

ANTIVIRAL PROPERTIES OF 1,3,4-OXADIAZOLE DERIVATIVES AGAINST SARS-2

Shama Parveen¹, Rishabh Bhardwaj², Madhu Sudan³, Vikas Gupta^{1*},

¹Department of Chemistry, Motherhood University, Roorkee

²Research Scholar, Department of Zoology and Environmental Science, Gurukula Kangri
(Deemed to be University), Haridwar

³Department of Chemistry, R.S.M. Degree College, Dhampur, Bijnor, Uttar Pradesh

*Corresponding Author: Dr. Vikas Gupta (drvikasgupta2510@gmail.com)

Article History

Received: 26/07/2025

Accepted: 05/08/2025

Article ID: RRBB/02_2025

Corresponding Author:

E-Mail:

drvikasgupta2510@gmail.com

ABSTRACT

We describe the manufacture and biological evaluation of new, potentially effective antibiotics against viruses that are 1,3,4-oxadiazole substituted derivatives. Using a combination of mass spectrometry, ¹H NMR, IR, UV, and ¹³C NMR techniques, the structures of the recently manufactured derivatives were established. Since there aren't many clinically authorized treatments for the COVID-19 pandemic yet, new ones must be developed by mid-2022. This study examined three 1,3,4-oxadiazole derivatives' ability to stop the SARS-2 virus from multiplying in the cultivation of cells. One of the compounds described did not effectively stop the SARS-2 virus from reproducing.

Keywords: antiviral action, 1,3,4-oxadiazole, &1,3,4-oxadiazole derivatives.

INTRODUCTION

Heterocyclic compounds known as oxadiazoles include one O atom and two N atoms in a 5-membered ring [1, 2] and have several positive effects on life [3]. Oxadiazole is believed to be the final result of furan by substituting two nitrogen atoms of the pyridine type (-N=) for two methane (-CH=) groups [2]. The literature has described 1,3,4-oxadiazoles is a thermally stable, impartial heterocyclic molecular structure (65JA5800).

Among these aromatic systems are mesoionic, exocyclic-conjugated 1,3,4-oxadiazolium cations

(2), 1,3,4-oxadiazoles (3), and 1,3,4-oxadiazolines (4). Additionally, 2,3-dihydro-1,3,4-oxadiazol is a nonaromatic reduced system. Furthermore, 2,3-dihydro-1,3,4-oxadiazole (Δ^2 -1,3,4-oxadiazoline; 5), 2,5-dihydro-1,3,4-oxadiazole (Δ^3 -1,3,4-oxadiazoline; 6), & 2,3,4,5-tetrahydro-1,3,4-oxadiazole (1,3,4-oxadiazolidine; 7) are derivatives of the non-aromatic reduced systems.

A thorough review < 66AHC(7)183 > surveys the literature previous to 1965; further helpful reviews < B-61MI42300, 62HC(17)263, 64RCR508 > are also accessible.

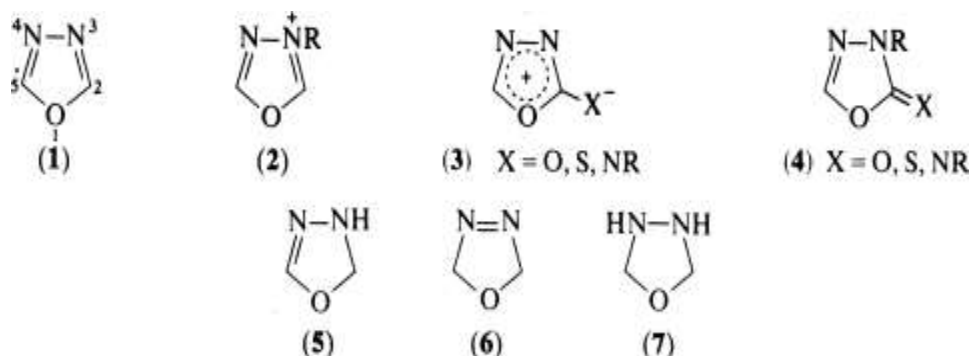


Figure 1; derivatives of 1,3,4 oxadiazoles derrivatives

The most common methods for creating 1,3,4-oxadiazoles are either cyclizing diacylhydrazines with a range of dehydrating substances, including polyphosphoric acid [7], polyphosphoric acid [8–11], the phosphorous pentoxide [5], thionyl chloride [4], phosphorous oxychloride [3], trifolic anhydride [6], and acid hydrazides (or hydrazine) reacting alongside acid chlorides/carboxylic acidic substances.

There is an almost endless supply of medications and other physiologically active molecules available from heterocyclic compounds.

This new coronavirus infection, known as COVID-19, is caused by the highly pathogenic severe acute breathing syndrome virus coronavirus-2 (SARS-2) which has resulted in a significant worldwide emergency. [12]

]. The COVID-19 epidemic continued until the middle of 2022, which made the creation of innovative preventative and treatment strategies necessary. Relatively few drugs with direct antiviral effectiveness are accessible in effective treatment towards SARS-2 [13].

The U.S. FDA has granted Remdesivir comprehensive clinical approval, making it the only medicine to do so [14] (Figure 1).

Moreover, paxlovid, a medication that combines nirmatrelvir and ritonavir, was given an Emergency Use Authorization (EUA) [15] and

Figure 1. However, Paxlovid's EUA was recently revoked since it appeared to be unaffected for the Omicron variant of SARS -2, which is the most prevalent strain in circulation presently.

Favipiravir is toxic at therapeutic levels [17] and it has demonstrated limited therapeutic efficacy in clinical studies; yet, many historical countries have licensed the medicine [16] (Figure 1). By specifically identifying viral proteins, attaching to them, and preventing the activities of viral enzymes, The drugs listed above, directly act as antivirals (DAA) that fight SARS-2.

Finding novel lead compounds that fall into the practical need is to look for novel lead compounds that fall within the DAA class. In order to discover new lead compounds, we tested compounds from the epoxy benzo oxocino pyridine class for SARS-2 viral activity in this work.

CHEMISTRY

The substituted aromatic acids served as a flexible platform to prepare derivatives of 1,3,4-oxadiazoles, which involved the hydrazide and ester production of related compounds. The Fischer esterification procedure was used to create ethyl esters, which then combined with the hydrate of hydrazine in the absence of ethanol to produce the appropriate hydrazide equivalent. This method of production replaced aromatic acids. The resulting final compounds 6(a–h) (Fig

1) were obtained by the reaction of the hydrazide derivatives products with β -benzoyl propionic acid and water with phosphorus oxychloride, a cyclodehydrating agent.

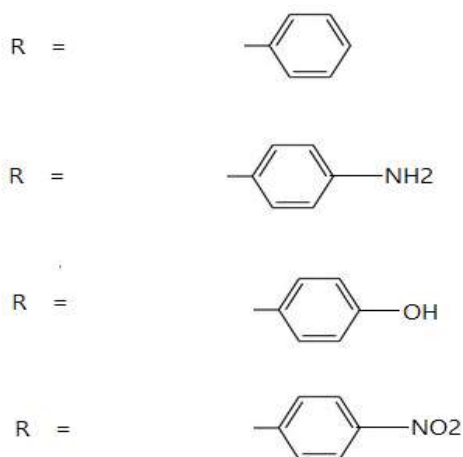
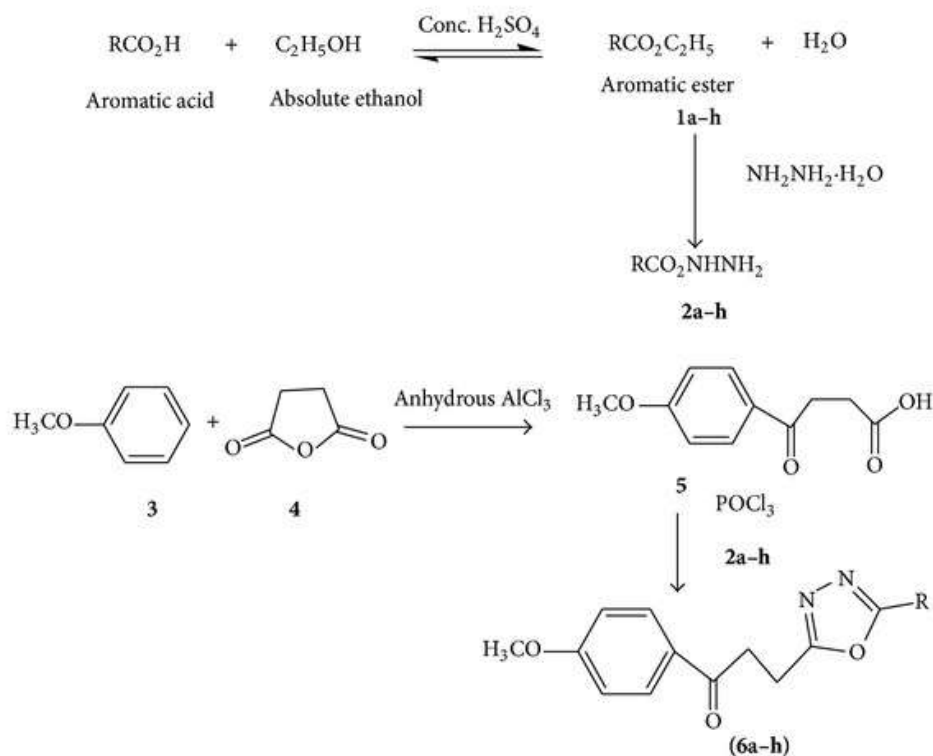
A Comprehensive Process for Manufacturing 1-(4-Methoxyphenyl)-3-(5-phenyl-1,3,4-oxadiazol-2-yl)propan-1-one (6a-h). After Aryl Hydrazide 2a (1M) was dissolved in 5 mL of phosphorous oxychloride, compound 5 (1M) was added. Following six to seven hours of refluxing, the

resultant fluid was subjected to cooling at ambient temperature and then poured on top of crushed ice.

A solid mass emerged following the neutralization of the contents with a 20% sodium bicarbonate solution. This underwent a water wash after filtering. It was crystallized using methanol, yielding 6a. The production of compounds (6b-h) was similar (Figure 1)[18-19].

Table 1 gives the corresponding R for 6a.

Compound	R	CC ₅₀ ^a (μ M)	IC ₅₀ ^b (μ M)	SI ^c	IC ₅₀ SARS-2 M ^{pro} (μ M)
1-(4-Methoxyphenyl)-3-(5-phenyl-1,3,4-oxadiazol-2-yl)propan-1-one	Methoxy phenyl	275.03	15.2	18.09	2.4+0.1
1-(4-Methoxyphenyl)-3-[5-(4-aminophenyl)-1,3,4-oxadiazol-2-yl]propan-1-one	Amino phenyl	451.5	15.7	28.75	3.1+0.1
1-(4-Methoxyphenyl)-3-[5-(4-hydroxyphenyl)-1,3,4-oxadiazol-2-yl]propan-1-one	Hydroxyl phenyl	153.4	49.2	3.11	5.2+0.2
1-(4-Methoxyphenyl)-3-[5-(4-nitrophenyl)-1,3,4-oxadiazol-2-yl]propan-1-one	Nitro phenyl	352.2	14.3	24.6	2.5+0.1



BIOLOGICAL ACTIVITIES

Identification of cytotoxicity in vitro using an MTT assay that is colorimetric,, the cytotoxicity of compounds 6a, 6b, 6c, and 6h on Vero-E6

lines was assessed [20]. Table 1 showed the range that their safety and CC50 indices, which

National Press Associates. (2025). ANTIVIRAL PROPERTIES OF 1,3,4-OXADIAZOLE DERIVATIVES AGAINST SARS-2 . *Research & Reviews in Biotechnology & Biosciences*, 12(2), 25–32.

<https://doi.org/10.5281/zenodo.16742908>

were 2.40 to 3.10 and 153.2 and 451.5 μM , respectively. It has been found that 6a and 6b, with IC_{50} values of 15.2 and 15.7 μM and SI values of 18.09 and 28.75, are two of the best SARS-2 inhibitors.

Following previously published processes, in vitro bioassays of cytotoxicity [33], antiviral activity [21], [22], [23], [24], method of action(s) [23], and interfering immunoassay against SARS-2 Mpro [25] were carried out.

^a CC_{50} : half maximal cytotoxic conc.

^b IC_{50} : half maximal inhibitory conc.

^cSI: Safety index = $\text{CC}_{50}/\text{IC}_{50}$ for the compound.

ANTI-SARS-2 ACTIVITY

As demonstrated by Table 1's results, the majority of the substances examined considerably and dose-dependently reduced the NRC-03-nhCoV virus's ability to replicate. With IC_{50} values of 15.2, 15.7, 49.2, and 24.6 μM , respectively, compounds 6a, 6b, and 6(a-c) exhibited the greatest antiviral efficacy against SARS-2. The most successful synthetic derivative against Vero-E6 cells was the unsubstituted variant 6a (R = phenyl), with an IC_{50}

value of 15.2 μM . With an IC_{50} of 15.7 μM , the modified derivative 6b (R = amino phenyl) shown potential action towards Vero-E6 cells, corresponding to 6c (R = hydroxyl phenyl) and 6d (nitro phenyl). The previously described methodology [40] was used to assess the inhibitory action of compounds 6a, 6b, 6c, and 6h against SARS-2 Mpro. According to the results displayed in Table 1, the 6a demonstrated encouraging inhibitory effect against SARS-2 Mpro, with a The IC_{50} value of 2.42 μM . The information in Table 1 shows that groupings lower the biological Based on the results shown in Table 1, the 6a demonstrated encouraging inhibition of SARS-2 Mpro, with an IC_{50} of 2.42 μM . According to the data of Table 1, groups alter 6b, 6c, and 6h, lowering the biological activity of the substances with IC_{50} s of 15.2 μM , 15.7 μM , 49.2, and 14.6 μM , respectively. When the 6b, 6c, and 6h are altered, the compounds' individual IC_{50} s are 15.2 μM , 15.7 μM , 49.2, and 14.6 μM .

How anti-SARS-2 activity works

The chemicals' inhibitory effect 6a, 6b, 6c, and 6h against SARS-2 Mpro was evaluated using the previously specified approach [40].

The ways in which compounds 13, 14, and 15a work

Compound	Conc. (μM)	Mode of action		
		Viral adsorption	Viral replication	virucidal
6a	15	41%	43%	48%
6b	10	10%	15%	18%
6c	5	00%	00%	10%
6h	2.5	44%	28%	52%

CONCLUSION

National Press Associates. (2025). ANTIVIRAL PROPERTIES OF 1,3,4-OXADIAZOLE DERIVATIVES AGAINST SARS-2 . Research & Reviews in Biotechnology & Biosciences, 12(2), 25–32.

<https://doi.org/10.5281/zenodo.16742908>

Seven chemicals were examined to assess their capability for suppression for their ability to suppress the SARS-2 virus after they were produced. The structural framework of epoxy benzo oxocino pyridine is the same for all seven compounds, but the connected side groups—dihydroquinoxalin-2-one, dihydrobenzooxazin-2-one, α,γ -diketo acid, and a nitro derivative of dihydrobenzooxazin-2-one—distinguish them from one another. Ethylidene hydrazide is used as a linker between pyridinyl and phenyl groups. The SARS-2 virus cannot be stopped from multiplying by six of the compounds, which show no cytotoxicity at all.

A cytotoxic compound that prevents the SARS-2 virus from replicating is present at a dosage that is within a pharmacologically attainable range. Epoxybenzoxocino pyridine is the compound in question, and it is joined to a dihydroquinoxalin-2-one group. This molecule can serve as a good starting point for future studies on natural antivirals.

REFERENCES

1. P. Sengupta, M. Mal, S. Mandal, J. Singh, and T. K. Maity, "Evaluation of antibacterial and antifungal activity of some 1, 3, 4 oxadiazoles," *Iranian Journal of Pharmacology and Therapeutics*, vol. 7, no. 2, pp. 165–167, 2008.
2. N. Bhardwaj, S. K. Saraf, P. Sharma, and P. Kumar, "Syntheses, evaluation and characterization of some 1, 3, 4-oxadiazoles as antimicrobial agents," *E-Journal of Chemistry*, vol. 6, no. 4, pp. 1133–1138, 2009.
3. A. A. Kadi, N. R. El-Brollosy, O. A. Al-Deeb, E. E. Habib, T. M. Ibrahim, and A. A. El-Emam, "Synthesis, antimicrobial, and anti-inflammatory activities of novel 2-(1-adamantyl)-5-substituted-1,3,4-oxadiazoles and 2-(1-adamantylamino)-5-substituted-1,3,4-thiadiazoles," *European Journal of Medicinal Chemistry*, vol. 42, no. 2, pp. 235–242, 2007.
4. V. Mickevičius, R. Vaickelionienė, and B. Sapijanskaitė, "Synthesis of substituted 1,3,4-oxadiazole derivatives," *Chemistry of Heterocyclic Compounds*, vol. 45, no. 2, pp. 215–218, 2009.
5. F. Bentiss and M. Lagrenée, "A new synthesis of symmetrical 2,5-disubstituted 1,3,4-oxadiazoles," *Journal of Heterocyclic Chemistry*, vol. 36, no. 4, pp. 1029–1032, 1999.
6. S. Liras, M. P. Allen, and B. E. Segelstein, "A mild method for the preparation of 1,3,4-oxadiazoles: triflic acid promoted cyclization of diacylhydrazines," *Synthetic Communications*, vol. 30, no. 3, pp. 437–443, 2000.
7. D. Gomes, C. P. Borges, and J. C. Pinto, "Study of the synthesis of poly(4,4'-diphenylether-1,3,4-oxadiazole) in solutions of poly(phosphoric acid)," *Polymer*, vol. 42, no. 3, pp. 851–865, 2001.
8. A. Souldozi and A. Ramazani, "The reaction of (N-isocyanimino)triphenylphosphorane with benzoic acid derivatives: a novel synthesis of 2-aryl-1,3,4-oxadiazole derivatives," *Tetrahedron Letters*, vol. 48, no. 9, pp. 1549–1551, 2007.
9. A. Ramazani, B. Abdian, F. Z. Nasrabadi, and M. Rouhani, "The reaction of N-isocyanimino triphenyl phosphorane with biacetyl in the

- presence of (E)-cinnamic acids: synthesis of fully substituted 1,3,4-oxadiazole derivatives via intramolecular aza-Wittig reactions of in situ generated imino phosphoranes," *Phosphorus, Sulfur and Silicon and the Related Elements*, vol. 188, no. 5, pp. 642–648, 2013.
10. A. Ramazani, F. Zeinali Nasrabadi, and Y. Ahmadi, "One-pot, four-component synthesis of fully substituted 1,3,4-oxadiazole derivatives from (isocyanoimino)triphenyl phosphorane, a primary amine, an aromatic carboxylic acid, and chloroacetone," *Helvetica Chimica Acta*, vol. 94, no. 6, pp. 1024–1029, 2011.
 11. A. Ramazani and A. Souldozi, "Iminophosphorane-mediated one-pot synthesis of 1,3,4-oxadiazole derivatives," *Arkivoc*, vol. 2008, no. 16, pp. 235–242, 2008. [12] R. Frañski, "Biological activities.
 12. Li, J.; Lai, S.; Gao, G.-F.; Shi, W. The emergence, genomic diversity and global spread of SARS-2. *Nature* 2021, 600, 408–418. [CrossRef] [PubMed]
 13. Tran, A.; Witek, T.J. The emergency use authorization of pharmaceuticals: History and utility during the COVID-19 pandemic. *Pharm. Med.* 2021, 35, 203–213. [CrossRef] [PubMed]
 14. COVID-19 updates: Remdesivir (Veklury) in high-risk outpatients with COVID-19. *Med. Lett. Drugs Ther.* 2022, 64, 31–32.
 15. COVID-19 updates: NIH outpatient treatment guidelines. *Med. Lett. Drugs Ther.* 2022, 64, 32.
 16. Finberg, R.W.; Ashraf, M.; Julg, B.; Ayoade, F.; Marathe, J.G.; Issa, N.C.; Epstein, C. US201 Study: A Phase 2, Randomized Proof-of-Concept Trial of Favipiravir for the Treatment of COVID-19. *Open Forum Infect. Dis.* 2021, 8, ofab563. [CrossRef]
 17. AlQahtani, M.; Kumar, N.; Aljawder, D.; Abdulrahman, A.; Alnashaba, F.; Fayyad, M.A.; Atkin, S.L. Randomized controlled trial of favipiravir, hydroxychloroquine, and standard care in patients with mild/moderate COVID-19 disease. *Sci. Rep.* 2022, 12, 1–10.
 18. L. Fu, F. Ye, Y. Feng, F. Yu, Q. Wang, Y. Wu, C. Zhao, H. Sun, B. Huang, P. Niu, H. Song, Y. Shi, X. Li, W. Tan, J. Qi, G.F. Gao, Both Boceprevir and GC376 efficaciously inhibit SARS-2 by targeting its main protease, *Nat. Commun.* 11 (2020) 4417.
 19. M.A. Abdalla, L.J. McGaw, Natural cyclic peptides as an attractive modality for therapeutics: a mini review, *Molecules* 23 (2018) 2080–2099.
 20. T. Mosmann, Rapid colorimetric assay for cellular growth and survival: application to proliferation and cytotoxicity assays, *J. immunol. Methods* 65 (1983) 55–63.
 21. A. Mostafa, E.M. Abdelwhab, T.C. Mettenleiter, S. Pleschka, Zoonotic potential of influenza A viruses: a comprehensive overview, *Viruses* 10 (2018) 497–534.
 22. Radwan Alnajjar, Ahmed Mostafa, Ahmed Kandeil, Ahmed A. Al-Karmalawy, Molecular docking, molecular dynamics, and in vitro studies reveal the potential of angiotensin II receptor blockers to inhibit the COVID-19 main protease, *Heliyon*. 6 (12) (2020)

e05641,
<https://doi.org/10.1016/j.heliyon.2020.e05641>.

23. A. Mostafa, A. Kandeil, Y. AMME, O. Kutkat, Y. Moatasim, A. A. Rashad, FDA- Approved Drugs with Potent In Vitro Antiviral Activity against Severe Acute Respiratory Syndrome Coronavirus 2, *Pharma.13* (2020) 443-466.
24. Mohammed Al-Rabia, Nabil Alhakamy, Osama Ahmed, Khalid Eljaaly, Ahmed Aloafi, Ahmed Mostafa, Hani Asfour, Ahmed Aldarmahi, Khaled Darwish, Tarek Ibrahim, Usama Fahmy, Repurposing of sitagliptin- melittin optimized nanoformula against SARS-2; antiviral screening and molecular docking studies, *Pharmaceutics* 13 (3) (2021) 307, <https://doi.org/10.3390/pharmaceutics13030307>. [