Short Note

N'-(5-bromofuran-2-carbonyl)isonicotinohydrazide

Ersya Yanu Ramadhani , Nur Pasca Aijijiyah , Eko Santoso , Lukman Atmaja , Mardi Santoso *

Posted Date: 3 July 2023

doi: 10.20944/preprints202307.0049.v1

Keywords: furan carboxamide; organic synthesis; furoic acid; isoniazid; molecular docking; COX-2 inhibitor

Short Note

N'-(5-bromofuran-2-carbonyl)isonicotinohydrazide

Ersya Yanu Ramadhani, Nur Pasca Aijijiyah, Eko Santoso, Lukman Atmaja and Mardi Santoso *

Department of Chemistry, Faculty of Science and Analytical Data, Institut Teknologi Sepuluh Nopember, Kampus ITS Sukolilo, Surabaya 60111, Indonesia

* Correspondence: tsv09@chem.its.ac.id

Abstract: *N'*-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)** was obtained as a white solid in 83% yield by 2-methyl-6-nitrobenzoic anhydride (MNBA)/4-dimethylaminopyridine (DMAP)-catalyzed reaction of 5-bromofuran-2-carboxylic acid and isoniazid in dichloromethane at room temperature. The structure of *N'*-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)** was elucidated by ^{1}H NMR, ^{13}C NMR, FTIR, and the high resolution mass spectrometers. Molecular docking screening of the title compound **(1)** on the cyclooxygenase-2 (COX-2) protein (PDB ID: 5IKR) exhibited that compound **(1)** has a good binding affinity, suggesting that it is very interesting for further structure optimization and in-depth research as a potential COX-2 inhibitor.

Keywords: furan carboxamide; organic synthesis; furoic acid; isoniazid; molecular docking; COX-2 inhibitor

1. Introduction

Furan carboxamides are furan amides with various pharmacological activities, including antitumor [1], antifungal [2], antimicrobial [3], anticancer [4], and antidiabetic [5]. The synthesis of amides commonly using thionyl chloride as coupling agent [6], which not only produce toxic gas sulfur dioxide but also one of the chemicals that included in the list of Chemical Weapons Conventions (CWC) and the Law of the Republic of Indonesia Number 9 of 2008 concerning the Use of Chemicals and Prohibitions on the Use of Chemicals as Chemical Weapons [7,8]. Furan carboxamides have successfully synthesized using a hydroxybenzotriazol coupling reagent [4], but this material is explosive so the use of it should be avoided [9]. More environmentally-friendly synthesis of furan carboxamides can be carried out using 1,1'-carbonyldiimidazole (CDI) coupling agent [10], however, it has low reactivity, especially when compared to similar materials [11]. Alternately, synthesis of carboxamides by utilizing 2-methyl-6-nitrobenzoic anhydride (MNBA) and 4-dimethylaminopyridine (DMAP) as a coupling agent and a catalyst respectively, showing several advantages, including the reaction can be carried out in one pot at room temperature, and the resulting compounds having high yields and purity [12]. Therefore, this study aimed to utilize MNBA/DMAP for the synthesis of furan carboxamide from furoic acid and, namely *N*'-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)** with a molecular docking study to understand its molecular behavior against cyclooxygenase-2 (COX-2) protein.

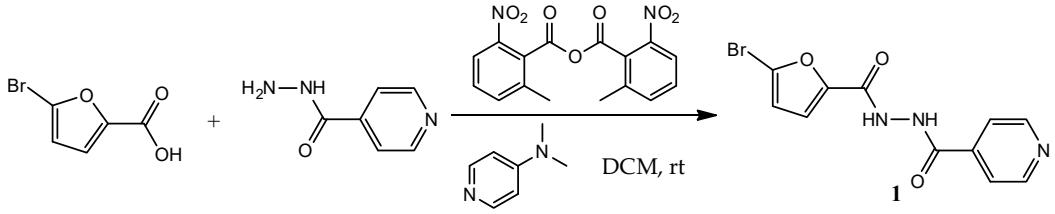
2. Results and Discussion

2.1. Chemistry

The synthesis of *N'*-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)** was carried out by utilizing the reaction of 5-bromofuran-2-carboxylic acid and isoniazid with the presence of MNBA and DMAP in dichloromethane at room temperature (Scheme 1). The crude product was purified by dry-column flash chromatography to afford pure product *N'*-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)** as a white solid in 83% yield. The ^{1}H NMR spectrum of the product clearly showed two doublet signals at δ 6.84 and 7.32 ppm indicating two furanyl protons, two doublet of doublets signals at δ 7.80 and 8.79 ppm indicating four pyridinyl protons, and two broad singlet signals at δ 10.62 and 10.81 ppm indicating proton signals of the NH groups. The presence of

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 3 July 2023 doi:10.20944/preprints202307.0049.v1

the NH group was confirmed by the IR spectrum which showed a single absorption at a wavenumber of 3169 cm^{-1} indicating the presence of a secondary NH group. It is also supported by the ^{13}C NMR spectrum, which exhibited nine signals which correspond to the nine carbon types in the structure of *N'*-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)**. The carbon of the two carbonyl groups gave signals at δ 156.71 and 164.85 ppm; and aromatic carbons signals at δ 114.69, 117.87, 121.84, 126.21, 139.80, 148.41, and 151.05 ppm. The presence of these carbonyl groups was confirmed by the IR spectrum which showed absorption at a wavenumber of 1644 and 1682 cm^{-1}. The high-resolution mass spectrum further supported that the reaction product as *N'*-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)** based on the molecular ion peak at *m/z* 309,9885 $[M+H]^+$ in a positive ionization mode.



Scheme 1. Synthesis of N'-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)**.

2.2. Molecular Docking Study

Molecular docking was used to investigate the molecular behavior of *N'*-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)** at the COX-2 protein binding site. Human cyclooxygenase-2 complexed with mefenamic acid (PDB ID: 5IKR) was chosen as a molecular target to examine the interaction of compound **(1)** with COX-2 protein. Redocking of mefenamic acid, as the native ligand, at the COX-2 binding site resulted in a binding energy of -7.61 kcal/mol and an RMSD value of 0.55 Å (Figure 1).

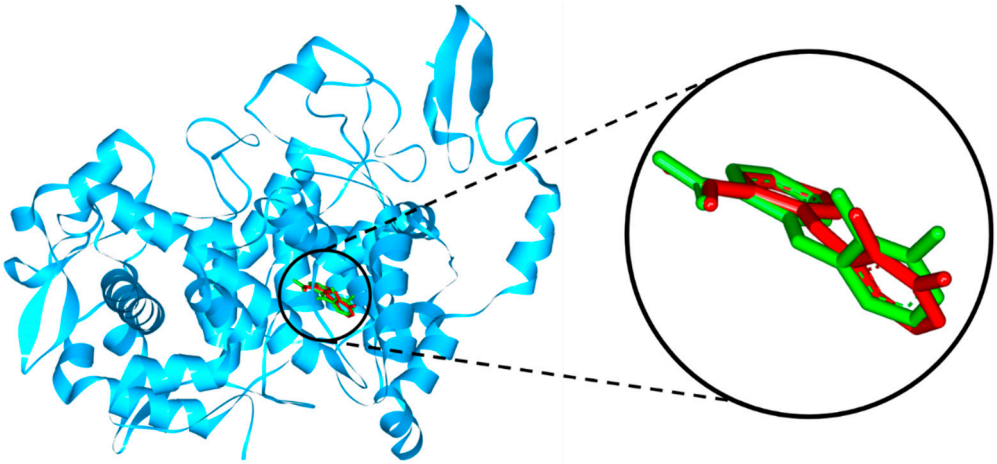


Figure 1. Redocked mefenamic acid (green) was superimposed onto the crystallographic mefenamic acid (red) in the binding site of the COX-2 protein (PDB ID: 5IKR).

The docking result showed that the title compound **(1)** had a binding energy of -7.89 kcal/mol, which was not significantly different from mefenamic acid. Compound **(1)** exhibited hydrogen bonding, electrostatic interaction, and hydrophobic interaction (Figure 2). The pyridine core of compound **(1)** generated hydrophobic interactions with Val349 and Leu352 via μ-alkyl interactions. One of the carbonyl groups and two NH groups formed hydrogen bonds with Gly526, Met522, and Ala527, respectively. The furan core formed hydrophobic interactions with Val523, Leu352, and Ala527 in the form of μ-sigma and μ-alkyl interactions. Furthermore, electrostatic interaction was established through μ-cation interaction between the furan core and Arg120. Meanwhile, the bromo atom on the furan moiety formed hydrophobic interactions with Tyr385 and Ala527 in the form of μ-alkyl and alkyl interactions, respectively.

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 3 July 2023 doi:10.20944/preprints202307.0049.v1

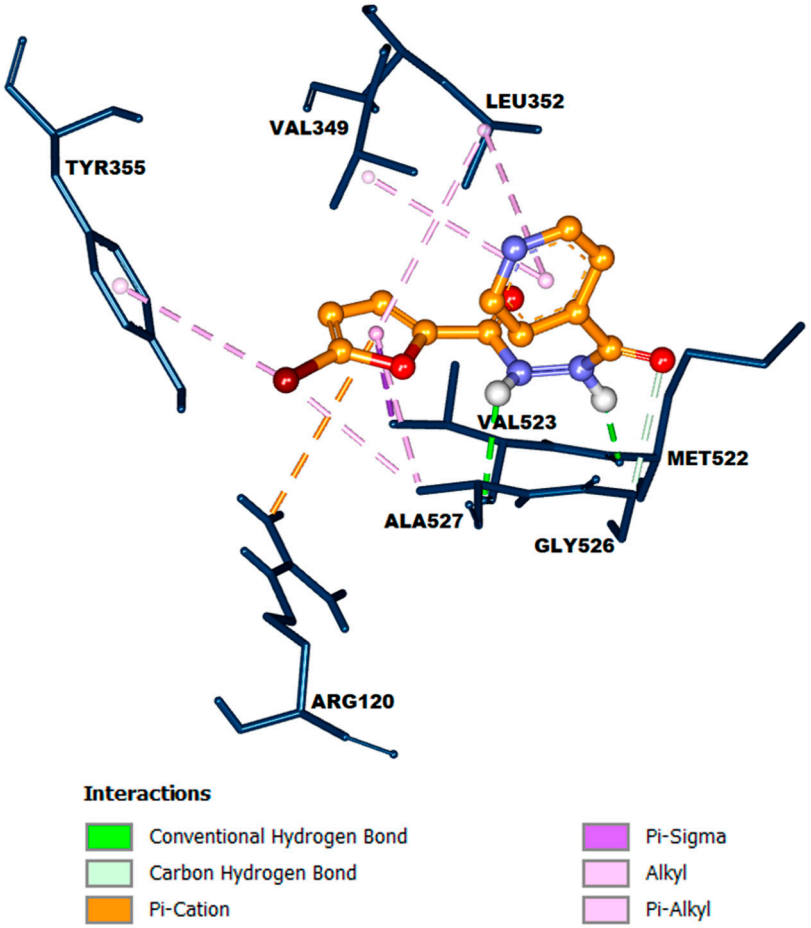


Figure 2. Binding pose and interaction of the compound **(1)** in the binding site of the COX-2 protein (PDB ID: 5IKR).

3. Materials and Methods

The starting materials and reagents used in this study were obtained from Sigma-Aldrich (St. Louis, MO, USA) and Merck (Rahway, NJ, USA) which were used without further purification. Thin layer chromatography was carried out with Merck 0.20 mm precoated silica gel aluminum plates (Kieselgel 60, F_{254}) and was visualized using a 245 nm UV lamp. Dry-column flash chromatography was carried out with Merck 60H. NMR spectra were obtained in DMSO-d_6, with a Jeol JNM-ECS400 spectrometer (400 MHz). A high-resolution mass spectrum was recorded on a Thermo Scientific TSQ Vantage Triple State Quadrupole, and an infrared spectrum was obtained on a Thermo Scientific Nicolet iS10 FTIR spectrophotometer.

2.1. Synthesis of N'-(5-bromofuran-2-carbonyl)isonicotinohydrazide (1)

The solution of 5-bromofuran-2-carboxylic acid (0.30 g; 1.62 mmol), MNBA (0.39 g; 1.16 mmol), and DMAP (0.26 g; 2.12 mmol) in dichloromethane (10 mL) was stirred at room temperature for 60 minutes. The solution was added with isoniazid (0.13 g; 0.96 mmol), and stirred further for 5 days (the reaction was monitored by TLC with ethyl acetate:methanol (5:1) as an eluent). The reaction product was evaporated under reduced pressure, and the residue was purified by dry-column flash chromatography with ethyl acetate:*n*-hexane (3:1) as eluent to give *N*'-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)** as a white solid (0.25 g, 83%); mp: 203°C; FTIR (KBr) υ_{max} (cm^{-1}): 3168 (N-H), 1682 (C=O amide), 1644 (C=O amide); ^{1}H NMR (DMSO-d_6, 400 MHz): δ 6.84 (1H, d, *J*=3.6 Hz, ArH), 7.32 (1H, d, *J*=3.6 Hz, ArH), 7.80 (2H, dd, *J*=4.4, 1.6 Hz, ArH), 8.79 (2H, dd, *J*=4.4, 1.6 Hz, ArH), 10.62 (1H, bs, NH), 10.81 (1H, bs, NH); ^{13}C NMR (DMSO-d_6, 100 MHz): δ 114.69, 117.87, 121.84,

126.21, 139.80, 148.41, 151.05, 156.71, 164.85; HRESIMS *m/z* (pos): 309,9885 $C_{11}H_9N_3O_3Br$ (calcd. 309,9827) (Supplementary Materials).

2.2. Molecular Docking Study

The crystal structure of the COX-2 protein was obtained from the Protein Data Bank (PDB ID: 5IKR) and prepared using MGLTools 1.5.6 [13]. The 3D structure of *N*'-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)** was generated using the MarvinSketch tool [14]. The location of the receptor binding site was set using a grid box placed at the position of the native ligand (mefenamic acid) with a centering of x: 38.042; y: 2.131; z: 61.280, *xyz* dimensions of 30x30x30, and a spacing of 0.375 Å. The 200 iterations of the Lamarckian genetic algorithm were performed on the Autodock4.2 program in order to obtain the best binding pose of *N*'-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)** at the binding site of COX-2 protein [15]. The complex formed between compound **(1)** and COX-2 was analyzed using Biovia Discovery Studio 2020 [16].

4. Conclusions

N'-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)** was successfully synthesized from the reaction of 5-bromofuran-2-carboxylic acid and isoniazid in the presence of MNBA and DMAP, which obtained a good yield (83%). Molecular docking showed that *N*'-(5-bromofuran-2-carbonyl)isonicotinohydrazide **(1)** interacted with the residues of the COX-2 protein through several interactions, including hydrogen bonding, electrostatic interaction, and hydrophobic interaction, and had a good binding affinity with a value of -7.89 kcal/mol. Isonicotinohydrazide has the potential to serve as a lead structure in the development of a COX-2 inhibitor.

Supplementary Materials: The following supporting information can be downloaded online, Figure S1: IR spectrum of the title compound **(1)**; Figure S2: ^{1}H NMR spectrum of the title compound **(1)**; Figure S3: ^{13}C NMR spectrum of the title compound **(1)**; Figure S4: Mass spectrum of the title compound **(1)**.

Author Contributions: Conceptualization, M.S.; methodology, M.S.; software, N.P.A.; validation, E.S., E.Y.R.; formal analysis, E.Y.R.; investigation, E.Y.R.; resources, M.S.; data curation, L.A.; writing—original draft preparation, E.Y.R.; writing—review and editing, M.S. and N.P.A.; visualization, N.P.A.; supervision, M.S.; project administration, M.S.; funding acquisition, M.S. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by Institut Teknologi Sepuluh Nopember (Penelitian Keilmuan Dana ITS grant number 1690/PKS/ITS/2023).

Data Availability Statement: The data presented in this study are available in the Supplementary Materials.

Acknowledgments: The authors acknowledge Institut Teknologi Sepuluh Nopember for funding.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Tikhomirov, A.S.; Lin, C.Y.; Volodina, Y.L.; Dezhenkova, L.G.; Tatarskiy, V. V.; Schols, D.; Shtil, A.A.; Kaur, P.; Chueh, P.J.; Shchekotikhin, A.E. New Antitumor Anthra[2,3-b]Furan-3-Carboxamides: Synthesis and Structure-Activity Relationship. *European Journal of Medicinal Chemistry* **2018**, *148*, 128–139, doi:10.1016/j.ejmech.2018.02.027.
2. Yang, Z.; Sun, Y.; Liu, Q.; Li, A.; Wang, W.; Gu, W. Design, Synthesis, and Antifungal Activity of Novel Thiophene/Furan-1,3,4-Oxadiazole Carboxamides as Potent Succinate Dehydrogenase Inhibitors. *J Agric Food Chem* **2021**, *69*, 13373–13385, doi:10.1021/acs.jafc.1c03857.
3. Zanatta, N.; Alves, S.H.; Coelho, H.S.; Borchhardt, D.M.; Machado, P.; Flores, K.M.; da Silva, F.M.; Spader, T.B.; Santurio, J.M.; Bonacorso, H.G.; Martins, M.A.P. Synthesis, Antimicrobial Activity, and QSAR Studies of Furan-3-Carboxamides. *Bioorganic and Medicinal Chemistry* **2007**, *15*, 1947–1958, doi:10.1016/j.bmc.2007.01.003.
4. Zhan, W.; Xu, L.; Dong, X.; Dong, J.; Yi, X.; Ma, X.; Qiu, N.; Li, J.; Yang, B.; Zhou, Y.; Hu, Y. Design, Synthesis and Biological Evaluation of Pyrazol-Furan Carboxamide Analogues as Novel Akt Kinase Inhibitors. *European Journal of Medicinal Chemistry* **2016**, *117*, 47–58, doi:10.1016/j.ejmech.2016.03.074.

5. He, M.; Li, Y.-J.; Shao, J.; Li, Y.-S.; Cui, Z.-N. Synthesis and Biological Evaluation of 2,5-Disubstituted Furan Derivatives Containing 1,3-Thiazole Moiety as Potential α-Glucosidase Inhibitors. *Bioorg Med Chem Lett* **2023**, *83*, 129173, doi:10.1016/j.bmcl.2023.129173.
6. Monier, M.; El-Mekabaty, A.; Elattar, K.M. Five-Membered Ring Systems with One Heteroatom: Synthetic Routes, Chemical Reactivity, and Biological Properties of Furan-Carboxamide Analogues. *Synthetic Communications* **2018**, *48*, 839–875, doi:10.1080/00397911.2017.1421227.
7. Narasimhan, B.; Belsare, D.; Pharande, D.; Mourya, V.; Dhake, A. Esters, Amides and Substituted Derivatives of Cinnamic Acid: Synthesis, Antimicrobial Activity and QSAR Investigations. *European Journal of Medicinal Chemistry* **2004**, *39*, 827–834, doi:10.1016/j.ejmech.2004.06.013.
8. Zhang, M.; Lu, X.; Zhang, H.J.; Li, N.; Xiao, Y.; Zhu, H.L.; Ye, Y.H. Synthesis, Structure, and Biological Assay of Cinnamic Amides as Potential EGFR Kinase Inhibitors. *Medicinal Chemistry Research* **2013**, *22*, 986–994, doi:10.1007/s00044-012-0093-z.
9. Dunetz, J.R.; Magano, J.; Weisenburger, G.A. Large-Scale Applications of Amide Coupling Reagents for the Synthesis of Pharmaceuticals. *Organic Process Research and Development* **2016**, *20*, 140–177, doi:10.1021/op500305s.
10. Shwetha, B.; Sudhanva, M.S.; Jagadeesha, G.S.; Thimmegowda, N.R.; Hamse, V.K.; Sridhar, B.T.; Thimmaiah, K.N.; Ananda Kumar, C.S.; Shobith, R.; Rangappa, K.S. Furan-2-Carboxamide Derivative, a Novel Microtubule Stabilizing Agent Induces Mitotic Arrest and Potentiates Apoptosis in Cancer Cells. *Bioorganic Chemistry* **2021**, *108*, 104586, doi:10.1016/j.bioorg.2020.104586.
11. Métro, T.X.; Martinez, J.; Lamaty, F. 1,1′-Carbonyldiimidazole and Mechanochemistry: A Shining Green Combination. *ACS Sustainable Chemistry and Engineering* **2017**, *5*, 9599–9602, doi:10.1021/acssuschemeng.7b03260.
12. Shiina, I.; Kawakita, Y.I. The Effective Use of Substituted Benzoic Anhydrides for the Synthesis of Carboxamides. *Tetrahedron* **2004**, *60*, 4729–4733, doi:10.1016/j.tet.2004.03.066.
13. Kurumbail, R.G.; Stevens, A.M.; Gierse, J.K.; McDonald, J.J.; Stegeman, R.A.; Pak, J.Y.; Gildehaus, D.; Iyashiro, J.M.; Penning, T.D.; Seibert, K.; Isakson, P.C.; Stallings, W.C. Structural Basis for Selective Inhibition of Cyclooxygenase-2 by Anti-Inflammatory Agents. *Nature* **1996**, *384*, 644–648, doi:10.1038/384644a0.
14. Aijijiyah, N.P.; Wati, F.A.; Rahayu, R.; Srilistiani, A.; Mahzumi, F.; Aulia, T.; Santoso, L.; Pamela, E.; Ramadhani, E.Y.; Ilfahmi, Y.A.; Purnomo, A.S.; Putra, S.R.; Santoso, E.; Ningsih, S.; Firdausi, N.; Santoso, M. Synthesis, α-Glucosidase Inhibitory Activity, and Molecular Docking of Cinnamamides. *Med Chem Res* **2023**, *32*, 723–735, doi:10.1007/s00044-023-03032-y.
15. Morris, G.M.; Huey, R.; Lindstrom, W.; Sanner, M.F.; Belew, R.K.; Goodsell, D.S.; Olson, A.J. AutoDock4 and AutoDockTools4: Automated Docking with Selective Receptor Flexibility. *J. Comput. Chem.* **2009**, *30*, 2785–2791, doi:10.1002/jcc.21256.
16. Santoso, M.; Pamela, E.; Ramadhani, E.Y.; Ilfahmi, Y.A.; Aijijiyah, N.P.; Purnomo, A.S.; Putra, S.R. 4-Methoxyphenethyl (E)-3-(o-Tolyl)Acrylate. *Molbank* **2022**, *2022*, M1519, doi:10.3390/M1519.