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In silico study of the potential interactions of 4'-acetamidochalcones with protein targets in SARS-CoV-2

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ABSTRACT

The sanitary emergency generated by the pandemic COVID-19, instigates the search for scientific strategies to mitigate the damage caused by the disease to different sectors of society. The disease caused by the coronavirus, SARS-CoV-2, reached 216 countries/territories, where about 20 million people were reported with the infection. Of these, more than 740,000 died. In view of the situation, strategies involving the development of new antiviral molecules are extremely important. The present work evaluated, through molecular docking assays, the interactions of 4'-acetamidochalcones with enzymatic and structural targets of SARS-CoV-2 and with the host’s ACE2, which is recognized by the virus, facilitating its entry into cells. Therefore, it was observed that, regarding the interactions of chalcones with Main protease (Mpro), the chalcone N-[(2E)-3-(4-fluorophenyl)-1-(phenyl)prop-2-en-1-one] acetamide (PAAPF) has the potential for coupling in the same region as the natural inhibitor FJC through strong hydrogen bonding. The formation of two strong hydrogen bonds between N-[(2E)-3-(phenyl)-1-(phenyl)prop-2-en-1-one] acetamide (PAAB) and the NSP16-NSP10 heterodimer methyltransferase was also noted. N-[(2E)-3-(4-methoxyphenyl)-1-(phenyl)prop-2-en-1-one] acetamide (PAAPM) and N-[(2E)-3-(4-ethoxyphenyl)-1-(phenyl)prop-2-en-1-one] acetamide (PAAPE) chalcones showed at least one strong intensity interaction of the SPIKE protein. N-[(2E)-3-(4-dimetilaminophenyl)-1-(phenyl)prop-2-en-1-one] acetamide (PAAPA) chalcone had a better affinity with ACE2, with strong hydrogen interactions. Together, our results suggest that 4'-acetamidochalcones inhibit the interaction of the virus with host cells through binding to ACE2 or SPIKE protein, probably generating a steric impediment. In addition, chalcones have an affinity for important enzymes in post-translational processes, interfering with viral replication.

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1. Introduction

The health problem caused by the COVID-19 pandemic can be measured by the numbers of cases and deaths confirmed by the disease globally. In Brazil, community transmission is observed,
with the collapse of the Health System in some regions. As it is an infection triggered by a new coronavirus (SARS-CoV-2), the pathophysiology of COVID-19 is little known and there is no specific treatment for the disease [1,2].

The search for new candidates for antiviral drugs has made great progress in recent years with the discovery of molecular targets, the development of organic synthesis and the discovery of new bioactive substances. A big number of techniques have been used in the search for new antiviral drugs. Despite the great progress, the arsenal of antiviral drugs is still small [1]. In this sense, strategies involving the development and validation of new antiviral molecules have been considered.

Chalcones, known as \( \alpha \), \( \beta \)-unsaturated ketones (1,3-diaryl-2-propene-1-one) are a class of naturally occurring compounds belonging to the flavonoid family. They can be obtained from natural sources or by synthesis, and are widely distributed in fruits, vegetables, and tea [3]. The double connection together with carbonyl group are possibly responsible for diverse biological activities such as antibacterial, antioxidant, anti-inflammatory and antiparasitic [4].

Antiviral properties of chalcones have been recorded in studies with plant viruses and human rhinoviruses [5]. Antiviral studies [6] with chalcones containing hydroxy and methoxy groups, confirm that the activity is dependent on the nature of the group and its positions in the aromatic rings. Santos [7] reports in a recent study the evaluation of the antiviral activity of hydroxychalcones and synthetic curcuminoids against infection caused by HPV in vitro.

Therefore, in this work, for the first time acetamide chalcones will be study theoretically by the Molecular Docking to characterize the inhibition power of the chalcones with the enzyme Mpro, methyltransferase, the SPIKE, and ACE2 proteins by the interaction energy and the distance of the compounds and the target protein's amino acids.

2. Material and methods

2.1. Chalcones

Using the methodological principle of synthesis the Claisen-Schmith reaction (in basic medium) [8], chalcones were synthesized from benzaldehydes and 4-aminoacetophenone, both at a concentration of 2 mmol. The reagents were added in a volumetric flask (25 mL), to which 5 mL of ethanolic NaOH solution (50%) were added. After adding the ethanol solution, the mixtures were kept under stirring for 48 h (at room temperature). TLC (n-hexane: ethyl acetamidecalcones 0-aminocalcones 0-aminocalcones were used, in which heat maps were generated to identify the ligand-residue interaction and similarity profiles by the Pearson statistical test [18].

Based on observations of the interactions of the molecules with the enzyme, the hydrogen bonds were plotted and classified according to previous studies that group interactions with distances between 2.5 and 3.1 Å as strong, from 3.1 to 3.55 Å as average and >3.55 Å as weak [19].

3. Results

3.1. Interaction between the 4-acetamidochalcones molecules and the Main protease (Mpro)

The positions shown in Fig. 2A were obtained from the simulations of interactions of 4-acetamidochalcones with Mpro by molecular docking. After comparative analyzes, it was noted that the PAAPF interacted with the enzyme at a site like that of the FJC inhibitor. Likewise, PAAB interacted at the same site as the antibiotic Azithromycin. Also, the reference drugs Anakinra and Remdesivir interacted in common sites, the same as for PAAPA. Additionally, PAAPM, PAACN, and PAAPF interacted with distinct sites from each other and different from any other reference inhibitor.

To analyze the intensity and affinity of the interactions obtained in the molecular docking simulations, the values of the interaction energy and the distance of the compounds and the target protein's amino acids.
energy and RMSD were collected, plotted, and compared (Table S2, Supplementary material). Interactions with energy \(< -6.0\ \text{kcal mol}^{-1}\) and RMSD \(< 2.0\ \text{Å}\) were considered satisfactory. Therefore, based on these criteria, only the chalcone PAACN showed an affinity of \(-6.1\ \text{kcal mol}^{-1}\) with the enzyme Mpro, although this molecule does not have a binding site in common with the inhibitors, especially the natural inhibitor FJC. Concerning the reference inhibitor, azithromycin did not show good affinity when presenting energy of \(-5.8\ \text{kcal mol}^{-1}\).

To examine and describe the intrinsic characteristics of the interactions of \(4'\)-acetamidechalcones molecules with the enzyme, the hydrogen bonds, and hydrophobic interactions were highlighted and compared with the reference ligands (Fig. 2B). Since PAAPM, PAACN, and PAAPE did not show similarities with the reference ligands, those molecules were not considered for analysis. Table S3 (Supplementary material) shows the interaction distances between chalcones and the amino acid residues of the enzyme Mpro. Thus, detailed information about the interactions of the molecules with the enzyme was obtained. Initially, for the PAAPF chalcone, which has the potential for coupling in the same region as the FJC inhibitor, it has two significant interactions with the GLU166 residue, one of which is a strong hydrogen bond. The PAAB chalcone presented only interaction of hydrogen with the residue ARG298, which the Azithromycin did not interact; moreover, the Azithromycin presents most hydrophobic interactions, all with a distance higher than 3.5 Å. The PAAPA chalcone interacted with the enzyme through five hydrogen bonds, mostly of moderate to strong intensity.

3.2. Interaction with the methyltransferase heterodimer NSP16-NSP10 SARS-CoV-2

The interactions of chalcones with the NSP16-NSP10 are illustrated in Fig. 3A. Among the studied structures, PAACN and PAAPE did not show significant interaction with the enzyme. Besides, the other chalcones interacted in similar places. Table S4 (Supplementary material) lists the energies of the interactions and the respective RMSD; thus, it was possible to observe affinity values \(< -6.0\ \text{kcal mol}^{-1}\) for the four chalcones, obtaining values of up to \(-8.2\ \text{kcal mol}^{-1}\) for PAAPF. All RMSD values were less than 2.0 Å, suggesting a satisfactory interaction between the molecules and the enzyme.

The specification of the interactions of chalcones with the enzyme is illustrated in the bi-dimensional maps contained in
in the molecular docking simulations (Fig. 4C). Besides, the specific interactions of the chalcones derivatives with the Spike protein are shown in Table S5 (Supplementary material). It is possible to observe that all chalcones had connections with similar regions of the target cells, as illustrated in the bi-dimensional maps contained in Fig. 4A. It was possible to observe that all chalcones had interactions with the SER371 residue, demonstrating the link of greater stability between the chalcones with the Spike protein.

3.3. Interactions with the SPIKE protein of the SARS-CoV-2 and with the ACE2

To study the potential of the chalcone acetamide derivatives to inhibit the interaction of the virus that causes COVID-19 with the target cells, a simulation of the interaction of the chalcones with the heterodimeric proteins of the virus, the Spike proteins, was carried out. The simulations of the coupling simulations are contained in Fig. 4A. It was possible to observe that all chalcones had interactions with similar regions of the protein. According to data of interaction energy and RMSD contained in Table S2 (Supplementary material), it was observed that all chalcones had satisfactory affinity < −6.0 kcal/mol. In particular, the PAAPA and PAACN derivatives showed the best affinity values (−7.0 and −6.9, respectively), demonstrating the link of greater stability between the chalcones.

The interactions with the amino acid residues of Spike protein heterodimer and the chalcones molecules were represented in bi-dimensional maps, corroborating with the information collected in the molecular docking simulations (Fig. 4C). Besides, the specific interactions of the chalcones derivatives with the Spike protein are shown in Table S5 (Supplementary material). It is possible to observe that all chalcones showed a pattern of interaction with the protein, as with the SER371 residue, which the derivatives interacted through the hydrogen of moderate intensity. Furthermore, the PAAPM and PAAPE chalcones showed, at least, one interaction of strong intensity, and the chalcone PAAPA did not show hydrogen bonds with the Spike protein.

Additionally, it is known that the interaction of the SPIKE protein with ACE2 is necessary for the virus to enter the host cells, hence the simulations of the interactions of the chalcones derivatives with this enzyme were performed, as shown in Fig. 4B. It is possible to observe that the chalcones interacted in similar sites, except for PAACN, which may have interacted at a non-specific site. Those data are reinforced by the affinity energy of the connections and RMSD, which the PAACN presented higher values, indicating an interaction of lower stability. PAAB and PAAPA had lower affinity values, with PAAPA having the best-suggested interaction, with an affinity of −8.0 kcal/mol associated with a low RMSD value (1355 Å).

Those results are reinforced when characterizing the interactions of the chalcones derivatives with the ACE2 protein, as illustrated in the bi-dimensional maps (Fig. 4D) and the interaction distances are shown in Table S5 (Supplementary material). The chalcones PAAPA and PAAPF stood out for presenting two interactions of strong hydrogen bonds. Also, the chalcones derivatives had common binding sites, such as the ILE291 residue, in which the PAAPA even had a hydrogen bond.

4. Discussion

Overall, the present work evaluated, through molecular docking assays, the interactions of chalcone acetamide derivatives with enzymatic and structural targets of SARS-CoV-2 and with the host’s ACE2, which is recognized by the virus, facilitating its entry into cells. Therefore, it was observed that, regarding the interactions of chalcones with Main protease (Mpro), the PAAPF derivative has the potential for coupling in the same region as the natural inhibitor FJC through strong hydrogen bonding. The formation of two strong hydrogen bonds between PAAB and the NSP16-NSP10 heterodimer methyltransferase was also noted. PAAPM and PAAPF showed at least one strong intensity interaction of the SPIKE protein. PAAPA had a better affinity with ACE2, with strong hydrogen interactions.

These results are relevant, as several chalcones have been described as having antiviral activity. A work performed by Park et al. [20] showed that chalcones isolated from Angelica Keiskei inhibit the chymotrypsin protease (3CL (pro)) and a papain protease (PL (pro)) in SARS-CoV. Proteases are important for post-translational modifications of structural proteins in the viral particle. Therefore, the inhibition of proteases interferes with the viral replication process [20]. Literature data suggest the inhibition of viral proteases by flavonoids and related compounds. For example, flavonoids such herbacetin, isohavachalcone, quercetin 3-β-D-glucoside and helicrysetin have been described as potent inhibitors of the Middle Eastern respiratory syndrome–coronavirus protease (MERS-CoV 3CLpro), indicating that flavonol and chalcones are
favorite structures for binding to the catalytic site, suggesting that modifications in the more hydrophobic molecules or with carbohydrates attached to their main structures have a good inhibitory effect [21].

In fact, synthetic flavonoids and chalcones are described for potential antiviral properties. In a previous study, substituted chalcones, showed inhibition of viral translation in cells infected with hepatitis C virus (HCV) by the ablation of ribosomal protein phosphorylation 6 (rps6) [22]. Additionally, several reports in the literature demonstrate that derivatives of chalcones present antiviral activity better than the reference drugs in experimental models, including synergistic effects with these. For example, a previous study showed that thienyl-chalcone derivatives showed moderate to excellent antiviral activity, with higher in vitro potency against human cytomegalovirus compared to the standard drug Ganciclovir [23]. These data corroborate the findings of the present study, in which the chalcone derivatives interacted with the virus protease at sites and with similar affinities to the clinically used drugs and the theoretical inhibitor FJC.

Binding and inhibiting the enzymatic activity of proteases and methyltransferases can lead to a disruption in the viral capsid construction process, interrupting the flow of transmission. In this sense, the importance of in silico screening of phytochemical compounds is ratified, allowing a preliminary and rational analysis of a high number of molecules. For example, an investigation of phytochemicals as antiviral agents against dengue has shown that secondary phenolic plant metabolites such as alkaloids, terpenoids, chalcones, flavonoids, coumarins, and quinones have the potential to bind to targets such as

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**Fig. 4.** Calculated interaction positions of the studied chalcones with the SPIKE (A) protein and the ACE2 protein of the human host (B). The bi-dimensional map of the hydrogen bonds and hydrophobic interactions between the chalcones and the SPIKE protein of the SARS-CoV-2 (C) and between the ACE2 protein of the human host (D).
5. Conclusion

The 4'-acetamidochalcones presented inhibitory potential over the SARS-CoV-2 proteins, detached the PAAP that has the potential to couple to Mpro (same region as the natural inhibitor FJC through strong hydrogen bonds), PAAB that can bind to NSP16 methyltransferase, PAAPM and PAAPE that can interact with the protein Spike and PAAPA that demonstrated a strong affinity for important enzymes in post-translational processes, interfering with viral replication.

In addition, chalcones have an affinity for important enzymes in post-translational processes, interfering with viral replication.

Conclusion

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at https://doi.org/10.1016/j.bbrc.2020.12.074.

References

[1] A.E. Gorbalenya, S.C. Baker, R.S. Baric, R.J. de Groot, C. Drosten, A.A. Gulyaeva, B.L. Haagmans, C. Lauber, A.M. Leontovich, B.W. Neuman, D. Penzar, S. Perlman, L.M. Poon, D.V. Samborskiy, I.A. Sidorov, I. Sola, J. Ziebuhr, V. Coronaviridae Study Group, International Committee on Taxonomy of Viruses, The species SARS-CoV-2, a novel coronavirus: classification of 2019-nCoV and naming it SARS-CoV-2, Nat Microbiol 5 (2020) 536–544, https://doi.org/10.1038/s41564-020-0695-z.

[2] Y. Chen, Q. Liu, D. Guo, Emerging coronaviruses: genome structure, replication, and pathogenesis, J. Med. Virol. 92 (2020) 418–423, https://doi.org/10.1002/jmv.25681.

[3] A.A. Siddiqui, A. Rahman, Shaharyar, R. Mishra, Synthesis and antiviral activity of some substituted 2,4-diphenyl-3-pyrazoline-5-carboxamide derivatives, J. Bioorg. Med. Chem. Lett. 22 (2012) 1250–1254, https://doi.org/10.1016/j.bmcl.2012.02.114.

[4] P.T. da Silva, J. daCunhaXavier, T.S. Freitas, M.M. Oliveira, H.D.M. Coutinho, A.L.A.B. Leal, H.M. Barreto, P.N. Bandeira, C.E.S. Nogueira, D.M. Senajr, F.W.Q. Almeida-Neto, E.S. Marinho, H.S. Santos, A.M.R. Teixeira, Synthesis, spectrophotometric characterization and antibacterial evaluation of derivatives derived of acetonaphthon isolated from Croton anisodontus Mull.Arg., J. Mol. Struct. 1226 (2021), https://doi.org/10.1016/j.molstruc.2020.129403.

[5] Z. Nowakowska, A review of anti-angiogenic and anti-inflammatory chalcones, Eur. J. Med. Chem. 42 (2007) 125–137, https://doi.org/10.1016/j.ejmech.2006.09.019.

[6] J.C. Onyilagha, B. Malhotra, M. Elder, C.J. French, G.H.N. Towers, Comparative studies of inhibitory activities of chalcones on tomato ringspot virus (ToRSV), Indian Dent. Assoc. 19 (1997) 133–137, https://doi.org/10.1007/978-0-306-66997-05541.

[7] I.A. Santos, Atividade antiviral de compostos naturais brasileiros e hidroxichalconas e curcuminoides sintéticos, Universidade Federal de Uberlândia-Instituto de Ciências Biomedicas, 2018.

[8] B.A. Bhat, K.L. Dhar, S.C. Puri, A.K. Saxena, M. Shanmugavel, G.N. Qazi, Synthesis of a 4'-acetamidochalcone and its derivatives as potential cytotoxic agents, Bioorg. Med. Chem. Lett. 15 (2005) 3177–3180, https://doi.org/10.1016/j.bmcl.2005.03.121.

[9] P.N. Bandeira, T.L.G. Lemos, H.S. Santos, M.C.S. de Carvalho, D.P. Pinheiro, M.O. de Moraes, C. Pessoa, F.W.A. Barros-Nepomuceno, T.H.S. Rodrigues, P.R.V. Ribeiro, H.S. Magalhães, A.M.R. Teixeira, Synthesis, structural characterization, and cytotoxic evaluation of chalcone derivatives, Med. Chem. Res. 28 (2019) 2037–2049, https://doi.org/10.1007/s00014-019-04243-1.

[10] J. Jin, X. Du, Y. Xu, Y. Deng, M. Liu, Y. Zhao, B. Zhang, X. Li, L. Zhang, C. Peng, Y. Duan, J. Yu, L. Wang, K. Yang, F. Liu, J. Wang, X. Yang, T. You, X. Li, X. Yang, F. Bai, H. Liu, X. Liu, L.W. Guddat, W. Xu, X. Xiao, C. Qin, Z. Shi, H. Jiang, Z. Rao, H. Yang, Structure of Mpro from SARS-CoV-2 and discovery of its inhibitors, Nature 582 (2020) 289–293, https://doi.org/10.1038/s41586-020-2223-y.

[11] M.A. Basas-Lemus, G. Minakov, L. Shuvalova, N.L. Imnis, O. Kuyukhova, G. Wiersum, Y. Kim, R. Jridzejczak, N.L. Malteva, M. Endres, L. Jaroszewski, A. Godzik, A. Joachimiak, K.J.F. Satchell, The Crystal Structure of Nsp16 from SARS-CoV-2 Complexed with 2,4-Diselenylsulfoxphene, bioRxiv (2020), 047408, https://doi.org/10.1101/2020.04.07.47408, 2020.2004.2017.

[12] J. Lan, J. Ge, Y. Yu, S. Shan, H. Zhou, S. Fan, Q. Zhang, X. Shi, Q. Wang, L. Zhang, X. Wang, Structure of the SARS-CoV-2 spike receptor-binding domain bound to the ACE2 receptor, Nature 581 (2020) 215–220, https://doi.org/10.1038/s41586-020-2180-5.

[13] O. Trott, A.J. Olson, AutoDock Vina, Improving the speed and accuracy of docking with a new scoring function, efficient optimization, and multi-threading, J. Comput. Chem. 31 (2010) 455–461, https://doi.org/10.1002/jcc.21334.

[14] S. Shiyakov, C. Förster, In silico predictive model to determine vaccine-mediated transport properties for the blood-brain barrier choline transporter, Adv Appl Biomater Chem 7 (2014) 23–36, https://doi.org/10.2147/aabc.s36349.

[15] D. Yusuf, A.M. Davis, G.J. Kleywegt, S. Schmitt, An alternative method for the evaluation of docking performance: RSR vs RMSD, J. Chem. Inf. Model. 48 (2008) 1411–1422, https://doi.org/10.1021/ci080064x.

[16] D.S. Biovia, H.M. Berman, J. Westbrook, Z. Feng, G. Gilliland, T.N. Bhat, H. Weissig, I.N. Shindyalov, P.E. Bourne, J. Med. Virol. 92 (2020) 10110, https://doi.org/10.1002/jmv.25901.

[17] E.F. Pettersen, T.D. Goddard, C.C. Huang, G.S. Couch, D.M. Greenblatt, C.A. Meng, E. Ferrin, UCSC Chimera—a visualization system for exploratory research and analysis, J. Comput. Chem. 25 (2004) 1605–1612, https://doi.org/10.1002/jcc.20084.

[18] F.A. Ruf€orster, In silico predictive model to determine vector-mediated transport properties for the blood-brain barrier choline transporter, Adv Appl Biomater Chem 7 (2014) 23–36, https://doi.org/10.2147/aabc.s36349.

[19] I.A. Santos, Atividade antiviral de compostos naturais brasileiros e hidroxichalconas e curcuminoides sintéticos, Universidade Federal de Uberlândia-Instituto de Ciências Biomedicas, 2018.

[20] F.A. Ruf€orster, In silico predictive model to determine vector-mediated transport properties for the blood-brain barrier choline transporter, Adv Appl Biomater Chem 7 (2014) 23–36, https://doi.org/10.2147/aabc.s36349.

[21] F.A. Ruf€orster, In silico predictive model to determine vector-mediated transport properties for the blood-brain barrier choline transporter, Adv Appl Biomater Chem 7 (2014) 23–36, https://doi.org/10.2147/aabc.s36349.
K.H. Park, W.S. Lee, Y.B. Ryu, Chalcones isolated from Angelica keiskei inhibit cysteine proteases of SARS-CoV, J. Enzym. Inhib. Med. Chem. 31 (2016) 23–30, https://doi.org/10.3109/14756366.2014.1001216.

[21] S. Jo, H. Kim, S. Kim, D.H. Shin, M.-S. Kim, Characteristics of flavonoids as potent MERS-CoV 3C-like protease inhibitors, Chem. Biol. Drug Des. 94 (2019) 2023–2030, https://doi.org/10.1111/cbdd.13604.

[22] N. Mateeva, S.V.K. Eyunni, K.K. Redda, U. Ononuju, T.D. Hansberry, C. Aikens, A. Nag, Functional evaluation of synthetic flavonoids and chalcones for potential antiviral and anticancer properties, Bioorg. Med. Chem. Lett 27 (2017) 2350–2356, https://doi.org/10.1016/j.bmcl.2017.04.034.

[23] V. Patil, S.A. Patil, R. Patil, A. Bugarin, K. Beaman, S.A. Patil, Exploration of (hetero)aryl derived thiophylchalcones for antiviral and anticancer activities, Med. Chem. 15 (2019) 150–161, https://doi.org/10.2174/1573406414666180524074648.

[24] C.N. Powers, W.N. Setzer, An in-silico investigation of phytochemicals as antiviral agents against dengue fever, Comb. Chem. High Throughput Screen. 19 (2016) 516–536, https://doi.org/10.2174/1386207319666160506123715.

[25] L. Yi, Z. Li, K. Yuan, X. Qu, J. Chen, G. Wang, H. Zhang, H. Luo, L. Zhu, P. Jiang, L. Chen, Y. Shen, M. Luo, G. Zuo, J. Hu, D. Duan, Y. Nie, X. Shi, W. Wang, Y. Han, T. Li, Y. Liu, M. Ding, H. Deng, X. Xu, Small molecules blocking the entry of severe acute respiratory syndrome coronavirus into host cells, J. Virol. 78 (2004) 11334, https://doi.org/10.1128/JVI.78.20.11334-11339.2004.

[26] M. Wyganowska-Swiatkowska, M. Nohawica, K. Grocholiewicz, G. Nawak, Influence of herbal medicines on HMGB1 release, SARS-CoV-2 viral attachment, acute respiratory failure, and sepsis. A literature review, Int. J. Mol. Sci. 21 (2020), https://doi.org/10.3390/ijms21134639.