2025
12(1), 1153

Biomedical and
Therapeutics LETTERS

Synthesis, DNA binding and molecular docking studies of 2H-benzo[b] [1,4] oxazines

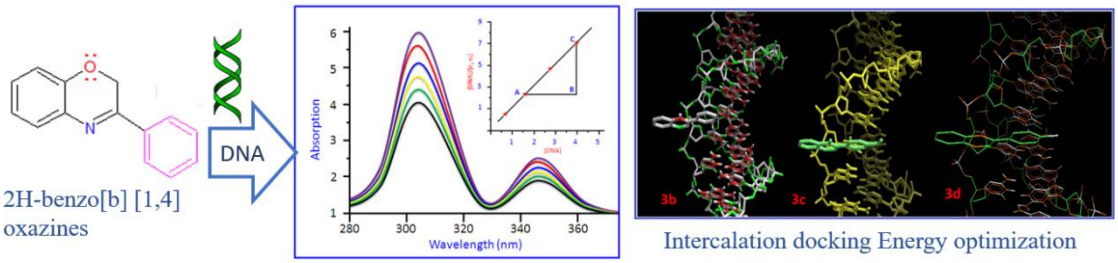
P.J. Bindu[1]*, T.R. Ravikumar Naik[2], G. Krishnamurthy[1,3]

[1]Department of Chemistry, A. S. M. Women's College, Ballari, Women's University, Karnataka. India. [2]Department of P. G. Studies in Chemistry, Veerashaiva College, Ballari, VSK University, Karnataka. India. [3]Department of P. G. Studies in Chemistry, Sahayadri Science College, Shivamoga, Karnataka. India.

Submitted on: 12-Dec-2024, Accepted and Published on: 27-Jan-2025

Article

ABSTRACT



A convenient method for synthesis of 2H-benzo[b] [1,4] oxazines has been achieved by cyclocondensation of substituted phenacyl bromides with aminophenols in $ZnFe_2O_4$ as a bimetallic and eco-friendly catalyst. The benefit of the current protocol includes mild reaction conditions, shorter reaction time, reusability of catalyst, ambient temperature, a simple work-up procedure, good yields. The synthesized 2H-benzo[b] [1,4] oxazines were characterized by IR, 1H NMR, and mass spectra. To understand the mechanism of action and design-specific DNA binders, the evaluation of DNA–ligand interactions is critical. Among products, molecular docking studies revealed that 3b, 3c and 3d have the best interactions with the ct-DNA via the minor groove binding. The interaction profiles of the selected compound (3d) with DNA were evaluated by UV–Visible titration. UV–Visible titration data confirm this interaction. According to the molecular docking results, the Structure–Activity relationships for all synthesized 2H-benzo[b] [1,4] oxazines were proposed. It was observed that 3d have better DNA interactions than other derivatives.

Keywords: 1,4-Oxazines, ZnFe2O4 catalyst, DNA binding, Molecular docking.

INTRODUCTION

Nowadays, the development of innovative methodology in the field of synthetic organic chemistry prompts the synthesis of various heterocyclic scaffolds due to their prominent biological activities and importance in medicinal chemistry.[1,2] Heterogeneous $ZnFe_2O_4$ magnetite nanoparticle (MNPs) catalysts can be simply separated from the reaction mixture by an external permanent magnet. Additionally, the catalyst can be easily recovered from the reaction mixture and reused, making the method more cost effective. Recently, $ZnFe_2O_4$ magnetite nanoparticle (MNPs) showed good catalytic behavior in organic transformations.[1,2]

DNA represents a traditional target for chemotherapeutic intervention in human cancers, especially for those where high proliferation rates of some tumor cell types have resulted in sensitivity to drugs, which block replication and transcription of their DNA.[3] Molecular recognition of DNA by small molecules is a fundamental problem in drug design. Polycyclic heterocycles having a planar structure can be effective pharmacophore moieties for DNA-interactive drugs because they can insert between the stacked base paired oligonucleotides. Moreover, if they bear suitable side chains, further interactions of these ligands with the other important architectural feature of DNA, its minor groove, can be envisaged.[4]

Generally, the intercalative molecules should contain an extended heteroatom, as its stacking between base pairs is considered to be a major driving force that leads to binding.[5] The binding mode and strength are sensitively dependent on the shape, planar area, size and electron density of the interacting aromatic rings. So, a systematic study on the influence of varying parameters on the interaction of small molecules with DNA would be valuable in the rational design of new drugs and therapeutic reagents targeted to DNA.[6-12]

Molecular docking has been a focus of attention for many years. In the current scenario, the flexible docking program is able to predict protein ligand complex structures with reasonable accuracy and speed.[12-14] There has been very little application of docking studies to interpretation of the DNA binding affinity, in modern chemistry and biochemistry. To obtain a significant correlation, it is essential that appropriate descriptors are employed, whether they are theoretical, empirical or derived

*Corresponding Author: Prof. P.J. Bindu
Email: bindu12_naik@rediffmail.com

URN:NBN:sciencein.btl.2025.v12.1153
DOI: 10.62110/sciencein.btl.2025.v12.1153

https://pubs.thesciencein.org/btl

from readily available experimental characteristics of structures. Many descriptors reflect simple molecular properties and can thus provide insight into the physicochemical nature of the activity/property under consideration. Subsequently, several researchers have reported correlations for a wide variety of chemical properties such as including DNA binding studies.[12-14]

Further, The strategy towards the synthesis of compounds bearing oxygen and nitrogen in a ring is growing attention because the resulting molecules are present in numerous biologically important compounds.[15,16] The morpholine moiety has a greater impact on human well-being as it presents in approved CNS drugs such as doxapram,[17] phendimetrazine,[18] moclobemide,[19] aprepitant,[20] reboxetine,[21] and antifungal compound fenpropimorph.[22] The most important 1,4-benzoxazine derivatives that are currently in clinical are Ofloxacin and Levofloxacin.[23.24]

The large number of papers on the synthesis of oxazines that have appeared in recent years attests to the huge contemporary interest owing to its usefulness.[24-26] However, most of the earlier published techniques have at least one of the following drawbacks: low yields resulting from the fairly lengthy, laborious workup, hazardous catalysts used and difficult reaction assemblage etc. Despite the growing need and challenges, the literature on the syntheses and biological evaluation of 1,4-Oxazines are limited in number as compared to 1,3-benzoxazines, research on 1,4-Oxazines should be scaled up to achieve desired outcome.

Oxazine derivatives have been evaluated extensively and reported to possess good anticancer potential in various preclinical tumor models. The nitrogen and oxygen containing heterocyclic compounds reported as good DNA binding molecules. Interestingly, oxazine contains both nitrogen and oxygen hetero atom in a ring system and showed good DNA binding and anticancer activities.[27,28]

Based on all the above consideration and adding to our research endeavor, we have been interested in developing the new anticancer drugs. Taking inspiration from the above and as a part of our continuing research on the synthesis and anticancer, antibacterial, DNA photocleavage and molecular docking studies of biologically active compounds.[29-35] Hence, the need to be replaced by newer drugs periodically. In this perspective, we proposed the synthesis and development of 1,4-Oxazines unit targeting DNA binding and molecular docking studies.

MATERIALS AND METHODS

Materials and equipment

All the chemicals used in the present study are of AR grade. M/s Sigma-Aldrich, and Merck, USA and M/s S.D. Fine Chem. Pvt. Ltd, Mumbai, India. Calf thymus DNA (CT DNA) and supercoiled pUC19 DNA (cesium chloride purified) was obtained from Bangalore Genei (India). Agarose (low melt, 65°C, molecular biology grade for DNA gels), ethidium bromide, bromophenol blue, Tris(hydroxymethyl)aminomethane (Tris), sodium chloride, ethylene diamine tetraacetic acid disodium salt (EDTA-Na2), sodium azide, were of molecular biology grade, obtained from Himedia (India).

Melting points were recorded on an open capillary tube with a Buchi melting point apparatus and are uncorrected. Elemental analyses were carried out using Perkin-Elmer 240C CHN-analyzer. IR spectra were recorded on a FT-IR infrared spectrophotometer. 1H- NMR spectra were obtained using a 300 MHz and 400 MHz on a Bruker spectrometer (chemical shifts in δ ppm). Mass spectra were recorded using a micro spray Q-TOF MS ES Mass spectrometer.

EXPERIMENTAL

General procedure for the preparation 2H-benzo[b][1,4] oxazines:

A mixture of substituted 2-aminophenol (5 mmol) in ethanol (10 mL) was stirred for 20 minutes at room temperature, phenacyl bromide/substituted phenacyl bromide (5 mmol) then potassium carbonate (5 mmol) and $ZnFe_2O_4$ (30 mg) and were added and continued the stirring for 3 h at ambient temperature. The reaction progress was monitored by TLC using Silica coated aluminium TLC using 20% ethyl acetate in hexane as the solvent system. After the completion of the reaction, as indicated by TLC, the resultant mixture was extracted from the vial using ethyl acetate. The solvent was removed under reduced pressure using a rotary evaporator. The crude product was purified by column chromatography using n-hexane/EtOAc 3:1 as eluent. The structures of all the synthesized molecules were established by spectral analyses (IR, 1H NMR, 13CNMR, and mass) data. All the synthesized compounds showed appropriate characteristic signals, which confirm their structures.

The representative spectral data is given below.

3-(4-Bromophenyl)- 2H-benzo[b][1,4]oxazine (3a). Mp 162-164℃. IR (KBr) cm^{-1} : 3030 (Ar-H str.), 2923 (Al-H str.), 16687 (C=N str.), 1230 (C-N str.), 1080 (C-O-C str.), 587 (C-Br str.); 1H NMR (400 MHz, CDCl3) δ: 4.9 (s, 2H, CH2); 6.80 (dd,1H); 7.1-7.7 (m, 7H, Ar); 13C NMR= δ: 60.5, 116.0, 115.5, 120.7, 124.0, 129.0, 129.7, 130.0, 134.5, 135.0, 152.5, 155.3; MS: m/z 288 (M)+ .

3-(p-Tolyl)-2H-benzo[b][1,4]oxazine (3b). Mp 88-90℃. IR (KBr) cm^{-1}: 3062 (Ar-H str.), 2927(Al-H str.), 1580 (C=N str.), 1277(C-N str.), 1081 (C-O-C str.); (1H NMR (400 MHz, CDCl3) δ: 2.38 (s, 3H); 5.85 (s, 2H, CH2); 7.0-7.65 (m, 8H) ; 13C NMR = δ:21.1, 60.50, 109.6, 118.6, 119.7,123.6, 126.6, 128.2, 130.4, 131.5, 132.1, 140.4, 152.4; MS: m/z 224 (M)+.

3-(4-Bromophenyl)-6-methyl-2H-benzo[b][1,4]oxazine (3c). Mp 141-143℃. IR (KBr) cm^{-1}: 3034 (Ar-H str.), 1074 (C-O-C str.), 640 (C-Br str.); (1H NMR (400 MHz, CDCl3) δ:2.35 (s, 3H, Al); 4.89 (s, 2H, CH2); 7.1-7.61(m, 7H, Ar); 13C NMR = δ:20.8, 60.8, 115.5, 116.7, 124.4, 125.3, 129.4, 129.5, 130.2, 131.4, 141.7, 150.1, 153.5; MS: m/z 302 (M)+.

3-(4-Methoxyphenyl)-6-methyl-2H-benzo[b][1,4]oxazine (3d). Mp 120-122℃. IR (KBr) cm^{-1}: 3033 (Ar-H str.), 2890(Al-H str.), 1574(C=N str.), 1267(C-N str.), 1089(C-O-C str.); (1H NMR (400 MHz, CDCl3) δ: 2.30(s,3H); 4.3(s, 3H, Al); 5.01 (s, 2H, CH2); 7.0-7.94 (m, 7H, Ar) ; 13C NMR = δ: 22.3, 57.7, 71.9, 118.8, 123.9, 124.5, 126.96, 127.4, 129.9, 132.5, 136.4, 137.7, 140.3, 154.5; MS: m/z 254 (M)+.

6-Chloro-3-(p-tolyl)-2H-benzo[b][1,4]oxazine (3e). Mp 121-123℃ IR (KBr) cm^{-1}: 3020 (Ar-H str.), 2928 (Al-H str.), 1605 (C=N str.), 1380 (C-N str.), 1097 (C-O-C str.), 720 (C-Cl str.); (1H NMR (400 MHz, CDCl3) δ: 2.2 (s,3H, Al); 4.8 (s, 2H, CH2); 7.0-7.31 (m, 7H, Ar); 13C NMR = δ: 29.7, 61.8, 116.2, 116.7, 124.3, 124.4, 129.5, 129.9, 135.1, 140.5, 149.8, 151.4, 155.2; MS: m/z 258 (M)+.

3-Phenyl-2H-benzo[b][1,4]oxazine (3f). Mp 105-107℃ IR (KBr) cm^{-1} : 2980 (Ar-H str.), 2924 (Al-H str.), 1650 (C=N str.), 1230 (C-N str.), 1080 (C-O-C str.), 790 (C-Cl str.); (1H NMR (400 MHz, CDCl3) δ: 4.80 (s, 2H, CH2); 6.50 (d, 1H, Ar); 6.80-7.80 (m, 8H, Ar); 13C NMR = δ: 61.3, 115.1, 116.3, 120.5, 128.9, 128.3, 129.2, 130.0, 135.0, 135.2, 152.3, 157.3; MS: m/z 209 (M)+.

3-(4-Chlorophenyl)-2H-benzo[b][1,4]oxazine (3g). Mp 158-160℃ IR (KBr) cm^{-1}: 2930 (Ar-H str.), 1600 (C=N str.), 1478 (C=C str.), 1250 (C-N str.), 1020 (C-O-C str.), 745 (C-Cl str.); (1H NMR (400 MHz, CDCl3) δ: 4.80(s, 2H, CH2); 6.61(d, 1H, Ar); 7.10-7.95 (m, 7H, Ar); 13C NMR = δ: 56.2, 116.4, 117.9, 124.8, 125.0, 127.0, 127.7, 128.6, 130.0, 136.8, 154.4, 158.3 ; MS: m/z 245 (M)+.

3-(4-Chlorophenyl)-6-methyl-2H-benzo[b][1,4]oxazine (3h). Mp 108-110℃ IR (KBr) cm^{-1}: 2980 (Ar-H str.), 2930 (AlH str.), 1464 (C=N str.), 1270 (C-N str.), 1020 (C-O-C str.); (1H NMR (400 MHz, CDCl3) δ: 2.30 (s,3H, Al); 4.90 (s, 2H, CH2); 6.85 (d, 1H, Ar); 7.10-7.60 (m, 6H, Ar) ; 13C NMR = δ: 21.0, 60.5, 117.9, 123.3, 127.5, 128.9, 130.5, 130.7, 131.0, 134.4, 139.9, 152.4, 152.5; MS: m/z 257 (M)+.

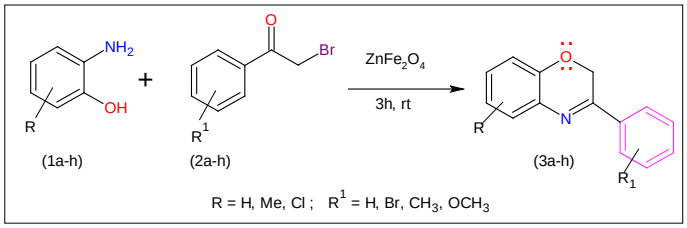
RESULTS AND DISCUSSION

In our attempts to search for novel antitumor agents, we extended the interest to biological active nitrogen heterocylces and synthesized series 2H-benzo[b][1,4] oxazines derivatives with the aim of evaluating their diverse biological activity.

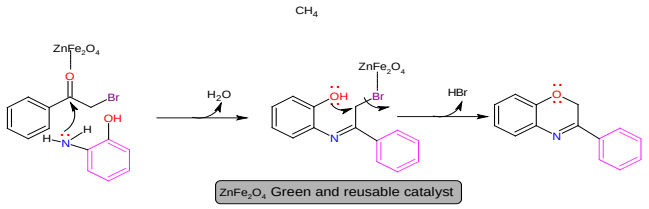
Further, 1, 4-benzoxazine is also widely used as a building block for the synthesis of more complex molecular structures, active pharmaceutical ingredients, herbicides and pesticides and thus, have attracted considerable interest of chemists in designing and developing new greener synthetic strategies to construct different functionally substituted 1,4-benzoxazine scaffolds. Some reports are available in the literature on the synthesis of 1,4-benzoxazine scaffolds. However, the available methods had drawbacks such as high temperature, long reaction time, low yield, lack of diastereomer selectivity, use of expensive catalyst and hazardous solvents, non-eco-friendly and most importantly the methods were not versatile enough to synthesize differentially substituted 1,4-benzoxazine.

4.1. Chemistry

The reaction conditions were optimized on the reaction of 2-aminophenol (5 mmol) in ethanol (10 mL) was stirred for 20 minutes at room temperature, phenacyl bromide/substituted phenacyl bromide (5 mmol) then potassium carbonate (5 mmol) and $ZnFe_2O_4$ (30 mg) and were added and continued the stirring for 3 h at ambient temperature. After an extractive workup, to afford a pure product (yield 90%), which was identified as 3-(4-methoxyphenyl)-2H-benzo[b] [1,4] oxazine (3a). In general, the use of $ZnFe_2O_4$ acts as a base catalyst combined with potassium carbonate was easier to workup and yields were higher which can be attributed to its high solubility in solvents. The reaction proceeds efficiently with both electron-donating and electron withdrawing substituent's (Table 1).



Scheme 1: Synthesis of 2H-Benzo[b] [1,4] Oxazines.



Scheme 2. A plausible mechanism for the synthesis of 2H-Benzo[b] [1,4] Oxazines.

To expand the scope of this protocol, aliphatic and various substituted 2-aminophenols (1) possessing both electron donating (-CH_3) and withdrawing groups (-Cl) were reacted with methyl, methoxy and bromo substituted phenacyl bromide (2a) under optimized reaction condition. The methoxy, metyl and chloro substituted compounds gave excellent results yielding desired products (Table 2) in moderate to excellent yields (80-90%).

4.2. Biological Activity

In recent years, many studies on the binding of organic compounds to DNA have been performed. Studying drug–DNA interaction mechanisms is crucial for designing the beneficial drugs that specifically target DNA. In General, drugs bind to the DNA through two types of covalent and non-covalent interactions. Drug–DNA Covalent binding is irreversible, leading to complete inhibition of the cell division process and eventual cell death. Cyclophosphamide, Carmustine and Busulfan are examples of drugs that covalently bind to DNA. Non-covalent interactions cause some drugs to be located between adjacent nucleotide bases to form the DNA–ligand complex. Popular interactions that stabilize the DNA–ligand complex include van der Waals forces, hydrogen bonding, hydrophobic attractions, and charge-transfer forces. DNA-binding agents stop replication in the DNA chain of cancer cells and induce cell death.[34] Actinomycin, Doxorubicin, and Daunorubicin are examples of drugs that bind non-covalently to DNA. One of the applications of spectrophotometric methods is to determine the constant binding of the drug to DNA. UV absorption spectroscopy is the simplest and most common method of studying DNA stability and its interactions with small molecules.[34]

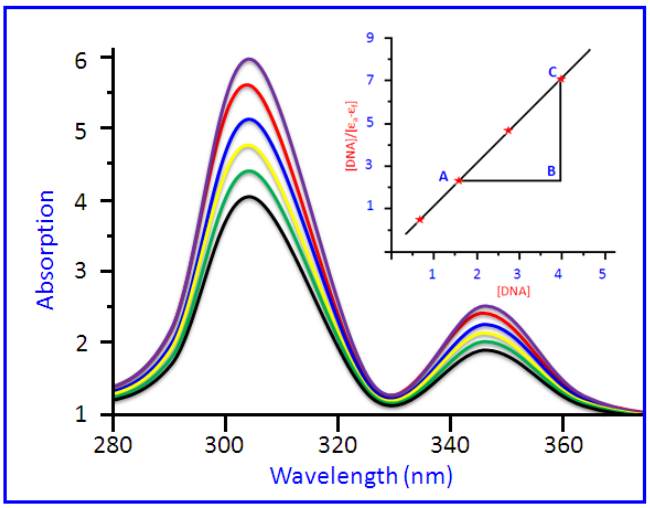


Figure 1. DNA binding studies of 1,4-oxazines (3d). UV absorption spectra of (3d) upon addition of calf thymus (ds) DNA. Concentration of control [DNA] = 0.5 µM and increased the Concentration of [DNA] =10 µM, 20 µM, 30 µM, 40 µM and 50 µM respectively. Arrow shows the absorbance changing upon the increase of DNA concentration. The inner plot of [DNA]/(εa-εf) vs [DNA] for the titration of DNA with 1,4-oxazines.

In this study, the absorption bands of the tested 1,4-oxazines (3a–3h) are recorded in the absence and presence of CT-DNA. The DNA binding spectra for 3a is as shown in Figure 1. The UV–vis spectral data for (3a–3h) are summarized in Table 2. The association 3d to DNA in buffered solution was monitored by spectrophotometeric titrations. The effect of progressively increasing higher concentrations of DNA on the absorption spectra of 1,4-oxazines (3d) is shown in Figure 1. The spectral change involved essentially a progressive red shift and hypochromicity, i.e., the interaction of 1,4-oxazines (3d) with DNA resulted in a strong decrease of the absorption intensity. The absorption of 1,4-oxazines (3d) in the peak was decreased by about 22.00 % at 350 nm peak maximum along with a bathochromic shift of 5–6 nm. This hypochromism and bathochromic shift may be due to strong interaction between the electronic states of the interacalating chromophore and that of the DNA base pairs.[32] Such changes are obvious evidence of the formation of DNA adducts. It is believed that these hypochromic shifts are probably due to the covalent and noncovalent bonds, which were observed in all target compounds.[34]

Table 1. Synthesis of 2H-Benzo[b] [1,4] Oxazines

Sl. No	Product	R	R^1	Time taken (hours)	Yield (%)	M. P. (°C)[a,b]
1	3a	H	Br	6	80	162-164
2	3b	H	CH_3	6	85	88-90
3	3c	CH_3	Br	6	80	141-143
4	3d	CH_3	OCH_3	6	90	120-122
5	3e	Cl	CH_3	6	85	121-123
6	3f	H	H	6	80	105-107
7	3g	H	Cl	6	80	158-160
8	3h	CH_3	Cl	6	80	108-110

[a]All the products were characterized by elemental analysis, ^{1}H NMR, ^{13}C NMR and mass spectral data. [b]Yields of isolated products

Table 2. DNA binding and Molecular Docking studies of 1,4-oxazines

Products	Docking Energy (Kcal/mol)	Inhibition Constant (M)	RMSD	DNA binding constant K_b
3a	-7.57	5.08 x 10^{-16}	2.5	4.8 x 10^4 M^{-1}
3b	-7.48	4.58 x 10^{-16}	2.5	4.2 x 10^4 M
3c	-7.61	4.30 x 10^{-16}	2.5	4.5 x 10^4 M
3d	-10.23	3.64 x 10^{-7}	2.5	3.8 x 10^4 M^{-1}
3e	-7.30	5.11 x 10^{-16}	2.5	5.2 x 10^4 M
3f	-7.45	6.23 x 10^{-16}	2.5	5.5 x 10^4 M
3g	-7.37	6.15 x 10^{-16}	2.5	5.6 x 10^4 M
3h	-7.56	6.29 x 10^{-16}	2.5	5.8 x 10^4 M

4.2.2. Molecular docking studies

Molecular docking has been an increasingly pivotal tool for drug discovery. The ability to predict binding affinity for different DNA target is useful in characterizing genetic regulatory pathway. This enables us to characterize the behaviour of small molecules in the binding site of target DNA as well as to elucidate fundamental biochemical processes. [35,36] In this sense, it is of high relevance taking into account that, the previous non-covalent binding between drug and DNA has a strong influence on the subsequent photoreaction and therefore on their biological activity.[35]

The 1,4-oxazines (3a–3h) were prepared using ChemDraw 12.0 and Autodock tools software. The ligand structures were drawn in 2D conformation using ChemDraw Ultra 12.0. Conversion of 2D to 3D conformation and energy minimization were carried out using Chem3D Pro 12.0. The energy-minimized structure (3D conformation) was open in Autodock Tools to view the geometry and bond flexibility, and finally saved in pdbqt format. In order to estimate the conformation of the nucleic acid base-organic molecule and to increase accuracy, repeatability, and reliability of docking results, AutoDock Vina 4.2 docking program was used as described in our previous reports.[35]

In order to rationalize the observed spectroscopic results and to get more insight into the intercalation modality, the 1,4-oxazines (3a–3h) were successively docked,[35] within the DNA duplex of sequence d(CGCGAATTCGCG)2 dodecamer (PDB ID: 1BNA) (Figure 2, Table 2) in order to predict the chosen binding site along with preferred orientation of the ligand inside the DNA minor groove. All synthesized 1,4-oxazines (3a–3h) derivatives were drawn in ChemSketch and structures were saved in .mol format. Afterwards the .mol format was used in Hyperchem-7, to adjust their fragments, followed by total energy minimization of ligands so that they can attain a stable conformation and the file was saved in .pdb format.

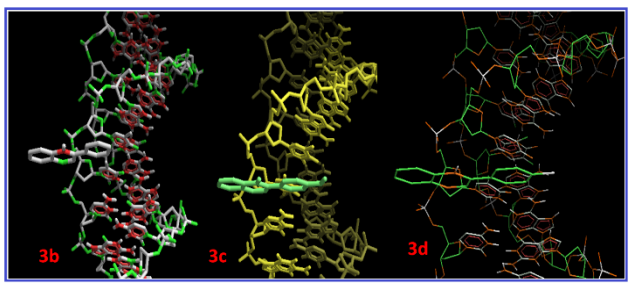


Figure 2. View of the energy minimized docked poses of 1,4-oxazines (3b, 3c and 3d) with DNA d(CGCGAATTCGCG)2 (PDB ID: 1BNA).

Generally, the result obtained indicated that the reactions were spontaneous (Table 2). From the result obtained 1,4-oxazines (3h) has the lowest binding affinity to CT-DNA and Interestingly 1,4-oxazines (3b, 3c and 3d) demonstrated had the highest binding affinity towards CT-DNA. The compound exhibited 8.26% better affinity (−10.3 KJ/mol) for CT-DNA compared to other compounds. One potential reason for this phenomenon we propose may be that in the case of compound 3d, it is structurally more suited to interact with the observed sites. As a consequence, its molecular recognition with active site in the CT-DNA was more than the others. This is indicative of its specificity for such sites in comparison with the other compounds. The binding of 1,4-oxazines (3b, 3c and 3d) with CT-DNA is illustrated in Figure 2.

CONCLUSION

In conclusion, we report a technique that is more simplified, following the principles of green chemistry for the synthesis of 1,4-oxazines, resulting in excellent yields and evaluated their biological activity. The present study provides evidence that the synthesized 1, 4 Oxazine compounds can be good nominees for future investigations to synthesize new anticancer drugs. Few compounds showed good DNA binding and DNA Photoclevage studies. It is concluded from the results that substituents on the 1,4-oxazines skeleton are responsible for the enhancement of the DNA binding studies. Results obtained from our present work would be very useful to understand the DNA interaction with compound. Further studies on the different application of the synthesized compounds are currently at different stages and emphasis on continued research is going on to design more efficient methods for organic syntheses of oxazines.

ACKNOWLEDGMENTS

Author P. J. B. thank to Department of Chemistry, A. S. M. Women's College, Ballari, for lab facility to carry out this work and V.V. Sangha for financial support and Department of Organic Chemistry, Indian Institute of Science for spectral facilities.

CONFLICT OF INTEREST STATEMENT

Authors do not have conflict of interest for this work.

ETHICAL DECLARATIONS

Not applicable.

CLINICAL TRIAL INFORMATION

Not applicable.

REFERENCES AND NOTES

1. T. R. Ravikumar Naik, S. A. Shivashankar. Heterogeneous bimetallic ZnFe2O4 nanopowder catalyzed synthesis of Hantzsch 1,4-dihydropyridines in wate. *Tetrahedron Letters*, **2016**, 57, 4046-4049.
2. A. Khazaei, A. Ranjbaran, F. Abbasi, M. Khazaei, A. R. Moosavi-Zare. Synthesis, characterization and application of ZnFe2O4 nanoparticles as a heterogeneous ditopic catalyst for the synthesis of pyrano[2,3-d] pyrimidines. *RSC Adv*. **2015**, 5, 13643.
3. R. Kumar, J. W. Lown. Design, synthesis and in vitro cytotoxic studies of novel bis-pyrrolo[2,1][1,4] benzodiazepine-pyrrole and imidazole polyamide conjugates. *Eur. J. Med. Chem*. **2005**, 40, 641.
4. A. Lauria, P. Diana, P. Barraja, A. M. Almerico. G. Cirrincione, G. Dattolo. *J. Heterocycl. Chem.* **2000**, 37, 747.
5. T. R. Ravikumar Naik, H. S. Bhojya Naik, H. R. Prakash Naik, P. J. Bindu, B. G. Harish, V. Krishna. Synthesis, DNA binding, docking and photoclevage studies of novel benzo[b][1,8]naphthyridines, *Med. Chem.* **2009**, 5, 411-418.
6. P.J. Bindu, T. R. Ravikumar Naik, K. M. Mahadevan, G. Krishnamurthy, Synthesis, DNA photocleavage, molecular docking and anticancer studies of 2-methyl-1,2,3,4-tetrahydroquinolines. *Chem. Bio. Lett.*, **2019**, 6, 8-13.
7. X.-Q. Zhou, Li. D.-Y. Y, Z-Y. Nie, Z.-J. Li, W. Gu. Copper complexes based on chiral Schiff-base ligands: DNA/BSA binding ability, DNA cleavage activity, cytotoxicity and mechanism of apoptosis. *Eur. J. Med. Chem.* **2016**, 114, 244–256.
8. P. R. Inamdar, R. Chauhan, J. Abraham. A. Sheela, DNA interaction and cytotoxic activity of copper complex based on tridentate hydrazone derived ligand and nitrogen donor heterocycle. *Inorg. Chem. Commun.* **2016**, 67, 67–71.
9. Z. Shokohi-Pour, H. Chiniforoshan, A. A. Momtazi-Borojeni, B. Notash, A novel Schiff base derived from the gabapentin drug and copper (II) complex: Synthesis, characterization, interaction with DNA/protein and cytotoxic activity. *J. Photochem. Photobiol. B.* **2016**, 162, 34–44.
10. R. Pradhan, M. Banik, D. B. Cordes, A. M. Z. Slawin, N. C. Saha, Synthesis, characterization, X-ray crystallography and DNA binding activities of Co (III) and Cu (II) complexes with a pyrimidine-based Schiff base ligand. *Inorg. Chim. Acta.* **2016**, 442, 70–80.
11. L. H. Abdel-Rahman, A. M. Abu-Dief, M. Ismael, M. A. A. Mohamed, N. A. Hashem, Synthesis, structure elucidation, biological screening, molecular modeling and DNA binding of some Cu (II) chelates incorporating imines derived from amino acids. *J. Mol. Struct.* **2016**, 1103, 232–244.
12. V. Rustagi, S.R.R. Gupta, A. Singh, I.K. Singh. Beyond trial and error: Leveraging advanced software for Therapeutic discovery. *Chem. Biol. Lett.* **2025**, 12 (1), 1251.
13. O. Trott, A. J. Olson. Auto Dock Vina: improving the speed and accuracy of docking with a new scoring function, efficient optimization, and multithreading. *J. Comput. Chem.* **2010**, 31, 455-461.
14. R. Kumar, M.P. Chaudhary, N. Chauhan. Recent advances and current strategies of cheminformatics with artificial intelligence for development of molecular chemistry simulations. *J. Mol. Chem.* **2022**, 2 (2), 440.
15. D. Faulkner. Marine natural products. *J. Nat. Prod. Rep.* **1986**, 3, 1.
16. T. Yasumoto, M. Murata, Marine toxins. *Chem. Rev.* **1993**, 93, 1897.
17. J. F. Cotten, B. Keshavaprasad, M. J. Laster, E. I. Eger, C. S. Yost. The Ventilatory Stimulant Doxapram Inhibits TASK Tandem Pore (K2P) Potassium Channel Function but Does Not Affect Minimum Alveolar Anesthetic Concentration. *Anesth. Analg.* **2006**, 102, 779-785.
18. W. W. Stoops, J. C. Strickland, J. L. Alcorn, L. R. Hays, A. O. Rayapati, J. A. Lile, C. R. Rush. Influence of phendimetrazine maintenance on the reinforcing, subjective, performance, and physiological effects of intranasal cocaine. *Psychopharmacology*. **2019**, 236, 2569-2577.
19. U. Bonnet. Moclobemide: Therapeutic Use and Clinical Studies. *CNS Drug Rev.* **2003**, 9, 97−140.
20. R. Hargreaves, J. C. Arjona Ferreira, D. Hughes, J. Brands, J. Hale, B. Mattson, S. Mills. Development of aprepitant, the first neurokinin-1 receptor antagonist for the prevention of chemotherapy-induced nausea and vomiting. *Ann. N. Y. Acad. Sci.* **2011**, 1222, 40-48.
21. C. Naidu, J. Kulkarni. Psychosis induced by antidepressant treatment with reboxetine. *Aust. N. Z. J. Psychiatry*. **2019**, 53, 1227-1228.
22. H. Dieckmann, M. Stockmaier, R. Kreuzig, M. Bahadir, Fresenius' Simultaneous determination of fenpropimorph and the corresponding metabolite fenpropimorphic acid in soil. *J. Anal. Chem.* **1993**, 345, 784.
23. L. A. Mitscher, P. N. Sharma, D. T. Chu, L. L. Shen, A. G. Pernets. Chiral DNA gyrase inhibitors. 2. Asymmetric synthesis and biological activity of the enantiomers of 9-fluoro-3-methyl-10-(4-methyl-1-piperazinyl)-7-oxo-2,3-dihydro-7H-pyrido[1,2,3-de]-1,4-benzoxazine-6-carboxylic acid. *J. Med. Chem.* **1987**, 30, 2283-2286.

24. I. Hayakawa, S. Atarashi, S. Yokohama, Synthesis and antibacterial activities of optically active ofloxacin. *Antimicrobial agents and chemotherapy*. **1986**, 163-164.
25. S. L. Gaonkar, A Review on Current Synthetic Strategies of Oxazines. *Mini-Rev. Org. Chem.* **2019**, 16, 43-58.
26. B. K. Zhimomi, P. Imchen, T. Phucho. Recent advances in strategies of green synthesis of 1,3-oxazines- a brief review. *Tetrahedron*. **2022**, 109, 132672.
27. Y. Kwok, D. Sun, J. J. Clement, L. H. Hurley, The quinobenzoxazines: relationship between DNA binding and biological activity, *Anticancer Drug Des*, **1999**,14, 443-50.
28. H. Bharathkumar, C. D. Mohan, S. Rangappa, T. Kang, H. K. Keerthy, J. E. Fuchs, N. H. Kwon, A. Bender, S. Kim, K. Basappa, S. Rangappa, Screening of quinoline, 1,3-benzoxazine, and 1,3-oxazine-based small molecules against isolated methionyl-tRNA synthetase and A549 and HCT116 cancer cells including an in silico binding mode analysis, *Org. Biomol. Chem*, **2015**, 13, 9381-9387.
29. T. R. Ravikumar Naik, S. A. Shivashankar, P. J. Bindu. In-vitro antioxidant and anticancer activities of MnFe2O4 nanoparticles synthesized using spinach leaves extract. *Appl. Nanomed.* **2022**, 2(1), 330.
30. P. J. Bindu, T. R. Ravikumar Naik, S. Chandrashekar, K. M. Mahadevan. Synthesis, DNA binding, photocleavage, molecular docking studies of 2-oxo-3-quinoline-3-oximes. *J. Mol. Chem.* **2022**, 2(2), 444.
31. P. J. Bindu, K. M. Mahadevan, N. D. Satyanarayan, T. R. Ravikumar Naik. Synthesis and DNA cleavage studies of novel quinoline oxime esters. *Bioorg. Med. Chem. Lett*. **2012**, 22, 898-900.
32. P. J. Bindu, K. M. Mahadevan, T. R. Ravikumar Naik. An efficient one-pot synthesis and photoinduced DNA cleavage studies of 2-chloro-3-(5-aryl-4,5-dihydroisoxazol-3-yl)quinolines. *Bioorg. Med. Chem. Lett.* **2012**, 22(2), 6095-8.
33. P. J. Bindu, K. M. Mahadevan, T. R. Ravikumar Naik. Sm(III)nitrate-catalyzed one-pot synthesis of furano[3,2c]-1,2,3,4-tetrahydroquinolines and DNA photocleavage studies. *J. Mol. Struct*. **2012**, 1020, 142-147.
34. P. J. Bindu, T. R. Ravikumar Naik, K. M. Mahadevan. Synthesis, DNA binding, docking and photocleavage studies of 2-chloro-3-quinolinyl-3-phenylpropen-2-ones, *Med. Chem. Comm*, **2014**, 5, 1708-1717.
35. M. A. Abdel-Sayed, S. M. Bayomi, M. A. El-Sherbeny, N. I. Abdel-Aziz, K. E. H. ElTahir, G. S. G. Shehatou, A. A.-M. Abdel-Aziz. Synthesis, anti-inflammatory, analgesic, COX-1/2 inhibition activities and molecular docking study of pyrazoline derivatives. *Bioorg. Med. Chem.* **2016**, 24, 2032–2042.
36. Y. N. Mabkhot, M. S. Al-Har, A. Barakat, F. D. Aldawsari, A. Aldalbahi, Z. Ul-Haq. Synthesis, anti-microbial and molecular docking studies of quinazolin-4(3H)-one derivatives. *Molecules*. **2014**, 19, 8725-8739.