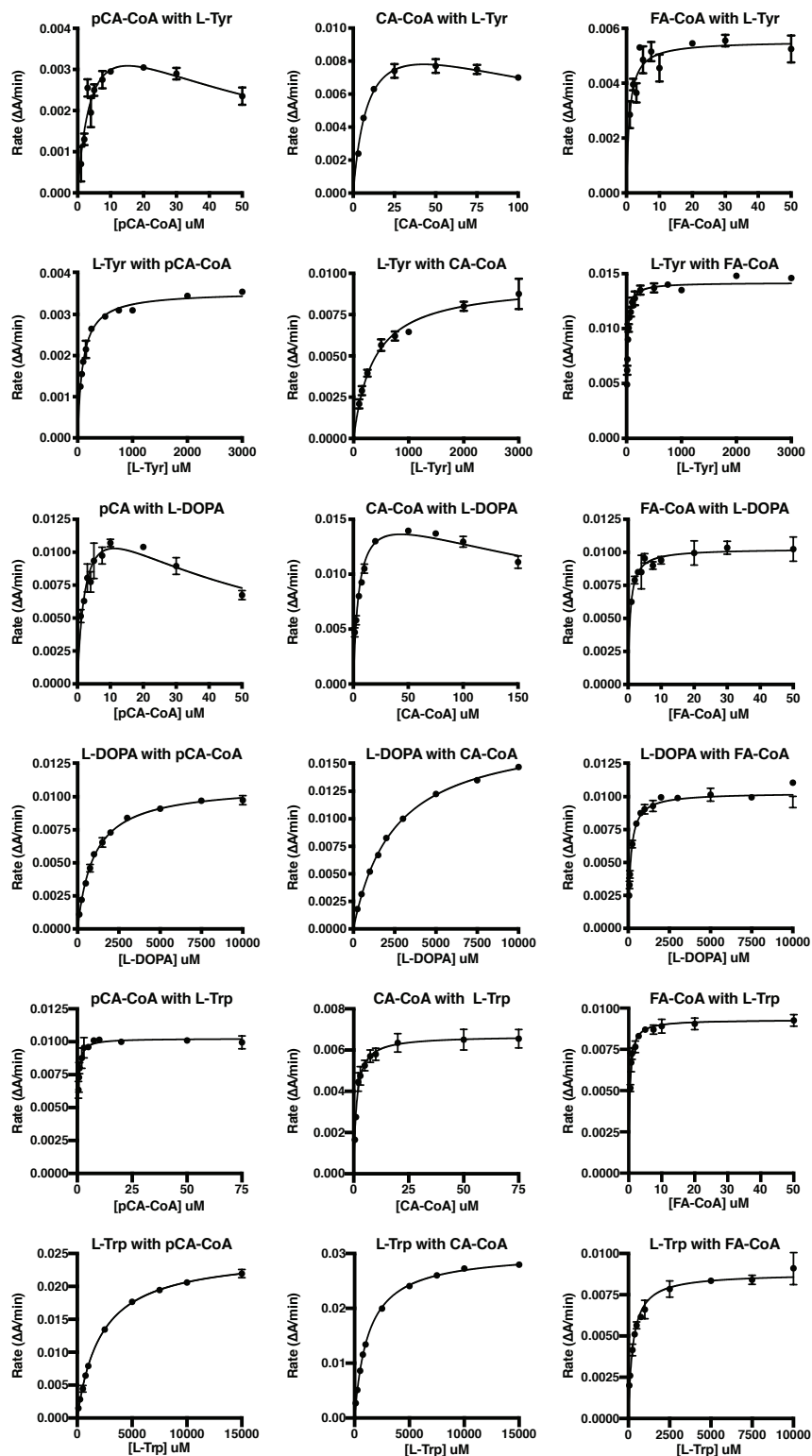


Figure S3. Kinetic curves showing rate (as $\Delta A_{412\text{nm}}/\text{min}$) versus substrate concentration. Donor and acceptor substrates are as indicated. $\Delta A_{412\text{nm}}/\text{min}$ values were converted to katal as described in Sullivan and Bonawitz, 2018 for determination calculation of k_{cat} .