FABAD J. Pharm. Sci., 50, 1, 15-20, 2025
Doi: 10.55262/fabadeczacilik.1577453
RESEARCH ARTICLE

N-Alkylation of Some Imidazopyridines

Fatima DOGANC[*], Hakan GOKER[**°]

N-Alkylation of Some Imidazopyridines

SUMMARY

6-Bromo-2-(4-(4-fluorophenoxy)phenyl)-4H-imidazo[4,5-b] pyridine (I) and 2-[4-(4-fluorophenoxy)phenyl]-5H-imidazo[4,5-c] pyridine (III) were prepared by the reaction of 5-bromo-2,3-diaminopyridine and 3,4-diaminopyridine with sodyum metabisulfite adduct of 4-(4-fluorophenoxy)benzaldehyde (1), respectively. Alkylation of these compounds with 1-(chloromethyl)-4-methoxybenzene under basic conditions (K2CO3 in DMF) was formed as mainly N4 regioisomer (II) and N5 regioisomer (IV). Their regioisomeric structures were assigned with 2D-NOESY (Nuclear Overhauser Effect Spectroscopy) spectra.

Key Words: *Imidazopyridines, NOESY, regioisomers*

Bazı İmidazopiridinlerin N-Alkilasyonu

ÖZ

5-Bromo-2,3-diaminopiridin ve 3,4-diaminopiridinin, 4-(4-florofenoksi)benzaldehitin (1) sodyum metabisülfit tuzuyla ile reaksiyona sokulmasıyla sırasıyla 6-bromo-2-(4-(4-florofenoksi) fenil)-4H-imidazo[4,5-b]piridin (I) ve 2-[4-(4-florofenoksi)fenil]-5H-imidazo[4,5-c]piridin (III) bileşikleri elde edildi. Bu bileşiklerin 1-(klorometil)-4-metoksibenzen ile bazik koşullar altında (DMF içinde K2CO3) alkilasyonuyla esas olarak N4 regioizomeri (II) ve N5 regioizomeri (IV) oluştu. Regioizomerik yapıları 2D-NOESY (Nükleer Overhauser Etki Spektroskopisi) spektrumları ile belirlendi.

Anahtar Kelimeler: *İmidazopiridin, NOESY, regioizomer*

Received: 01.11.2024
Revised: 12.11.2024
Accepted: 26.11.2024

[*] ORCID: 0000-0002-1832-587X: Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Ankara University, Ankara, Türkiye.
[**] ORCID: 0000-0002-9366-6949: Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Ankara University, Ankara, Türkiye.

[°] Corresponding Author; Hakan Göker
E-mail: goker@ankara.edu.tr

INTRODUCTION

Imidazopyridines, one of the most common heterocycles, are primarily used in medicinal chemistry, since they have potent several biological activities. The pharmacological profiles of imidazopyridines have been mentioned in many literatures, such as antibacterial, anti-inflammatory, antipyretic, analgesic, antiapoptotic, antitumor, antifungal, hypnotic, antiviral, and antiprotozoal agents (Dyminska, 2015; Krause et al., 2017; Volpi, et al., 2024). Imidazopyridine scaffolds are formed by condensing imidazole and pyridine rings. In these condensed systems, the nitrogen bears a hydrogen atom ($N^{1,3}$) as a pyrrole-like *N*-atom; the others ($N^{4,5}$) resembles a pyridine-like *N*-atom. Hydrogen atom attached to nitrogen in the 1, 3, 4 and 5th position readily tautomerise in several positions depicted in Figure 1.

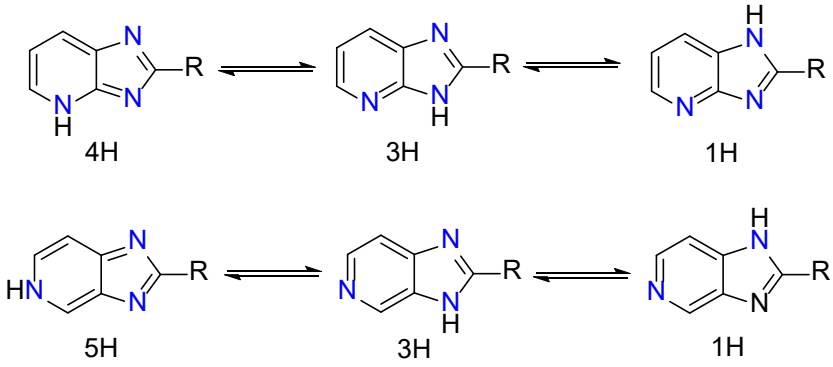


Figure 1. Tautomeric forms of imidazo[4,5-*b*]pyridine and imidazo[4,5-*c*]pyridine moieties

These relocations are entirely lost when the mobile hydrogen in imidazopyridines is replaced by any alkyl groups.

In our recently published papers, we have characterized the occurrence and structures of various regioisomers of imidazopyrimidines, imidazopyridines, imidazopyrazines, benzimidazoles and indazoles (Göker and Özden, 2019; Doganc et al., 2020; Karaaslan et al., 2020; Puskullu et al., 2021; Doganc and Göker, 2024). For this purpose, we used advanced 2D-NMR techniques for the structural elucidation. In continuation of these works, we now report, *N*-alkylation reaction of some imidazopyridines with 4-methoxybenzyl chloride, for investigation of the forming possible regioisomers. The 2D-NOESY (Nuclear Overhauser Effect Spectroscopy) technique was used for the structural elucidation.

MATERIAL AND METHODS

Uncorrected melting points were measured on a Büchi B-540 capillary melting point apparatus. ^{1}H (500 MHz) and ^{13}C (125 MHz) NMR spectra were recorded employing BRUKER AVANCE NEO 500 MHz FT spectrometer, chemical shifts (δ) are in ppm relative to TMS. The samples (5-10 mg) were prepared in 0.75 ml of DMSO-d_6. The liquid chromatography mass spectrometry (LC-MS) spectra were taken on a Waters Micromass ZQ connected with Waters Alliance HPLC (Waters Corporation), using the ESI (+) method with a C-18 column (XTerra®, 4.6 X 250 mm, 5 µm).

Synthesis of sodium metabisulfite adduct of 4-(4-fluorophenoxy)benzaldehyde (1)

4-(4-Fluorophenoxy)benzaldehyde (30 mmol) was dissolved in EtOH (100 ml) and sodium metabisulfite (3.2 g) (in 5 ml of water) was added in portions. The reaction mixture was stirred vigorously. The mixture was kept in a refrigerator for a while. The precipitate was gained by filtration, dried and used for the further steps without purification and characterisation.

General Synthesis of I and III

The mixture of related *o*-pyridine-diamine derivatives (1 mmol) and $Na_2S_2O_5$ adduct of 4-(4-fluorophenoxy)benzaldehyde (1 mmol) in DMF (0.5 ml) were heated at 130°C, for 4 h. The reaction mixture was cooled, poured into water. The resulting precipitate was collected by filtration washed with water and dried. The resulting precipitate was crystallized from EtOH.

6-Bromo-2-(4-(4-fluorophenoxy)phenyl)-4*H*-imidazo[4,5-*b*]pyridine (I)

Prepared from 5-bromopyridine-2,3-diamine (0.188 g) and $Na_2S_2O_5$ adduct of 4-(4-fluoro-phenoxy) benzaldehyde (0.320 g) as described in the general method. It was triturated with hot EtOH. Yield 0.286 g, 75%, m.p. = 310-312°C. **^{1}H-NMR** δ ppm (DMSO-d_6+one drop of TFA) : 7.18-7.19 (m, 4H), 7.23-7.26 (m, 2H), 8.24 (d, 2H, *J*=8.95 Hz), 8.49 (d, 1H, *J*=2 Hz), 8.64 (d, 1H, *J*=2Hz) ; **^{13}C-NMR** δ ppm (DMSO-d_6 + one drop of TFA) : 162.35, 159.6 (d, *J*=240 Hz), 152.8, 150.9 (d, *J*=2.5 Hz), 146.29, 146.21, 130.8, 128.6, 125.9, 122.5 (d, *J*=8.7 Hz), 118.0, 117.2 (d, *J*=23 Hz, 115.5. **MS** (ESI+) m/z : 384 (M+H, 100%), 386 (M+H+2, 98%), $C_{18}H_{11}BrFN_3O$.

2-[4-(4-fluorophenoxy)phenyl]-5*H*-imidazo[4,5-*c*]pyridine (III)

Prepared from pyridine-3,4-diamine (0.109 g) and $Na_2S_2O_5$ adduct of 4-(4-fluoro-phenoxy)benzaldehyde (0.320 g) as described in the general method. It was triturated with hot EtOH. Yield 0.12 g, 39%, m.p. = 263-266°C. **^{1}H-NMR** δ ppm (DMSO-d_6+one drop of TFA) : 7.16-7.22 (m, 4H), 7.26-7.29 (m, 2H), 8.11 (d, 1H, *J*=6.45 Hz), 8.29 (d, 2H, *J*=8.9 Hz), 8.58 (d, 1H, *J*=6.55 Hz), 9.40 (s, 1H) ; **^{13}C-NMR** δ ppm (DMSO-d_6+ one drop of TFA) : 161.3, 159.4 (d, *J*=239 Hz), 158.9, 151.4 (d, *J*=2 Hz), 147.5, 139.3, 133.7, 132.5, 130.4, 122.6, 122.4 (d, *J*=8.44 Hz), 118.8, 117.2 (d, *J*=23.2 Hz, 115.15. **MS** (ESI+) m/z : 306 (M+H, 100%), $C_{18}H_{12}FN_3O$.

General Synthesis of II and IV

K_2CO_3 (1 mmol, 0.138 g) was added to a suspension of the **I** or **III** (0.5 mmol) in DMF (0.7 ml) and stirred. One hour later, 1-(chloromethyl)-4-methoxybenzene (0.6 mmol, 0.094 g) was added. After overnight stirring at room temperature, water was added and precipitate was filtered.

6-Bromo-2-(4-(4-fluorophenoxy)phenyl)-4-(4-methoxybenzyl)-4*H*-imidazo[4,5-*b*]pyridine (II)

Prepared from **I** (0.192 g), 1-(chloromethyl)-4-methoxybenzene and K_2CO_3 as described in the general method. The crude product was crystallised from EtOH. Yield 0.181 g, 72%, m.p. = 202-204°C. **^{1}H-NMR** δ ppm (DMSO-d_6): 3.70 (s, 3H, -OC$\underline{H}_3$), 5.78 (s, 2H, benzylic -C$\underline{H}_2$), 6.92 (d, 2H, *J*=8.65 Hz, H-3‴,5‴), 7.09 (d, 2H, *J*=8.75 Hz, H-3',5'), 7.17-7.19 (m, 2H, H-2",6"), 7.27-7.30 (m, 2H, H-3",5"), 7.61 (d, 2H, *J*=8.65 Hz, H-2‴,6‴), 8.36 (d, 1H, *J*=1.45 Hz, H-7), 8.40 (d, 2H, *J*=8.75 Hz, H-2',6'), 8.65 (d, 1H, *J*=1.4 Hz, H-5) ; **COSY** : [H-2',6'/ H-3',5'], [H-2",6"/ H-3",5"], [H-2‴,6‴/ H-3‴,5‴] ; **NOESY** : [N-C$\underline{H}_2$ / H-5 & H-2‴,6‴], [-OC$\underline{H}_3$ / H-3‴,5‴] ; **^{13}C-NMR & HSQC** δ ppm (DMSO-d_6) : 169.6, 159.9, 159.55, 159.0 (d, *J*=239 Hz, C-4"), 153.55, 152.3 (d, *J*=2 Hz, C-1"), 146.5, 130.93 (C-5H), 130.86 (C-2‴,6‴H), 130.3 (C-2',6'H), 129.7 (C-7H), 129.6, 127.9, 121.9 (d, *J*=7.5 Hz, C-2",6"H), 118.1 (C-3',5'H), 117.2 (d, *J*=22.5 Hz, C-3",5"H), 114.6 (C-3‴,5‴H), 105.8, 56.2 (benzylic -$\underline{C}H_2$), 55.6 (-O$\underline{C}H_3$). **MS** (ESI+) m/z : 504 (M+H, 100%), 506 (M+H+2, 98%), $C_{26}H_{19}BrFN_3O_2$.

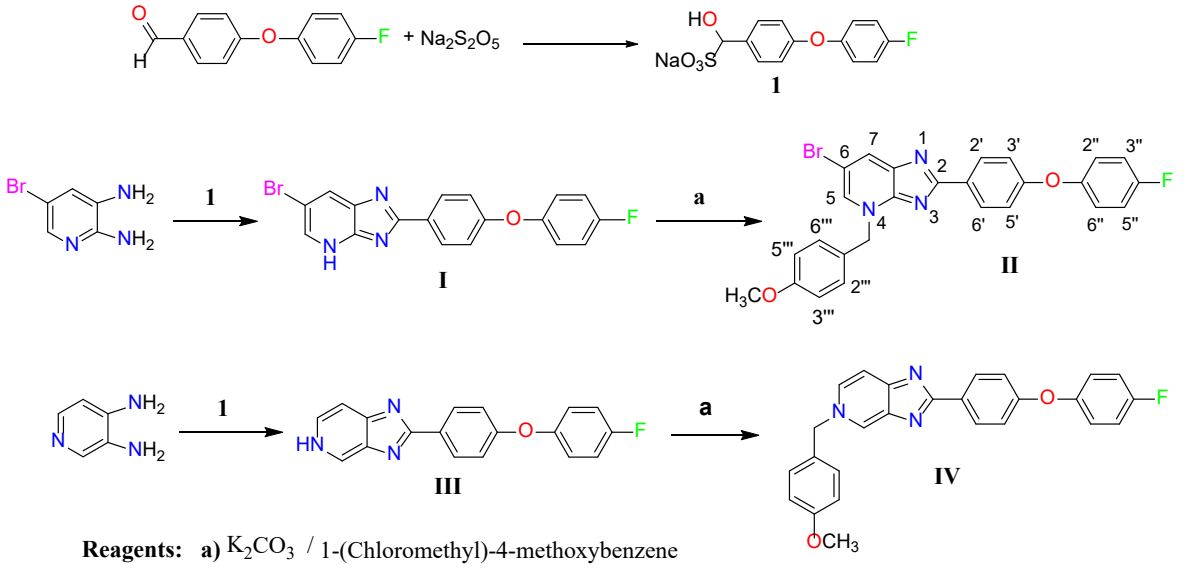
2-(4-(4-Fluorophenoxy)phenyl)-5-(4-methoxybenzyl)-5*H*-imidazo[4,5-*c*]pyridine (IV)

Prepared from **III** (0.153 g), 1-(chloromethyl)-4-methoxybenzene and K_2CO_3 as described in the general method. The crude product was crystallized from EtOAc : *n*-Hexane. Yield 0.11 g, 52 %, m.p. = 196-198°C. **^{1}H-NMR** δ ppm (DMSO-d_6): 3.73 (s, 3H, -OC$\underline{H}_3$), 5.56 (s, 2H, benzylic -C$\underline{H}_2$), 6.95 (d, 2H, *J*=8.7 Hz, H-3‴,5‴), 7.05 (d, 2H, *J*=8.85 Hz, H-3',5'), 7.15-7.17 (m, 2H, H-2",6"), 7.25-7.28 (m, 2H, H-3",5"), 7.46 (d, 2H, *J*=8.65 Hz, H-2‴,6‴), 7.69 (d, 1H, *J*=6.75 Hz, H-7), 8.16 (dd, 1H, *J*=6.75 & 1.4 Hz, H-6), 8.36

(d, 2H, *J*=8.7 Hz, H-2',6'), 9.06 (s, 1H, H-4) ; **COSY** : [H-6 / H-7], [H-2',6'/ H-3',5'], [H-2",6"/ H-3",5"], [H-2"',6"'/ H-3"',5"'] ; **NOESY** : [N-C$\underline{H}_2$ / H-4 & H-6 & H-2"',6"'], [-OC$\underline{H}_3$ / H-3"',5"'] ; **^{13}C-NMR & HSQC** δ ppm (DMSO-d_6) : 171.2, 159.9, 158.94 (d, *J*=239 Hz, C-4"), 158.87, 156.2, 152.5 (d, *J*=2 Hz, C-1"), 145.9, 131.3 (C-4H), 131.1 (C-6H), 130.6, 130.2 (C-2"',6"'H), 129.9 (C-2',6'H), 128.9, 121.7 (d, *J*=8.6 Hz, C-2",6"H), 118.0 (C-3',5'H), 117.1 (d, *J*=23.1 Hz, C-3",5"H), 114.8 (C-3"',5"'H), 112.55 (C-7H), 61.2 (benzylic -$\underline{C}H_2$), 55.6 (-O$\underline{C}H_3$). **MS** (ESI+) m/z : 426 (M+H, 100%), $C_{26}H_{20}FN_3O_2$.

RESULTS AND DISCUSSION

Targeted compounds were prepared using the methods outlined in Scheme 1. Cyclization of 5-bromo -2,3-diaminopyridine and 3,4-diaminopyridine with sodium metabisulfite adduct of 4-(4-fluorophenoxy)benzaldehyde (**1**) gave required imidazo[4,5-*b*] pyridines (**I**) and imidazo[4,5-*c*]pyridines (**III**), respectively. This group exhibits rapid prototrophic tautomerism, resulting in equilibrium mixtures. Due to these tautomeric forms (Figure 1.) both ^{1}H and ^{13}C-NMR spectra of unsubstituted analogues (**I** and **III**) may not be sufficiently clear. It is typical for some proton and carbon signals to appear as broad peaks and even certain hinge carbon signals may be undetectable. To address this, we utilized trifluoroacetic acid for running proton and carbon NMR spectra. The removal of the NH proton and subsequent substitution of this nitrogen atom would inhibit rapid tautomerism, leading to a separable mixture of regioisomers. When we have attempted alkylation of **I** and **III** with 1-(chloromethyl)-4-methoxybenzene under basic conditions (K_2CO_3, DMF), alkylation were formed as only N^4 **(II)** and N^5 **(IV)** position. Interestingly, we have never detected other possible regioisomers. The characterization of the individual isomeric products was achieved by observing the 2D-NOESY enhancements between the *N*-CH_2 and pyridine aromatic protons. As it is well known, NOESY is a valuable 2D NMR technique for identifying signals of protons in close spatial proximity (4-5 A° in distance) even if they are not directly bonded. In the NOESY spectrum of compound **II,** strong correlations have been observed between the benzylic and H-5 protons (Figure 2.). This finding showed us that, the synthesized final targeted compound **II** is the N^4 regioisomer form. Similarly, very strong NOE enhancements were seen between *N*-CH_2 and H-4,6 in the NOESY spectra of **IV** (Figure 3.).



Scheme 1. Synthesis of targeted compounds

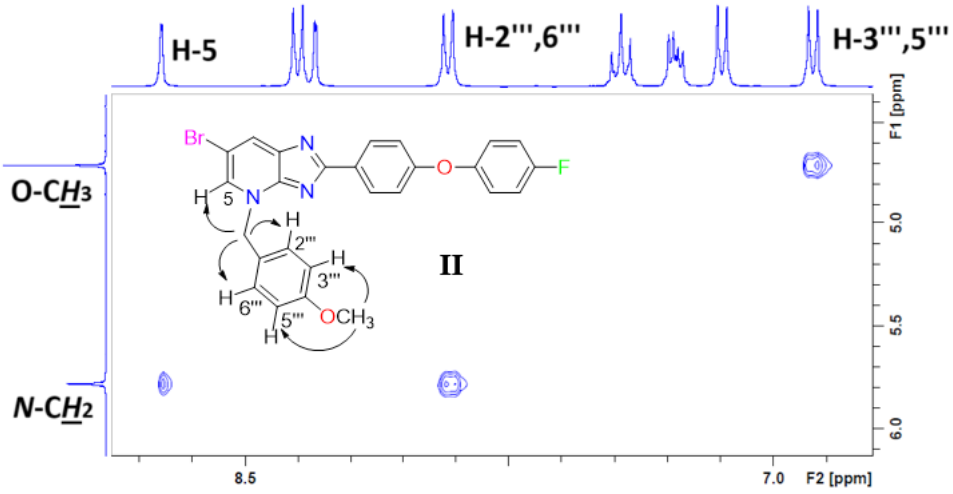


Figure 2. Partial NOESY spectrum of compound **II**

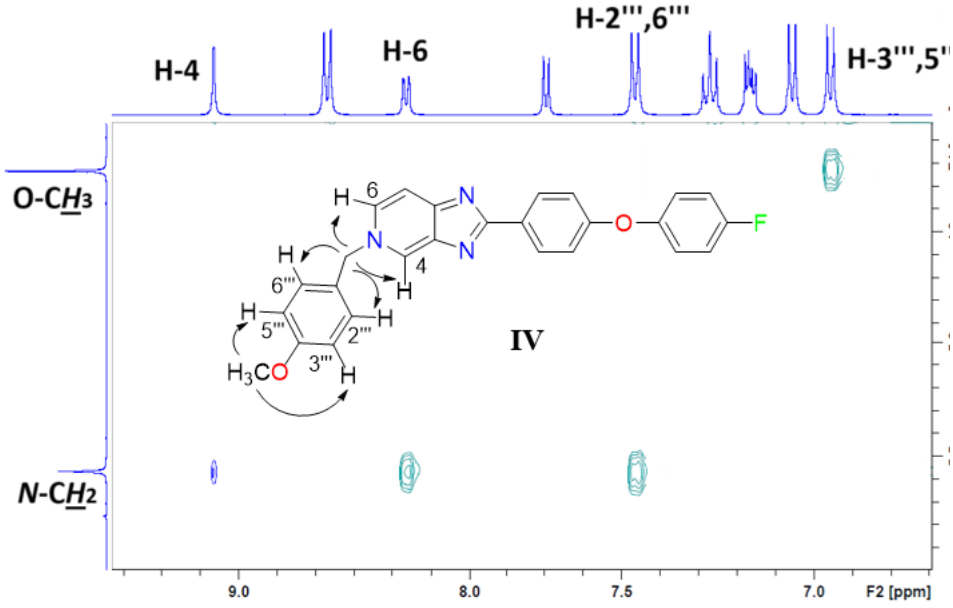


Figure 3. Partial NOESY spectrum of compound **IV**

CONCLUSION

N-alkylation of 4*H*-imidazo[4,5-*b*]pyridines and 5*H*-imidazo[4,5-*c*]pyridines were mainly realized on the nitrogen atoms of pyridine ring, in the presence of anhydrous K_2CO_3 in DMF with 4-methoxybenzyl chloride. 2D-NOESY experiment is the best method for structural elucidation of these types of regioisomers. The structure elucidation of synthesized compounds were performed using 1D and 2D NMR experiments including COSY, NOESY, gHSQC.

ACKNOWLEDGEMENTS

Central Laboratory of Pharmacy, Faculty of Ankara University provided support for acquisition of NMR and mass spectrometer used in this work.

AUTHOR CONTRIBUTION STATEMENT

Fatima Doganc and Hakan Goker were responsible for conducting developing the hypothesis, literature research, performing experiments, preparing and reviewing manuscript.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

REFERENCES

Doganc, F., Aydin, A. S., Şahin, E., Göker, H. (2020). Regioselective *N*-alkylation of some 2 or 6-chlorinated purine analogues. *Journal of Molecular Structure, 1272,* 134200. doi : 10.1016/j.molstruc.2022.134200

Doganc, F., Göker, H. (2024). Differentiation of regioisomeric *N*-alkylation of some indazoles and pyrazolopyridines by advanced NMR techniques. *Magnetic Resonance in Chemistry, 62,* 765-774. doi : 10.1002/mrc.5471

Dyminska, L. (2015). Imidazopyridines as a source of biological activity and their pharmacological potentials infrared and raman spectroscopic evidence of their content in pharmaceuticals and plant materials. *Bioorganic & Medicinal Chemistry, 23,* 6087-6099. doi : 10.1016/j.bmc.2015.07.045

Göker, H., Özden, S. (2019). Regioselective *N*-alkylation of 2-(3,4-dimethoxyphenyl)imidazo[4,5-*b*] and [4,5-*c*]pyridine oxide derivatives: Synthesis and structure elucidation by NMR. *Journal of Molecular Structure, 1197,* 183-195. doi : 10.1016/j.molstruc.2019.07.058

Karaaslan, C., Doganc, F., Alp, M., Koc, A., Karabay, A. Z., Göker, H. (2020). Regioselective *N*-alkylation of some imidazole-containing heterocycles and their *in vitro* anticancer evaluation. *Journal of Molecular Structure, 1205,* 127673. doi : 10.1016/j.molstruc.2019.127673

Krause, M., Foks, H., Gobis, K. (2017). Pharmacological potential and synthetic approaches of imidazo[4,5-*b*]pyridine and imidazo[4,5-*c*]pyridine derivatives. *Molecules 22,* 399. doi : 10.3390/molecules22030399

Puskullu, M. O., Doganc, F., Ozden, S., Sahin, E., Celik, I., Göker, H. (2021). Synthesis, NMR, X-ray crystallography and DFT studies of some regioisomers possessing imidazole heterocycles. *Journal of Molecular Structure, 1243,* 130811. doi : 10.1016/j.molstruc.2021.130811

Volpi, G., Laurenti, E., Rabezzana, R. (2024). Imidazopyridine family: Versatile and promising heterocyclic skeletons for different applications. *Molecules, 29(*11*),* 2668. doi : 10.3390/molecules29112668