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## **ELECTRONIC SUPPLEMENTARY INFORMATION**

for the paper entitled

Synthesis, Physicochemical Characterization and Neuroprotective Evaluation of Novel 1-hydroxypyrazin-2(1*H*)-one Iron Chelators in an *In Vitro* Cell Model of Parkinson's Disease

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## **1. Organic Synthesis**

#### Discussion

The target 1-hydroxypyrazin-2(1H)-ones 6 were synthesized in two steps from amino acid ethyl esters following the literature procedures as shown below in Scheme S1.<sup>1</sup> Initially, reaction of glycine ethyl ester hydrochloride 4a with hydroxylamine hydrochloride in alkaline water afforded the known glycine hydroxamic acid **5a** in 64 % yield.<sup>1a,2</sup> Condensation reaction of **5a** with 2,3-butanedione afforded the known 1-hydroxypyrazin-2(1*H*)-one  $6a^{1b,3-5}$  in 24 % yield (Scheme S1). Unfortunately, application of this two-step procedure to the synthesis of **6b** from alanine ethyl ester **4b** failed to give the desired product, due to the high solubility of the hydroxamic acid **5b** in water. We subsequently modified this procedure by using methanol as the solvent and we were able to obtain 5a from 4a in 56 % yield (Scheme S1). However, application of this modified procedure to the synthesis of alanine hydroxamic acid **5b**<sup>2b,2c,6</sup> from alanine ethyl ester **4b** gave a mixture of **5b** and another compound (presumed to be the corresponding diketopiperazine) in low yield as judged by <sup>1</sup>H NMR spectroscopy. Reaction of this mixture with 2,3-butanedione gave an intractable mixture of products from which the novel 1-hydroxypyrazin-2(1H)-one **6b** could not be isolated by chromatography. However, 1hydroxypyrazin-2(1H)-ones **6c** and **6d** were successfully obtained by this modified procedure from the known hydroxamic acids  $5c^{1b,2b,7}$  and 5d,<sup>8</sup> albeit in only 13 % and 14 % overall yields from 4c and 4d, respectively (Scheme S1). There are some reports of multifunctional hydroxypyridinone metal chelators containing phenolic antioxidant moieties that show promising efficacy against neurodegenerative diseases by acting as radical traps as well as metal chelators.<sup>9</sup> Accordingly, we synthesized 1-hydroxypyrazin-2(1H)-one **6d** that contains a phenol moiety which could provide a beneficial antioxidant mode of action in addition to iron chelation. Unfortunately, all our attempts to isolate hydroxamic acids 5e-5g from amino esters 4e-4g met with no success.



H-Gly-OEta $R_1 = Et$ ,  $R_2 = H$ H-Ala-OEtb $R_1 = Et$ ,  $R_2 = Me$ H-Phe-OBnc $R_1 = Bn$ ,  $R_2 = PhCH_2$ H-Tyr-OEtd $R_1 = Et$ ,  $R_2 = 4-HOC_6H_4CH_2$ H-Val-OEte $R_1 = Et$ ,  $R_2 = i$ -PrH-Leu-OEtf $R_1 = Et$ ,  $R_2 = i$ -PrCH2H-Ile-OEtg $R_1 = Et$ ,  $R_2 = CH_3CH_2CH(Me)$ 

Scheme S1. Synthesis of 1-hydroxypyrazin-2(1*H*)-ones 6a–6g.

Due to the low yields obtained above and the failure to synthesize certain 1-hydroxypyrazin-2(1H)ones **6** by the procedure shown in Scheme S1, we sought a more general synthetic method which could
be applied to the synthesis of a broader range of these compounds. The synthesis of 1-hydroxypyrazin-2(1H)-ones **6** in 4 steps from *N*-Boc amino acids *via* their protected hydroxamic acid benzyl esters was
previously reported.<sup>3–5,10</sup> Inspired by this approach, we explored a new synthesis of 1-hydroxypyrazin-2(1H)-ones **6** from activated *N*-Boc amino acid *N*-hydroxysuccinimide esters **7** as shown below in
Scheme S2.

Reaction of *N*-Boc-protected *N*-hydroxysuccinimide esters **7a**, **7b**, **7c**, **7e** and **7f** with *O*benzylhydroxylamine generated the Boc-protected aminohydroxamic acid benzyl esters **8a**, **8b**, **8c**, **8e** and **8f** in high yields. Subsequent *N*-Boc deprotection (TFA in DCM) gave the free aminohydroxamic acid benzyl esters **9b**, **9c**, **9e** and **9f** in excellent yields. However, despite the known formation of **6'a** from **9a** (as HCl salt) and 2,3-butanedione reported in the literature,<sup>5,11</sup> attempted condensation reactions of compounds **9b**, **9c**, **9e** and **9f** with 2,3-butanedione in our hands failed to generate the desired 1-benzyloxypyrazin-2(1*H*)-ones **6'b**, **6'c**, **6'e** and **6'f**. This synthetic approach was subsequently abandoned in favour of the approach outlined above in Scheme S1.



Scheme S2. Attempted synthesis of 1-hydroxypyrazin-2(1*H*)-ones 6a–6f.

We also explored the reactions of glycine hydroxamic acid **5a** with both aromatic and aliphatic  $\alpha$ -ketoaldehydes (glyoxals) as shown below in Scheme S3. Reaction of **5a** with phenylglyoxal in ethanol/water at reflux afforded the novel 1-hydroxypyrazin-2(1*H*)-one **10a** in 30 % yield as a single regioisomer. Similarly, reaction of **5a** with 4-methoxyphenylglyoxal and 4-fluorophenylglyoxal gave **10b** and **10c** as single regioisomers in 27 % and 24 % yields, respectively. As with 1-hydroxypyrazin-2(1*H*)-one **6d**, we sought to convert **10b** into a 1-hydroxypyrazin-2(1*H*)-one bearing a phenol moiety with potential antioxidant activity. Accordingly, deprotection of the methoxy group of **10b** with boron tribromide in DCM afforded the novel 1-hydroxypyrazin-2(1*H*)-one **10a** in 21 % yield. Reaction of **5a** with pyruvaldehyde gave the novel 1-hydroxypyrazin-2(1*H*)-ones **11a** and **12a** as a 12:1 mixture of regioisomers, as judged by <sup>1</sup>H NMR spectroscopy (Scheme S3). The major regioisomer was tentatively assigned as **11a** on the basis that the free primary amino group of **5a** would preferentially react with the aldehyde carbonyl group of the glyoxal, rather than the less electrophilic ketone carbonyl group. These regioisomers proved inseparable by recrystallisation or chromatography, and were studied without further purification.



Scheme S3. Synthesis of 1-hydroxypyrazin-2(1*H*)-ones 10a–10d, 11a and 12a.

## 2. Experimental Procedures

Synthesis of N-Boc hydroxamic acid benzyl esters 8a-8f: General procedure



To a solution of the appropriate *N*-Boc amino acid *N*-hydroxysuccinimide (OSu) ester **7** (1.47 mmol) in DCM (20 mL) at room temperature was added *O*-benzylhydroxylamine (1.47 mmol, 1 eq). The solution was allowed to stir at room temperature for 24 hours. The solvent was evaporated to afford the crude *N*-Boc hydroxamic acid benzyl ester **8** as an oil that crystallised contaminated with *N*-hydroxysuccinimide. This mixture was used in the next step without further purification.

*N*-Boc glycine hydroxamic acid benzyl ester  $8a^{3-5,11} \delta_{H}(399.8 \text{ MHz}, \text{CDCl}_3, \text{Me}_4\text{Si})$  1.40 (9H, s, (CH<sub>3</sub>)<sub>3</sub>), 3.67 (2H, s, CH<sub>2</sub>NHBoc), 4.82 (1H, br s, NH), 4.88 (2H, s, OCH<sub>2</sub>Ph), 5.25 (1H, br s, NHBoc), 7.36 (5H, s, ArH).

*N***-Boc alanine hydroxamic acid benzyl ester 8b**<sup>11</sup> δ<sub>H</sub>(399.8 MHz, CDCl<sub>3</sub>, Me<sub>4</sub>Si) 1.28 (3H, d, *J* 6.8, CH<sub>3</sub>CH), 1.38 (9H, s, (CH<sub>3</sub>)<sub>3</sub>), 4.05 (1H, app t, *J* 6.8, CH<sub>3</sub>CH), 4.86 (2H, s, OCH<sub>2</sub>Ph), 5.29 (1H, d, *J* 6.4, NHBoc), 7.27–7.36 (5H, m, ArH).

*N*-Boc phenylalanine hydroxamic acid benzyl ester 8c<sup>12,13</sup> δ<sub>H</sub>(399.8 MHz, CDCl<sub>3</sub>, Me<sub>4</sub>Si) 1.36 (9H, s, (CH<sub>3</sub>)<sub>3</sub>), 2.96–3.06 (2H, m, CHCH<sub>2</sub>Ph), 4.20 (1H, q, *J* 7.6, C*H*NHBoc), 4.62–4.84 (2H, m, OCH<sub>2</sub>Ph), 5.26 (1H, d, *J* 7.6, CHNHBoc), 7.17–7.36 (10H, m, Ar*H*).

*N***-Boc valine hydroxamic acid benzyl ester 8e**<sup>12,13</sup> δ<sub>H</sub>(399.8 MHz, CDCl<sub>3</sub>, Me<sub>4</sub>Si) 0.88 (3H, d, *J* 6.4, (C*H*<sub>3</sub>)<sub>2</sub>CH), 0.90 (3H, d, *J* 6.4, (C*H*<sub>3</sub>)<sub>2</sub>CH), 1.39 (9H, s, (C*H*<sub>3</sub>)<sub>3</sub>), 1.96–2.02 (1H, m, (CH<sub>3</sub>)<sub>2</sub>CH), 3.70 (1H, t, *J* 8.4, C*H*NHBoc), 4.88 (2H, s, OC*H*<sub>2</sub>Ph), 5.28 (1H, d, *J* 8.4, CHNHBoc), 7.28–7.37 (5H, m, Ar*H*), 9.45 (1H, br s, N*H*OCH<sub>2</sub>Ph).

*N*-Boc leucine hydroxamic acid benzyl ester 8f<sup>11</sup> δ<sub>H</sub>(399.8 MHz, CDCl<sub>3</sub>, Me<sub>4</sub>Si) 0.85–0.87 (6H, m, (CH<sub>3</sub>)<sub>2</sub>CH), 1.39 (9H, s, (CH<sub>3</sub>)<sub>3</sub>), 1.41–1.47 (1H, m, (CH<sub>3</sub>)<sub>2</sub>CH), 1.54–1.60 (2H, m, (CH<sub>2</sub>CH(CH<sub>3</sub>)<sub>2</sub>),

3.97 (1H, q, *J* 8.0, *CH*NHBoc), 4.88 (2H, s, OC*H*<sub>2</sub>Ph), 5.17 (1H, d, *J* 8.0, CHNHBoc), 7.24–7.37 (5H, m, Ar*H*), 9.45 (1H, br s, N*H*OCH<sub>2</sub>Ph).

### Synthesis of hydroxamic acid benzyl ester TFA salts 9b–9f: General procedure



The appropriate crude *N*-Boc hydroxamic acid benzyl ester **8** (1.47 mmol) was dissolved in DCM (10 mL) and trifluoroacetic acid (10 mL) was added. The solution was allowed to stir at room temperature for 24 hours. The solvents were evaporated to afford the crude TFA salt **9** as a clear oil. The oil was triturated with diethyl ether (10 mL) and the resulting white solid was filtered and washed with diethyl ether (10 mL) and allowed to dry in air to afford the pure TFA salt **9** as a white solid.

Alanine hydroxamic acid benzyl ester TFA salt  $9b^{4,11}$  Obtained from 7b in 87% overall yield.  $\delta_{H}(399.8 \text{ MHz}, \text{DMSO-}d_6)$  1.24 (3H, d, *J* 7.2, *CH*<sub>3</sub>CH), 3.64 (1H, br s, CH<sub>3</sub>CH), 4.77 (1H, d, *J* 11.2, OCH<sub>2</sub>Ph), 4.81 (1H, d, *J* 11.2, OCH<sub>2</sub>Ph), 7.34–7.38 (5H, m, ArH).

**Phenylalanine hydroxamic acid benzyl ester TFA salt 9c**<sup>12,13</sup> Obtained from **7c** in 78% overall yield.  $\delta_{\rm H}(399.8 \text{ MHz}, D_2O)$  2.91 (1H, dd, *J* 14.0 and 8.4, CHC*H*<sub>2</sub>Ph), 2.99 (1H, dd, *J* 14.0 and 6.8, CHC*H*<sub>2</sub>Ph), 3.82 (1H, dd, *J* 8.4 and 6.8, CHCH<sub>2</sub>Ph), 4.41 (1H, d, *J* 11.0, OC*H*<sub>2</sub>Ph), 4.59 (1H, d, *J* 11.0, OC*H*<sub>2</sub>Ph), 7.06–7.13 (4H, m, Ar*H*), 7.20–7.26 (6H, m, Ar*H*).

Valine hydroxamic acid benzyl ester TFA salt  $9e^{12,13}$  Obtained from 7e in 91% overall yield.  $\delta_{H}(399.8 \text{ MHz}, D_2O) 0.69 (3H, d, J 6.4, (CH_3)_2)CH)$ , 0.73 (3H, d, J 6.4, (CH<sub>3</sub>)\_2)CH), 1.88 (1H, sp, J 6.4, (CH<sub>3</sub>)\_2CH), 3.38 (1H, d, J 6.4, CHNH<sub>3</sub><sup>+</sup>), 4.75 (1H, d, J 11.2, OCH<sub>2</sub>Ph), 4.80 (1H, d, J 11.2, OCH<sub>2</sub>Ph), 7.27–7.32 (5H, m, ArH).

Leucine hydroxamic acid benzyl ester TFA salt 9f<sup>11</sup> Obtained from 7f in 84% overall yield.  $\delta_{H}(399.8 \text{ MHz}, D_2O)$  0.65 (3H, d, J 6.0, (CH<sub>3</sub>)<sub>2</sub>CH), 0.67 (3H, d, J 6.0, (CH<sub>3</sub>)<sub>2</sub>CH), 1.01–1.10 (1H, m, (CH<sub>3</sub>)<sub>2</sub>CH), 1.37 (2H, t, J 7.2, CH<sub>2</sub>CH(CH<sub>3</sub>)<sub>2</sub>), 3.56 (1H, t, J 7.2, CHNH<sub>3</sub><sup>+</sup>), 4.74 (1H, d, J 11.2, OCH<sub>2</sub>Ph), 4.82 (1H, d, J 11.2, OCH<sub>2</sub>Ph), 7.25–7.34 (5H, m, ArH).

## 3. NMR Spectra



Glycine hydroxamic acid **5a** 



Alanine hydroxamic acid **5b** 











1-Hydroxy-5,6-dimethylpyrazin-2(1*H*)-one **6a** 









1-Hydroxy-3-benzyl-5,6-dimethylpyrazin-2(1*H*)-one **6c** 





1-Hydroxy-3-(4-hydroxybenzyl)-5,6-dimethylpyrazin-2(1*H*)-one **6d** 





*N*-Boc glycine hydroxamic acid benzyl ester **8a** 



*N*-Boc alanine hydroxamic acid benzyl ester **8b** 



*N*-Boc phenylalanine hydroxamic acid benzyl ester **8c** 



*N*-Boc valine hydroxamic acid benzyl ester **8e** 

87.57 5.0 4.0 53.56 3.0 48 25 2 6 abundance 0 4.91 √ **№** 5.0 4,002 3,982 3,945 3,945 3,945 3,945 3.0 9.0 8.0 6.0 2.0 1.0 Ζ I III 11 X : parts per Million : Proton 7.562 7.385 7.388 7.388 7.388 7.388 6.984 6.664 6.196 8.118 684 2.432 58 8 3.471 610 ci ci

 $\mathit{N}\text{-}\mathsf{Boc}$  leucine hydroxamic acid benzyl ester  $\mathbf{8f}$ 



Alanine hydroxamic acid benzyl ester TFA salt **9b** 



Phenylalanine hydroxamic acid benzyl ester TFA salt **9c** 



Valine hydroxamic acid benzyl ester TFA salt **9e** 



Leucine hydroxamic acid benzyl ester TFA salt 9f



1-Hydroxy-6-phenylpyrazin-2(1*H*)-one **10a** 





1-Hydroxy-6-(4-methoxyphenyl)-pyrazin-2(1*H*)-one **10b** 







1-Hydroxy-6-(4-fluorophenyl)-pyrazin-2(1*H*)-one **10c**






1-Hydroxy-6-(4-hydroxyphenyl)-pyrazin-2(1*H*)-one **10d** 









1-Hydroxy-6-methylpyrazin-2(1*H*)-one **11a** and 1-Hydroxy-5-methylpyrazin-2(1*H*)-one **12a** 







## 4. Mass Spectra



1-Hydroxy-3-(4-hydroxybenzyl)-5,6-dimethylpyrazin-2(1*H*)-one 6d









1-Hydroxy-6-(4-methoxyphenyl)-pyrazin-2(1*H*)-one **10b** 







### 1-Hydroxy-6-(4-fluorophenyl)-pyrazin-2(1*H*)-one **10**c





## 1-Hydroxy-6-(4-hydroxyphenyl)-pyrazin-2(1*H*)-one **10d**



1-Hydroxy-6-methylpyrazin-2(1*H*)-one **11a** and 1-Hydroxy-5-methylpyrazin-2(1*H*)-one **12a** 

# **5.** Determination of pKa Values of the Ligands and Stability Constants of the Complexes



#### Protonation studies with ligand 11a

Figure S1. Spectrophotometric titrations vs pH of ligand 11a between (A) -0.5 < pH < 2.08 (batch titration,  $[11a] = 2.57 \times 10^{-4}$  M) and (B) 2.61 < pH < 10.17 (direct titration,  $[11a] = 2.55 \times 10^{-4}$  M). (C) Electronic spectra and (D) distribution curves ( $[11a] = 2.55 \times 10^{-4}$  M) of the protonated species of ligand 11a. Solvent: H<sub>2</sub>O, I = 0.1 M (NaClO<sub>4</sub>), T= 25.0 °C.





Figure S2. Spectrophotometric titrations vs pH of ligand 10a between (A) -0.75 < pH < 2 (batch titration, [10a] =  $1.95 \times 10^{-4}$  M) and (B) 2.12 < pH < 11.79 (direct titration, [10a] =  $1.02 \times 10^{-4}$  M). (C) Electronic spectra and (D) distribution curves ([10a] =  $1.95 \times 10^{-4}$  M) of the protonated species of ligand 10a. Solvent: H<sub>2</sub>O, I = 0.1 M (NaClO<sub>4</sub>), T= 25.0 °C.





Figure S3. Spectrophotometric titrations vs pH of ligand 10a between (A) -0.37 < pH < 0.63 (batch titration,  $[10a] = 1.02 \times 10^{-4}$  M) and (B) 2.43 < pH < 11.88 (direct titration,  $[10a] = 1.01 \times 10^{-3}$  M). (C) Electronic spectra and (D) distribution curves ( $[10a] = 1.02 \times 10^{-4}$  M) of the protonated species of ligand 10a. Solvent: MeOH/H<sub>2</sub>O (80/20 w/w), I = 0.1 M (NaClO<sub>4</sub>), T= 25.0 °C.

#### Protonation studies with ligand 6d



Figure S4. Spectrophotometric titrations vs pH of ligand 6d between (A) -0.36 < pH < 2.36 (batch titration, [6d] =  $3.0 \times 10^{-4}$  M) and (B) 1.78 < pH < 11.47 (direct titration, [6d] =  $9.98 \times 10^{-5}$  M). (C) Electronic spectra and (D) distribution curves ([6d] =  $3.0 \times 10^{-4}$  M) of the protonated species of ligand 6d. Solvent: MeOH/H<sub>2</sub>O (80/20 w/w), I = 0.1 M (NaClO<sub>4</sub>), T= 25.0 °C.

#### **Protonation studies with ligand 6c**



Figure S5. Spectrophotometric titrations vs pH of ligand 6c between (A) -0.04 < pH < 2.11 (batch titration, [6c] =  $3.0 \times 10^{-4}$  M) and (B) 2.49 < pH < 11.74 (direct titration, [6c] =  $9.98 \times 10^{-5}$  M). (C) Electronic spectra and (D) distribution curves ([6c] =  $1.54 \times 10^{-4}$  M) of the protonated species of ligand 6c. Solvent: MeOH/H<sub>2</sub>O (80/20 w/w), I = 0.1 M (NaClO<sub>4</sub>), T= 25.0 °C.



Fe<sup>3+</sup> complexation studies with ligand 6a

Figure S6. Spectrophotometric titration vs pH of Fe<sup>3+</sup> complexes of ligand **6a** between (A)  $-0.5 \le$  pH  $\le 1.25$  (batch titration, [**6a**] =  $3.78 \times 10^{-4}$  M, [Fe<sup>3+</sup>] =  $1.26 \times 10^{-4}$  M) and (**B**)  $1.97 \le$ pH  $\le 12.04$  (direct titration, [**6a**] =  $2.38 \times 10^{-4}$  M, [Fe<sup>3+</sup>] =  $7.14 \times 10^{-5}$  M). (**C**) Electronic spectra and (**D**) distribution curves ([**6a**] =  $2.38 \times 10^{-4}$  M, [Fe<sup>3+</sup>] =  $7.14 \times 10^{-5}$  M) of the Fe<sup>3+</sup> complexes of **6a**. Solvent: H<sub>2</sub>O, I = 0.1 M (NaClO<sub>4</sub>), T= 25.0 °C.



## Fe<sup>3+</sup> complexation studies with ligand 10a

Figure S7. Spectrophotometric titration vs pH of Fe<sup>3+</sup> complexes of ligand **10a** between (A)  $-0.36 \le$  pH  $\le 1.80$  (batch titration, [**10a**] =  $1.02 \times 10^{-4}$  M, [Fe<sup>3+</sup>] =  $3.20 \times 10^{-5}$  M) and (B)  $2.34 \le$ pH  $\le 8.03$  (direct titration, [**10a**] =  $1.02 \times 10^{-3}$  M, [Fe<sup>3+</sup>] =  $3.12 \times 10^{-4}$  M). (C) Electronic spectra and (D) distribution curves ([**10a**] =  $1.02 \times 10^{-4}$  M, [Fe<sup>3+</sup>] =  $3.20 \times 10^{-5}$  M) of the Fe<sup>3+</sup> complexes of **10a**. Solvent: MeOH/H<sub>2</sub>O (80/20 w/w), I = 0.1 M (NaClO<sub>4</sub>), T= 25.0 °C.



Fe<sup>3+</sup> complexation studies with ligand 6d

Figure S8. Spectrophotometric titration vs pH of Fe<sup>3+</sup> complexes of ligand 6d between (A)  $-0.36 \le$  pH  $\le 2.36$  (batch titration, [6d] =  $3.0 \times 10^{-4}$  M, [Fe<sup>3+</sup>] =  $8.0 \times 10^{-5}$  M) and (B)  $1.97 \le$  pH  $\le 12.04$  (direct titration, [6d] =  $1.04 \times 10^{-4}$  M, [Fe<sup>3+</sup>] =  $3.23 \times 10^{-5}$  M). (C) Electronic spectra and (D) distribution curves ([6d] =  $1.04 \times 10^{-4}$  M, [Fe<sup>3+</sup>] =  $3.23 \times 10^{-5}$  M) of the Fe<sup>3+</sup> complexes of 6d. Solvent: MeOH/H<sub>2</sub>O (80/20 w/w), I = 0.1 M (NaClO<sub>4</sub>), T= 25.0 °C.



Fe<sup>3+</sup> complexation studies with ligand 6c

Figure S9. Spectrophotometric titration vs pH of Fe<sup>3+</sup> complexes of ligand 6c between (A)  $-0.36 \le$  pH  $\le 1.83$  (batch titration, [6c] =  $3.0 \times 10^{-4}$  M, [Fe<sup>3+</sup>] =  $9.38 \times 10^{-5}$  M) and (B)  $1.97 \le$ pH  $\le 12.04$  (direct titration, [6c] =  $1.02 \times 10^{-4}$  M, [Fe<sup>3+</sup>] =  $3.12 \times 10^{-5}$  M). (C) Electronic spectra and (D) distribution curves ([6c] =  $1.02 \times 10^{-4}$  M, [Fe<sup>3+</sup>] =  $3.12 \times 10^{-5}$  M) of the Fe<sup>3+</sup> complexes of 6c. Solvent: MeOH/H<sub>2</sub>O (80/20 w/w), I = 0.1 M (NaClO<sub>4</sub>), T= 25.0 °C.

# 6. BBB Penetration Scores

6a		
Property	Value	T <sub>0</sub>
Number of Aromatic Rings (Aro_R)	1	0.82
Number of Heavy Atoms (HA)	10	0.65
Molecular Weight (MW)	140.14	
Number of Hydrogen Bond Acceptor (HBA)	3	
Number of Hydrogen Bond Donor (HBD)	1	
MWHBN [MWHBN = (MW^(-0.5)*HBN),	0.34	0.71
where HBN=HBA+HBD]		
Topological Polar Surface Area(TPSA)	55.12	0.62
рКа	4.58	0.23
BBB SCORE		3.88

 Table S1. Predicted BBB score of compound 6a.

6с		
Property	Value	To
Number of Aromatic Rings (Aro_R)	2	1.00
Number of Heavy Atoms (HA)	17	0.98
Molecular Weight (MW)	230.26	
Number of Hydrogen Bond Acceptor (HBA)	3	
Number of Hydrogen Bond Donor (HBD)	1	
MWHBN [MWHBN = (MW^(-0.5)*HBN),	0.26	0.93
where HBN=HBA+HBD]		
Topological Polar Surface Area(TPSA)	65.42	0.54
рКа	5.53	0.47
BBB SCORE		4.70

# Table S2. Predicted BBB score of compound 6c.

6d		
Property	Value	T <sub>0</sub>
Number of Aromatic Rings (Aro_R)	2	1.00
Number of Heavy Atoms (HA)	18	0.99
Molecular Weight (MW)	246.26	
Number of Hydrogen Bond Acceptor (HBA)	4	
Number of Hydrogen Bond Donor (HBD)	2	
MWHBN [MWHBN = (MW^(-0.5)*HBN),	0.38	0.55
where HBN=HBA+HBD]		
Topological Polar Surface Area(TPSA)	75.35	0.47
рКа	5.96	0.58
BBB SCORE	1	4.05

# Table S3. Predicted BBB score of compound 6d.

10a		
Property	Value	To
Number of Aromatic Rings (Aro_R)	2	1.00
Number of Heavy Atoms (HA)	14	0.89
Molecular Weight (MW)	188.18	
Number of Hydrogen Bond Acceptor (HBA)	3	
Number of Hydrogen Bond Donor (HBD)	1	
MWHBN [MWHBN = (MW^(-0.5)*HBN),	0.29	0.87
where HBN=HBA+HBD]		
Topological Polar Surface Area(TPSA)	51.48	0.64
рКа	3.18	0.00
BBB SCORE		4.47

# Table S4. Predicted BBB score of compound 10a.

10d		
Property	Value	T <sub>0</sub>
Number of Aromatic Rings (Aro_R)	2	1.00
Number of Heavy Atoms (HA)	15	0.93
Molecular Weight (MW)	204.18	
Number of Hydrogen Bond Acceptor (HBA)	4	
Number of Hydrogen Bond Donor (HBD)	2	
MWHBN [MWHBN = (MW^(-0.5)*HBN),	0.42	0.40
where HBN=HBA+HBD]		
Topological Polar Surface Area(TPSA)	53.5	0.63
рКа	4	0.11
BBB SCORE		3.83

# Table S5. Predicted BBB score of compound 10d.

11a		
Property	Value	To
Number of Aromatic Rings (Aro_R)	1	0.82
Number of Heavy Atoms (HA)	9	0.56
Molecular Weight (MW)	126.11	
Number of Hydrogen Bond Acceptor (HBA)	3	
Number of Hydrogen Bond Donor (HBD)	1	
MWHBN [MWHBN = (MW^(-0.5)*HBN),	0.36	0.65
where HBN=HBA+HBD]		
Topological Polar Surface Area(TPSA)	31.01	0.78
рКа	3.98	0.11
BBB SCORE		3.97

# Table S6. Predicted BBB score of compound 11a.

2		
Property	Value	To
Number of Aromatic Rings (Aro_R)	1	0.82
Number of Heavy Atoms (HA)	10	0.65
Molecular Weight (MW)	141.12	
Number of Hydrogen Bond Acceptor (HBA)	3	
Number of Hydrogen Bond Donor (HBD)	2	
MWHBN [MWHBN = (MW^(-0.5)*HBN),	0.42	0.39
where HBN=HBA+HBD]		
Topological Polar Surface Area(TPSA)	34.37	0.76
рКа	6.02	0.60
BBB SCORE		3.87

 Table S7. Predicted BBB score of compound 2.

3		
Property	Value	To
Number of Aromatic Rings (Aro_R)	1	0.82
Number of Heavy Atoms (HA)	11	0.72
Molecular Weight (MW)	155.11	
Number of Hydrogen Bond Acceptor (HBA)	4	
Number of Hydrogen Bond Donor (HBD)	2	
MWHBN [MWHBN = (MW^(-0.5)*HBN),	0.48	0.00
where HBN=HBA+HBD]		
Topological Polar Surface Area(TPSA)	79.53	0.44
рКа	3.7	0.06
BBB SCORE		2.46

 Table S8. Predicted BBB score of compound 3.
DFP 1			
Property	Value	To	
Number of Aromatic Rings (Aro_R)	1	0.82	
Number of Heavy Atoms (HA)	10	0.65	
Molecular Weight (MW)	139.15		
Number of Hydrogen Bond Acceptor (HBA)	2		
Number of Hydrogen Bond Donor (HBD)	1		
MWHBN [MWHBN = (MW^(-0.5)*HBN),	0.25	0.95	
where HBN=HBA+HBD]			
Topological Polar Surface Area(TPSA)	38.95	0.73	
рКа	3.68	0.06	
BBB SCORE		4.37	

#### Table S9. Predicted BBB score of DFP 1.

 $\label{eq:stables} \begin{array}{l} \mbox{Table S10. Comparison of predicted BBB scores with percentage neuronal rescue from 6-OHDA} \\ & \mbox{neurotoxicity at 100 $\mu$M dose of the compound.} \end{array}$ 

Compound	BBB Score	% 6-OHDA Rescue
		(at 100 μM)
ба	3.88	100
6с	4.70	89
6d	4.05	64
10a	4.47	63
10d	3.83	76
11a	3.97	60
DFP 1	4.37	117
2	3.87	93
3	2.46	84



Figure S10. Plot of predicted BBB scores versus percentage neuronal rescue from 6-OHDA neurotoxicity at 100 µM dose of the compound, showing no clear correlation between the two properties (● = 6a–6d, 10a, 10d and 11a, ■ = 2 and 3, ▲ = DFP 1).

## 7. DPPH Antioxidant Assay



**Figure S11**. Percentage inhibition of the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical by ligand **6a** after 24 hours (24h) and 48 hours (48h).



Figure S12. Percentage inhibition of the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical by ligand 11a after 1 hour (1h), 24 hours (24h) and 48 hours (48h).



Figure S13. Percentage inhibition of the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical by ligand **10a** after 1 hour (1h), 24 hours (24h) and 48 hours (48h).



**Figure S14**. Percentage inhibition of the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical by ligand **6d** after 24 hours (24h) and 48 hours (48h).



**Figure S15**. Percentage inhibition of the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical by ligand **6c** after 1 hour (1h), 24 hours (24h), 48 hours (48h) and 72 hours (72h).

### 8. Trolox Equivalent Antioxidant Capacity (TEAC) Assay



**Figure S16**. Percentage of ABTS inhibition (TEAC) by ligand **6a** (ABTS = 2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)).



**Figure S17**. Percentage of ABTS inhibition (TEAC) by ligand **11a** (ABTS = 2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)).



**Figure S18**. Percentage of ABTS inhibition (TEAC) by ligand **10a** (ABTS = 2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)).



**Figure S19**. Percentage of ABTS inhibition (TEAC) by ligand **6d** (ABTS = 2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)).



**Figure S20**. Percentage of ABTS inhibition (TEAC) by ligand **6c** (ABTS = 2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)).



**Figure S21**. Percentage of ABTS inhibition (TEAC) by Trolox (ABTS = 2,2'-azinobis-(3ethylbenzothiazoline-6-sulfonic acid)).

# 9. Neuroprotection against 6-OHDA Neurotoxicity



Figure S22. Comparison of the percentage neuroprotection against 6-hydroxydopamine (6-OHDA) neurotoxicity in SH-SY5Y neuroblastoma cells by compounds **6a**, **6c**, **2**, **3** and DFP **1** (at 100  $\mu$ M compound dose).

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