Electronic Supplementary Material (ESI) for Metallomics.
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Supplementary information

Preparation of Aβ samples

Non-labeled and ^{15}N isotopically enriched Aβ(1-40) samples were purchased from EZBiolab and Alexotech, respectively. Both companies dispose the samples in vials containing 1 mg of lyophilized Aβ. The peptide was prepared as suggested by the suppliers and described previously, using an alkaline dissolution protocol[1], which reduces the formation of pre-aggregated species during the solvation of the peptide. The lyophilized peptide was dissolved in 400 μL of NaOH 10 mM, incubated during 30 min in ice, aliquoted, and stored at –80 °C. This NaOH Aβ peptide solution constituted the starting material for all subsequent structural experiments. Immediately before recording the NMR experiments, one or more Aβ aliquots were diluted in TRIS 20 mM. The pH was then adjusted to 7.4 and the samples were centrifuged at 20,000 *x g* to eliminate potentially pre-formed aggregates. All procedures were conducted at 4 °C. The peptide concentration was measured by UV spectroscopy using a molar extinction coefficient $\varepsilon_{280nm} = 1490\ M^{-1}\ cm^{-1}$.

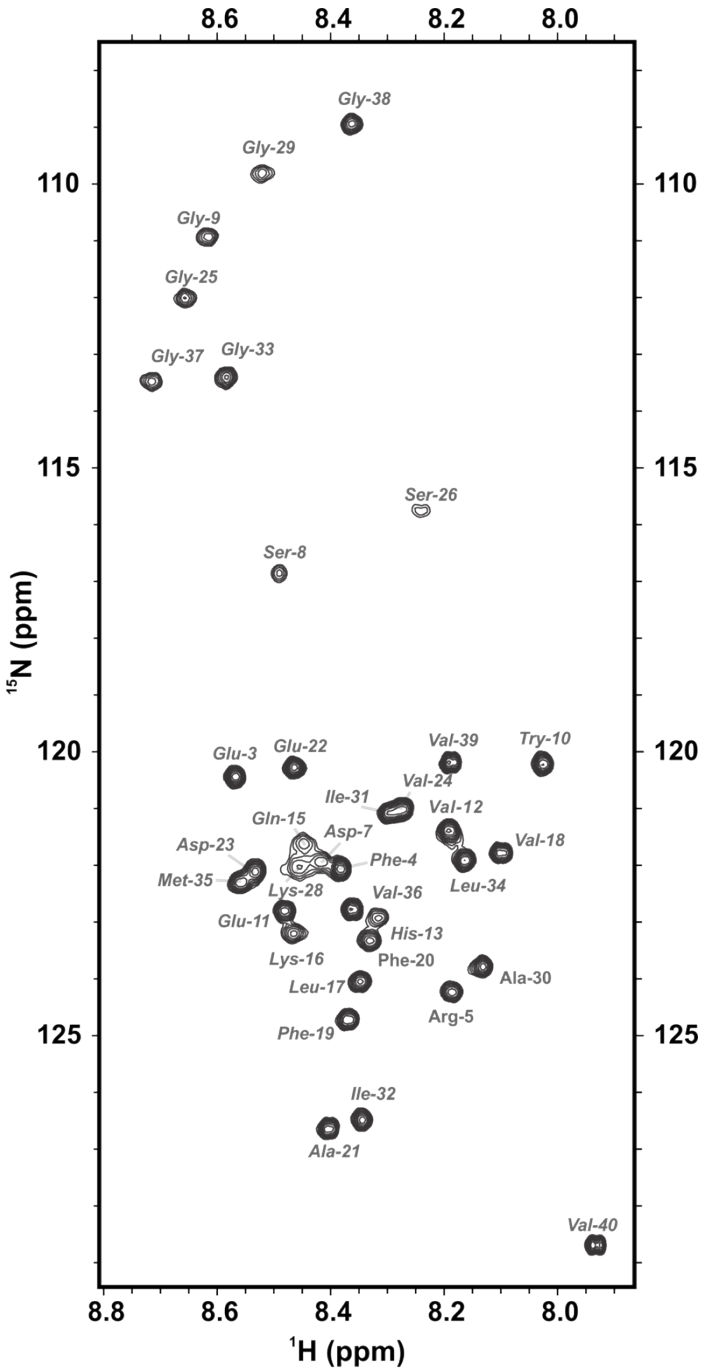

Figure S1. Assignment of backbone amide Aβ(1-40) peptide resonances. The ^{1}H–^{15}N HSQC spectrum was acquired at 5 ºC on 50 µM peptide samples dissolved in TRIS buffer 20 mM, pH 7.4. Resonance assignments were based on the literature[1, 2] and further confirmed by 2D ^{1}H–^{1}H TOCSY and 2D ^{1}H–^{1}H NOESY experiments following standard strategies[3].

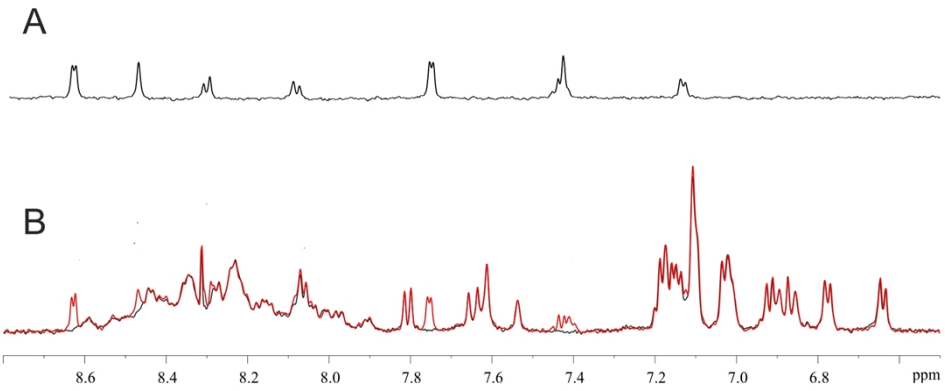

Figure S2. A. 1D ^{1}H-NMR spectrum of 100 µM INHHQ registered at 5 ºC on samples dissolved in buffer TRIS 20 mM, pH 7.4. **B.** 1D ^{1}H-NMR spectrum of 50 µM Aβ samples in the absence (black) and presence (red) of 100 µM INHHQ in TRIS 20 mM, pH 7.4, at 5 ºC. Addition of INHHQ did not affect Aβ(1-40) peptide amide and aromatic resonances.

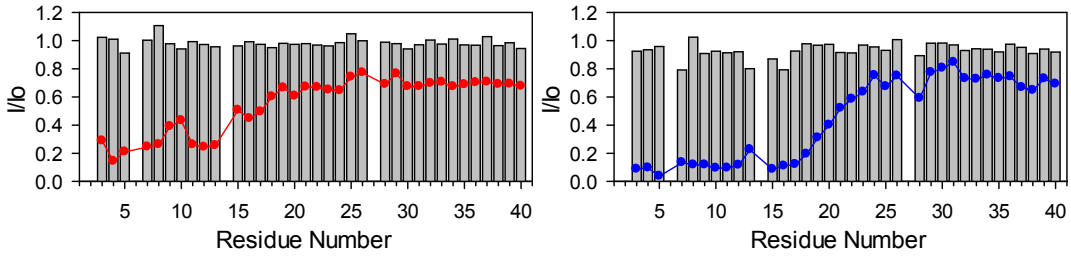

Figure S3. Effect of EDTA on the Aβ-Zn^{2+} (left) and Aβ-Cu^{2+} (right) interactions. I/Io intensity profiles of 50uM Aβ samples measured in the presence of 1 equivalent of Zn^{2+} (red circles) and Cu^{2+} (blue circles) ions. The grey bars correspond to data measured after the addition of 1 equivalent of EDTA to the solutions containing the Aβ metal-complexes.

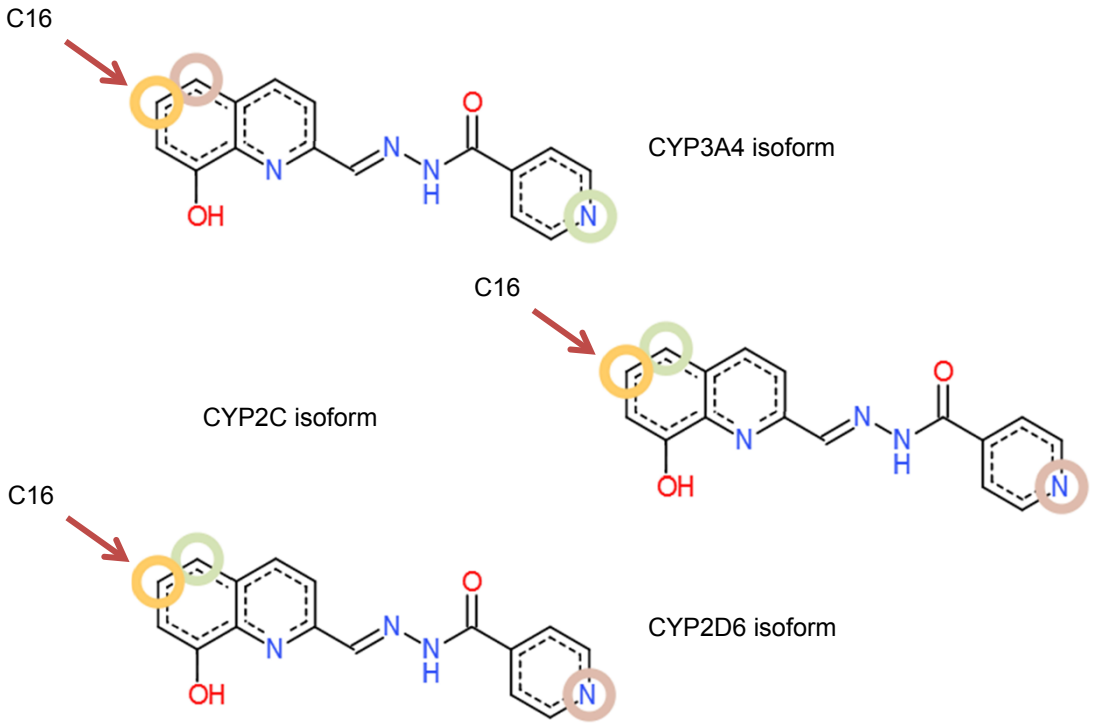

Figure S4. INHHQ atoms most susceptible to oxidation by different isoforms of the CYP superfamily of enzymes, circled, as calculated by the SMARTCyp V. 2.4.2 software package. Yellow circles show the sites with highest oxidizing probability, followed by salmon and, finally, green circles. C16 is the most reactive site for all the studied isoforms.

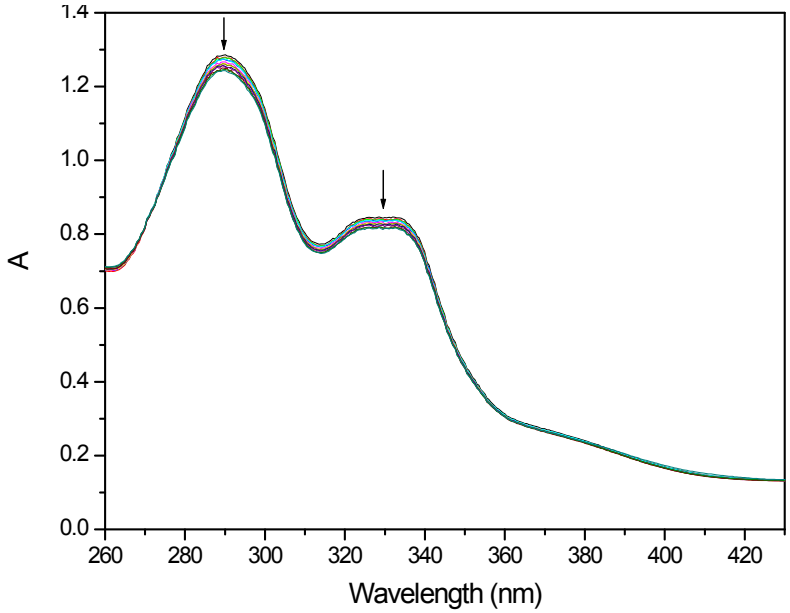

Figure S5. UV-Vis spectra of a 3.0 x10^{-7} M INNHQ solution in 10% DMSO/saline solution vehicle. Readings were taken every hour, for 12 h. The arrows stand for decreases in the intensity of the two absorption bands over time, related to the partial hydrolysis of INHHQ.

Additional references

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