BIOSENSOR: CONSTRUCTION AND APPLICATION STUDY IN THE RECOGNITION OF CHEMICAL WARFARE AGENTS AND IONIZING RADIATIONS

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ABSTRACT

Given the prospect of increasing innovation and technological input, it is necessary to take a special look at issues of instrumentation and measurements for the chemical and nuclear sectors. In this context, the idea of developing research that combines knowledge from different areas, such as nuclear physics, chemistry, physical chemistry, biochemistry, biotechnology, electronics, enables the construction of new biosensors capable of detecting chemical warfare agents (QWA) and producing proportional electrical signals, and from the interaction with ionizing radiation produce radiological and nuclear effects (ERN) to be measured. In this work, *in silico* studies were carried out for the development of enzymatic biosensors, which will later be built and tested *in vitro* and *in loco* on environmental samples. The concepts for the development of the biosensor are initially composed of the theoretical study of biochemical systems aimed at this application, in parallel with modeling and computational simulations identifying the boundaries of the systems involved, resulting in the practical action and uniqueness of the tool.

Keywords: Biotechnology. Biosensor. Organophosphate. Nuclear. Ionizing-Radiation.

1 INTRODUCTION

The development of national biosensors can contribute to increasing the country's independence in terms of importing defense products for detecting chemical warfare agents and ionizing radiation. In this new technological condition, in a real scenario of attacks or incidents, efficient detection can be carried out in a timely manner, guaranteeing national sovereignty in the face of the issue addressed. In this work, molecular modeling techniques in biological systems were applied to verify the interactions between chemical warfare agents (QGA) and ionizing radiation (ERN) with the enzymes acetylcholinesterase and butyrylcholineterease. These enzymes are the main targets of these agents in the human and animal organism. These biochemical interactions will be transduced into electrical signals or information, in order to promote the detection of these compounds in a timely manner and assist emergency response teams and healthcare teams in containing possible damage to people, animals, materials and the environment.

2 MATERIAL & METHODS

The present work is divided into three main stages: (a) the first stage, already underway, consists of molecular docking studies between the enzymes acetylcholinesterase and butyrylcholinesterase with the main AGQ and two Aflatoxins (B1 and M1). These agents are organophosphate compounds and aflatoxins, capable of inhibiting the molecular targets in question and causing cholinergic syndrome in humans and animals. To this end, two receptor models were built and validated based on crystallographic structures deposited in the *Protein Data Bank* database (Computational Structure Models (CSM))[1]. The three-dimensional structure of the organophosphates and toxins were constructed and the partial atomic charges were calculated using the *Spartan'08* program (Version 08 ®, Wavefunction, Irvine, CA, USA, collaboration with Q-Chem, 2009)[2]. Figures 01, 02 and 03 represent the flat structural formulas of the organophosphates Ciclosarin and Tabun built in Spartan'08. Then, docking studies were carried out using the *Molegro Virtual Docker* (MVD) ® program (version 6.0, CLC bio, Aarhus, Denmark, 2013)[3], with the results being evaluated according to the interaction energy and interaction residues with both enzymes. This modeling stage was carried out in the Laboratory of Molecular Modeling applied to Chemical and Biological Defense (LMDQB) of the Chemical Engineering Section of IME; - (b) second stage, also in progress, was carried out for the purpose of detecting ionizing radiation. Computational simulations were performed with the MCNPX code (general purpose Monte Carlo radiation transport code, continuous energy, generalized geometry, time dependent) (MCNPX EXTENSIONS VERSION 2.6.0, Los Alamos National Laboratory, 2008)[4]. The code generated in MCNPX had its geometry evaluated in the Vised software (which is an extension of the MCNPX code). This simulation stage was carried out at the Nuclear Simulation Laboratory (LSN) of the IME Nuclear Engineering Section; (c) the third stage will be the construction of the biosensor, using the previous results as a starting point.

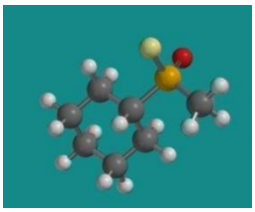

Fig. 01: Cyclosarin

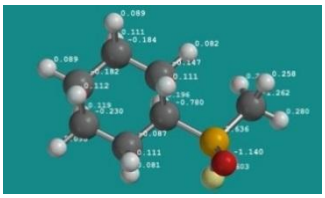

Fig. 02: Cyclosarin with charges

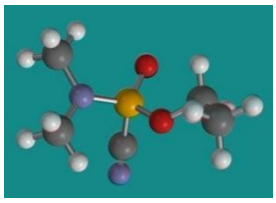

Fig. 03: Tabun

3 RESULTS & DISCUSSION

In the present study, 4680 iterations of 11 organophosphates and 2 aflatoxins were performed with acetylcholinesterase. The best docking results were for the organophosphates VR and Novichok-A232 and for the toxin Aflatoxin M1. These results are presented in figures 04, 05 and 06.

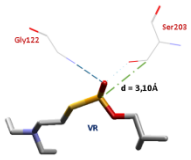

Fig. 04: VR

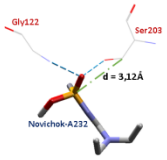

Fig. 05: Novichok-A232

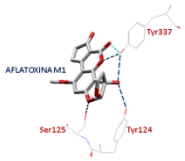

Fig. 06: Aflatoxin M1

It is possible to verify that there is agreement between the results obtained through molecular docking simulations with AGQ and toxins with the simulations via MCNPX. The observed agreement results in information that characterizes a representative model and increases the possibility of building efficient equipment by concentrating due focus on the schematic representation. In this condition, it is plausible to verify a certain trend among the resulting data, which covers the dispersion of energy around a specific target, the absorbed energy, the energy resulting from the Compton effect, scattering and other related properties. Table 01 presents the interaction energy results between the compounds studied with the enzyme acetylcholinesterase (AchE) and with the main hydrogen bond residues. The distance between phosphorus (P_i) and serine-203 (Ser-203).

Table 01 Results of Dockings for VR, Novichok-A232 and Aflatoxin M1.

Molecule	SCORE [kcal/mol] (AChE)	Distance [Å] (P_i – Ser203)	Residue (hydrogen bonds)
VR	-100,21	3,10	Gly - 122 / Ser - 203
Novichok-A232	-78,43	3,12	Gly - 122 / Ser - 203
Aflatoxin M1	-139,05	-	Ser - 125 / Tyr - 124 / Tyr - 337

It is noted that organophosphates and aflatoxin M1, when approaching the target molecule, are capable of generating an energy fluctuation presented in [kcal/mol]. This energetic fluctuation is sufficient to be quantified and with the appropriate mathematical correlations[5] (Equation 01) to be expressed as a short-term increase in the system's electrical current of the order of $1\,[Cal] \sim 2{,}611x10^{19}[eV]$. This correlation obtains the same value found in the literature, validating the mathematical modeling.

$$\frac{4{,}18\,[J]}{1\,[Cal]} * \frac{1\,[C]}{34\,[J]} * \frac{1\,[ion]}{1{,}6 \cdot 10^{-19}\,[C]} * \frac{34\,[eV]}{1\,[ion]} \quad (1)$$

The graph in figure 07 displays information on the number of particles (gamma) generated and the number of particles that escape (that do not collide with the target - ERN).

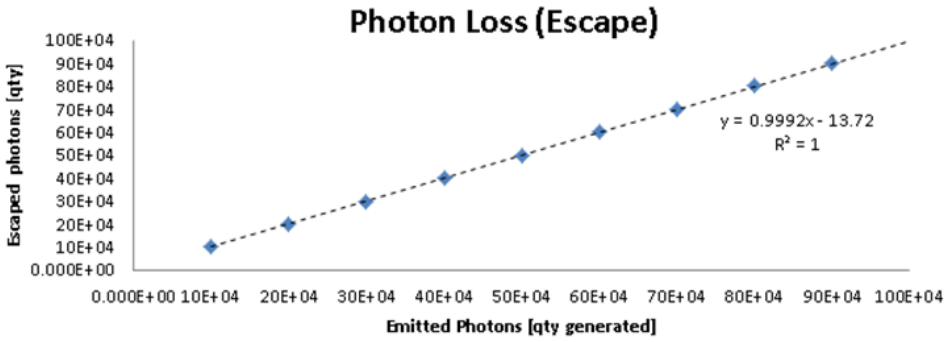

Fig. 07: Photon Tracking Chart

The graph in Fig 07 shows the results obtained in simulations with the MCNPX code, revealing a linear distribution of data, highlighting the low probability of interaction between photons and the biosensor under development. This linearity suggests that the rate of photons detected by the biosensor is proportional to the intensity of the incident radiation, and the low probability of interaction indicates that the majority of photons generated are not absorbed by the control volume. Considering that photons that do not interact with the biosensor are not absorbed, it is plausible to infer that they remain in the environment and may be subject to other effects of ionizing radiation[6]. This finding raises questions about the sensitivity and specificity of the biosensor, in addition to the implications of the presence of undetected photons. Understanding these interaction mechanisms and the influence of other factors, such as biosensor geometry and material composition, are crucial to improving device performance. Carrying out experimental tests in real conditions of radiation exposure will make it possible to validate the simulations and deepen research into the effects of ionizing radiation, contributing to the development of more efficient and safe biosensors.

CONCLUSION

The data collected and the observations carried out lead to the formalization and materialization of the construction of the biosensor, since the subtle variation in the electrical potential can be measured through the correlation between [Cal] and [eV]. In this way, conclusions and inferences can be drawn, generating relevant impacts, both due to the innovative potential and the resource to be used in the field against systems that use radionuclides for other studies. The partial results indicate that the proposed equipment has the potential to perform detection quickly, accurately and at a lower cost due to high sensitivity, contributing to the development of national technology.

REFERENCES

[1] RCSB PROTEIN DATA BANK (RCSB - PDB). Computed Structure Models (CSM). *ELETROPHORUS ELETRICUS ACETYLCHOLINESTERASE.* Link for acess: < https://www.rcsb.org/>. Accessed on 20, Nov, 2023.
[2] PC *Spartan'08* for windows, macintosh and linux. Molecular modeling for desktop. Wavefunction, Inc., Japan Branch Office. Spartan'08 is a collaboration with Q-Chem, Inc. 2006 - 2009.
[3] Thomsen, R.; Christensen, M.H. MolDock: A New Technique for High-Accuracy Molecular Docking. *J. Med. Chem.* 2006, *49*, 3315–3321.
[4] John S. Hendricks et. al., *MCNPX EXTENSIONS VERSION 2.6.0*, Los Alamos National Laboratory, LA-UR-08-2216 (2008).
[5] Material obtido a partir dos apontamentos do prof. Luiz Ferraz Netto. *Sinais Elétricos: Formas de Ondas.* Acesso virtual. Disponível em: <https://www.facom.ufu.br/~jamil/eletronica/sinais_eletricos.htm>. Acesso em: 15 de jan. de 2024.
[6] TAUHATA, L. *et al*. *RADIOPROTEÇÃO E DOSIMETRIA: FUNDAMENTOS*. 10ª revisão. Rio de Janeiro-Brasil (2014).

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