

## *Supporting Information for*

### **Evaluation and comparison of antioxidant abilities of five bioactive molecules with C-H and O-H in thermodynamics and kinetics**

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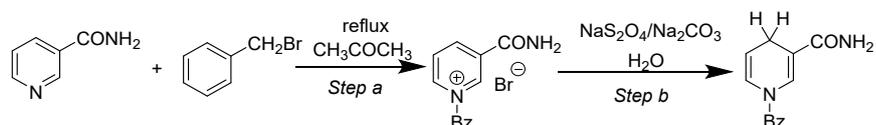
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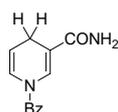
## SI. Syntheses of compounds

### Syntheses of BNAH:



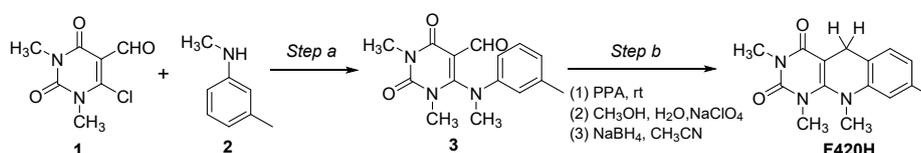
*Step a:* Nicotinamide (2.44 g, 0.02 mol) was dissolved in acetone (50 ml), and 3.80 g (0.03 mol) PhCH<sub>2</sub>Br was added in the solution, then the mixture was refluxed for 7 h. The reaction was monitored by TLC until the reaction was finished. When the mixture was cooled to room temperature, the precipitation was filtered and recrystallized to give BNA<sup>+</sup> (yield 85%).

*Step b:* 16.63 g (0.096 mol) Na<sub>2</sub>S<sub>2</sub>O<sub>4</sub> and 5.55 g (0.052 mol) Na<sub>2</sub>CO<sub>3</sub> were added in 40 ml H<sub>2</sub>O, the aqueous solution (mark as solution A) was removed oxygen by bubbling with pure argon gas for 15 min. 3.0 g (0.012 mol) BNA<sup>+</sup> was dissolved in 20 ml H<sub>2</sub>O (mark as solution B), and solution B was removed oxygen by bubbling with pure argon gas for 15 min. Solution B was slowly dropped into solution A within 30 min, then the mixture was stirred for 4 h at room temperature to get crude product BNAH. The crude product was recrystallized from mixed solvent (H<sub>2</sub>O and ethonal) to give pure BNAH (yield 40%).



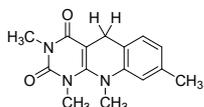
<sup>1</sup>H NMR (CD<sub>3</sub>Cl, 400MHz): δ (ppm) 7.25-7.45(q, 5H), 7.00(s, 1H), 5.86 (d, 1H), 5.58(s, 2H), 4.75(t, 1H), 4.33 (s, 2H), 3.08 (s, 2H).

### Syntheses of F420H:



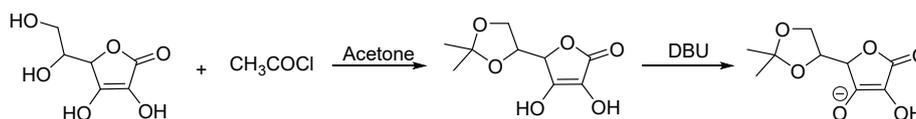
*Step a:* 5mmol 6-chloro-5-formyl-1,3-dimethyluracil (1) was solved in CH<sub>2</sub>Cl<sub>2</sub> in a round-bottomed flask, add 5 mmol substituted N-methylanilines (2) into the flask, then 0.7ml of dried Et<sub>3</sub>N was added, stirred the solution for 10 h at room temperature. Then the reaction mixture was evaporated in vacuo and will get yellow products. The yellow residues are pure enough for the next step, so they do not need any purification (yield 70%).

*Step b:* Compounds 3 without purification are placed in a 50ml flask, then add polyphosphoric acid (PPA) to cover the yellow compounds, the mixture was stirring at room temperature for 12 h by using a mechanical stirrer. Then, 30 ml of cold methanol was added into the flask slowly to prevent violent exothermic. When the mixture became liquid from viscous, the mixture was poured into a 500 ml flask which contains 200 ml water in it. Add sodium hydroxide to adjust pH to 7 under ice bath, adding sodium perchlorate (NaClO<sub>4</sub>) slowly while adjusting the pH, and precipitate of the models' salt will appear, add sodium perchlorate until the precipitate stop to increase. The precipitate was filtered. The pale yellow powder collected was then dissolved in acetonitrile directly. Monitored by TLC, the salt has a bright fluorescence under UV lamp. Then add NaBH<sub>4</sub> into the solution, monitored by TLC until the bright fluorescence disappeared. The solution was removed by a rotary evaporator, and the white reduced F420 model will be got. Purification through column chromatography will get pure target products F420H (yield 65%).

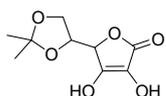


<sup>1</sup>H NMR (CDCl<sub>3</sub>, 400 MHz): δ (ppm) 7.05 (d, 1H), 6.90 (d, 1H), 6.86 (s, 1H), 3.71 (s, 2H), 3.49 (s, 3H), 3.37 (s, 3H), 3.33 (s, 3H), 2.34 (s, 3H).

### Syntheses of iAscH<sup>-</sup>:



The 5g L- ascorbic acid, 30ml of anhydrous acetone and 2.2ml of acetyl chloride were added to 100ml single mouth flask, refluxed for 2h at 40-50°C. Then cooled at -10°C and filtered to give a white solid, washed with cold acetone and dried under vacuum (yield 87%).



<sup>1</sup>H NMR (400MHz, *d*<sub>6</sub>-DMSO): δ (ppm) 1.23 (s, 6H), 3.85 (m, 1H), 4.07 (m, 1H), 4.24 (m, 1H), 4.68 (d, 1H), 8.47 (s, 1H), 11.28 (s, 1H).

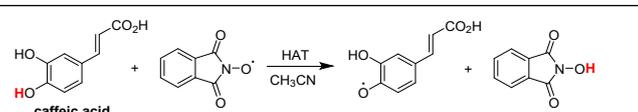
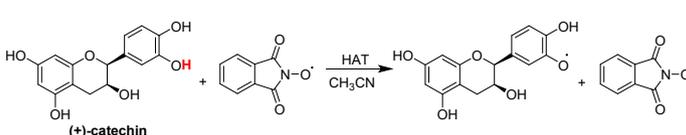
### Syntheses of caffeic acid, (+)-catechin and DPPH<sup>•</sup>:

These three compounds were commercially available.

### SII. Calculation process of parameters $\Delta G^{\ddagger}_{XH/X}$ and $\Delta G^{\ddagger 0}(XH)$ for caffeic acid and (+)-catechin

According to the rate constants  $k_H$  of two hydrogen transfer reaction (HAT) between caffeic acid and (+)-catechin with phthalimide-N-oxyl radical (PINO) respectively in literature (*J. Org. Chem.* 2014, 79, 11, 5209–5218),<sup>S1</sup> the activation free energies of the HAT reactions can be obtained by using Eyring equation [ $k_2 = (k_B T/h) \exp(-\Delta G^{\ddagger}/RT)$ ], as shown in Table S1.

**Table S1.** Second-order rate constants  $k_H$  ( $M^{-1}s^{-1}$ ) for the HAT reactions of PINO radical with caffeic acid and (+)-catechin measured at 298 K in acetonitrile.

HAT Reaction	$k_H$ ( $M^{-1}s^{-1}$ ) <sup>a</sup>	$\Delta G^{\ddagger}$ (kcal/mol)
 caffeic acid	$8.2 \times 10^4$	10.74
 (+)-catechin	$1.2 \times 10^5$	10.52

<sup>a</sup>  $k_H$  is derived from reference S1.

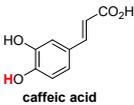
According to the previous work,<sup>S2</sup> the activation free energy of HAT reaction (eq 1) is equal to the sum of the thermo-kinetic parameters  $\Delta G^{\ddagger 0}$  of the two reactants (eq 2). The definition of thermo-kinetic parameter  $\Delta G^{\ddagger 0}(XH)$  is listed in eq 3. As the thermo-kinetic parameter  $\Delta G^{\ddagger 0}(\text{PINO}) = -34.94$  kcal/mol of PINO has already been determined in our previous work<sup>S3</sup>, the thermo-kinetic parameter  $\Delta G^{\ddagger 0}(XH) = 46.84$  kcal/mol of caffeic acid can be derived according to eq 2. As the bond dissociation free energy of caffeic acid is known,<sup>S1</sup> the self-exchange HAT reaction  $\Delta G^{\ddagger}_{XH/X}$  of caffeic acid can be determined according to eq 3. Using the same method, the parameters of (+)-catechin can be determined, too. The data are listed in Table S2. In order to conveniently calculate the activation free energies of the self-exchange HAT reactions  $\Delta G^{\ddagger}_{XH/X}$  of the two antioxidants, the data of bond dissociation free energies  $\Delta G^0(XH)$  are also listed in Table S2.



$$\Delta G^{\ddagger}_{XH/Y} = \Delta G^{\ddagger 0}(XH) + \Delta G^{\ddagger 0}(Y) \quad (2)$$

$$\Delta G^{\ddagger 0}(XH) \equiv 1/2[\Delta G^{\ddagger}_{XH/X} + \Delta G^0(XH)] \quad (3)$$

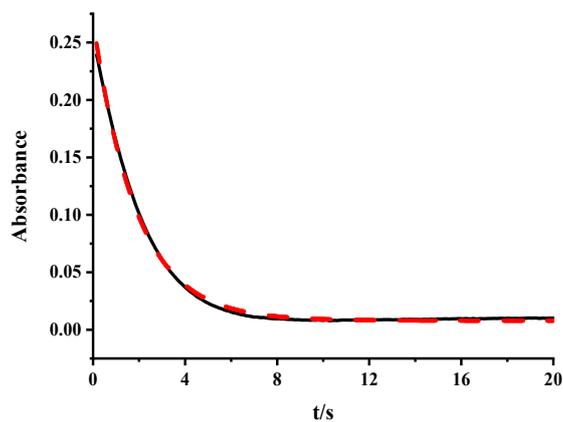
**Table S2.** Three parameters of caffeic acid and (+)-catechin at 298 K (kcal/mol).

Compound	$\Delta G^{\circ}(\text{XH})^a$	$\Delta G^{\ddagger}_{\text{XH}/\text{X}}$	$\Delta G^{\ddagger 0}(\text{XH})$
	kcal/mol		
 caffeic acid	77.0	16.68	46.84
 (+)-catechin	76.2	17.02	46.61

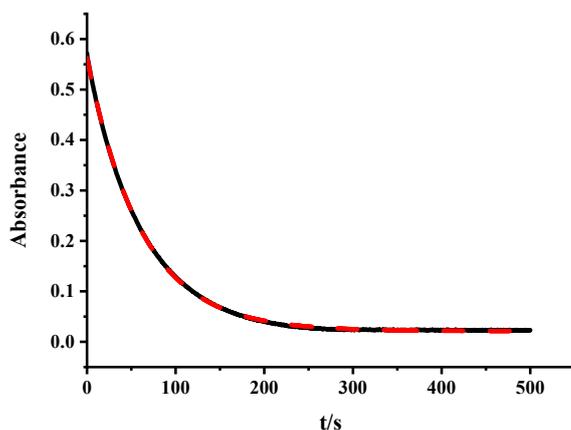
<sup>a</sup>  $\Delta G^{\circ}$  is derived from BDE of the compound,  $\Delta G^{\circ} = \text{BDE} - 4.9$ .<sup>S4</sup>

### SI. Kinetic absorbance decay curves of four HAT reactions BNAH/DPPH<sup>•</sup>, (+)-catechin/DPPH<sup>•</sup>, F420H/DPPH<sup>•</sup> and caffeic acid/DPPH<sup>•</sup> in acetonitrile at 298 K

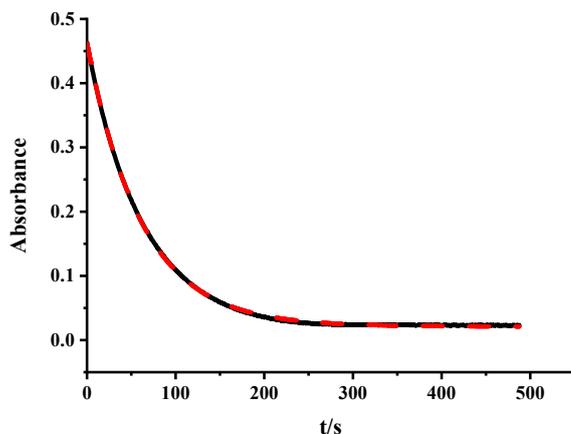
In order to verify whether the order of thermo-kinetic parameters of these five antioxidants is accurate, the second-order rate constants  $k_{\text{H}}$  ( $\text{M}^{-1}\text{s}^{-1}$ ) of the HAT reactions BNAH/DPPH<sup>•</sup>, (+)-catechin/DPPH<sup>•</sup>, F420H/DPPH<sup>•</sup> and caffeic acid/DPPH<sup>•</sup> were researched using stopped-flow method by monitoring the absorbance decay of DPPH<sup>•</sup> at 518 nm using pseudo-first-order kinetic model. The kinetic absorbance decay curves of these four HAT reactions were shown as follows. The concentration of the antioxidant was maintained at more than 20-fold excess of the oxidant to attain pseudo-first-order condition. The second-order rate constants ( $k_2$ ) were derived from plots of the pseudo-first-order rate constants versus the concentrations of the excessive reactants. In each case, it was confirmed that the rate constants derived from three to five independent measurements agreed within an experimental error of  $\pm 5\%$ .



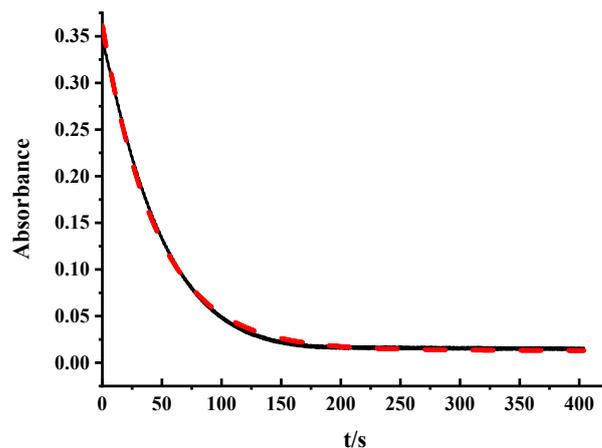
**Figure S1.** Absorbance decay of DPPH<sup>•</sup> (0.1 mM) in acetonitrile at  $\lambda_{\text{max}} = 518$  nm after addition of BNAH (2.0 mM) in deaerated anhydrous acetonitrile at 298 K (black line) and the fit (red line) using pseudo-first-order kinetic model. ( $k_2 = 1.07 \times 10^2 \text{ M}^{-1}\text{s}^{-1}$ )



**Figure S2.** Absorbance decay of DPPH<sup>•</sup> (0.1 mM) in acetonitrile at  $\lambda_{\text{max}} = 518$  nm after addition of (+)-catechin (2.0 mM) in deaerated anhydrous acetonitrile at 298 K (black line) and the fit (red line) using pseudo-first-order kinetic model. ( $k_2 = 2.35 \text{ M}^{-1}\text{s}^{-1}$ )

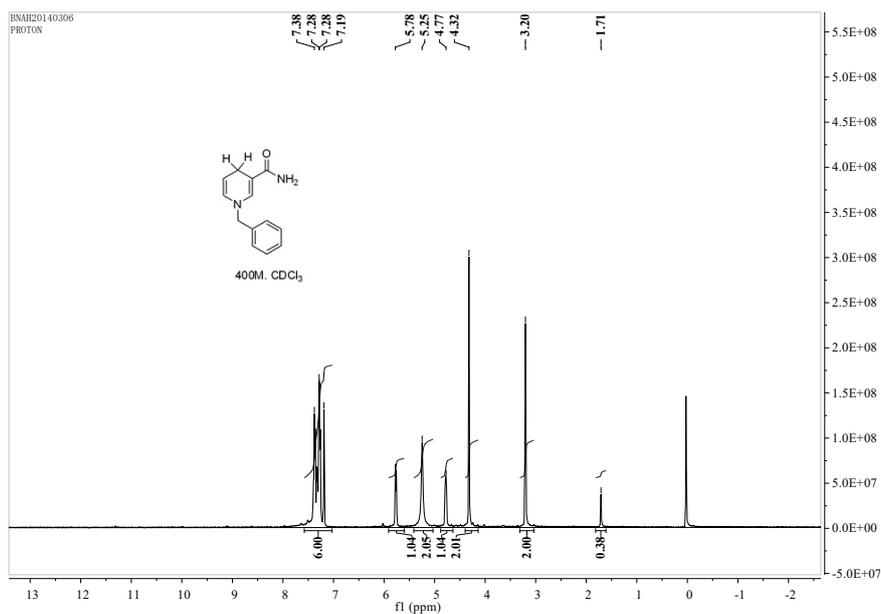


**Figure S3.** Absorbance decay of DPPH<sup>•</sup> (0.1 mM) in acetonitrile at  $\lambda_{\text{max}} = 518$  nm after addition of F420H (2.0 mM) in deaerated anhydrous acetonitrile at 298 K (black line) and the fit (red line) using pseudo-first-order kinetic model. ( $k_2 = 2.02 \text{ M}^{-1}\text{s}^{-1}$ )



**Figure S4.** Absorbance decay of DPPH<sup>•</sup> (0.1 mM) in acetonitrile at  $\lambda_{\text{max}} = 518 \text{ nm}$  after addition of caffeic acid (2.0 mM) in deaerated anhydrous acetonitrile at 298 K (black line) and the fit (red line) using pseudo-first-order kinetic model. ( $k_2 = 1.60 \text{ M}^{-1}\text{s}^{-1}$ )

#### SIV. <sup>1</sup>H NMR spectra of BNAH, F420H and iAsch<sub>2</sub>



**Scheme S1.** <sup>1</sup>H NMR spectrum of BNAH.

