Results in Chemistry 6 (2023) 101023

Contents lists available at ScienceDirect

Results in Chemistry

journal homepage: www.sciencedirect.com/journal/results-in-chemistry

$ProHSO_4$: An efficient catalyst for solvent-free synthesis of bis(indolyl) methanes and their *in silico* screening for potential biological activity

Vikrant Kumbhar [a,b], Rutik Raskar [b], Radha Chafle [c], Vandana Nikam [c], Avinash Kumbhar [b], Ramdas Pawar [a], Manohar Chaskar [a], Gulab Gugale [a,d], Bhushan Khairnar [a,b,*]

[a] Department of Chemistry, PDEA's Prof. Ramkrishna More College, Pune 411044, India
[b] Interdisciplinary School of Science (IDSS), Savitribai Phule Pune University, Ganeshkhind, Pune 411007, India
[c] Department of Pharmacology, STES's Smt. Kashibai Navale College of Pharmacy, Pune 411048, India
[d] Department of Chemistry, Haribhai V. Desai College, Pune 411002, India

ARTICLE INFO

Keywords:
Bis(indolyl)methanes
$ProHSO_4$
Eg5 kinesin
Human p38 α MAP Kinase
Human 3 alpha HSD type 3

ABSTRACT

A simple, mild, and efficient protocol has been developed for the synthesis of 3,3′-bis(indolyl)methanes (BIMs) using prolinium hydrogen sulfate ($ProHSO_4$) as a catalyst under solvent-free and support-free conditions. This mechanochemical method undergoes electrophilic substitution reaction of indole with diverse aldehydes to afford the desired BIMs in high yields at room temperature. Due to diverse pharmacological utility, the synthesized BIMs were subjected to in silico studies. The structure activity analysis of BIMs revealed their ability to bind Eg5 kinesin, Human p38 α MAP Kinase, and Human 3 alpha HSD type 3 as a potential protein target for anticancer activity. The compounds exhibited higher binding affinities with Eg5 kinesin having hydrophobic interactions with α2, α3 helices and loop5 amino acid residues. Moreover, molecular docking with Human p38 α MAP Kinase protein depicted compounds as ATP-competitive molecules interacting with hydrophobic region I, II and allosteric DFG motif. A total of 13 derivatives were synthesized and subjected for in silico screening for predicting potential biological activity. Among them, **3b**, **3d**, **3f**, **3 g**, **3i**, **3j**, and **3 k** compounds were identified as potential inhibitors of Eg5 kinesin, Human p38 α MAP Kinase, and Human 3 alpha HSD type 3, and their evaluation in the biological experimental system is warranted.

Introduction

Among the plethora of naturally occurring bioactive compounds, indole based alkaloids such as 3,3′-bis(indolyl)methanes (BIMs) have drawn significant interest due to their diverse biological activities (Fig. 1).[1] They are naturally available in cruciferous plants, terrestrial and marine sources.[2,3] BIMs are composed of two indole rings connected at 3 and 3′- positions by a single carbon atom. This scaffold have unique arrangement of indole rings which imparts remarkable pharmacological activities such as antibacterial, antihyperglycemic, antiinflammatory, anticancer, antimicrobial, antiviral, and antileishmanial properties.[4–6] Due to their ability to induce beneficial estrogen metabolism, they act as promoter-specific activators of estrogen receptors in the absence of 17β-estradiol.[7] Interestingly, BIMs have been reported to normalize the abnormal cell growth linked to cervical dysplasia.[8] They are also prescribed in treatment of fibromyalgia, chronic fatigue and irritable bowel syndrome.[9] Moreover, the oxidized form of BIMs have usage in dyes[10] and colorimetric sensors.[11,12].

Owing to their diverse applications, intensive studies are being done to functionalize BIMs in order to ascertain its biological activities. The construction of symmetrical and unsymmetrical BIMs are mainly based on nucleophilic attack of indole derivatives on substrates such as aldehydes, ketones, allenes, alkynes, alcohols, amino acids, etc.[13–16] As a result, numerous catalytic systems have been developed for synthesis of BIMs, either in solution phase or solid phase, and these include Lewis acids such as $CeCl_3$,[17] TiO_2,[18] $CuBr_2$,[19] $ZrCl_4$,[20] protic acids such as H_2SO_4[21] and H_3NSO_3,[22] solid acidic catalysts such as montmorillonite K-10,[23] Fe^{+3}-montmorillonite *K*10,[24] zeolites,[25] and amberlyst-15.[26] Moreover, preparation of BIMs by using silica gel itself[27,28] or silica as a support have been reported, such as silica-boric acid,[29] silica-diphenic acid,[30] HBF_4-SiO_2,[31] $AlCl_3$-SiO_2,[32] silica-sulfuric acid,[33] $HClO_4$-SiO_2,[34] and SiO_2-HCl.[35] Furthermore, oxalic acid dihydrate,[36] trichloroisocyanuric acid,[37]

* Corresponding author.
E-mail address: bhushan.khairnar@unipune.ac.in (B. Khairnar).

https://doi.org/10.1016/j.rechem.2023.101023
Received 2 May 2023; Accepted 22 June 2023
Available online 23 June 2023

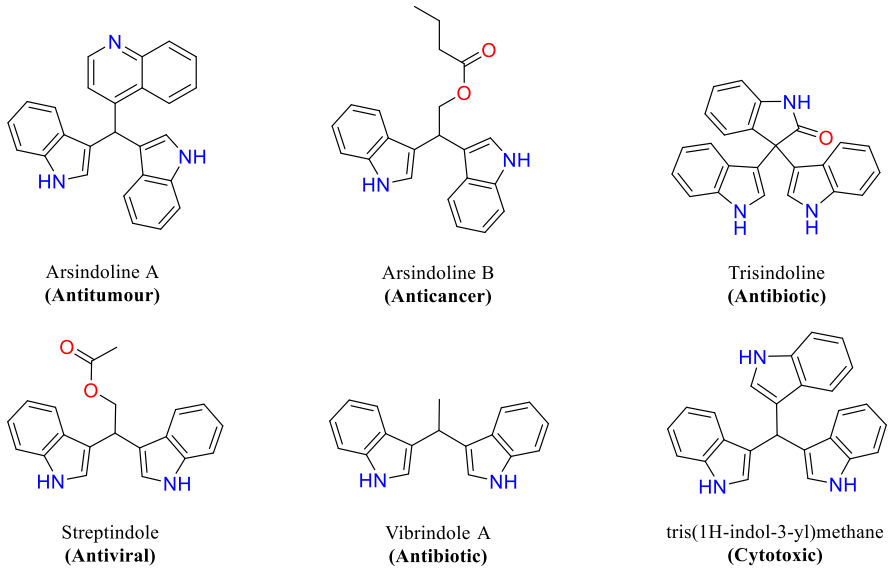


Fig. 1. Structures of some naturally derived 3,3′-bis(indolyl)methanes.

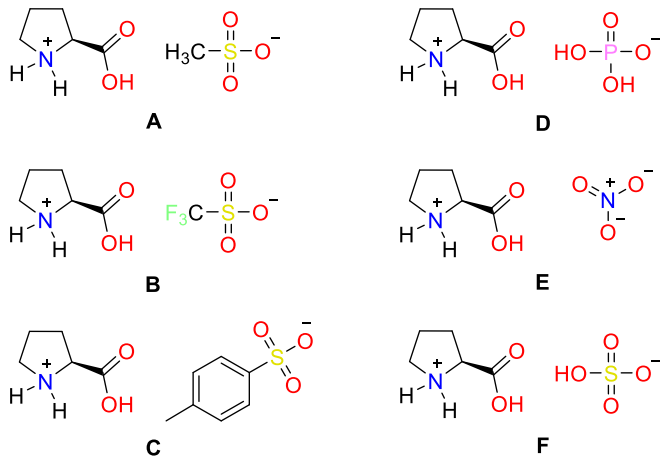


Fig. 2. L-proline based Brønsted amino acid ionic liquids.

NBS,[38] β-cyclodextrin sulfuric acid,[39] thiamine hydrochloride,[40] α-chymotrypsin,[41] [(NH_4) $H_2PW_{12}O_{40}$],[42] visible light,[43] $CuFe_2O_4$,[15] ionic liquids,[44,45] and enzymes[46] have also been investigated in the synthesis of BIMs. Despite their individual merits in moving the field forward, some of these protocols do have limitations, including long reaction times, expensive catalytic systems, low yields, or harsh reaction conditions; and these are particularly acute in light of the need for sustainable green chemistry methods.[47].

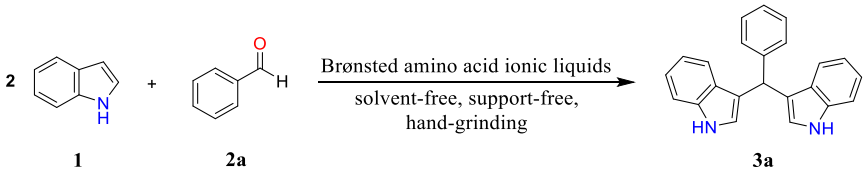
In 1995, F. Toda published an article which gave insight about the significance of organic transformations in the solid state,[48] and since then many solvent-free methods have emerged.[49] Whereas, ionic liquids have also received increasing attention for being a clean and environmentally benign alternative to volatile organic solvents due to their low vapor pressure, non-flammability and high thermo-chemical stability.[50] In this context, we are interested in developing an eco-friendly protocol towards synthesis of BIMs, which could be a potential anticancer drug. Hence, we decided to study the condensation of aldehydes with indole by employing ionic liquids as a source of acid under solvent-free and support-free conditions to afford BIMs. In view of diverse utility of BIMs in pharmacology, we planned to perform in silico structure activity studies of the synthesized BIMs towards anticancer drug targets.

Kinesins are biological motor proteins that are crucial for biological processes such as mitosis, microtubule movement, and intracellular transport. Among them, Eg5 kinesin is a vital spindle motor protein which is responsible for assembly and maintenance of the bipolar spindle during mitosis. This has made Eg5 kinesin an appealing therapeutic target that could impede cell cycle progression through mitosis and induce tumor growth regression.[51,52] Due to the lack of adverse effects and resistance mechanisms, kinesin Eg5 inhibitors are being studied as a chemotherapeutic drugs.[53] In silico studies have recently shown that BIMs have the potential to bind with allosteric inhibitory site of Eg5 kinesin and lock the Eg5 microtubule gliding activity by disengaging the nucleotide-driven conformational changes.[54].

Mitogen-activated protein kinase (MAPK) cascades are signaling pathways involved in several cellular processes like proliferation, differentiation, and growth.[55] There are three main pathways of MAPK: (a) ERK1/2, activated by mitogens; (b) Jun N-terminal kinase (JNK) and (c) p38 pathways are activated by stress and genotoxic stimuli. The p38 alpha is a well-studied target for inflammatory, autoimmune, and cancer disorders. The p38 is a multitasking kinase that regulates a wide range of cellular processes. The role of p38 alpha in cancer has been reported in later stage of tumor development.[56].

Human 3 alpha HSD type 3 belongs to the ketosteroid reductase or hydroxysteroid oxidase enzymes and converts potent hormones (androgen, estrogen) into inactive metabolites and reversely converts metabolites into hormones. Human 3 alpha HSD type 3 is expressed in several tissues, including the liver, lung, brain, prostate, testis, mammary gland, and adrenals. Human 3 alpha HSD type 3 plays a dominant role in the reduction of 5α-dihydrotestosterone to produce the inactive steroid 3α-diol (5α-androstane-3α,17β-diol) in the presence of NADPH, indicating that 3α-HSD3 acts as a pre-receptor regulator of AR (androgen

Table 1
Optimization of reaction conditions for synthesis of 3,3′-(phenylmethylene)bis(1H-indole) **3a** [a].



Entry	Catalyst	Catalyst (mol%)	Temp. (°C)	Time (min.)	Yield (%) [b]
1	–	–	RT	15	Trace
2	–	–	100	60	Trace [c]
3	Prolinium methanesulfonate	40	RT	5	85
4	Prolinium triflate	40	RT	5	90
5	Prolinium p-toluenesulfonate	40	RT	5	85
6	Prolinium nitrate	40	RT	5	84
7	Prolinium hydrogen sulfate	40	RT	5	92
8	Prolinium dihydrogen phosphate	40	RT	5	Trace
9	Prolinium hydrogen sulfate	50	RT	5	93
10	Prolinium hydrogen sulfate	30	RT	5	88
11	Prolinium hydrogen sulfate	20	RT	5	85

[a] Reaction conditions: Indole **1** (2.0 mmol,), benzaldehyde **2a** (1.0 mmol,) and catalyst (given mol%), grinding.
[b] Isolated yields.
[c] Solvent-free heating in a 25 mL round bottom flask.

receptor) in prostate cells.[57] An imbalance in the level of 5α-dihydrotestosterone (5α- DHT) has been implicated in prostate cancer. Currently, androgen ablation therapy with 5α- reductase type 2 inhibitors is used for prostate cancer. However, 3α-HSD being the major component, it can be targeted as an alternate drug target for prostate cancer therapy. Similarly, the down-regulation of Human 3 alpha HSD type 3 has been associated with a decrease in breast cancer cell growth.[58] As a result, an intricate in silico structure activity studies were performed on our series of BIMs in the light of Eg5 kinesin, Human p38 α MAP Kinase, and Human 3 alpha HSD type 3 as a potential protein target.

Results and discussion

Chemistry

As a part of continuing efforts to develop greener protocols in organic synthesis,[59] now we aim at designing an eco-friendly protocol for synthesis of BIMs using amino acid ionic liquids (AAILs). These AAILs are endowed with properties such as non-toxic, biodegradable, biocompatible and low-cost.[60] Among them, protic AAILs have previously been reported in many multicomponent reactions as an acidic catalyst and solvent system.[61–63] In this study, we have screened a series of Brønsted amino acid ionic liquids (Fig. 2) as an acidic catalyst for synthesis of BIMs. A model reaction was performed using indole **1** and benzaldehyde **2a** either in the presence or absence of amino acid ionic liquids to access 3,3′-(phenylmethylene)bis(1H-indole) **3a** and the results are mentioned in Table 1. Initially, no reaction was observed in the absence of acidic catalyst, either by grinding at room temperature or solvent-free heating at 100 °C (Table 1, entries 1, 2). Apparently, the trace amount of product was visible on TLC; this could be due to the fact that some of the benzaldehyde oxidizes to benzoic acid, which further acts as an acidic catalyst to generate the product **3a**.

In pursuit of best catalytic system, indole **1** and aldehyde **2a** were reacted by using a series of Brønsted amino acid ionic liquids under solvent-free conditions by hand grinding in a porcelain mortar and pestle for 5 min. Ionic liquids such as Prolinium methanesulfonate **A**, Prolinium triflate **B**, Prolinium p-toluenesulfonate **C** and Prolinium nitrate **E** gave yields in the range of 84% to 90% with no side reactions (Table 1, entries 3–6). Interestingly, highest yield was obtained using Prolinium hydrogen sulfate ($ProHSO_4$) **F** (Table 1, entry 7). Whereas, trace amount of product was obtained using Prolinium dihydrogen phosphate **D** (Table 1, entry 8). Encouraged by these results, $ProHSO_4$ was selected for further studies. It was found that, increasing the catalytic loading of $ProHSO_4$ did not increased the yield significantly (Table 1, entry 9). Whereas, lowering the catalytic loading resulted in some decrement of yield (Table 1, entries 10, 11). Although, most of the L-proline derived Brønsted amino acid ionic liquids were successful to afford BIM **3a**, but best results were obtained using $ProHSO_4$ under solvent-free grinding at ambient temperature.

With optimized condition in hand, we tried to generalize this protocol towards synthesis of a series of BIMs **3a-3 m** via electrophilic substitution reaction of substituted aryl aldehydes **2a-2 m** with indole **1** (Table 2). Here, aryl aldehydes bearing electron withdrawing groups were highly reactive and gave the corresponding BIMs in 5 min with 84% to 92% yields (Table 2, **3b-3h**). Whereas, aryl aldehydes bearing electron donating groups were relatively slower to afford the corresponding BIMs with 74% to 84% yields (Table 2, **3j-3m**). It was observed that, as the number of methoxy groups in an aldehyde increases, the electrophilicity of aldehyde decreases, which results in lower yield of product. To our delight, this protocol was well tolerated on thiophene-2-carboxaldehyde and resulted in 85% yield with no side reactions (Table 2, **3i**). At last, owing to the medicinal significance, all the synthesized BIMs **3a-3 m** were further subjected to in silico studies.

Note: The highest binding affinity compounds are highlighted in a blue background.

Biology

A target-based screening approach of BIMs have previously demonstrated their ability to bind the allosteric inhibitory site of Eg5 kinesin, a

Table 2
Prolinium hydrogen sulfate mediated synthesis of 3,3′-bis(indolyl)methanes **3a-3 m**.[a]

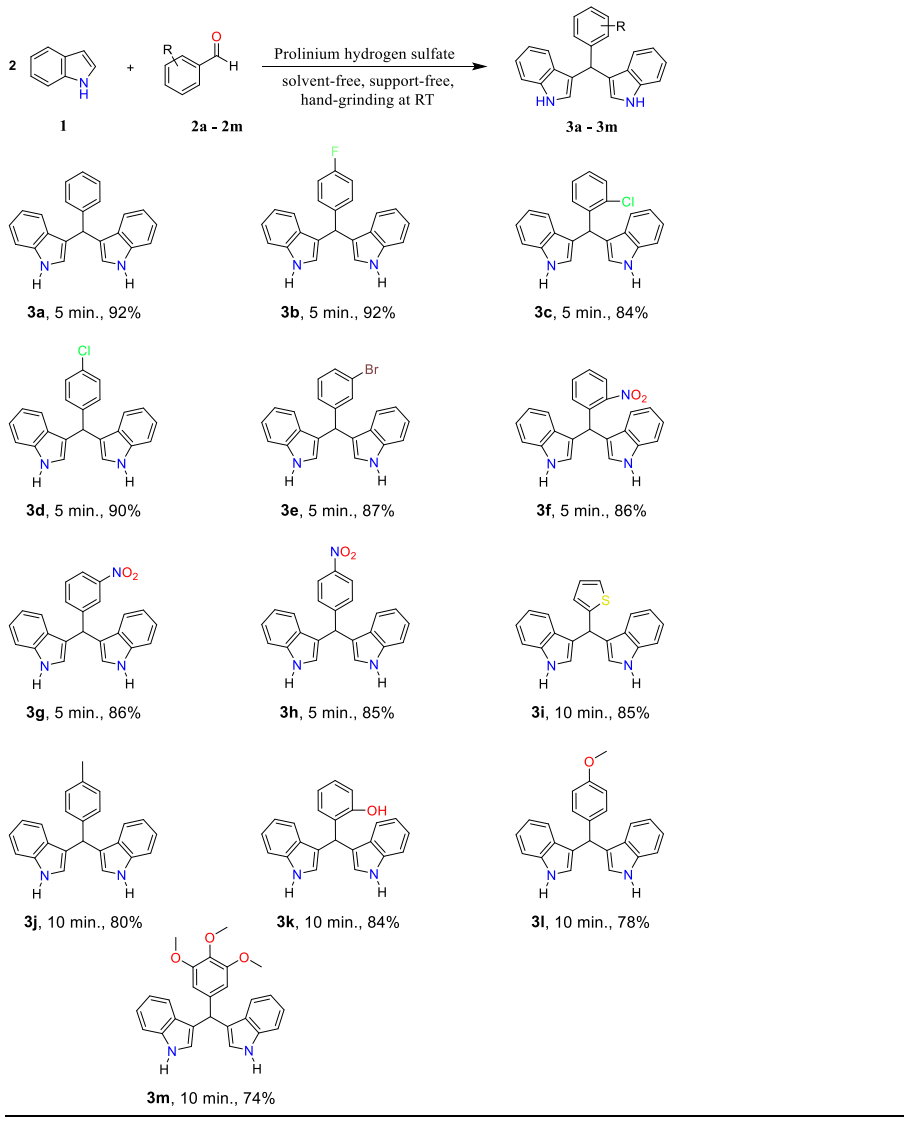


Table 3
Binding energy estimation of BIMS with three potential anticancer target proteins.

BIMs	MW (g/mol)	ΔG (kcal/mol) Eg5 Kinesin (PDB ID 2PG2)	Human MAPK13 (PDB ID 3HVC)	Human 3 alpha HSD type 3 (PDB ID 1 J96)
3a	322.41	−9.9	−8.1	−7.9
3b	340.4	−11	−8.5	−8.3
3c	356.85	−9.2	−8.1	−7.9
3d	356.85	−10	−8	−7.6
3e	401.31	−9.7	−7.8	−7.9
3f	367.41	−8.2	−8.4	−7.8
3g	367.41	−10.3	−8.3	−8.1
3h	367.41	−9.4	−8.3	−7.9
3i	328.43	−9.4	−8.7	−7.4
3j	336.44	−10	−8.2	−7.6
3k	338.41	−9.8	−8.4	−7.5
3l	352.44	−8.8	−8	−7.5
3m	412.49	−9.1	−7.9	−7.8

well-known target of anticancer drugs.[54] Hence, we selected Eg5 kinesin, along with Human p38 α MAP Kinase, and Human 3 alpha HSD type 3 as our target proteins for virtual screening of our library of BIMs belonging to the same chemical class. The 3D protein structures were retrieved from RCSB protein data bank (https://www.rcsb.org/). The protein crystal structures for Eg5 kinesin, Human p38 α MAP Kinase and Human 3 alpha HSD type 3 with PDB ID 2PG2, 3HVC and 1 J96 were selected respectively.[54] In silico studies on binding energy (ΔG) reveal that, the synthesized BIMs have strong binding affinities towards the selected pharmacological targets (Table 3).

Preparation of protein:

RCSB Protein Data Bank was chosen for selection of proteins. Proteins were downloaded with suitable resolutions to favour the docking of given ligands. The proteins were initialized in Biovia Discovery Studio Visualizer v 21.1.0.20298 to view the interactions. Typically, the downloaded proteins are complexed with ligands. The interactions of the ligands were viewed and the amino acid sequences were recorded. Amino acid sequences and residues on the target proteins were explored from the available literature.[64] Then, water molecules and ligand molecules were removed. The complexation of cofactors linked to

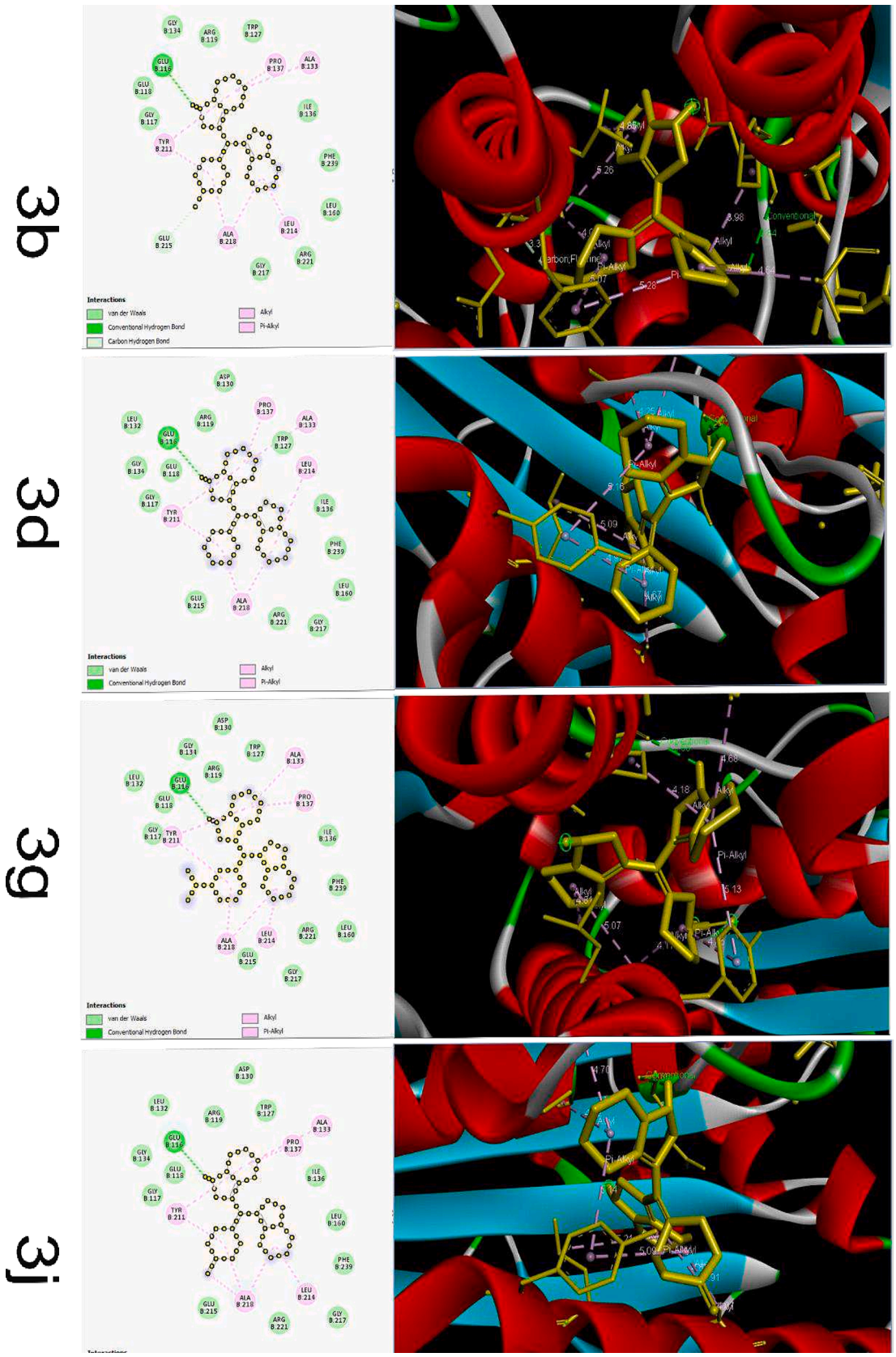


Fig. 3. Molecular docking of BIM derivatives with Eg5 kinesin. The left panel shows ligands and their 2D interactions with amino acid residues, while the right panel exhibits the 3D structure of compounds (yellow-colored), with types of bonds, and distance. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Table 4
Interactions of compounds **3b**, **3d**, **3g** and **3j** with the Eg5 kinesin target protein.

BIMs	Van Der Waals	Hydrogen bonds	Hydrophobic bonds
3b	Gly117, Gly134, Glu118, Phe239, Gly 217, Arg119, Trp127, Arg221, Ile136, Leu160	Glu116	Try211, Pro137, Ala133, Ala218, Leu214
3d	Gly117, Gly134, Glu118, Phe239, Gly217, Arg119, Trp127, Arg221, Ile136, Leu160, Asp130, Glu215	Glu116	Try211, Pro137, Ala133, Ala218, Leu214
3g	Gly117, Gly134, Glu118, Phe239, Gly217, Arg119, Trp127, Arg221, Ile136, Leu160, Asp130, Glu215, Leu132	Glu116	Try211, Pro137, Ala133, Ala218, Leu214
3j	Gly117, Gly134, Glu118, Phe239, Gly217, Arg119, Trp127, Arg221, Ile136, Leu160, Asp130, Glu215, Leu132	Glu116	Try211, Pro137, Ala133, Ala218, Leu214

proteins were kept intact while removing the ligand, as it is the critical step to mimic in vivo-biology. For example, Eg5 kinesin has two domains such as tail and motor domain. The allosteric binding pocket of the motor domain was retrieved through the literature.[64] Polar hydrogen atoms were added and proteins were saved in the format of a protein data bank (pdb) file. The protein data bank file was opened in the AutoDock Tools Version 1.5.7. The protein was saved in the.pdbqt extension. The noted amino acid sequences were earmarked; the grid was applied; and adjusted to the earmarked amino acid sequences. The output grid dimensions file was saved.

Energy minimization of compounds and preparation of ligand:

The structures of compounds were drawn using the Biovia Discovery Studio Visualizer v 21.1.0.20298 and the files were saved in the format of Sybyl MOL2 files. The Sybyl MOL2 files of compounds were opened using the UCSF Chimera Version 1.16. Hydrogen atoms were added to the structure. The Gasteiger charges were applied to the compounds. Structure was minimized and the files were saved in MOL2 format. The MOL2 files of compounds were opened in AutoDock Tools Version 1.5.7. The ligand was prepared after root selection and torsion number selection. The ligand file was saved in.pdbqt extension.

Molecular docking:

Docking studies were executed using the Biovia Discovery Studio Visualizer v 21.1.0.20298 and MGL tools Version 1.5.7. Atomic coordinates of the Eg5 complex were taken after a brief literature review. The amino acid sequences from the literature were considered as the reference for allosteric binding sites of Eg5 kinesin.[64] The ligand co-crystallized with the protein was removed. The allosteric binding sites were validated using the co-crystallised ligand of 2PG2 protein and the Monastrol which is considered as the first prototype small molecule inhibitor of Eg5 kinesin. The amino acid sequence and structure of Eg5 kinesin is a highly conserved sequence across most species. The Eg5 is homomeric protein having N terminal motor domain, central stalk domain and C terminal tail domain. The N terminal motor domain consists of ~ 330 amino acids with allosteric binding pockets having Glu116, Glu117, Glu118, Arg119 residue. Most Eg5 kinesin inhibitors use allosteric sites formed by alpha helix 2, loop5 and alpha helix 3. We used the same site for molecular docking after validating with Eg5 kinesin inhibitor Monastrol and co-crystallized ligand *K*01 (Fig. 3, Table 4).

Similarly, other two target proteins namely Human p38 α MAP Kinase and Human 3 alpha HSD type 3 were searched for their binding sites through the brief literature review. The p38 α MAP Kinase has ATP binding sites and non-ATP binding sites. The ATP binding site is covered by the N and C terminal which are connected with backbone residues 106– 110 of p38 α MAP Kinase and His107 and Met109 residues directly form hydrogen bonds with adenine ring of ATP. The pocket thus formed has conserved Gly-X-Gly-X-X-Gly sequence. There are three additional non ATP binding hydrophobic regions (HR) called HR I, HR II and allosteric site DFG. The HR I region is located behind ATP binding site having Ala51, Lys53, Leu75, Ile84, Leu104, Thr106, and Leu167 residue, while HR II region includes a solvent-exposed hydrophobic area marked by residues Val30, Ile108, Gly110, Ala111, and Asp112. The allosteric site adjacent to the HRI region has an Asp168-Phe169-Gly170 (DFG) motif which is flipped out (DFG in), an inactive state, exactly opposite to its 'DFG out' active confirmation state. We have applied docking at ATP binding site, since most co-crystallized ligands are targeted at this site, irrespective of DFG confirmation. The molecular docking was validated using co-crystallized ligand GG5 (Fig. 4, Table 5).

Similarly, the third protein Human 3 alpha HSD type 3 (PDB ID: 1 J96) was complexed with the ligand (Fig. 5, Table 6). The co-crystallized ligand (TES) was removed and amino acid sequences were made a note of. The binding site was validated and a given set of compounds were docked on the recorded sites. The molecular docking exhibited the highest binding affinity (Table 3) for the selected three anti-cancer target proteins and are found to be better or comparable to co-crystallized ligands or small molecules (**Supplementary Tables S1, S2, and S3**). The binding affinities of **3b**, **3d**, **3 g** and **3j** are higher (-11, −10, −10.3 and −10 kcal/mol respectively) compared to reported inhibitor Arry520 (Filanesib; targets Eg5 Monastrol binding site and co-crystallized ligand of Eg5 kinesin protein; PDB id: 6hky) with −8.2 kcal/mol.[54] The interactions of these compounds showed array of hydrophobic interactions with α2, α3 helices and loop5 amino acid residues.

Similarly, the BIM derivatives **3b**, **3f**, **3i** and **3 k** exhibited higher binding affinities −8.5, −8.4, −8.7 and −8.4 kcal/mol respectively compared to co-crystallized ligands (**Supplementary Table S2**). The bound inhibitor of 3HVC crystal structure used for docking classified as ATP site inhibitor and belongs to Type 1 inhibitors (T1I). T1I are ATP-competitive small molecules binding at ADP binding region forming hydrogen bond with Met 109 amino acid without altering DFG allosteric site. As shown in (Table 5) and (Fig. 4), BIM derivatives delineated backbone residues 106– 110 of p38 α MAP Kinase having hydrogen Met109 residues that directly forms hydrogen bonds with adenine ring of ATP. Moreover, derivatives **3b**, **3i** and **3 k** are found to disturb DFG activation state by establishing hydrogen bonds with Asp168 (Table 5). These compounds are found to interact with HR I, HRII amino acid residues along with DFG motif.

Molecular docking of these compounds with Human 3 alpha HSD type 3 target protein yielded two potential molecules **3b** and **3 g** having binding affinity of −8.3 and −8.1 kcal/mol respectively. These compounds revealed an array of hydrophobic interactions compared to its co-crystallized inhibitor ligand TES (**Supplementary Table S3**).

Conclusion

In summary, we have introduced Brønsted amino acid ionic liquid in the form of prolinium hydrogen sulfate as an efficient catalyst for the first time to afford functionalized BIMs. This method is operationally

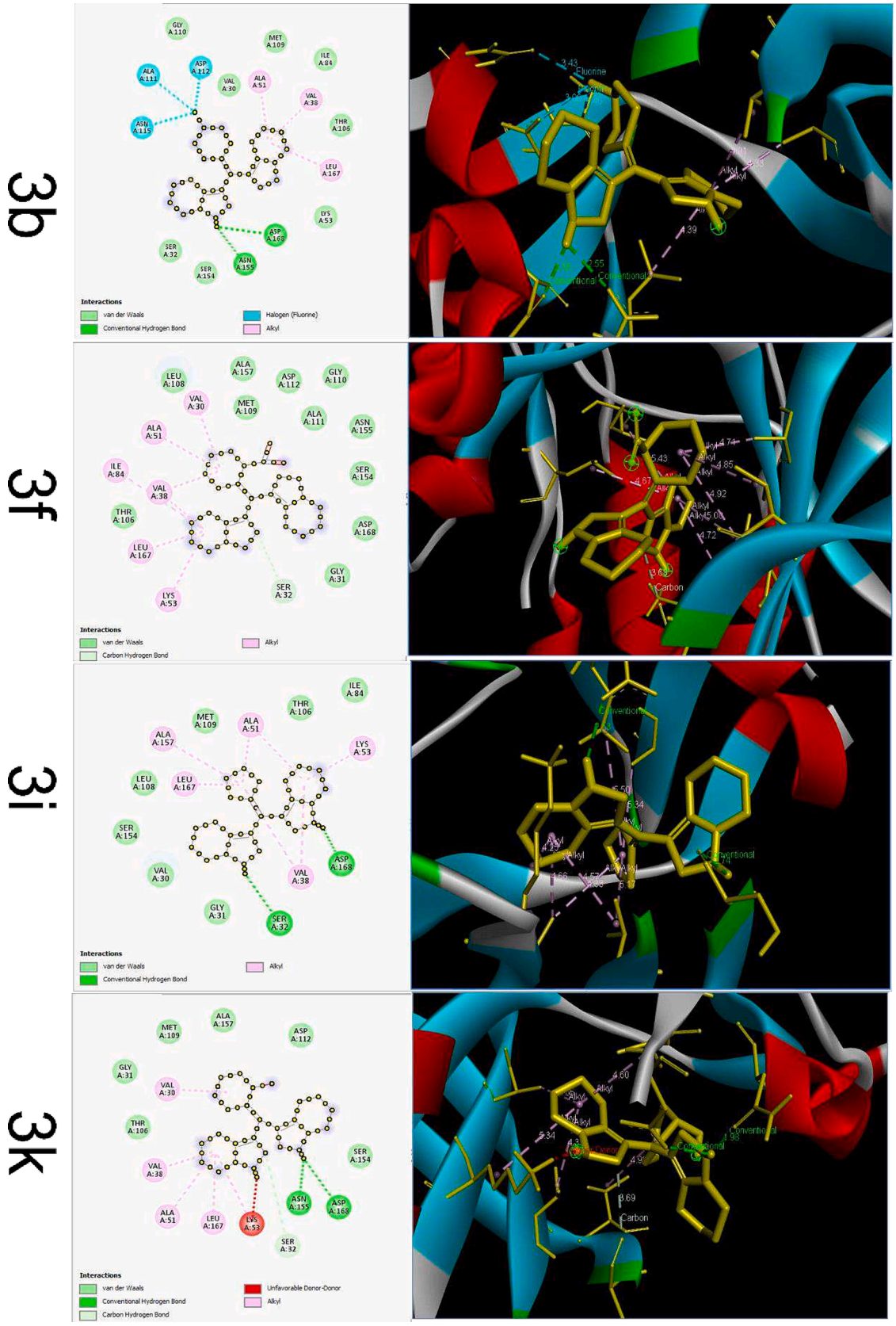


Fig. 4. Molecular docking of BIM derivatives with Human p38 α MAP Kinase. The left panel shows ligands and their 2D interactions with amino acid residues, while the right panel exhibits the 3D structure of compounds (yellow-colored), with types of bonds, and distance. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Table 5
Interactions of compounds **3b**, **3f**, **3i** and **3k** with the MAPK target protein.

BIMs	Van Der Waals	Hydrogen bonds	Hydrophobic bonds	Halogen bonds/ unfavourable bonds
3b	Gly110, Met109, Ile84, Val30, Ser32, Ser154, Thr106, Lys53	Asn155, Asp168	Leu167, Ala51, Val38	Asp112, Ala111, Asn115
3f	Thr106, Leu108, Ala157, Met109, Asp112, Ala111, Gly110, Asn155, Ser154, Asp168, Gly31	Ser32	Tyr53, Leu167, Val38, Ile84, Ala51, Val30	
3i	Leu108, Ser154, Met109, Thr106, Ile84, Val30, Gly31	Ser32, Asp168	Ala157, Leu167, Ala51, Lys53, Val38	
3k	Thr106, Met109, Ala157, Asp112, Ser154, Ser32	Asp168, Asn155	Val30, Val38, Ala51, Leu167	Lys53

Table 6
Interactions of compounds **3b**, and **3g** with the H3AHSD target protein.

BIMs	Van Der Waals	Hydrogen bonds	Hydrophobic bonds
3b	Pro26, Lys31, Asn56, Tyr55, Val54, Phe311, Ile310	Glu224	Tyr24, Ala27, Leu308, Trp227, Trp86, Val128, Ile129
3g	Pro26, Lys31, Val54, Tyr55, Asn56, His117 Val128, Leu308, Ile310, Phe311	Lys131, Glu224	Tyr24, Ala27, Trp86, Ile129, Trp227

simple, mild, rapid, and solvent-free with no need of conventional heating. Molecular docking studies presented compounds having higher affinities for three target proteins namely Eg5 kinesin, Human p38 α MAP Kinase and Human 3 alpha HSD type 3. These potential compounds need further validation for their pharmacological activity using biological screening method. With a simple and efficient green protocol, we have synthesized a series of BIM derivatives and their in-silico evaluation provided data about their potential pharmacological activity. We hope that the ease of preparation of the title compounds and their numerous applications will foster continued exploration of their unique properties and their development into lead molecules for therapeutic application.

CRediT authorship contribution statement

Vikrant Kumbhar: Investigation, Methodology, Writing – original draft. **Rutik Raskar:** Investigation, Methodology. **Radha Chafle:** Investigation, Methodology, Formal analysis. **Vandana Nikam:** Investigation, Methodology, Writing – original draft. **Avinash Kumbhar:** Supervision. **Ramdas Pawar:** Supervision. **Manohar Chaskar:** Supervision. **Gulab Gugale:** Supervision. **Bhushan Khairnar:** Validation, Supervision.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

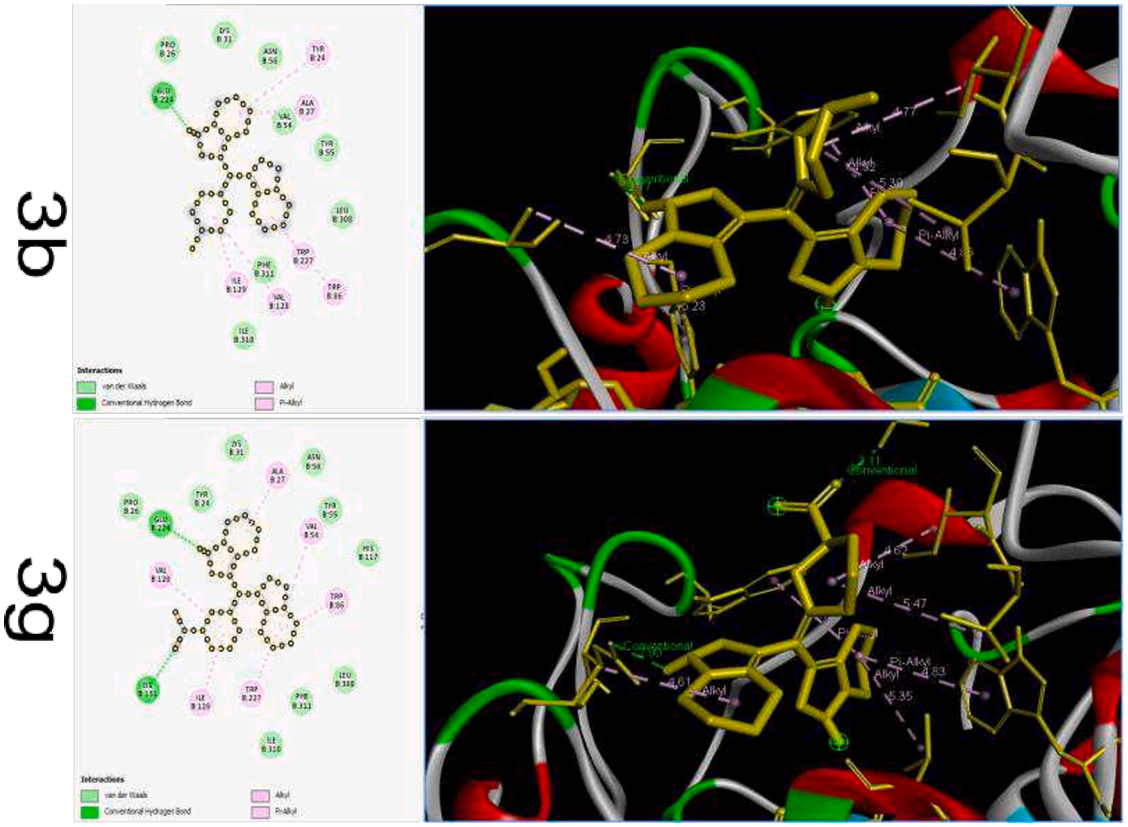


Fig. 5. Molecular docking of BIM derivatives with Human 3 alpha HSD type 3. The left panel shows ligands and their 2D interactions with amino acid residues, while the right panel exhibits the 3D structure of compounds (yellow-colored), with types of bonds, and distance. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Data availability

No data was used for the research described in the article.

Acknowledgment

Vikrant Kumbhar is thankful to UGC-New Delhi for providing a research fellowship.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.rechem.2023.101023.

References

[1] P. Praveen, P. Parameswaran, M. Majik, Synthesis (Stuttg) 47 (2015) 1827.
[2] R. Bell, S. Carmeli, N. Sar, J. Nat. Prod. 57 (1994) 1587.
[3] M.A. Zeligs, J. Med. Food 1 (1998) 67.
[4] S. Imran, M. Taha, N.H. Ismail, Curr. Med. Chem. 22 (2015) 4412.
[5] C. Hong, G.L. Firestone, L.F. Bjeldanes, Biochem. Pharmacol. 63 (2002) 1085.
[6] S. Hibino, T. Choshi, Nat. Prod. Rep. 18 (2001) 66.
[7] J.E. Riby, G.H.F. Chang, G.L. Firestone, L.F. Bjeldanes, Biochem. Pharmacol. 60 (2000) 167.
[8] Y. Jiang, Y. Fang, Y. Ye, X. Xu, B. Wang, J. Gu, M. Aschner, J. Chen, R. Lu, Front. Pharmacol. 10 (2019) 1.
[9] K. Arffmann, Treatment of fibromyalgia and related disorders, WO 99/17768 (1999).
[10] J.R. Majer, Tetrahedron 9 (1960) 106.
[11] R. Martínez, A. Espinosa, A. Tárraga, P. Molina, Tetrahedron 64 (2008) 2184.
[12] X. He, S. Hu, K. Liu, Y. Guo, J. Xu, S. Shao, Org. Lett. 8 (2006) 333.
[13] M.P. Muñoz, M.C. de la Torre, M.A. Sierra, Chem. - A Eur. J. 18 (2012) 4499.
[14] D. Xia, Y. Wang, Z. Du, Q.-Y. Zheng, C. Wang, Org. Lett. 14 (2012) 588.
[15] N.-K. Nguyen, M.-T. Ha, H.Y. Bui, Q.T. Trinh, B.N. Tran, V.T. Nguyen, T.Q. Hung, T.T. Dang, X.H. Vu, Catal. Commun. 149 (2021), 106240.
[16] J. Xiang, J. Wang, M. Wang, X. Meng, A. Wu, Org. Biomol. Chem. 13 (2015) 4240.
[17] B. Vijayakumar, N.D. Shakthi, Indian J. Adv, Chem. Sci. 1 (2013) 221.
[18] M. Hosseini-Sarvari, Acta Chim. Slov. 54 (2007) 354.
[19] L. Mo, Z. Ma, Z. Zhang, Synth. Commun. 35 (2005) 1997.
[20] Z.-H. Zhang, L. Yin, Y.-M. Wang, Synthesis (Stuttg) 2005 (2005) 1949.
[21] T. Pillaiyar, M. Dawood, H. Irum, C.E. Müller, Arkivoc 2018 (2017) 1.
[22] P.R. Singh, D.U. Singh, S.D. Samant, Synth. Commun. 35 (2005) 2133.
[23] A.K. Maiti, P. Bhattacharyya, *J. Chem. Res.*, **S** 424 (1997).
[24] L.Z. Fekri, M. Nikpassand, M. Kohansal, Russ. J. Gen. Chem. 85 (2015) 2861.
[25] M. Karthik, A. Tripathi, N. Gupta, M. Palanichamy, V. Murugesan, Catal. Commun. 5 (2004) 371.
[26] C. Ramesh, J. Banerjee, R. Pal, B. Das, Adv. Synth. Catal. 345 (2003) 557.
[27] S.R. Mendes, S. Thurow, M.P. Fortes, F. Penteado, E.J. Lenardão, D. Alves, G. Perin, R.G. Jacob, Tetrahedron Lett. 53 (2012) 5402.
[28] S.N. Mhaldar, K.S. Mandrekar, M.K. Gawde, R.V. Shet, S.G. Tilve, Synth. Commun. 49 (2019) 94.
[29] V. Kumar, C. Singh, U. Sharma, P. K. Verma, B. Singh, and N. Kumar, *Indian J. Chem. - Sect. B Org. Med. Chem.*, **53**, 83 (2014).
[30] R. Vaid, M. Gupta, O.S. Chambyal, R. Gupta, J. Chem. Sci. 127 (2015) 987.
[31] B.P. Bandgar, A.V. Patil, V.T. Kamble, Arkivoc 2007 (2008) 252.
[32] K.P. Boroujeni, K. Parvanak, Chinese Chem. Lett. 22 (2011) 939.
[33] D.M. Pore, U.V. Desai, T.S. Thopate, P.P. Wadgaonkar, Arkivoc 2006 (2006) 75.
[34] V.T. Kamble, K.R. Kadam, N.S. Joshi, D.B. Muley, Catal. Commun. 8 (2007) 498.
[35] P. Katrun, K. Atthawilai, C. Kuhakarn, Arkivoc 2021 (2021) 190.
[36] R. Ghorbani-Vaghei, H. Veisi, H. Keypour, A.A. Dehghani-Firouzabadi, Mol. Divers. 14 (2010) 87.
[37] H. Veisi, R. Gholbedaghi, J. Malakootikhah, A. Sedrpoushan, B. Maleki, D. Kordestani, J. Heterocycl. Chem. 47 (2010) 1398.
[38] H. Koshima, W. Matsusaka, J. Heterocycl. Chem. 39 (2002) 1089.
[39] H. Alinezhad, A.H. Haghighi, Casp. J. Chem 1 (2012) 1.
[40] S. Mathavan, K. Kannan, R.B.R.D. Yamajala, Org. Biomol. Chem. 17 (2019) 9620.
[41] Z.-B. Xie, D.-Z. Sun, G.-F. Jiang, Z.-G. Le, Molecules 19 (2014) 19665.
[42] B. Y. Giri, B. L. A. Prabhavathi Devi, K. Vijayalakshmi, R. B. N. Prasad, N. Lingaiah, and P. S. Sai Prasad, *Indian J. Chem. - Sect. B Org. Med. Chem.*, **51**, 1731 (2012).
[43] Z.M. Salem, J. Saway, J.J. Badillo, Org. Lett. 21 (2019) 8528.
[44] A.K. Chakraborti, S.R. Roy, D. Kumar, P. Chopra, Green Chem. 10 (2008) 1111.
[45] Y.-M. Ren, M.-D. Xu, X. Wang, Catalysts 7 (2017) 300.
[46] Y. Fu, Z. Lu, K. Fang, X. He, H. Xu, Y. Hu, RSC Adv. 10 (2020) 10848.
[47] V.G. Zuin, I. Eilks, M. Elschami, K. Kümmerer, Green Chem. 23 (2021) 1594.
[48] F. Toda, Acc. Chem. Res 28 (1995) 480.
[49] C. Espro, D. Rodríguez-Padrón, Curr. Opin. Green Sustain. Chem. 30 (2021), 100478.
[50] A.J. Greer, J. Jacquemin, C. Hardacre, Molecules 25 (2020) 1.
[51] D. Weil, L. Garçon, M. Harper, D. Duménil, F. Dautry, M. Kress, Biotechniques 33 (2002) 1244.
[52] B.C. Guido, L.M. Ramos, D.O. Nolasco, C.C. Nobrega, B.Y. Andrade, A. Pic-Taylor, B.A. Neto, J.R. Corrêa, BMC Cancer 15 (2015) 283.
[53] G.D. Marconi, S. Carradori, A. Ricci, P. Guglielmi, A. Cataldi, S. Zara, Molecules 24 (2019) 3948.
[54] K.A. Chavan, M. Shukla, A.N.S. Chauhan, S. Maji, G. Mali, S. Bhattacharyya, R. D. Erande, ACS Omega 7 (2022) 10438.
[55] M. Burotto, V.L. Chiou, J.-M. Lee, E.C. Kohn, Cancer 120 (2014) 3446.
[56] A. Martínez-Limón, M. Joaquin, M. Caballero, F. Posas, E. de Nadal, Int. J. Mol. Sci. 21 (2020) 1.
[57] T.L. Rizner, H.K. Lin, T.M. Penning, Chem. Biol. Interact. 143–144 (2003) 401.
[58] B. Zhang, X.-J. Hu, X.-Q. Wang, J.-F. Thériault, D.-W. Zhu, P. Shang, F. Labrie, S.-X. Lin, Biochem. J. 473 (2016) 1037.
[59] V. Kumbhar, S. Gaiki, R. Raskar, A. Kumbhar, B. Khairnar, Results Chem. 4 (2022), 100589.
[60] H. Ohno, K. Fukumoto, Acc. Chem. Res. 40 (2007) 1122.
[61] N. Rode, A. Tantray, A. Shelar, R. Patil, S. Terdale, Synth. Commun. 52 (2022) 190.
[62] N.R. Rode, A.A. Tantray, A.V. Shelar, R.H. Patil, S.S. Terdale, Res. Chem. Intermed. 48 (2022) 2391.
[63] R. Ratti, Adv. Chem. 2014 (2014) 1.
[64] X. Luo, M. Shu, Y. Wang, J. Liu, W. Yang, Z. Lin, Molecules 17 (2012) 2015.