Revista Facultad de Ciencias Básicas 2022 Vol. 18(2)
julio - diciembre ■ ISSN: 1900-4699 · e-ISSN: 2500-5316 ■ pp. 103-113

Editorial Neogranadina

DOI: https://doi.org/10.18359/rfcb.7200

Evaluation of the Xtb Semiempirical Method for the Prediction of Antioxidant Properties in Alzheimer's Disease: Salen-Type Ligands*

Sebastian Nieto-Alfonso[a] ■ Nicolás Puentes-Díaz[b] ■ Jorge Alí-Torres[c]

Abstract: Alzheimer's disease (AD) stands as the predominant form of dementia, accounting for up to 70% of all cases worldwide. AD is a complex disease with various contributing factors. Evidence suggests that the metallic complexes formed by the β-amyloid peptide (Aβ) and extraneuronal copper can catalyze the generation of reactive oxygen species, consequently increasing oxidative stress and contributing to the decline of neurons. This interaction underscores the significance of bioavailable copper as a crucial redox-active target in exploring protocols for multifunctional agents in AD treatment. In the field of computational chemistry, density functional theory (DFT) is widely accepted as a standard method across different disciplines. Despite this, DFT presents computational challenges, particularly in screening extensive molecular sets during the initial phases of drug research. Recent advances in semiempirical quantum mechanical methods (SQM) offer a promising alternative, providing rapid molecular geometry optimization and approximate estimation of thermodynamical properties, being at least two orders of magnitude faster than traditional DFT calculations. In this work, we present an evaluation of the GFNn-xTB SQM methods in the rapid screening of antioxidant properties in AD, performed on a set of salen ligands by calculating the standard reduction potentials of their copper complexes as key property. Results show that the implementation of GFNn-xTB SQM calculations before DFT evaluations is a useful technique to expedite the process and save computational time without sacrificing chemical accuracy.

Keywords: Virtual Screening; Semiempirical Quantum Mechanical Methods; Alzheimer's Disease; Copper Complexes; Standard Reduction Potentials

Recibido: 02/02/2024 **Aceptado:** 15/03/2024 **Disponible en línea:** 30/05/2024

Cómo citar: Nieto-Alfonso, S., Puentes Díaz, N., & Alí-Torres, J. (2024). Evaluation of the Xtb Semiempirical Method for the Prediction of Antioxidant Properties in Alzheimer's Disease: Salen-Type Ligands. *Revista Facultad de Ciencias Básicas, 18*(2), 103-113. https://doi.org/10.18359/rfcb.7200

* Research Article.

a Chemistry student. Universidad Nacional de Colombia, Bogotá, Colombia.
Email: snietoa@unal.edu.co; ORCID: https://orcid.org/0000-0002-7192-1195

b Master in Chemistry. Chemist. Universidad Nacional de Colombia, Bogotá, Colombia.
Email: ndpuentesd@unal.edu.co; ORCID: https://orcid.org/0000-0002-4638-229X.

c Ph.D. in Theoretical and Computational Chemistry. Master's in Theoretical and Computational Chemistry. Chemist. Universidad Nacional de Colombia, Bogotá, Colombia.
Email: jialit@unal.edu.co; ORCID: https://orcid.org/0000-0003-1354-8713

Evaluación del método semiempírico Xtb para la predicción de propiedades antioxidantes en la enfermedad de Alzheimer: ligandos tipo Salen

Resumen: La enfermedad de Alzheimer (EA) se presenta como la forma predominante de demencia, representando hasta el 70% de todos los casos a nivel mundial. La EA es una enfermedad compleja con diversos factores contribuyentes. La evidencia sugiere que los complejos metálicos formados por el péptido β-amiloide (Aβ) y el cobre extraneuronal pueden catalizar la generación de especies reactivas de oxígeno, aumentando consecuentemente el estrés oxidativo y contribuyendo al deterioro de las neuronas. Esta interacción subraya la importancia del cobre bioavailable como un objetivo redox activo crucial en la exploración de protocolos para agentes multifuncionales en el tratamiento de la EA. En el campo de la química computacional, la teoría del funcional de la densidad (TFD) es ampliamente aceptada como un método estándar en diferentes disciplinas. A pesar de esto, la TDF presenta desafíos computacionales, especialmente en el cribado de conjuntos moleculares extensos durante las fases iniciales de la investigación de fármacos. Los avances recientes en métodos cuánticos mecánicos semiempíricos (MCMSE) ofrecen una alternativa prometedora, proporcionando una optimización rápida de la geometría molecular y una estimación aproximada de las propiedades termodinámicas, siendo al menos dos órdenes de magnitud más rápido que los cálculos TFD tradicionales. En este trabajo, presentamos una evaluación de los métodos MCMSE GFNn-xTB en el cribado rápido de propiedades antioxidantes en la EA, realizado en un conjunto de ligandos salen mediante el cálculo de los potenciales de reducción estándar de sus complejos de cobre como propiedad clave. Los resultados muestran que la implementación de cálculos MCMSE GFNn-xTB antes de las evaluaciones TFD es una técnica útil para acelerar el proceso y ahorrar tiempo computacional sin sacrificar precisión química.

Palabras clave: cribado virtual; métodos cuánticos mecánicos semiempíricos; enfermedad de Alzheimer; complejos de cobre; potenciales de reducción estándar

Avaliação do método semiempírico Xtb para a previsão de propriedades antioxidantes na doença de Alzheimer: ligantes tipo Salen

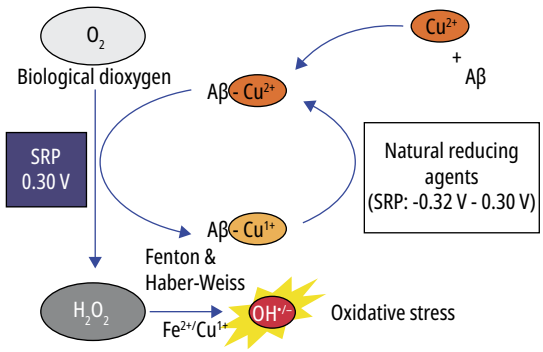
Resumo: A doença de Alzheimer (DA) se apresenta como a forma predominante de demência, representando até 70% de todos os casos a nível mundial. A DA é uma doença complexa com diversos fatores contribuintes. Evidências sugerem que os complexos metálicos formados pelo peptídeo β-amiloide (Aβ) e o cobre extraneuronal podem catalisar a geração de espécies reativas de oxigênio, aumentando consequentemente o estresse oxidativo e contribuindo para o deterioro dos neurônios. Esta interação destaca a importância do cobre biodisponível como um alvo redox ativo crucial na exploração de protocolos para agentes multifuncionais no tratamento da DA. No campo da química computacional, a teoria do funcional da densidade (DFT) é amplamente aceita como um método padrão em diferentes disciplinas. Apesar disso, a DFT apresenta desafios computacionais, especialmente na triagem de grandes conjuntos moleculares extensos durante as fases iniciais da pesquisa de medicamentos. Os avanços recentes em métodos quânticos mecânicos semiempíricos (SQM) oferecem uma alternativa promissora, proporcionando uma otimização rápida da geometria molecular e uma estimativa aproximada das propriedades termodinâmicas, sendo pelo menos duas ordens de magnitude mais rápido que os cálculos tradicionais de DFT. Neste trabalho, apresentamos uma avaliação dos métodos SQM GFNn-xTB na triagem rápida de propriedades antioxidantes na DA, realizada em um conjunto de ligantes Salen por meio do cálculo dos potenciais de redução padrão de seus complexos de cobre como propriedade chave. Os resultados mostram que a implementação de cálculos SQM GFNn-xTB antes das avaliações DFT é uma técnica útil para acelerar o processo e economizar tempo computacional sem sacrificar a precisão química.

Palavras-chave: triagem virtual; métodos quânticos mecânicos semiempíricos; Doença de Alzheimer; complexos de cobre; potenciais de redução padrão

Introduction

Alzheimer's disease (AD) is the most common form of dementia worldwide, accounting for up to 70% of all global cases. [1] AD presents both genetic and environmental risk factors and is considered a multifactorial disease, with the main leading cause yet to be clearly identified. One hypothesis regarding the disease involves oxidative damage mediated by redox-active metal ions and their interactions with biomolecules, such as the β-amyloid peptide (Aβ) commonly found in AD-affected brains [2]. Among these metallic ions, copper is recognized as a highly redox active metal whose complexes with Aβ can activate O_2 into reactive oxygen species through a catalytic cycle, as illustrated in Figure 1. This effect would increase oxidative stress in the brain and promote neuronal death [3].

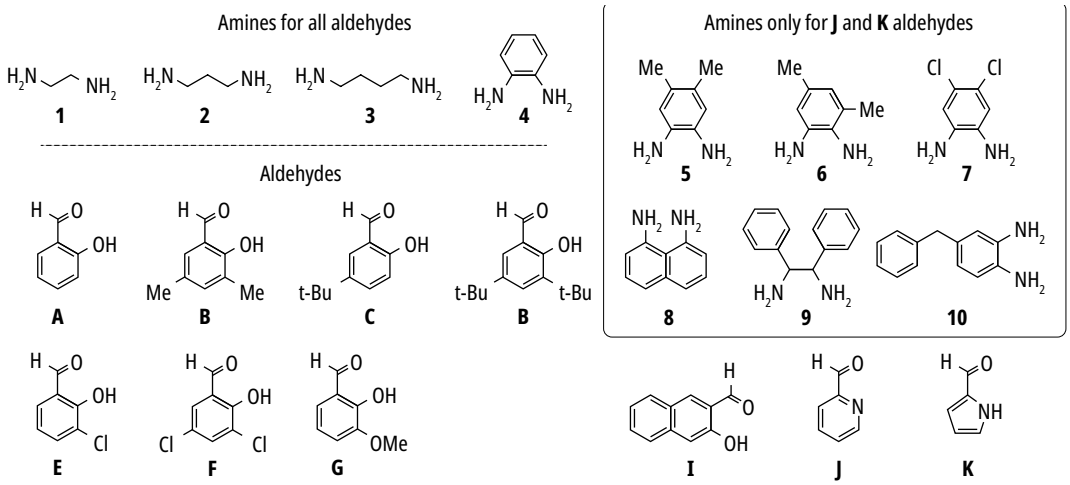
Figure 1. Schematic representation of oxidative stress mediated by copper cations and Aβ peptide. Adapted with permission from reference [2]



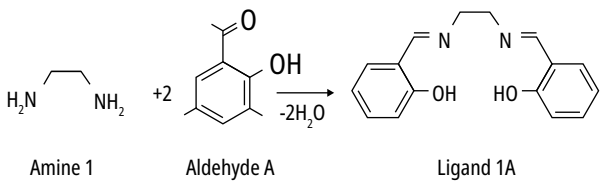
Strategies involving metal chelating constitute a significant area of study that has emerged as an option to prevent or reverse the metallic dysregulation and oxidative stress observed in AD [4], [5]. Many of these strategies are currently focused on the search for so-called multifunctional ligands, *i.e.*, molecules with more than one pharmacological target. Computational chemistry methods have become powerful tools for evaluating large sets of candidates while reducing time and costs [6]. In terms of quantum computational methods, Density Functional Theory (DFT) methodologies have become widely used in many fields due to their excellent balance between accuracy and computational cost compared to wave function-based methods [7]. However, DFT remains computationally intensive when applied to the screening of extensive molecular sets, a common procedure in the initial phases of drug research. Conversely, recent advancements in Semiempirical Quantum Mechanical methods (SQM) offer a compelling alternative, providing rapid molecular geometry optimization and approximate estimation of thermodynamic properties. SQM procedures are at least two orders of magnitude faster than conventional DFT methods [8].

In a previous study by Puentes-Díaz *et al.*, the potential pharmacological activity of a family of salen-type ligands in AD was investigated using DFT with the M06-2X functional. A method calibrated for an accurate description of the standard reduction potentials (SRP) of the salen-copper complexes was employed, with this value being the determining factor for classifying a ligand as an antioxidant candidate [3]. In this work, we assessed the performance of the SQM methods GFN1-XTB and GFN2-XTB, provided by Grimme *et al.* [9] in screening antioxidant candidates among a molecular set comprising 56 salen-type ligands previously evaluated by Puentes-Díaz *et al.* The precursors of these ligands are presented in Figure 2, and the nomenclature used throughout this work follows their respective precursor amine and aldehyde, outlined in Scheme 1. Accordingly, the ligands are formed by combining a diamine (numbered from **1** to **10**) with a single aldehyde (designated from **A** to **K**), generating a symmetrical tetradentate structure comprising a total of 36 2N2O-type ligands and 20 4N-type ligands.

Figure 2. 2D structure of the compounds used as precursors to construct the set of ligands under study. Adapted with permission from reference [3]



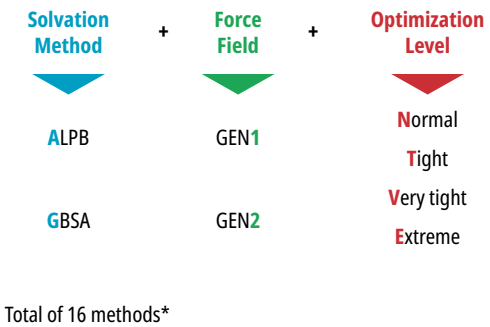
Scheme 1. Nomenclature used in this work for the set of salen-type ligands.



Computational methods

To assess the performance of the GFNn-xTB methods, equilibrium geometry and frequency calculations were conducted for all salen-copper complexes in both oxidation states of the metal, Cu^{+} and Cu^{2+}, using the standalone xTB version 6.6.1 code [8], [9], [10]. The GFN1-xTB and GFN2-xTB SQM methods available in the xTB code were utilized, with geometry convergence criteria set as *normal* (5 x 10^{-5} E_h), *tight* (1 x 10^{-6} E_h), *very tight* (1 x 10^{-7} E_h), and *extreme* (1 x 10^{-8} E_h). Additionally, the 0074wo solvation models included in the xTB code were considered: Analytical Linearized Poisson-Boltzmann (ALPB) and generalized Born surface area (GBSA) implicit solvation models [11]. Consequently, all possible combinations of these parameters - solvation method, force field, and geometry convergence criteria - were taken into account, resulting in a total of 16 methods. The nomenclature for these is illustrated in Figure 3.

Figure 3. Nomenclature code for the evaluated methods. These codes are derived from the combination of the respective letter/number assigned to the solvation method, force field, and optimization level.



The potential antioxidant behavior of the set of salen-type ligands was assessed by estimating the standard reduction potentials (SRP) using the thermochemical properties obtained from the frequency calculations for each Cu^{2+}-L/Cu^{+}-L pair with all methods. The optimized geometries obtained by each method were compared using

root-mean-square deviation (RMSD) [12] using the Kabsch algorithm [13] with the previous equilibrium geometries reported from DFT calculations [3]. For SRP calculation, two methodologies were employed: the direct method for **K** aldehyde derivatives, and the isodesmic method for all other ligands, following the procedure reported by Chaparro and Alí-Torres [14], utilizing phenylalanine as a reference pair for all 2N2O-type ligands and tris(pyridyl ethyl) amine for **J** aldehyde derivatives. A brief description of these methodologies is provided below.

- **Direct method.** This involves the reduction of the complex against the Standard Hydrogen Electrode (SHE) and is calculated according to the following equation:

$$E^{\circ}(Cu^{2+}/Cu^{+}) = -\frac{\Delta G^{Cu} - \Delta G^{SHE}}{F} + 0.158 \quad (1)$$

Where $E°(Cu^{2+}/Cu^{+})$ represents the SRP of the copper complex in volts, ΔG^{Cu} is the free energy change associated with the reduction of the Cu-complex (in kcal/mol), ΔG^{SHE} is the free energy change of the standard hydrogen electrode (99.9 kcal/mol [15]), and F is Faraday's constant (23.06 kcal/mol V).

The constant value of 0.158 V accounts for the error in the direct determination of the standard reduction potential (SRP) for the aqua-copper $[Cu(H_2O)_4]^{2+}/[Cu(H_2O)_3]^{+}$ couple [16], and is included as an empirical correction term.

- **Isodesmic method.** This method utilizes an external reference pair characterized by an experimental SRP value. The oxidation of the reference pair is coupled with the reduction of the complex of interest, as illustrated in Equation 2. It is crucial that this reference pair exhibits certain resemblances to the complex of interest. Specifically, it should share the same coordination sphere, possess an identical number of bonds in both oxidized and reduced species, and have a similar size. These similarities are essential for achieving effective error cancellations between the complexes. Ultimately, the potential value is determined through the application of Equation 3.

$$L_A Cu^{2+}_{(aq)} + L_B Cu^{+,ref}_{(aq)} \rightarrow L_A Cu^{+}_{(aq)} + L_B Cu^{2+,ref}_{(aq)} \quad (2)$$

$$E^{\circ} = E^{\circ}_{ref} + \Delta E^{\circ}_{rxn} \quad (3)$$

Where $L_ACu^{+/2+}$ represents the complex of interest with the obtained SRP value of $E°$, $L_BCu^{+/2+,}_{ref}$ denotes the reference pair with experimental SRP value of $E°_{ref}$, and $\Delta E°_{rxn}$ signifies the calculated change in potential for the reaction shown in Equation 2.

Chaparro and Alí-Torres demonstrated that the use of the isodesmic method significantly reduces errors associated with SRP calculations. In their study involving a set of 62 copper complexes, the direct method with empirical water correction resulted in errors of 0.39 V, whereas the application of the isodesmic method reduced the errors to 0.08 V placing the error margin below the experimental uncertainty associated with electrochemical methods for copper complexes [17].

Similar outcomes were observed by Orjuela *et al.* in their investigation of a series of iron complexes, further supporting the efficacy of the isodesmic method [18]. The SRP obtained from the direct and isodesmic method with the 16 methods evaluated were compared with the previous SRP reported from DFT calculations [3] through mean absolute error (MAE).

Results and discussion

Aqua-copper complex

To analyze the performance of XTB methods in modeling copper complexes, we considered the tetraaqua $[Cu(H_2O)_4]^{2+/+}$ complexes, a model representing the free copper state in biological conditions. The initial geometries of the coordination sphere for the optimization process were established in both planar and tetrahedral modes, as these are among the most common geometries exhibited by Cu^{2+} and Cu^{+} complexes, respectively [19].

The free energies of the optimized geometries derived from these two configurations were compared by calculating their difference as $\Delta G°_{diff} = G°_{planar} - °_{tetrahedral}$. The obtained free energy changes are illustrated in Figure 4. It can be observed that $\Delta G°_{diff}$ takes values near zero (-.19 ± 0.43 kcal/mol) for all methods, indicating that the starting

geometry did not significantly affect the stability of the optimized Cu^{+} complexes. The final geometries converged to tetrahedral configurations, accordingly to the expected outcome for $[Cu(H_2O)_4]^{+}$.

In the case of the Cu^{2+} complexes, the initial geometry significantly impacts both the free energies and final geometries. Initial planar geometries yielded optimized planar geometries by all GFN2-xTB related methods, while the GFN1-xTB methods generated tetrahedral optimized structures. Conversely, with tetrahedral starting geometries, only the GFN2-xTB methods coupled with the ALPB solvation model yielded planar configurations, while the remaining methods produced final tetrahedral geometries. This behavior is illustrated - in Figure 4, where the total free energy difference for the Cu^{2+} complexes vary considerable between -4.8 and 1.2 kcal/mol. These observations for the aqua-copper species suggest a greater difficulty and higher errors in the calculated thermodynamical properties for xTB methods in the modeling of open shell Cu^{2+} systems, highlighting the importance of beginning with an appropriate planar initial structure for Cu^{2+} geometry optimizations and frequency calculations.

Root- Mean- Square Deviation (RMSD)

The quality of geometry optimizations was evaluated by calculating the RMSD between the calculated values and the DFT reference data. Figure 5 presents the average RMSD for each complex across the 16 methods, indicating that lower RMSD values were achieved for Cu^{2+} complexes compared to Cu^{+} complexes in most cases. For Cu^{2+} complexes, RMSD values range from 0.03 Å (**4K** complex, G1N method) to 3.6 Å (**10J** complex, G1V method). In contrast, for Cu^{+} complexes, the minimum RMSD value was 0.15 Å (**1J** complex, G1V method), while the maximum reached 5.1 Å (**4C** complex, A2V method). These results demonstrate that all xTB methods performed better in estimating the planar geometry of Cu^{2+} complexes, compared to the more variable and higher RMSD values obtained for the tetrahedral geometry of Cu^{+} complexes. It is important to note that concerns associated with GFNn-xTB methods include the potential impact of approximate electrostatic interactions and/or the use of small Atomic Orbital (AO) basis sets [8], which may occasionally lead to qualitatively incorrect molecular geometries.

The methods with the lowest average RMSD and their corresponding standard deviations for the 2N2O and 4N complexes are presented in Table 1. It is noteworthy that the best methods for DFT geometry optimization utilize the GFN1-xTB SQM method. However, the RMSD values remain high, limiting the use of xTB-derived methods solely to rough estimations of which could be more beneficial for preoptimization steps within a DFT-oriented methodology.

Figure 4. Free energy differences between planar and tetrahedral initial geometries ($\Delta G°_{diff}$) for the $[Cu(H_2O)_4]^{2+/+}$ complexes

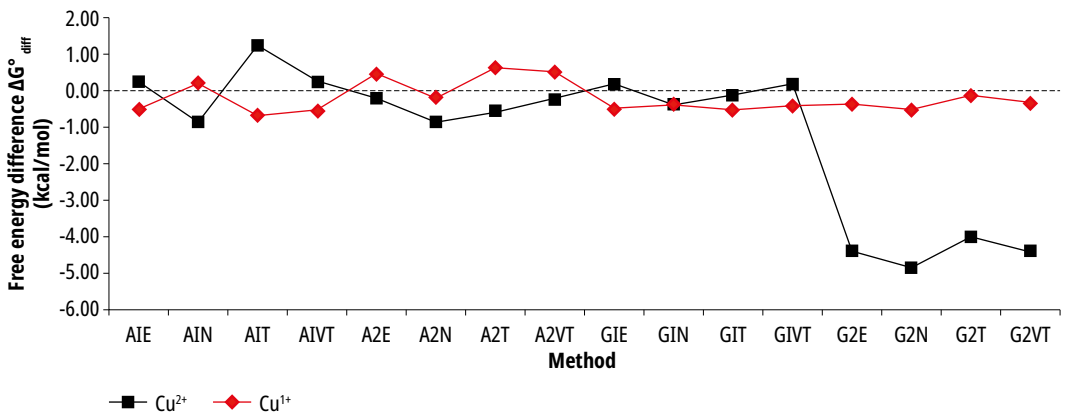


Figure 5. Average RMSD in angstroms (Å) of the salen-$Cu^{2+/+}$ complexes. The average was calculated using all RMSDs from the 16 methods evaluated for each complex

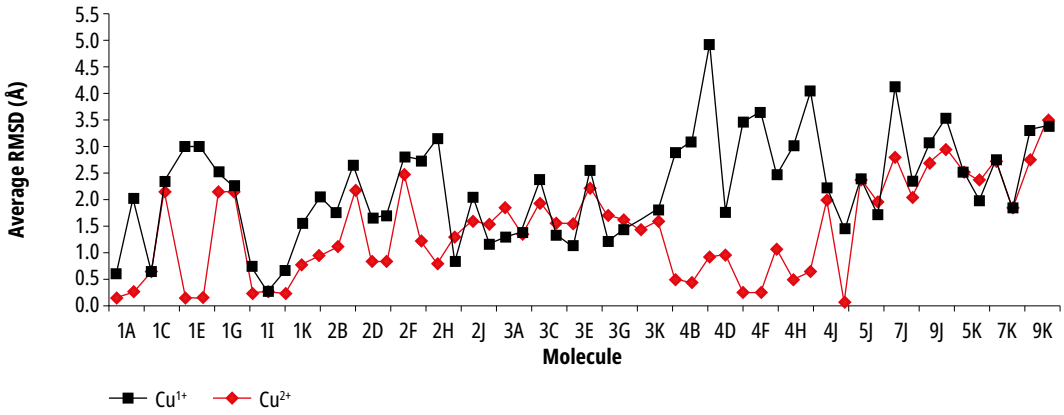


Table 1. Methods displaying the minimum average RMSD (Å) for all 2N2O and 4N complexes. Averages and deviations were computed individually for each method across the 36 2N2O and the 20 4N complexes.

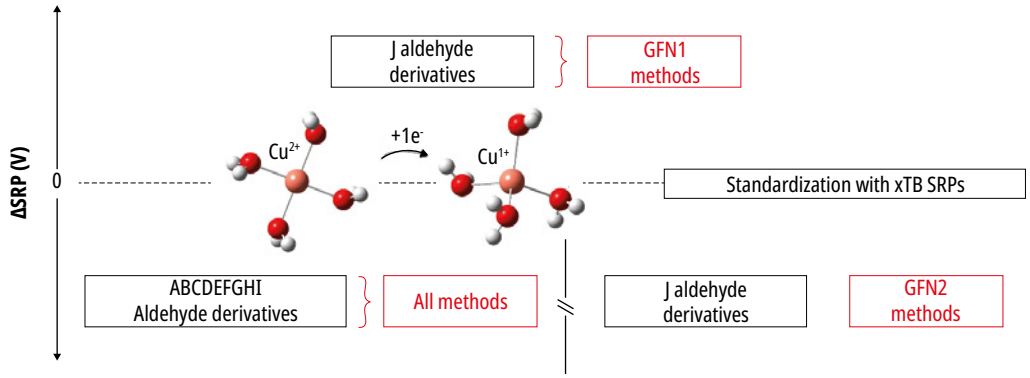
Cu^{2+}			
Coordination sphere	Method	Average RMSD (Å)	Standard deviation (Å)
2N2O	A1T	1.02	0.79
4N	G1T	1.96	0.95
Cu^{+}			
2N2O	G1T	2.09	1.10
4N	G1N	2.18	1.00

- **Prediction of antioxidant properties.** The primary objective of an antioxidant ligand in copper-associated oxidative stress is to mitigate the brain's deficit of natural antioxidants during the catalytic generation of reactive oxygen species, as depicted in Figure 1 [3]. To achieve this, their associated copper complexes must possess SRP values surpassing that of the O_2/H_2O_2 pair under biological conditions (0.30 V [20]).

As a rapid methods to screen the best antioxidant candidates from the ligand set, we propose standardizing all obtained salen-copper XTB-SRP values for all methods against their respective XTB-SRP values for the $[Cu(H_2O)_4]^{2+/+}$ calculated via direct method. Figure 6 illustrates the results, showing a consistent pattern across all **J** aldehyde derivatives with a positive ΔSRP when assessed using the GFN1-XTB methods. This aligns with the findings reported by Puentes-Díaz *et al*., indicating that **J** aldehyde derivatives were the most promising antioxidant candidates in this set, given their DFT-SRP values exceeding 0.30 V. In fact, recent experimental evidence attributes versatile antioxidant capability to the **2 J** ligand, demonstrating catalase and superoxide dismutase mimicking activity [21]. The suggested antioxidant capacity screening method provides a robust preliminary assessment, circumventing the conventional DFT method's 0.30 V limit. It reveals only two outliers in the trend (A1E and A1V methods for **9J** SRP standardization), displaying marginally negative ΔSRP value (-0.01 V). Detailed computed values for this section are provided in Table S2, accessible in the supporting information.

Analysis of the data reveals consistently positive ΔSRP values for the **J** aldehyde derivatives with GFN1-XTB methods. Notably, when utilizing the GBSA solvation model for GFN1-XTB methods, a significantly higher positive value was evident compared to the ALPB solvation model. This stands in contrast to the outcomes derived from the GFN2-XTB method, where the same complexes exhibited negative ΔSRP values. In general, the GFN2-XTB methods incorporating the GBSA solvation model displayed more pronouncedly negative ΔSRP values than those using the ALPB solvation model (refer to all standardized SRP values in the supporting information). As illustrated in Figure 6, ligands

Figure 6. Standardization of the xTB-SRP values of all salen-copper complexes with respect to the xTB-SRP value of the $[Cu(H_2O)_4]^{2+/1+}$ pair

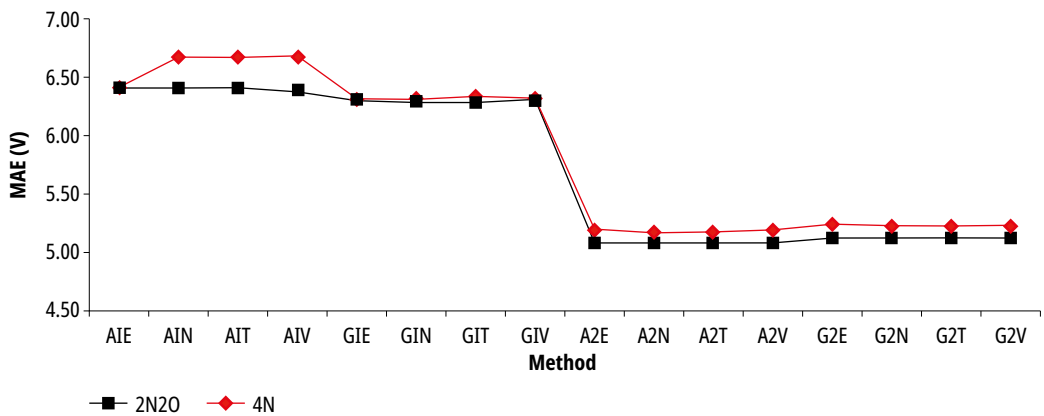


other than **J** aldehyde derivatives did not exhibit a positive ΔSRP. These findings are consistent with the results presented by Puentes-Díaz *et al.*, where these remaining ligands similarly displayed negative SRP values.

- **Standard reduction Potentials.** The mean absolute error (MAE) in xTB-SRP determination for the 2N2O-type and 4N-type complexes is depicted in Figure 7. The MAE values exhibit a consistent reduction of at least 1.16 V for the GFN2-xTB methods in comparison to the GFN1-xTB methods, with no significant difference between the ALPB and GBSA solvation models. This better performance of GFN2-xTB could be attributed to its anisotropic electrostatic and exchange-correlation terms, which are not included in the GFN1-xTB methods [22], [23]. Despite this, all xTB-SRP values calculated are still large compared to the DFT-SRP values previously reported in the reference[3].

In SQM methods, the error associated with the electronic structure component becomes notably influential for SRP calculations, as achieving precise modeling of redox processes relies on accurately depicting changes in the electronic structure of the target molecule in two oxidation states. The currently employed GFN*n*-xTB methods compromise a delicate balance between accuracy and computational cost, where the primary sources of error encompass self-interaction errors, the monopole description of electrostatic interactions (for GFN1-xTB), deficiencies in the atomic orbital (AO) basis set, and parametrization errors. Particularly, the tight binding methods tend to overestimate the delocalization of electrons, resulting in small orbital energy gaps [8]. In this sense, a reason for the

Figure 7. Mean (MAE) values in volts (V) for the xTB-SRP calculation of the 2N2O and 4N salen-copper complexes with all evaluated methods



large SRP values obtained could lie in the overestimation of the free energy of the Cu^{2+}-salen complexes, a problem that leads to higher free energy gaps between the oxidized and reduced species and consequently increases their calculated SRP. This hypothesis is supported by the difficulties discussed previously in the modeling of the oxidized aquo-copper complex ($[Cu(H_2O)_4]^{2+}$).

Additionally, even though the performance of GBSA and ALPB solvation methods has been proven to be sufficient for many practical purposes [11], [24], [25], the solvation treatment can play an important role in accurate calculating SRP values. Pantazis *et al*. identified the solvation contribution as the foremost source of error and a pivotal factor constraining the precision of redox potential calculations in organic aqueous systems[26]. According to Neugebauer *et al*., more elaborate solvation models such as COSMO-RS describe significant solvation effects better than the GBSA or the COSMO models used in SQM methods [22]. Nevertheless, the adoption of alternative solvation models such as COSMO-RS is not fully matured for SQM and proves to be more demanding compared to GBSA [22] or ALPB solvation models, thereby limiting their utility in screening studies.

To rectify these errors in SRP determination, we conducted a standardization process for the obtained XTB-SRP values, as illustrated in Figure 8. The reference complexes chosen to establish SRP = 0 were those with the least substituted structure.

The MAE analysis was conducted with the standardization for both XTB-SRP and DFT-SRP values. [3] The results are shown in Figure 9. Noticeably, the MAE of the standardized XTB-SRP values with respect to the standardized DFT-SRP ones is considerably lower than the raw SRP values presented in Figure 7. This occurs due to a proper cancellation of the XTB derived electronic and/or solvation errors, as the standardization process involves a comparison between similar pairs of oxidized and reduced species. Thus, it is critical to standardize only using similar structures to retrieve good results. This process highlights XTB methods as effective tools in the screening of changes in SRP values resulting from small substitutions in the scaffold of complexes with known SRP values

Figure 8. Standardization scheme for the obtained xTB-SRP and DFT-SRP values by referencing them to the SRP values of the reference complexes for each subset of salen-copper complexes

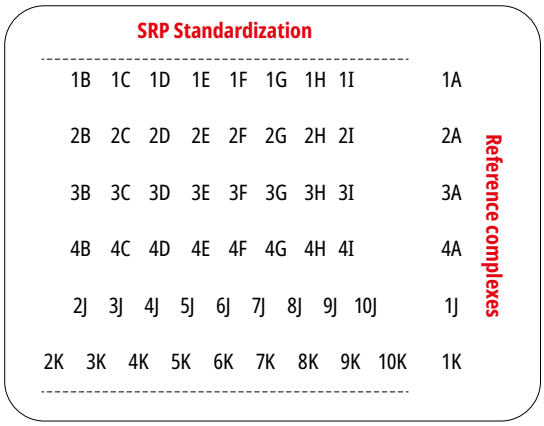
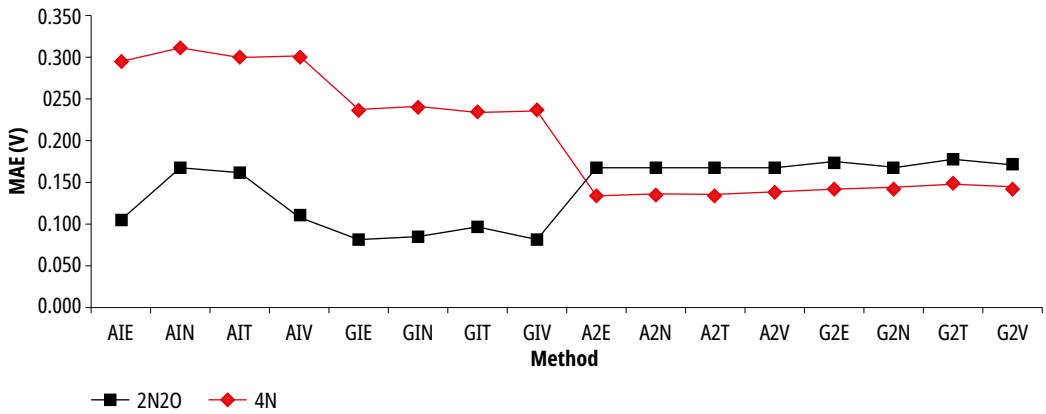


Figure 9. MAE values obtained in volts (V) for the SRP standardization of the 2N2O and 4N salen-copper complexes by all evaluated methods



(via experimentation or higher theory calculation methods).

In this context, the G1V method demonstrated superior performance for 2N2O complexes, achieving a minimal average error (MAE) of 0.08 V. Conversely, A2T emerged as the most suitable for 4N complexes, displaying a MAE of 0.13 V. As depicted in Figure 9, there is no notable distinction among the various GFN2-XTB methods across coordination types. In contrast, a substantial difference is observed with the GFN1-XTB methods, where higher MAE values were obtained for 4N complexes. These latter methods exhibited slightly lower efficiency, particularly when incorporating the ALPB solvation model.

Conclusions

GFNn-XTB SQM methods consistent produced tetrahedral final geometry in aqua-Cu^{+} complexes. GFN2-XTB with ALPB solvation accurately depicted planar geometry in aqua-Cu^{2+} complexes, regardless of the initial geometry. Despite the variability, XTB methods, with their cost-effectiveness and rapid computational turnaround, justified their use as an initial step for geometry optimization before transitioning to DFT. Specific complexes showcased remarkable RMSD values below 0.5 Å, contributing significantly to computational efficiency.

Regarding the screening of plausible antioxidant candidates in AD, **J** ldehyde derived complexes showed positive ΔSRP with GFN1-XTB methods respect to the SRP of the aquo-copper pair: $[Cu(H_2O)_4]^{2+/+}$, consistently indicating them as antioxidant candidates. We recommend using this aquo-copper SRP standardization screening employing all methods to verify consistent positive and negative behavior for the candidates.

The evaluation of the 56 salen-copper complexes using GFNn-XTB revealed elevated XTB-SRP values. Although GFN2-XTB exhibited improved results, it still deviated from reference SRP values. In this regard, the conducted XTB-SRP standardization, using complexes with similar structure as SRP reference, highlights GFNn-XTB methods as an effective pre-screening protocol for predicting ΔSRP values in salen-copper complexes. This could be particularly useful in calculating ΔSRP due to substitutions or other modifications upon a reference scaffold with known SRP value.

Acknowledgements

The authors express gratitude to Estefania Izquierdo-Bermeo for her assistance in designing Figure 6 and 8, as well as critically reviewing the manuscript.

References

[1] World Health Organization, "Dementia." Accessed: Jan. 24, 2024. [Online]. Available: https://www.who.int/es/news-room/fact-sheets/detail/dementia

[2] N. Puentes-Díaz, D. Chaparro, D. Morales-Morales, A. Flores-Gaspar, and J. Alí-Torres, "Role of Metal Cations of Copper, Iron, and Aluminum and Multifunctional Ligands in Alzheimer's Disease: Experimental and Computational Insights," *ACS Omega*, Jan. 2023, doi: 10.1021/acsomega.2c06939.

[3] N. Puentes-Díaz, D. Chaparro, V. Reyes-Marquez, D. Morales-Morales, A. Flores-Gaspar, and J. Alí-Torres, "Computational Evaluation Of The Potential Pharmacological Activity Of Salen-Type Ligands In Alzheimer's Disease." Doi: 10.3233/JAD-230542

[4] S. Lee *et al.*, "Rational design of a structural framework with potential use to develop chemical reagents that target and modulate multiple facets of Alzheimer's disease," *J Am Chem Soc*, vol. 136, no. 1, pp. 299–310, Jan. 2014, doi: 10.1021/ja409801p.

[5] K. Pradhan *et al.*, "Rhodamine-Based Metal Chelator: A Potent Inhibitor of Metal-Catalyzed Amyloid Toxicity," *ACS Omega*, vol. 5, no. 30, pp. 18958–18967, Jul. 2020, doi: 10.1021/acsomega.0c02235.

[6] W. Zhang, J. Pei, and L. Lai, "Computational Multitarget Drug Design," *J Chem Inf Model*, vol. 57, no. 3, pp. 403–412, Feb. 2017, doi: 10.1021/acs.jcim.6b00491.

[7] T. Van Mourik, M. Bühl, and M. P. Gaigeot, "Density functional theory across chemistry, physics and biology," *Philos Trans A Math Phys Eng Sci*, vol. 372, no. 2011, Mar. 2014, doi: 10.1098/RSTA.2012.0488.

[8] S. Grimme, C. Bannwarth, and P. Shushkov, "A Robust and Accurate Tight-Binding Quantum Chemical Method for Structures, Vibrational Frequencies, and Noncovalent Interactions of Large Molecular Systems Parametrized for All spd-Block Elements (Z = 1-86)," *J Chem Theory Comput*, vol. 13, no. 5, pp. 1989–2009, May 2017, doi: 10.1021/acs.jctc.7b00118.

[9] C. Bannwarth *et al.*, "Extended tight-binding quantum chemistry methods," *Wiley Interdisciplinary Reviews: Computational Molecular Science*, vol. 11, no. 2. Blackwell Publishing Inc., Mar. 01, 2021. doi: 10.1002/wcms.1493.

[10] "Semiempirical Extended Tight-Binding Program Package xtb," https://github.com/grimme-lab/xtb .

[11] S. Ehlert, M. Stahn, S. Spicher, and S. Grimme, "A robust and efficient implicit solvation model for fast semiempirical methods."

[12] "Calculate Root-mean-square deviation (RMSD) of Two Molecules Using Rotation," GitHub, http://github.com/charnley/rmsd, Version 1.5.1.

[13] W. Kabsch, "A solution for the best rotation to relate two sets of vectors," *Acta Crystallographica Section A*, vol. 32, no. 5, pp. 922–923, Sep. 1976, doi: 10.1107/S0567739476001873.

[14] D. Chaparro and J. Alí-Torres, "Assessment of the isodesmic method in the calculation of standard reduction potential of copper complexes," *J Mol Model*, vol. 23, no. 10, Oct. 2017, doi: 10.1007/S00894-017-3469-7.

[15] D. D. Wagman *et al.*, "Erratum: The NBS tables of chemical thermodynamic properties. Selected values for inorganic and C1 and C2 organic substances in SI units [J. Phys. Chem. Ref. Data 11, Suppl. 2 (1982)]," *J Phys Chem Ref Data*, vol. 18, no. 4, pp. 1807–1812, Oct. 1989, doi: 10.1063/1.555845.

[16] J. Alí-Torres, A. Mirats, J. D. Maréchal, L. Rodríguez-Santiago, and M. Sodupe, "3D structures and redox potentials of Cu2+-Aβ(1-16) complexes at different pH: A computational study," *Journal of Physical Chemistry B*, vol. 118, no. 18, pp. 4840–4850, May 2014, doi: 10.1021/JP5019718/SUPPL_FILE/JP5019718_SI_001.PDF.

[17] J. Alí-Torres, A. Mirats, J. D. Maréchal, L. Rodríguez-Santiago, and M. Sodupe, "Modeling Cu2+-Aβ complexes from computational approaches," *AIP Adv*, vol. 5, no. 9, p. 92402, Sep. 2015, doi: 10.1063/1.4921072/895713.

[18] A. L. Orjuela, F. Núñez-Zarur, and J. Alí-Torres, "A computational protocol for the calculation of the standard reduction potential of iron complexes: application to Fe2+/3+-Aβ model systems relevant to Alzheimer's disease," *RSC Adv*, vol. 12, no. 37, pp. 24077–24087, Aug. 2022, doi: 10.1039/D2RA03907A.

[19] R. R. Conry, "Copper: Inorganic & Coordination Chemistry Based in part on the article Copper: Inorganic & Coordination Chemistry by Rebecca R. Conry & Kenneth D. Karlin which appeared in the Encyclopedia of Inorganic Chemistry, First Edition.," *Encyclopedia of Inorganic Chemistry*, Sep. 2005, doi: 10.1002/0470862106.IA052.

[20] David L. Nelson. and Michael M. Cox, *Lehninger - Principles of Biochemistry*, Sexta Edición. New York: W.H. Freeman And Company, 2013. [Online]. Available: www.courses.bfwpub.com/lehninger6e.

[21] M. Richezzi *et al.*, "Versatile Activity of a Copper(II) Complex Bearing a N4-Tetradentate Schiff Base Ligand with Reduced Oxygen Species," *Eur J Inorg Chem*, vol. 2022, no. 8, p. e202101042, Mar. 2022, doi: 10.1002/EJIC.202101042.

[22] H. Neugebauer, F. Bohle, M. Bursch, A. Hansen, and S. Grimme, "Benchmark Study of Electrochemical Redox Potentials Calculated with Semiempirical and DFT Methods," *J Phys Chem A*, vol. 124, no. 35, pp. 7166–7176, Aug. 2020, doi: 10.1021/acs.jpca.0c05052.

[23] C. Bannwarth, S. Ehlert, and S. Grimme, "GFN2-XTB - An Accurate and Broadly Parametrized Self-Consistent Tight-Binding Quantum Chemical Method with Multipole Electrostatics and Density-Dependent Dispersion Contributions," *J Chem Theory Comput*, vol. 15, no. 3, pp. 1652–1671, Mar. 2019, doi: 10.1021/acs.jctc.8b01176.

[24] A. V. Onufriev and D. A. Case, "Generalized Born Implicit Solvent Models for Biomolecules," *Annu Rev Biophys*, vol. 48, pp. 275–296, May 2019, doi: 10.1146/ANNUREV-BIOPHYS-052118-115325.

[25] J. Gorges, S. Grimme, and A. Hansen, "Reliable prediction of association (free) energies of supramolecular complexes with heavy main group elements – the HS13L benchmark set," *Physical Chemistry Chemical Physics*, vol. 24, no. 47, pp. 28831–28843, Dec. 2022, doi: 10.1039/D2CP04049B.

[26] M. Isegawa, F. Neese, and D. A. Pantazis, "Ionization Energies and Aqueous Redox Potentials of Organic Molecules: Comparison of DFT, Correlated ab Initio Theory and Pair Natural Orbital Approaches," *J Chem Theory Comput*, vol. 12, no. 5, pp. 2272–2284, Apr. 2016, doi: 10.1021/acs.jctc.6b00252.