ANADOLU UNIVERSITY
FACULTY OF PHARMACY

European Journal of Life Sciences 2025; 1: 15-23

Research Article

Novel pyridine-thiazole hybrid: synthesis, structural characterisation and adme predictions

Aybüke Züleyha Kaya[1,2], Leyla Yurttaş[1]

[1]Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Anadolu University, Eskişehir, Türkiye
[2]Department of Pharmaceutical Chemistry, Institute of Graduate Education, Anadolu University, Eskişehir, Türkiye

✉ Aybüke Züleyha Kaya
aybukezuleyhakaya@anadolu.edu.tr

https://doi.org/10.55971/EJLS.1645175

Received: 24.02.2025
Accepted: 22.03.2025
Available online: 24.04.2025

ABSTRACT

In this study, novel 4-(4-chlorophenyl)-*N*-phenyl-3-(pyridin-4-yl)thiazol-2(*3H*)-imine derivative (**2**) has been synthesized and the structure of the compound has been investigated by spectral analysis methods. By ^{1}H-NMR and ^{13}C-NMR spectral analysis, it was determined that the compound was obtained purely and its structure was elucidated. Further characterization of the compound 2D-NMR has been used to confirm the ring closure of the thiazole and the positions of the substituents linked carbon atoms. *In silico* studies have been completed via SwissADME and pkCSM pharmacokinetics software programs. The SwissADME software predicted that compound **2** could cross the blood-brain barrier (BBB) and also enter the gastrointestinal system. pkCSM pharmacokinetics studies indicated that compound **2** has no hepatotoxicity and also shows no skin irritation.

Keywords: ADME properties, structure specification, thiazole

1. INTRODUCTION

The thiazole ring is an aromatic heterocyclic compound that is commonly found in both synthetic and natural compounds [1-4]. The best known example containing thiazole ring as a core structure is thiamine (vitamin B1), which is one of the most important vitamins for humans from the beginning of drug research studies to the current synthesis of drugs for various diseases [5-8]. Other drug molecules containing the thiazole ring in their structure are antifungals, cambendazole, ravucaonazole, and abafungin, non-steroidal anti-inflammatories meloxicam and fentiazac, histamine-2 receptor blocker nizatidine, antiprotozoal nitazoxanide, anticancer dasatinib and vosaroxin, antibiotic azteronam, and antiviral ritonavir can be seen in Figure 1 [9-13].

There are so many ways to achieve a thiazole ring. Researchers are always trying to find simple and high-yield methods for reactions. For the thiazole ring, Hantzsch (1887) and Robinson-Gabriel (1912) methods are aged but often used by pharmaceutical chemists. The Hantzsch synthesis is based on the condensation of alpha-haloketones and thioamides. The Robinson-Gabriel synthesis, on the other hand, is a cyclization reaction of acylaminocarbonyl compounds and a stoichiometric amount of phosphorus pentasulfide at 170 °C. In this way, 2- or 5-substituted thiazole derivatives can be synthesized. The Cook-Heilborn method is another method for the synthesis of 2-aminothiazole derivatives from 2-aminonitriles. Other than that, the thiazole ring can be closed using so many reagents and methods, such as methyl thioglycolate reacted with thiocarbamoylimidate, when methyl isothiocyanate reacted with lithium diisopropylamide, propargylamine reacted with CS_2 using palladium catalyst [9, 14-17].

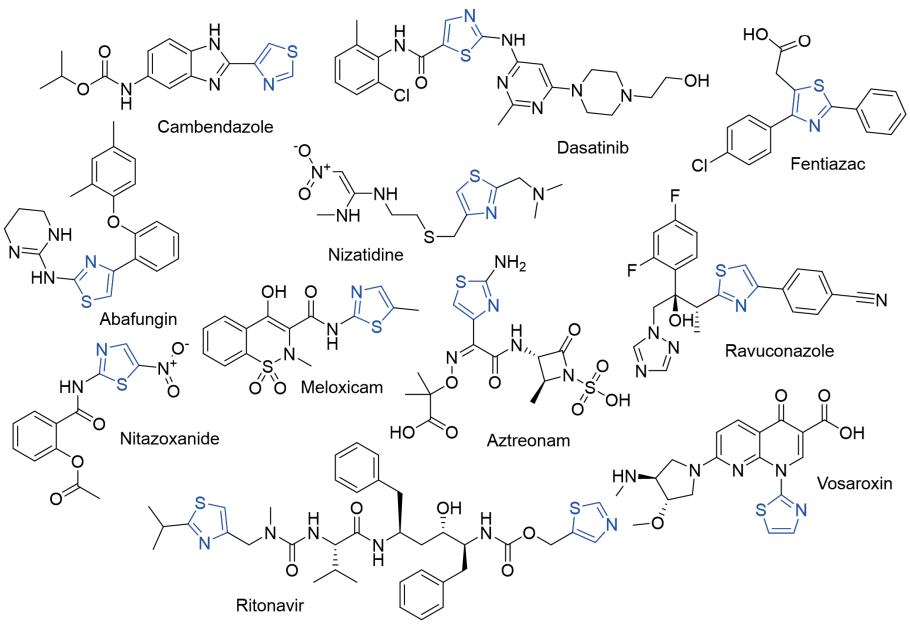


Figure 1. Various thiazole-based drugs

The pyridine ring also has many different therapeutic activities. These include antifungal, antibacterial, anticonvulsant, anti-inflammatory, antiviral and anticancer activities [18-21]. There are some many drugs that contain the pyridine ring such as pyridostigmine, isoniazid, piroxicam, omeprazole, delavirdine, sulfapyridine and metyrapone [22-25].

According to the previous studies, we have synthesized a pyridine-thiazole molecule and characterized it by spectral analysis [26-28]. In addition, studies have shown that the thiazole ring is associated with different activities. Many studies have looked at similar structures and found them to be effective against fungi, bacteria and viruses.

2. MATERIALS AND METHODS

2.1. Chemistry

All chemicals used in synthesis were supplied by Sigma-Aldrich Chemicals, USA and Merck Chemicals, Germany. Reactions and compound purities were monitored by thin layer chromatography (TLC) using silica gel 60 F_{254} aluminium plates purchased from Merck (Germany). MP90 digital melting point apparatus (Mettler-Toledo, USA) was used to record the uncorrected melting points of the synthesized compounds. The ^{1}H and ^{13}C NMR spectra in DMSO-d_6 were recorded using a Bruker 300 and 75 MHz digital nuclear magnetic resonance spectrometer (Bruker Bioscience, USA).

Synthesis of 1-phenyl-3-(pyridin-4-yl)thiourea derivative (1)

Appropriate amounts of pyridine-2-ylamine (1 eq) and phenyl isothiocyanate (1 eq) were dissolved in ethanol and refluxed for 3-4 hours. The end of the reaction was checked by thin layer chromatography. After cooling, the solid was filtered and recrystallised with ethanol.

Synthesis of final compound 4-(4-chorophenyl)-N-phenyl-3-(pyridin-4-yl)thiazol-2(3H)-imine (2)

1-Phenyl-3-(pyridin-4-yl)thiourea derivative (1 eq) and 2-bromo-4'-chloroacetophenone (1 eq) were dissolved in ethanol and refluxed for 4 to 5 hours. The end of the reaction was checked by thin layer chromatography. The solid was cooled, filtered and recrystallized in ethanol [29, 30].

4-(4-Chorophenyl)-N-phenyl-3-(pyridin-4-yl) thiazol-2(3H)-imine (2)

Yield: 60%. M.p. 285-288°C. ^{1}H NMR (300 MHz, DMSO-d_6, ppm): 7.20-7.22 (3H, m, Ar-H), 7.35-

7.42 (5H, m, Ar-H), 7.51-7.53 (4H, m, Ar-H), 7.59-7.61 (2H, m, Ar-H). ^{13}C NMR (75 MHz, DMSO-d_6, ppm): δ 105.51, 121.85, 124.79, 128.22, 128.93, 129.28, 129.59, 130.32, 130.60, 130.66, 130.94, 131.57, 132.50, 134.60, 134.85, 140.59, 168.76.

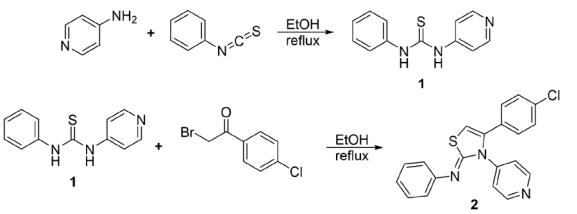
2.2. *In Silico* Studies

SwissADME and pkCSM-pharmacokinetics software programmes were used to study the physicochemical properties of the compound **2**. SwissADME software [31] and SwissTargetPrediction software [32, 33] were used to search the druglikeness and possible bioactivity profile of the compound **2**. Finally, the potential toxicity of our molecule was determined using the pkCSM pharmacokinetics software [34].

3. RESULTS AND DISCUSSIONS

3.1. Chemistry

In this study, we have synthesised (Scheme 1) novel 4-(4-chlorophenyl)-*N*-phenyl-3-(pyridin-4-yl)thiazol-2(*3H*)-imine. 1-Phenyl-3-(pyridin-4-yl) thiourea derivative synthesized from pyridin-2-amine and thiourea. 1-Phenyl-3-(pyridin-4-yl) thiourea and 2-bromo-4'-chloroacetophenone were



Scheme 1. General procedure for the synthesis of the final product 4-(4-chlorophenyl)-*N*-phenyl-3-(pyridin-4-yl) thiazol-2(*3H*)-imine (**2**)

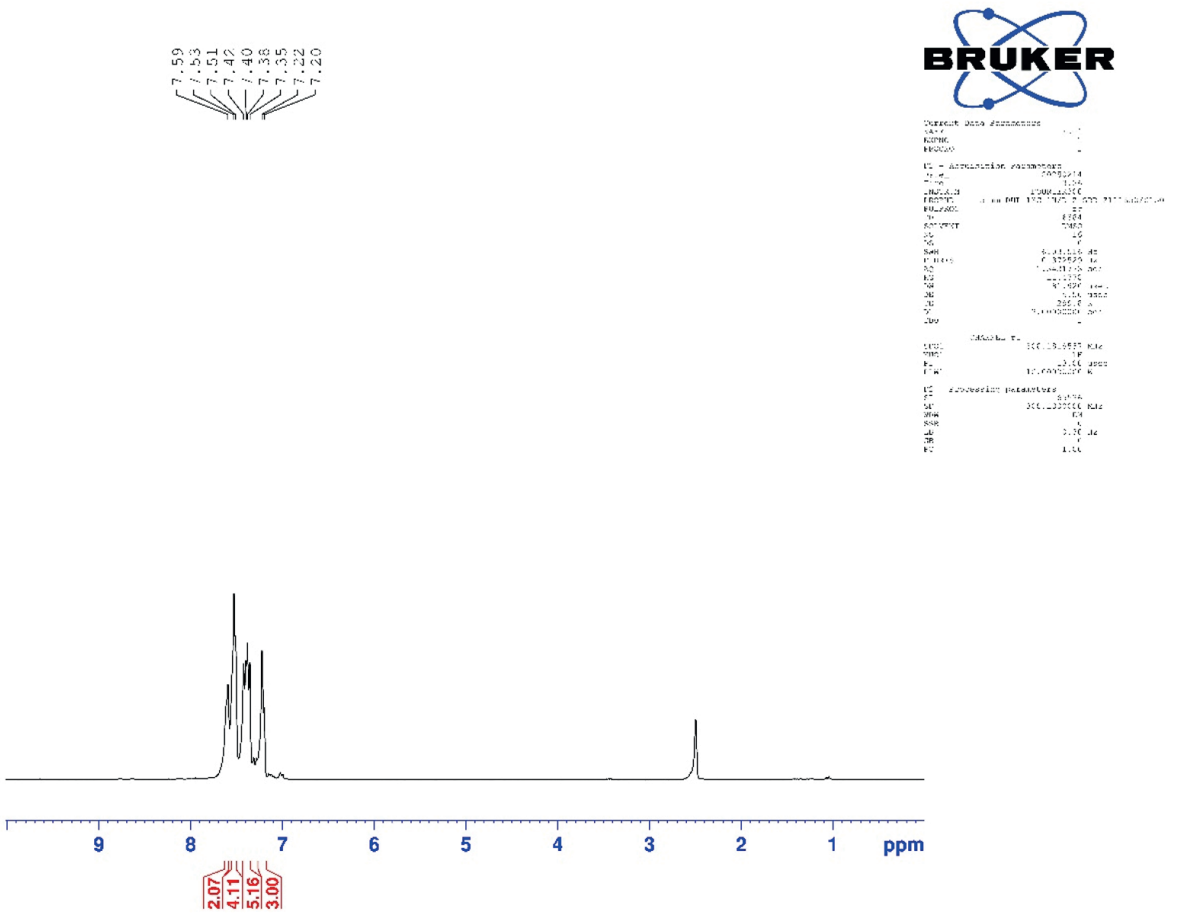


Figure 2. ^{1}H-NMR spectrum of compound **2**

reacted as described in the literature [29] to give the final product (**2**). To confirm the structure of compound **2**, various spectral analysis methods were used. ^{1}H-NMR, ^{13}C-NMR and 2D-NMR spectra were acquired and evaluated.

Compound **2** contains benzene, pyridine and thiazole rings in its structure. Aromatic -C-H proton peaks at 7.20-7.59 ppm were observed in the ^{1}H NMR spectra as shown in Figure 2. There are no aliphatic -C-H protons in the structure, therefore, there is no peak in the spectrum between 2.00-5.00 ppm. In the ^{13}C-NMR spectrum, carbon atoms were seen in the expected region, around 105.51-140.59 ppm (Figure 3).

Compound **2** was subjected to advanced 2D NMR (HSQC, NOESY, HMBC) studies. Firstly, the data obtained from the ^{1}H-NMR and ^{13}C-NMR spectra were analysed. Then, in order to match the carbons containing hydrogen(s), the HSQC spectrum was merged with the ^{1}H and ^{13}C NMR spectra. The HMBC data gives the spectrum of the carbons on the F1 axis, and the protons on the F2 axis, as in Figure 4. Then, using the data given, all the carbons are matched with the protons. According to HBMC spectrum at 168 ppm, there should be a single carbon interaction; this peak, compared with the HSQC spectrum and peaks are matched with one another. At 7.19 ppm on the F2 axis and at 105 ppm on the F1 axis, there were two peaks belonging to the thiazole ring in accordance with HSQC spectrum (Figure 5). These findings are proof that our ring has closed as designed.

3.2. *In Silico* Studies

The pharmacokinetic properties of the final compound were investigated using SwissADME and pkCSM pharmacokinetics software. Physicochemical properties, water solubility, pharmacokinetic properties (absorption, distribution, metabolism and excretion) and also druglikeness were calculated

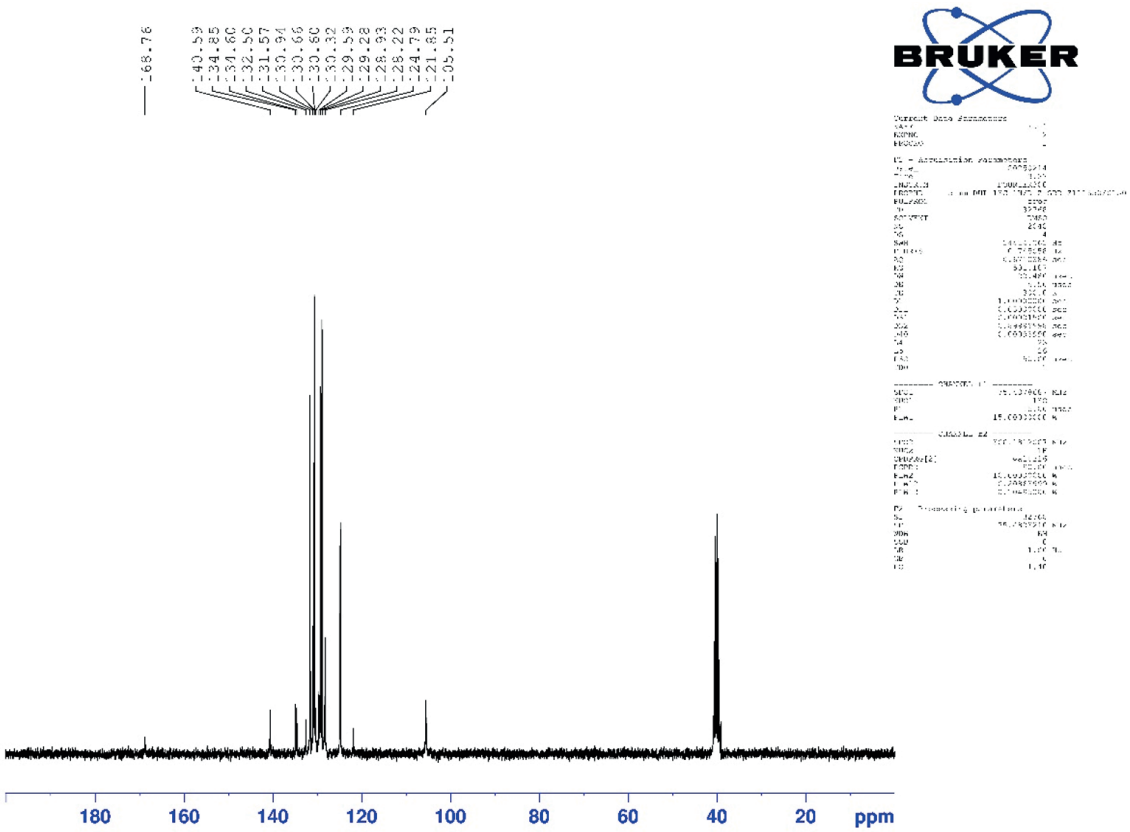


Figure 3. ^{13}C-NMR spectrum of compound **2**

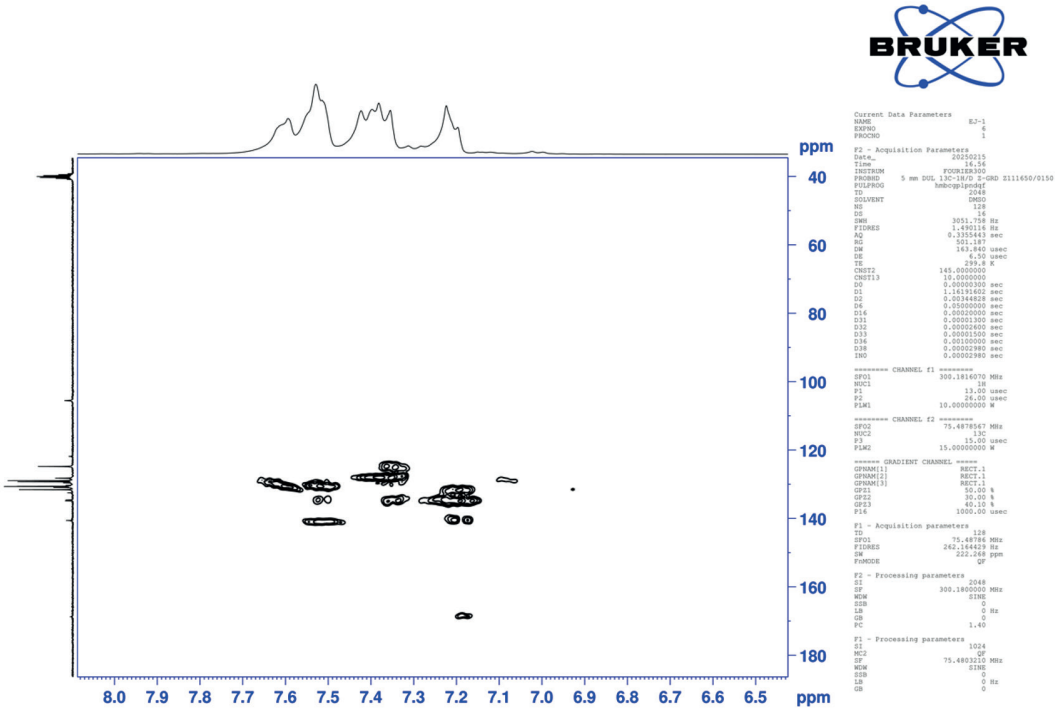


Figure 4. HBMC spectrum of compound **2**

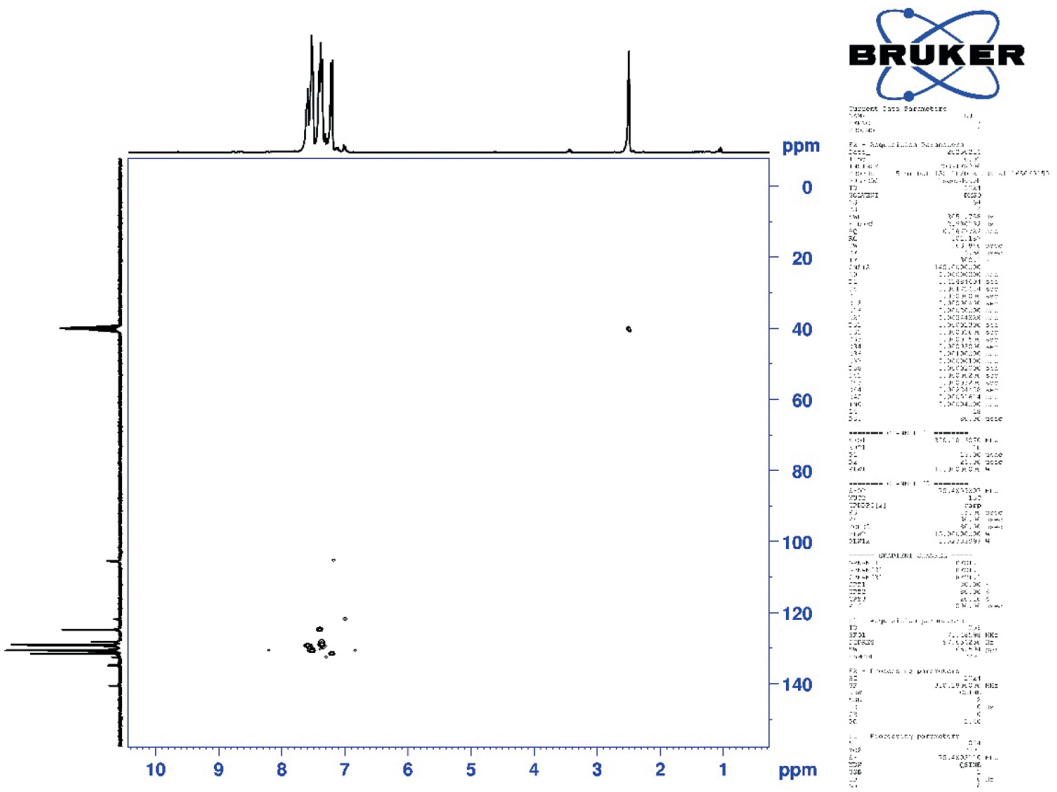


Figure 5. HSQC spectrum of compound **2**

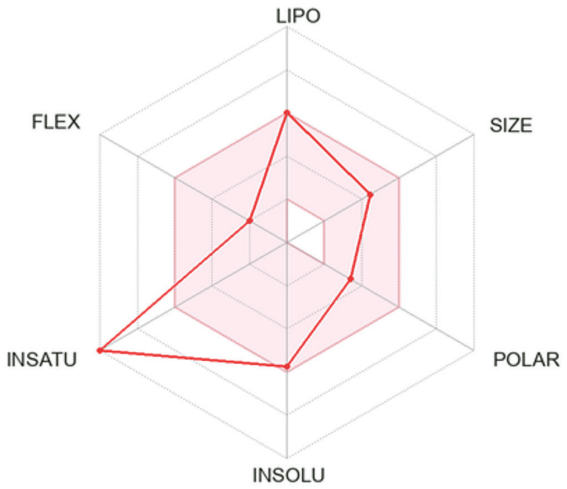


Figure 6. Physico-chemical field demonstration of the structure of compound **2** for the oral bioavailability

theoretically. Compound **2** has two hydrogen bond acceptor centres but no hydrogen bond donor centre. The topological polar surface area (TPSA) was 58.42 Å and the lipophilicity (cLogP) averaged over all five predictions was 4.75, close to the best range for these parameters. The solubility class of compound **2** in water was found to be moderately soluble according to logS value of 5.75 . Compound **2** can inhibit CYP1A2, CYP2C19, CYP2C9 and CYP3A4 enzymes. These features may lead to drug-drug interactions with our compound. Based on Lipinski's Rule of Five, compound **2** has a probability of being a orally bioavailable drug (Figure 6). In addition to Lipinski, there is also no violation according to the rules of Ghose, Veber and Egan.

To predict the gastrointestinal absorption and brain penetration of compound **2**, the boiled-egg plot between water partition coefficient (WLogP) and TPSA was also studied in SwissADME. As shown in Figure 7, Compound **2** was predicted to permeate the BBB and also to enter the gastrointestinal tract. Furthermore, red dots indicate molecules predicted not to be cleared by P-glycoprotein.

SwissTarget prediction software was used to identify the target prediction of the final compounds. The best 15 targets were predicted and the commonly predicted target regions are shown in Figure 8. Compound **2** was found to target phosphodiesterase, electrochemical transporter, lyase and enzyme with a percentage of 33.3%, 20.0%, 13.3% and 13.3%, respectively. Also with the same percentage of 6.7% compound **2** target on membrane receptor, writer and family A G protein-coupled receptor. With this, our compound may have a high level of attraction to the predicted targets.

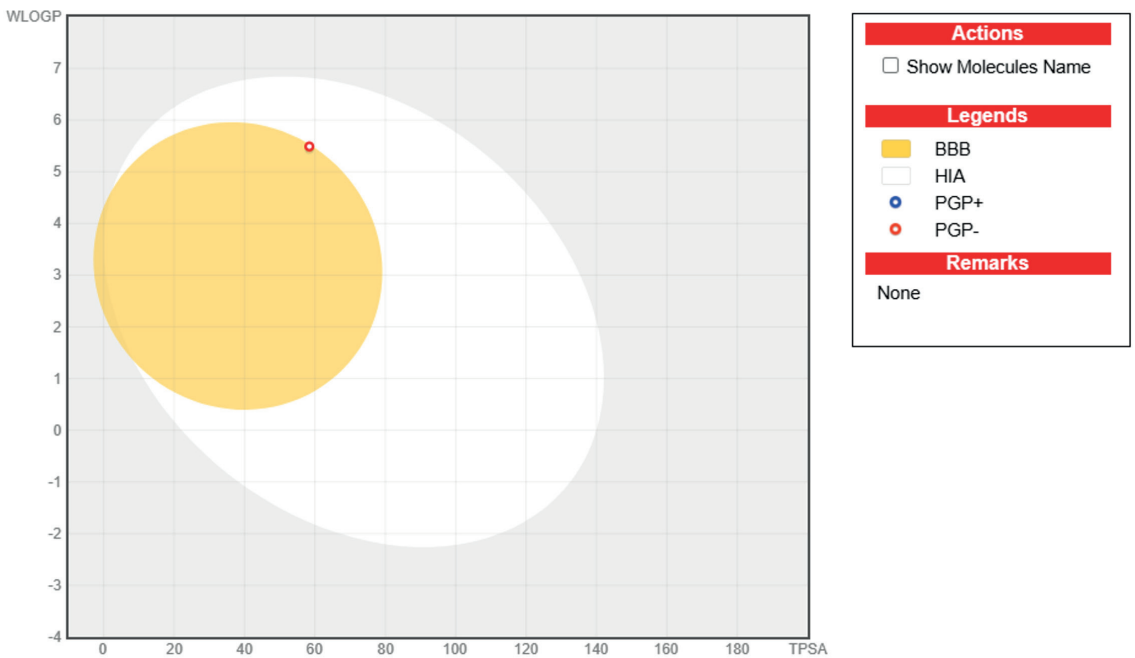


Figure 7. Boiled-egg screening of compound **2**

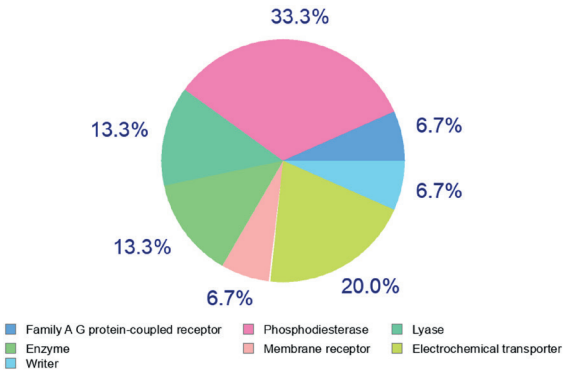


Figure 8. Pie chart showing the SwissTarget prediction of compound **2**

Table 1. Predicted toxic profiles using pkCSM software for compound **2**

	Compound 2
AMES toxicity (Yes/No)	Yes
Max. tolerated dose (human) (log mg/kg/day)	0.224
hERG I inhibitor (Yes/No)	No
hERG II inhibitor (Yes/No)	No
Oral Rat Acute Toxicity (LD_{50}) (mol/kg)	1.853
Oral Rat Chronic Toxicity (log mg/kg_bw/day)	0.359
Hepatotoxicity (Yes/No)	No
Skin Sensitisation (Yes/No)	No
T.Pyriformis toxicity (log ug/L)	0.304
Minnow toxicity (log mM)	0.355

The pkCSM software was also used to predict the toxicity profiles of the compounds and the results are shown in Table 1. The website can provide details of the toxicological effects in the following areas: AMES toxicity, human maximum tolerated dose, hERG-I inhibitor, hERG-II inhibitor, LD_{50} (lethal dose), chronic oral toxicity in rats, hepatotoxicity, skin toxicity, T. pyriformis toxicity and minnow toxicity. The results showed that our compound does not have any hepatotoxicity and is not skin sensitising either. However, it does have AMES toxicity, which means that it has a mutagenic effect. They don't inhibit hERG-I and hERG-II, which is a favorable finding which gives a non-cardiotoxic profile to the compounds. The acute oral toxicity (LD_{50}) of the compounds to rats was found to be 1.853 mol/kg, while the chronic oral toxicity (LOAEL) to rats was found to be 0.359 log mg/kg_bw/day. Toxicity to *T. pyriformis* has been reported in the range of 0.304 log µg/L.

4. CONCLUSION

A new 4-(4-chlorophenyl)-*N*-phenyl-3-(pyridin-4-yl)thiazol-2(*3H*)-imine derivative (**2**) was prepared following the literature knowledge of thiazole ring system synthesis methodology. Spectra of 2D NMR proved that our final product synthesized according to the synthesis scheme. The thiazole ring was closed from the thiourea structure as expected (Scheme 1). The final compound was evaluated for its physicochemical/pharmacokinetic properties as well as druglikeness properties. Lipinski's rule of five and other rules such as Ghose, Veber or Egan were not violated by compound **2**. Compound **2**'s lipophilicity is in the desired range of five different parameters. It was also predicted that compound **2** would not be hepatotoxic and would not be irritating to the skin. This synthesized compound needs to be investigated as a potential antifungal drug candidate because of the structure of the compund similar to the drugs against fungal infections. Further studies will include synthesizing new derivatives, *in vitro* and *in silico* studies.

Ethical approval

Ethics committee approval is not required as there are no in vivo or clinical studies.

Author contribution

Concept, L.Y.; Supervision, L.Y.; Methods, A.Z.K., and L.Y.; Data acquisition and/or processing, A.Z.K., L.Y.; Analysis and/or interpretation, A.Z.K.; Investigation, L.Y.; Writing - original draft preparation, A.Z.K., L.Y.; Critical review, L.Y. The

published version of the manuscript was read and approved by all authors.

Source of funding

This research received no grant from any funding agency/sector.

Conflict of interest

The authors declared that there is no conflict of interest.

REFERENCES

1. Gupta V, Kant V. A review on biological activity of imidazole and thiazole moieties and their derivatives. Science International. 2013;1(7):253-60. https://doi.org/10.17311/sciintl.2013.253.260
2. Kassem AF, Althomali RH, Anwar MM, El-Sofany WI. Thiazole moiety: A promising scaffold for anticancer drug discovery. Journal of Molecular Structure. 2024;1303:137510. https://doi.org/10.1016/j.molstruc.2024.137510
3. Kumari G, Dhillon S, Rani P, Chahal M, Aneja DK, Kinger M. Development in the synthesis of bioactive thiazole-based heterocyclic hybrids utilizing phenacyl bromide. ACS Omega. 2024;9(17):18709-46. https://doi.org/10.1021/acsomega.3c10299
4. Ashmawy FO, Gomha SM, Abdallah MA, Zaki MEA, Al-Hussain SA, El-Desouky MA. Synthesis, in vitro evaluation and molecular docking studies of novel thiophenyl thiazolyl-pyridine hybrids as potential anticancer agents. Molecules. 2023;28(11). https://doi.org/10.3390/molecules28114270
5. Chhabria MT, Patel S, Modi P, Brahmkshatriya PS. Thiazole: A review on chemistry, synthesis and therapeutic importance of its derivatives. Curr Top Med Chem. 2016;16(26):2841-62. https://doi.org/10.2174/1568026616666160506130731
6. Petrou A, Fesatidou M, Geronikaki A. Thiazole ring-a biologically active scaffold. Molecules. 2021;26(11). https://doi.org/10.3390/molecules26113166
7. Patil AR, Yallur BC, Chinnam S, Ananthnag GS, Santhosh CR, Pant G, et al. Pharmaceutical perspectives of thiazole analogues: An overview. Results in Chemistry. 2024;12:101820. https://doi.org/10.1016/j.rechem.2024.101820
8. Alamshany ZM, Nossier ES. New thiazole derivatives linked to pyridine, fused pyridine, pyrimidine and thiazolopyrimidine scaffolds with potential dual anticancer and antimicrobial activities: Design, synthesis and docking simulation. Journal of Molecular Structure. 2024;1316:138973. https://doi.org/10.1016/j.molstruc.2024.138973
9. Niu ZX, Wang YT, Zhang SN, Li Y, Chen XB, Wang SQ, et al. Application and synthesis of thiazole ring in clinically approved drugs. Eur J Med Chem. 2023;250:115172. https://doi.org/10.1016/j.ejmech.2023.115172
10. Ayati A, Emami S, Asadipour A, Shafiee A, Foroumadi A. Recent applications of 1,3-thiazole core structure in the identification of new lead compounds and drug discovery. Eur J Med Chem. 2015;97:699-718. https://doi.org/10.1016/j.ejmech.2015.04.015
11. Sharma PC, Bansal KK, Sharma A, Sharma D, Deep A. Thiazole-containing compounds as therapeutic targets for cancer therapy. Eur J Med Chem. 2020;188:112016. https://doi.org/10.1016/j.ejmech.2019.112016
12. Singh IP, Gupta S, Kumar S. Thiazole compounds as antiviral agents: An update. Med Chem. 2020;16(1):4-23. https://doi.org/10.2174/1573406415666190614101253
13. Helal MH, Salem MA, El-Gaby MS, Aljahdali M. Synthesis and biological evaluation of some novel thiazole compounds as potential anti-inflammatory agents. Eur J Med Chem. 2013;65:517-26. https://doi.org/10.1016/j.ejmech.2013.04.005
14. Duc DX, Chung NT. Recent Development in the Synthesis of Thiazoles. Curr Org Synth. 2022;19(6):702-30. https://doi.org/10.2174/1570179419666220216122637
15. Narasimhamurthy KH, Sajith AM, Joy MN, Rangappa KS. An overview of recent developments in the synthesis of substituted thiazoles. ChemistrySelect. 2020;5(19):5629-56. https://doi.org/10.1002/slct.202001133
16. Singh A, Malhotra D, Singh K, Chadha R, Bedi PMS. Thiazole derivatives in medicinal chemistry: Recent advancements in synthetic strategies, structure activity relationship and pharmacological outcomes. Journal of Molecular Structure. 2022;1266:133479. https://doi.org/10.1016/j.molstruc.2022.133479
17. Kassab AE, Shahin IG, Mohamed KO, Taher AT, Mayhoub AS. Recent advances in the synthesis of thiazole ring: Mini review. Mini-Reviews in Organic Chemistry. 2023;20(3):270-84. https://doi.org/10.2174/1570193x19666220413104255
18. Villa-Reyna AL, Perez-Velazquez M, Gonzalez-Felix ML, Galvez-Ruiz JC, Gonzalez-Mosquera DM, Valencia D, et al. The structure-antiproliferative activity relationship of pyridine derivatives. Int J Mol Sci. 2024;25(14). https://doi.org/10.3390/ijms25147640

19. Chemboli R, Kodali US, Taneja AK, Bandaru V, Mandava BT, Suryadevara V, et al. Sonochemical replacement of C-3 hydrogen of indole by a pyridine ring: Docking, synthesis and in vitro evaluation of 3-(6-aryl pyridin-2-yl)indoles against SIRT1. Journal of Molecular Structure. 2024;1298:137025. https://doi.org/10.1016/j.molstruc.2023.137025

20. Mohammad Abu-Taweel G, Ibrahim MM, Khan S, Al-Saidi HM, Alshamrani M, Alhumaydhi FA, et al. Medicinal importance and chemosensing applications of pyridine derivatives: A review. Crit Rev Anal Chem. 2024;54(3):599-616. https://doi.org/10.1080/10408347.2022.2089839

21. Elsayed MA, Elsayed AM, Sroor FM. Novel biologically active pyridine derivatives: Synthesis, structure characterization, in vitro antimicrobial evaluation and structure-activity relationship. Medicinal Chemistry Research. 2024;33(3):476-91. https://doi.org/10.1007/s00044-024-03188-1

22. Ling Y, Hao ZY, Liang D, Zhang CL, Liu YF, Wang Y. The expanding role of pyridine and dihydropyridine scaffolds in drug design. Drug Des Devel Ther. 2021;15:4289-338. https://doi.org/10.2147/DDDT.S329547

23. Abd El-Lateef HM, Khalaf MM, Gouda M, Kandeel M, Amer AA, Abdelhamid AA, et al. Functionalized pyridines: Synthesis and toxicity evaluation of potential insecticidal agents against aphis craccivora. ACS Omega. 2023;8(32):29685-92. https://doi.org/10.1021/acsomega.3c03831

24. Failla M, Lombardo GW, Orlando P, Fiorito D, Bombonato E, Ronchi P, et al. Late-stage functionalisation of pyridine-containing bioactive molecules: Recent strategies and perspectives. European Journal of Organic Chemistry. 2023;26(16). https://doi.org/10.1002/ejoc.202300074

25. Guchhait SK, Hura N, Sinha K, Panda D. Pyridine C3-arylation of nicotinic acids accessible via a multicomponent reaction: An entry to all-substituted-3,4-diarylated pyridines. RSC Advances. 2017;7(14):8323-31. https://doi.org/10.1039/c6ra28299g

26. Amer AA, Abdelhamid AA, Elnakeeb AS, Salah HA. One-pot multicomponent designing of novel 2-imino-4-arylidene-1,3-thiazolidin-4-one. Journal of Heterocyclic Chemistry. 2022;60(3):489-96. https://doi.org/10.1002/jhet.4604

27. Bodhak C, Mandal S, Dey P, Mukherjee SK, Pramanik A. Efficient synthesis of functionalized 2-iminothiazolines by ultrasonication under solvent-free conditions and access to 5-aryl-2-iminothiazolines. Results in Chemistry. 2022;4:100301. https://doi.org/10.1016/j.rechem.2022.100301

28. Berber N, Şahutoğlu AS, Gökçe B, Çıkrıkçı K, Gençer N. Synthesis, characterization, carbonic anhydrase inhibitor activity, and docking studies of phenylthiazol-2(3h)-ylidene-isoquinoline-5-amine derivatives. Journal of Molecular Structure. 2023;1291:136061. https://doi.org/10.1016/j.molstruc.2023.136061

29. Turan-Zitouni G, Özdemir A, Kaplancıklı ZA. Synthesis and antiviral activity of some (3,4-diaryl-3h-thiazol-2-ylidene)pyrimidin-2-yl amine derivatives. Phosphorus, Sulfur, and Silicon and the Related Elements. 2011;186(2):233-9. https://doi.org/10.1080/10426507.2010.494643

30. Haji Ali S, Osmaniye D, Saglik BN, Levent S, Ozkay Y, Kaplancikli ZA. Design, synthesis, investigation, and biological activity assessments of (4-substituted-phenyl)-n-(3-morpholinopropyl)-3-phenylthiazol-2(3h)-imine derivatives as antifungal agents. ACS Omega. 2024;9(38):39326-43. https://doi.org/10.1021/acsomega.3c07879

31. SwissADME SwissDrugDesign: Molecular Modelling Group of the University of Lausanne and the SIB Swiss Institute of Bioinformatics; [Available from: http://www.swissadme.ch/].

32. SwissTargetPrediction SwissDrugDesign: The Molecular Modelling Group of the University of Lausanne and the SIB Swiss Institute of Bioinformatics; [Available from: http://www.swisstargetprediction.ch/].

33. Daina A, Michielin O, Zoete V. SwissTargetPrediction: updated data and new features for efficient prediction of protein targets of small molecules. Nucleic Acids Res. 2019;47(W1):W357-W64. https://doi.org/10.1093/nar/gkz382

34. pkCSM. Pharmacokinetic properties: Instituto Rene Rachou Fiocruz Mınas, The University of Melbourne, University of Cambridge; [Available from: https://biosig.lab.uq.edu.au/pkcsm/].