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Protein-Ligand Binding Interactions of 4-Nitroimidazolium Salts with Breast Cancer Protein: A Computational Biology Study

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ABSTRACT

Breast cancer is the most foremost cause of the most cancer demises in women. In normal cells, BRCA1 and BRCA2 make certain the stability of DNA and also preclude hysterical cell progression. Metamorphosis of these genes is related to the expansion of hereditary breast and ovarian cancers. Bearing in mind the lacunae of consistent and prospective medications to remedy the lifetime intimidating most breast cancers, the present work has attention on molecular docking evaluation to ascertain the prospective binding sites and binding energies of 1-substituted-2-methyl-4-nitroimidazoles, nine protonated 4-nitroimidazolium cations and five aromatic carboxylate anions. Doxorubicin and vinorelbine were also docked with breast cancer protein (PDB code: **3K0K**) and the protein binding sites of these standard drugs were also identified. The results exposed that among the docked 4-nitroimdazoles, 4-nitroimidazolium cations and organic anions were found efficient in binding interactions and in wrecking the protein liable towards breast cancer.

KEYWORDS

4-Nitroimidazole, 4-Nitroimidazolium salt, Benzoate, Breast cancer, Docking, Protein-ligand interaction, Computational biology.

INTRODUCTION

Cancer remains the world as one of the utmost painstaking illnesses globally and is the second principal cause of human transience [1,2]. The cancer responsible number of fatalities has amplified from around 5.7 million in 1990 to 9.6 million or about one in six deaths in 2018. In 2020, 16-20 cancer deaths have been registered every 1 minute globally and based on the present forecasts, it is believed that cancer deaths will continue to escalate to an estimated 11.4 million dying in 2030 [3]. The load of the most frequent forms of cancer is in downward order of breast (female), lung and bronchus, prostate (both male and female), colon and rectum, melanoma of the skin, bladder, non-Hodgkin lymphoma, kidney and renal pelvis, endometrial, leukemia, pancreatic, thyroid and liver cancer [4]. Rapidly separating cells, for instance, skin, breast and uterine cells are at greater peril of changes contrasted and cells, which constraint partition and this way are progressively inclined to create cancer. Breast cancer (BC) is one of the furthermost happening tumours prompting critical dreariness and mortality among

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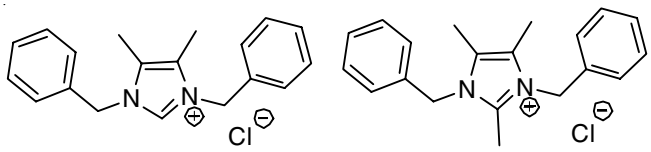
females [5,6]. An understanding of cancer disease statistical data points intended for 2016-2017 uncovered an expected numeral of passings of 41,070 concluded BC amongst men and women in the United States [7]. The circumstance is more dreadful in India than in other countries, wherever breast cancer accounts 25-32% of altogether malignant growth cases. As per the ICMR (Indian Council of Medical Research) study description of 2016 and expected newfangled aggregate of malignant growth inpatients was 14.5 million out of 2016, which will prone to stretch nearby 17.3 million by 2020 [8].

Amongst the vital clinical procedures for the cure of breast cancer involves specifically restricting the binding of estrogen to estrogen receptor alpha (ER-α) and beta (ER-β) and brought about the discerning of SERMs (selective estrogen receptor modulators). Estrogen has been accounted for to assume a crucial job in the development and advancement of mammary organs. Communication of estrogen together with ER-α and ER-β invigorates the expansion of mammary cells. Michigan cancer foundation-7 (MCF-7) cells, being ERα subordinate, are seen as delicate to SERMs while Monroe Dunaway Anderson-Metastasis breast cancer-231 (MDA-MB-231) cells, which are ER-β subordinate are described by the skiving of immune-histochemical articulation of estrogen, progesterone and HER2 (human epidermal growth factor receptor 2) receptors [9-12]. Triarylethylenes subordinates, alike tamoxifen (TAM), raloxifene and toremifene are endorsed SERMs by US FDA (Food and Drug Administration) together with critical anti-BC likeness [13]. TAM, a rival, is the chief-line medication utilized for the cure of breast cancer. Be that as it may, just 70% of ER-α cases react to TAM, although 30-40% sufferers through adjuvant TAM treatment in the long run relapse [14-16]. Moreover, TAM negatively affects the endometrium generating endometrial tumour and causes hot flashes, nausea, while raloxifene causes hot flashes, joint or muscle pain, white vaginal discharge, melancholy, insomnia and dizziness and toremifene causes hot flashes, white vaginal discharge and vomiting [17].

Imidazoles and their derivatives have pulled in incredible interests in medicinal chemistry on account of their flexible pharmacological characteristics, for instance, antifungal, antibacterial, antituberculosis, anticancer and sedative properties [18-23]. For example, two naturally available imidazolium salts based alkaloids namely Lepidiline A and B (Fig. 1) were isolated from the *Lepidium meyenii* Walpers (Brassicaceae) roots and revealed their prospective cytotoxic activity in contradiction of the human cancer cell lines [24]. For example, in antibreast cancer activity, 2,1,3-benzothiadiazole (BTD) moiety containing imidazole and imidazolium subordinates [25], pyrene-imidazolium derivatives [26], imidazolium-based amino acid ionic liquids [27], 1-(carboxyalkyl)-3-(12-mercapto-dodecyl)-1*H*-imidazolium ionic liquids with different inorganic anions (Br^-, BF_4^-, PF_6^-, ClO_4^- and NTf_2^-) [28], 3-methyl-1-sulfonic acid imidazolium chloride [29], 1-butyl-3-methyl imidazolium chloride and tetrafluoroborate and 1-methyl-3-octyl imidazolium chloride [30], benzothiazole amide functionalized imidazolium ionic liquids [31], 1-((indol-3-yl)methyl)-1*H*-imidazolium salts [32] were also investigated. Given this background, they are evidenced the importance of imidazole(ium) derivatives results to a large extent against breast cancer cells.



Lepidiline A Lepidiline B

Fig. 1. Representative structures of lepidiline A and lepidiline B

A series of 4-nitroimidazole derivatives (**1a**, **3a-i**) and their protic 4-nitroimidazolium derivatives are synthesized and characterized. 4-Nitroimidazolium salts with five different aromatic carboxylate based organic anions [33-35]. The current research focuses on the molecular docking studies of the previously synthesized nine 4-nitroimidazoles and their protonated 4-nitroimidazolium salts, as well as five aromatic carboxylate based organic anions, with the ultimate goal of developing strong anti-breast cancer drugs using a bio-computational technique (PDB code: 3K0K).

EXPERIMENTAL

4-Nitroimidazoles (**1a**, **3a-i**) and their 4-nitroimidazolium salts were employed in this study. 4-Nitroimidazolium salts were synthesized with five different aromatic carboxylate based organic anions [33-35]. Sigma Aldrich Chemical Co. Ltd., Mumbai, provided the 2-methyl-4(5)-nitroimidazole (**1a**). PubMed was used to get the structures of standard breast cancer medications such as doxorubicin and vinorelbine [36,37]. RCSB provided the 3D crystal structure of the breast cancer type 1 (BRCA1) susceptibility protein (PDB code: 3K0K) [38].

Molecular docking: The 1-click docking tool was used to simulate binding interactions of compounds **1a**, **3a-i**, protic 4-nitroimidazolium cations and aromatic carboxylate anions and the docking poses and binding modes were displayed [39,40].

RESULTS AND DISCUSSION

The molecular docking study was simulated to inspect the binding sites, binding mode and binding energies of 2-methyl-4(5)-nitroimidazole (**1a**), nine 4-nitroimidazole derivatives (**3a-i**), nine protic based 4-nitroimidazolium cations [**3a**-H]$^+$-[**3i**-H]$^+$, five different aromatic carboxylate organic anions (Fig. 2) and the standard drugs (doxorubicin and vinorelbine) with the breast cancer type 1 (BRCA1) susceptibility protein (PDB code: 3K0K).

Binding affinity: The binding affinities for the molecule **1a** are -3.5 kcal/mol, the 1-benzylated 4-nitroimidazoles **3a-f** have from -4.8 to -5.2 kcal/mol and the 1-butylated 4-nitro-imidazoles **3g-i** have from -3.9 to -4.2 kcal/mol, according to molecular modelling data (Table-1). Because of the substitution of 4-methylbenzyl and 4-nitrobenzyl groups in **1a**, 4-nitro-imidazoles **3e** & **3f** have a higher affinity (-5.2 kcal/mol). The kcal/mol range of the protic 4-nitroimidazolium salts [**3a**-H]$^+$-[**3i**-H]$^+$ is -4.0 to -5.2 (Table-2). The cation [**3h**-H]$^+$ shares the same binding affinity as its parent molecule **3h**. 4-Nitroimidazolium cations [(**3a,b,d,g,i**)-H]$^+$ have binding affinities of -0.1 to -0.4 kcal/mol, while the remainder of the cations have binding affinities of -0.1 to -0.3 kcal/mol. When compared to their parent 4-nitroimidazoles, five cations [(**3a,b,d,g,i**)-H]$^+$

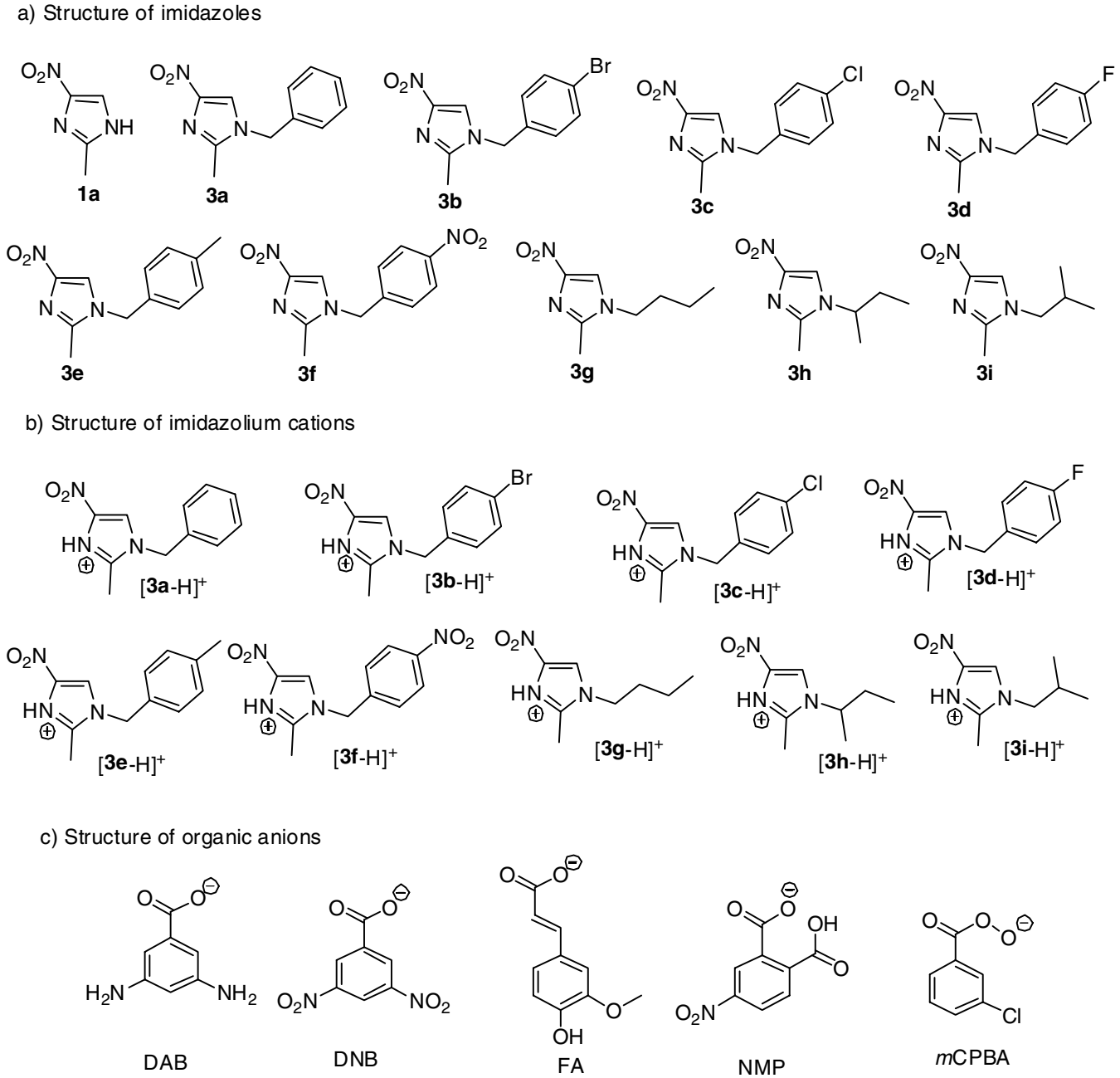


Fig. 2. Representative structures [33-35] of (a) 4-nitroimidazoles, (b) 4-nitroimidazolium cations, (c) aromatic carboxylic anions were used for docking studies in this work

interacted considerably with the targeted breast cancer protein (**3a, b, d, g & i**). Aromatic carboxylate anions have binding affinities of -4.0, -5.0, -4.7, -5.0 and -4.6 kcal/mol for DAB, DNB, FA, NMP and *m*CPBA, respectively (Table-3). The anions DNB and NMP bind to the protein more strongly than the other anions. Doxorubicin and vinorelbine, two common chemotherapy drugs, have the same binding affinity for the protein (-6.8 kcal/mol) (Table-3).

Hydrophobic binding interactions: The number of hydrophobic binding contacts, binding residues, binding distance, implicated ligand and protein atoms and standard medications of 4-nitroimidazoles, protic 4-nitroimidazolium cations, aromatic carboxylate anions and standard pharmaceuticals are all shown in Tables 1-3. 2-Methyl-4(5)-nitroimidazole (**1a**) has two hydrophobic interactions with the VAL-92A and VAL-93A breast cancer protein residues, with distances of 3.79 and 3.94, respectively (Fig. 3). The organic anions DNB and *m*CPBA have a hydrophobic interaction, 4-nitroimidazole **3a**, **3i**, 4-nitroimidazoleium cation [**3g**-H]$^+$ & anion DAB have a pair of hydrophobic binding contacts and the cation [**3i**-H]$^+$ has three hydrophobic binding interactions with the protein PHE-14A (Figs. 3-5). Vinorelbine, a routinely prescribed drug, has a hydrophobic interaction with the PHE-14A protein location. The LYS-54A residue of the breast cancer protein has been discovered to interact with doxorubicin (Fig. 6). It also revealed three interactions, with vinorelbine interacting with the protein's LEU-53A residue being one of them. The hydrophobic interaction of imidazole **3b**, **3e**, **3h** and cations [(**3a-c,e,f**)-H]$^+$ with the residue LYS-54A has been demonstrated. Imidazoles **3b**, **3e** and cations [(**3a,e,f**)-H]$^+$ exhibited a

TABLE-1
HYDROPHOBIC BINDING INTERACTIONS OF 4-NITROIMIDAZOLES (**1a**, **3a-i**)

Imidazole(s)	Binding affinity	Binding index	Protein residue	Amino acid	Distance (Å)	Atom	
						Ligand	Protein
1a	-3.5	1	92A	VAL	3.79	2065	924
		2	93A	VAL	3.94	2065	932
3a	-4.8	1	14A	PHE	3.74	2076	123
		2	14A	PHE	3.44	2072	125
3b	-4.9	1	32A	ILE	3.90	2076	318
		2	53A	LEU	3.65	2076	523
		3	54A	LYS	3.48	2078	530
3c	-5.1	1	31A	LEU	3.81	2074	310
		2	52A	THR	3.76	2065	513
		3	54A	LYS	3.90	2078	531
		4	54A	LYS	3.55	2076	530
3d	-4.8	1	56A	PHE	3.39	2076	560
3e	-5.2	1	31A	LEU	3.88	2077	310
		2	53A	LEU	3.68	2076	523
		3	54A	LYS	3.46	2078	530
3f	-5.2	1	56A	PHE	3.76	2071	560
		2	191A	LEU	3.58	2071	1872
3g	-3.9	1	51A	ARG	3.83	2065	494
		2	56A	PHE	3.62	2065	560
		3	126A	ASN	3.65	2072	1274
3h	-4.0	1	53A	LEU	3.74	2073	523
		2	53A	LEU	3.80	2072	520
		3	54A	LYS	3.49	2073	530
3i	-4.2	1	6A	VAL	3.95	2065	59
		2	11A	PRO	3.98	2073	97
		3	14A	PHE	3.74	2074	125
		4	14A	PHE	3.68	2065	127

TABLE 2
HYDROPHOBIC BINDING INTERACTIONS OF PROTONATED 4-NITROIMIDAZOLIUM CATIONS

Imidazolium cation(s)	Binding affinity	Binding index	Protein residue	Amino acid	Distance (Å)	Atom	
						Ligand	Protein
[**3a**-H]$^+$	-4.9	1	31A	LEU	3.96	2077	310
		2	32A	ILE	3.63	2075	318
		3	53A	LEU	3.74	2077	523
		4	54A	LYS	3.74	2075	530
[**3b**-H]$^+$	-5.0	1	52A	THR	3.68	2065	513
		2	53A	LEU	3.71	2075	520
		3	53A	LEU	3.76	2077	523
		4	54A	LYS	3.75	2075	530
[**3c**-H]$^+$	-5.0	1	52A	THR	3.68	2065	513
		2	53A	LEU	3.69	2075	520
		3	53A	LEU	3.83	2077	523
		4	54A	LYS	3.62	2075	530
[**3d**-H]$^+$	-5.2	1	51A	ARG	3.69	2077	494
		2	56A	PHE	3.55	2065	560
		3	93A	VAL	3.77	2077	932
		4	191A	LEU	3.68	2065	1872
[**3e**-H]$^+$	-4.9	1	31A	LEU	3.87	2075	310
		2	53A	LEU	3.64	2076	523
		3	54A	LYS	3.76	2079	530
[**3f**-H]$^+$	-5.1	1	31A	LEU	3.70	2075	310
		2	53A	LEU	3.59	2077	523
		3	54A	LYS	3.90	2078	530
[**3g**-H]$^+$	-4.1	1	11A	PRO	3.87	2065	97
		2	14A	PHE	3.76	2075	128
		3	14A	PHE	3.71	2074	129
[**3h**-H]$^+$	-4.0	1	191A	LEU	3.79	2073	1872
[**3i**-H]$^+$	-4.4	1	11A	PRO	3.94	2065	97
		2	14A	PHE	3.85	2075	124
		3	14A	PHE	3.48	2073	127
		4	14A	PHE	3.88	2074	129

TABLE-3
HYDROPHOBIC BINDING INTERACTIONS OF AROMATIC CARBOXYLATE ANIONS AND STANDARD DRUG

Organic anion/ Standard drug	Binding affinity	Binding index	Protein residue	Amino acid	Distance (Å)	Atom	
						Ligand	Protein
DAB	-4.0	1	14A	PHE	3.43	2068	125
		2	14A	PHE	3.83	2067	127
DNB	-5.0	1	6A	VAL	3.90	2068	59
		2	14A	PHE	3.50	2070	125
FA	-4.7	1	191A	LEU	3.51	2069	1872
NMP	-5.0	1	56A	PHE	3.52	2068	560
		2	191A	LEU	3.61	2069	1872
*m*CPBA	-4.6	1	6A	VAL	3.72	2065	59
		2	14A	PHE	3.93	2070	127
Doxorubicin	-6.8	1	53A	LEU	3.79	2069	523
		2	53A	LEU	3.60	2071	523
		3	53A	LEU	3.33	2092	520
		4	54A	LYS	3.62	2068	530
		5	128A	PRO	3.75	2079	1295
Vinorelbine	-6.8	1	6A	VAL	3.76	2087	59
		2	11A	PRO	3.90	2105	97
		3	14A	PHE	3.51	2089	125
		4	30A	ASN	3.63	2101	296
		5	31A	LEU	3.81	2100	310
		6	53A	LEU	3.64	2072	523

contact with LEU-53A of the breast cancer protein, whereas **3h**, [**3b**-H]$^+$ and [**3c**-H]$^+$ showed two interactions (Figs. 3 and 4). These forms of protein interactions have not been observed in the docked organic anions. Doxorubicin interacts with the protein PRO-128A, while vinorelbine interacts with the proteins VAL-6A, PRO-11A, ASN-30A and LEU-31A (Fig. 6). Imidazole

TABLE-4
HYDROGEN BOND BINDING INTERACTIONS OF 4-NITROIMIDAZOLES (**1a**, **3a-i**)

Imidazole(s)	Binding index	Protein residue	Amino acid	Distance (Å)		Donor angle	Atom	
				H-A	D-A		Donor	Acceptor
1a	1	51A	ARG	2.13	3.16	177.77	497 [Ng+]	2070 [N2]
	2	51A	ARG	2.39	3.25	140.30	500 [Ng+]	2074 [O2]
3a	1	7A	SER	2.70	3.48	140.06	66 [O3]	2080 [O2]
	2	8A	GLY	2.05	2.99	155.87	69 [Nam]	2079 [O-]
	3	54A	LYS	3.04	3.76	129.20	533 [N3+]	2080 [O2]
3b	1	7A	SER	2.19	3.03	145.93	66 [O3]	2080 [O-]
	2	8A	GLY	2.85	3.73	146.62	69 [Nam]	2081 [O2]
	3	54A	LYS	2.47	3.48	172.72	533 [N3+]	2070 [N2]
3c	1	7A	SER	3.07	3.71	125.64	66 [O3]	2070 [N2]
	2	8A	GLY	2.22	3.18	158.39	69 [Nam]	2070 [N2]
	3	9A	LEU	2.26	3.21	157.55	74 [Nam]	2081 [O2]
	4	54A	LYS	3.29	3.94	124.04	533 [N3+]	2070 [N2]
3d	1	165A	ASP	3.48	4.00	117.16	1626 [O3]	2070 [N2]
	2	187A	ARG	2.23	3.02	131.97	1825 [Ng+]	2080 [O-]
	3	187A	ARG	2.51	3.24	127.74	1822 [Ng+]	2080 [O-]
	4	188A	GLU	3.15	3.86	133.31	1840 [O3]	2080 [O-]
3e	1	7A	SER	2.23	3.07	146.43	66 [O3]	2081 [O2]
	2	8A	GLY	2.74	3.62	147.18	69 [Nam]	2080 [O-]
	3	54A	LYS	2.29	3.29	174.14	533 [N3+]	2070 [N2]
3f	1	51A	ARG	2.21	3.16	158.83	490 [Nam]	2073 [O-]
	2	51A	ARG	2.73	3.52	133.05	500 [Ng+]	2069 [N2]
	3	51A	ARG	2.05	3.02	156.14	497 [Ng+]	2069 [N2]
	4	53A	LEU	2.24	3.18	157.34	516 [Nam]	2082 [O2]
3g	1	51A	ARG	2.56	3.25	124.02	500 [Ng+]	2070 [N2]
	2	187A	ARG	2.09	3.11	168.22	1825 [Ng+]	2077 [O2]
	3	188A	GLU	3.35	3.67	102.70	1840 [O3]	2077 [O2]
3h	1	7A	SER	1.96	2.84	152.83	66 [O3]	2076 [O-]
	2	8A	GLY	2.09	3.07	166.22	69 [Nam]	2077 [O2]
	3	54A	LYS	2.16	3.16	172.48	533 [N3+]	2070 [N2]
3i	1	7A	SER	3.36	4.05	130.77	66 [O3]	2076 [O-]
	2	8A	GLY	1.89	2.87	165.46	69 [Nam]	2077 [O2]
	3	54A	LYS	2.40	3.15	130.49	533 [N3+]	2076 [O-]

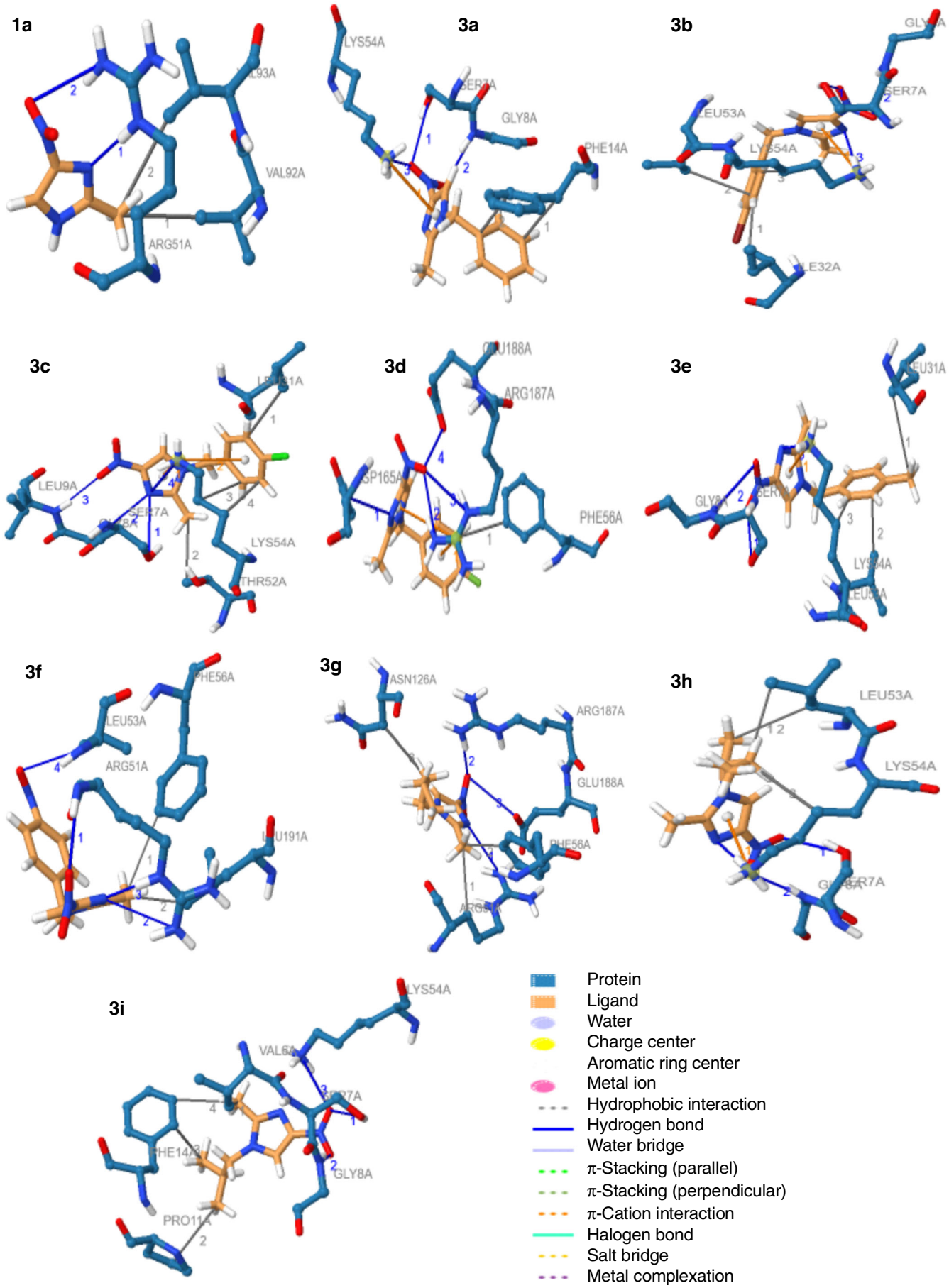


Fig. 3. Docking poses and binding modes of 4-nitroimidazoles (**1a**, **3a-i**) and representation mode of interaction

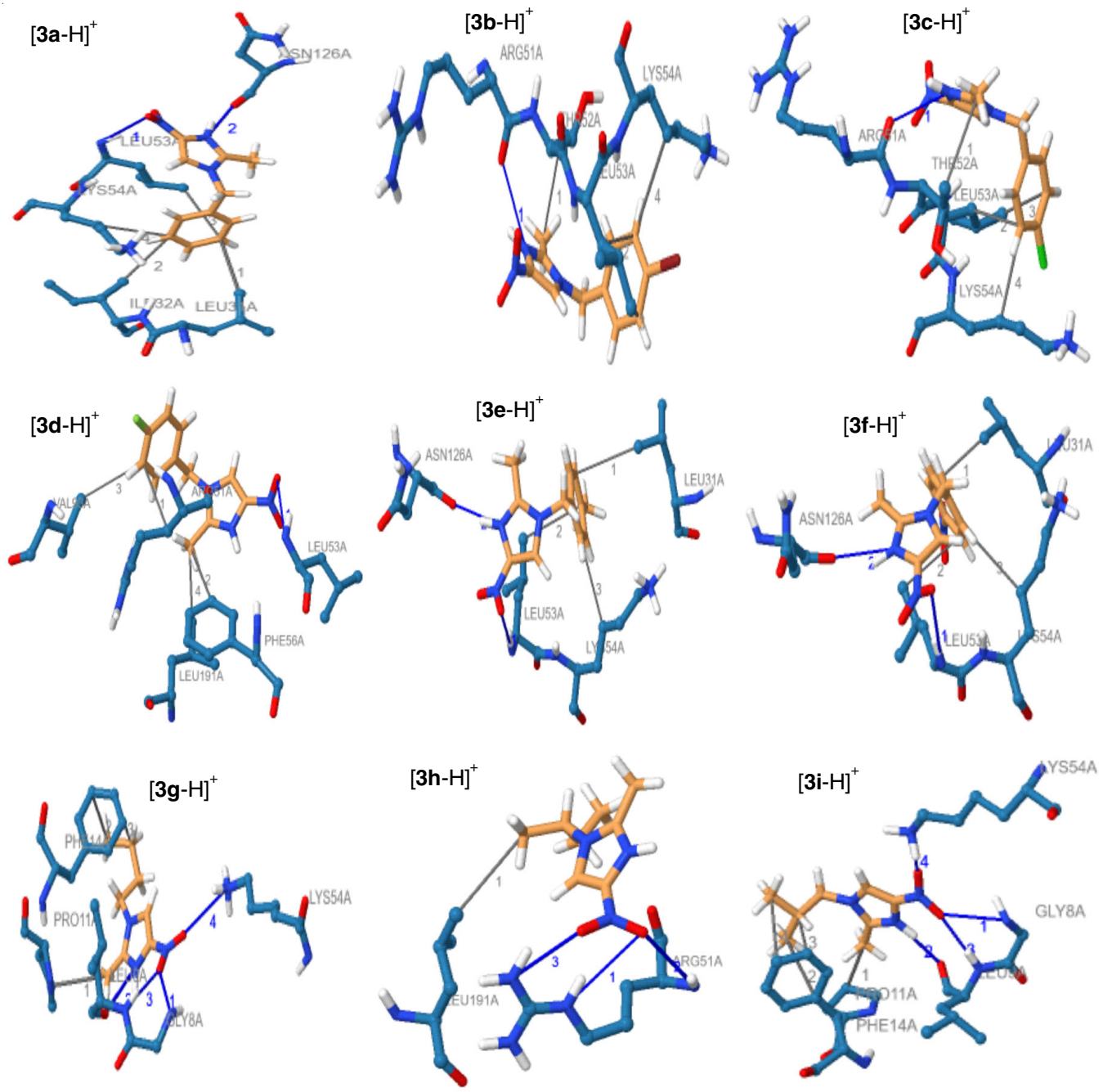


Fig. 4. Docking poses and binding modes of 4-nitroimidazolium cations [**3a**-H]$^+$-[**3i**-H]$^+$

3i, anion DNB & *m*CPBA interacted with VAL-6A, imidazole **3i**, cation [**3g**-H]$^+$ & [**3i**-H]$^+$ interacted with PRO-11A and imidazole **3c**, **3e**, cation [**3a**-H]$^+$, [**3e**-H]$^+$ & [**3f**-H]$^+$ interacted with LEU-31A (Figs. 3-5). In comparison with traditional medications, docked compounds, cations and anions have been shown some new hydrophobic interactions with the breast cancer protein. Imidazole **3b** & cation [**3a**-H]$^+$ exhibited an interaction with ILE-32A, imidazole **3g** & [**3d**-H]$^+$ revealed an interaction with ARG-51A, imidazole **3c**, cation [**3b**-H]$^+$ & [**3c**-H]$^+$ shown an interaction with THR-52A and **3d**, **3f**, **3g**, cation [**3d-H**]$^+$ & anion NMP have exposed an interaction with PHE-56A (Figs. 3-5). The hydrophobic interactions of imidazole **3f**, cation [**3d**-H]$^+$, [**3h**-H]$^+$, anion FA & NMP, imidazole **3g** has an interaction and the cation [**3d**-H]$^+$ has an interaction with the breast cancer protein residues LEU-191A, ASN-126A and VAL-93A, respectively (Figs. 3-5).

Hydrogen bond interactions: Tables 4-6 summarize the hydrogen bond (H-bond) interactions, binding residues, H-A and D-A distances, donor angle, donor atoms and acceptor atoms of docked 4-nitroimidazoles, 4-nitroimidazolium cations, organic anions and conventional pharmaceuticals. 4-Nitroimidazoles **3a-c**, **3e**, **3h** and **3i** have a significant hydrogen bonding interaction with the breast cancer protein residues SER-7A and GLY-8A (Fig. 3). Only 4-nitroimidazolium cations [**3g**-H]$^+$ and [**3i**-H]$^+$ have been demonstrated to interact with GLY-8A *via* hydrogen bonding (Fig. 4). The organic anions DAB, DNB and *m*CPBA showed an interaction with the residue GLY-8A (Fig. 5). Among the docked standard drugs, the

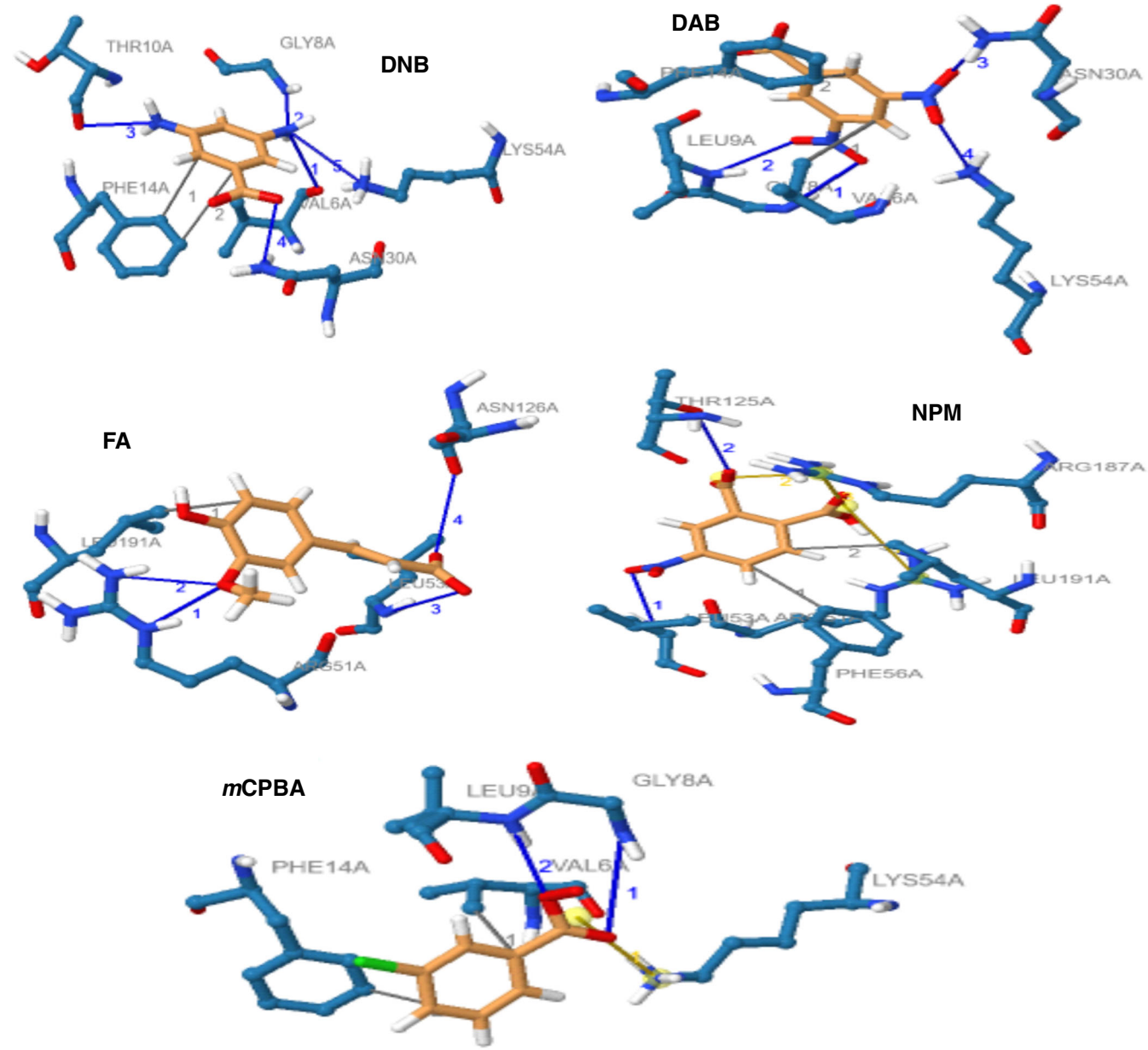


Fig. 5. Docking poses and binding modes of aromatic carboxylate anions (DAB, DNB, FA, NMP & *m*CPBA)

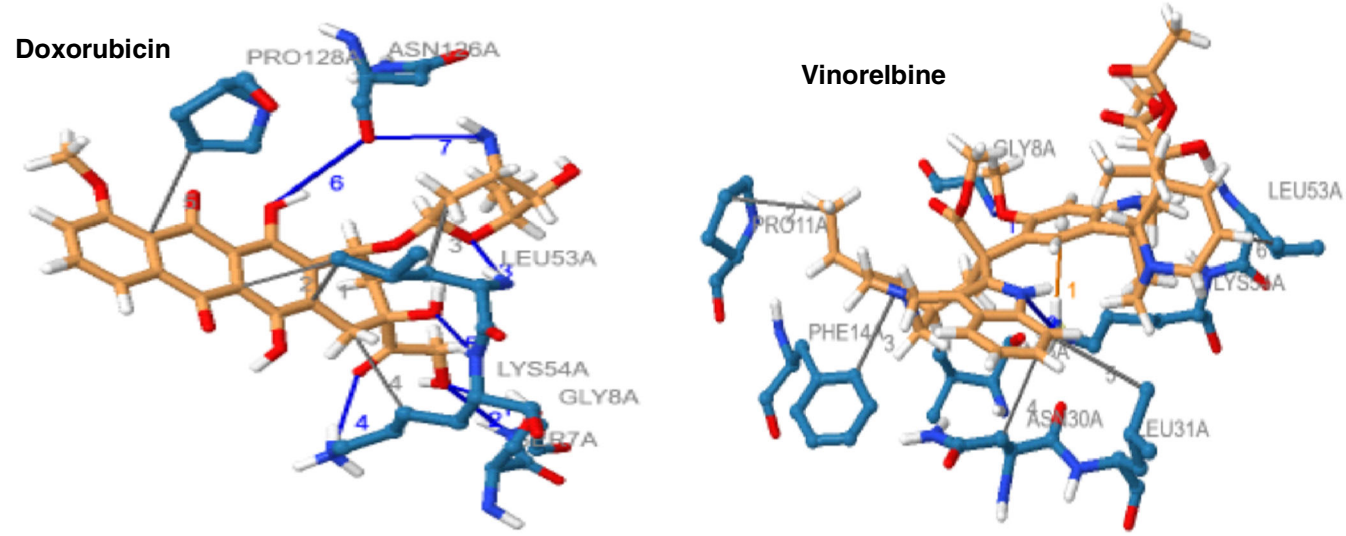


Fig. 6. Docking poses and binding modes of the standard drugs (doxorubicin and vinorelbine)

TABLE-5
HYDROGEN BOND BINDING INTERACTIONS OF 4-NITROIMIDAZOLIUM CATIONS

Imidazolium cation(s)	Binding index	Protein residue	Amino acid	Distance (Å)		Donor angle	Atom	
				H-A	D-A		Donor	Acceptor
[**3a**-H]$^+$	1	53A	LEU	2.97	3.91	157.44	516 [Nam]	2081 [O2]
	2	126A	ASN	2.01	2.97	155.58	2067 [N2]	1273 [O2]
[**3b**-H]$^+$	1	51A	ARG	2.38	3.10	126.43	2067 [N2]	493 [O2]
[**3c**-H]$^+$	1	51A	ARG	2.41	3.08	122.31	2067 [N2]	493 [O2]
[**3d**-H]$^+$	1	53A	LEU	2.36	3.27	151.24	516 [Nam]	2082 [O2]
[**3e**-H]$^+$	1	53A	LEU	2.71	3.66	158.32	516 [Nam]	2081 [O-]
	2	126A	ASN	1.97	2.95	160.28	2067 [N2]	1273 [O2]
[**3f**-H]$^+$	1	53A	LEU	3.00	3.94	156.46	516 [Nam]	2083 [O-]
	2	126A	ASN	2.10	3.05	153.26	2067 [N2]	1273 [O2]
[**3g**-H]$^+$	1	8A	GLY	2.50	2.96	107.91	69 [Nam]	2078 [O2]
	2	9A	LEU	2.30	3.00	124.65	2070 [N2]	77 [O2]
	3	9A	LEU	2.13	3.06	154.36	74 [Nam]	2078 [O2]
	4	54A	LYS	2.91	3.31	104.36	533 [N3+]	2077 [O-]
[**3h**-H]$^+$	1	51A	ARG	2.86	3.82	156.19	497 [Ng+]	2077 [O-]
	2	51A	ARG	3.02	3.87	143.32	490 [Nam]	2077 [O-]
	3	51A	ARG	2.16	3.05	142.64	500 [Ng+]	2078 [O2]
[**3i**-H]$^+$	1	8A	GLY	2.59	3.02	105.80	69 [Nam]	2078 [O2]
	2	9A	LEU	2.16	3.12	157.63	2070 [N2]	77 [O2]
	3	9A	LEU	2.12	3.07	158.10	74 [Nam]	2078 [O2]
	4	54A	LYS	2.80	3.19	103.60	533 [N3+]	2077 [O-]

TABLE-6
HYDROGEN BOND BINDING INTERACTIONS OF AROMATIC CARBOXYLATE ANIONS AND STANDARD DRUG

Organic anion/ Standard drug	Binding index	Protein residue	Amino acid	Distance (Å)		Donor angle	Atom	
				H-A	D-A		Donor	Acceptor
DAB	1	6A	VAL	2.57	3.14	115.25	2071 [Npl]	56 [O2]
	2	8A	GLY	2.93	3.64	128.95	69 [Nam]	2071 [Npl]
	3	10A	THR	2.99	3.87	146.52	2077 [Npl]	86 [O2]
	4	30A	ASN	2.39	3.06	124.19	299 [Nam]	2076 [O.co2]
	5	54A	LYS	3.16	3.69	114.30	533 [N3+]	2071 [Npl]
DNB	1	8A	GLY	2.23	3.15	151.02	69 [Nam]	2076 [O2]
	2	9A	LEU	2.48	3.10	119.16	74 [Nam]	2075 [O-]
	3	30A	ASN	2.30	3.00	126.11	299 [Nam]	2079 [O2]
	4	54A	LYS	2.33	3.06	128.09	533 [N3+]	2078 [O-]
FA	1	51A	ARG	2.00	2.87	140.62	497 [Ng+]	2076 [O3]
	2	51A	ARG	2.30	3.08	131.63	500 [Ng+]	2076 [O3]
	3	53A	LEU	3.04	3.95	152.27	516 [Nam]	2074 [O.co2]
	4	126A	ASN	3.27	3.66	107.21	2075 [O.co2]	1273 [O2]
NMP	1	53A	LEU	3.28	4.02	131.66	516 [Nam]	2080 [O2]
	2	125A	THR	3.27	4.03	138.77	1266 [O3]	2077 [O.co2]
*m*CPBA	1	8A	GLY	3.22	4.01	137.55	69 [Nam]	2073 [O2]
	2	9A	LEU	2.06	2.96	149.09	74 [Nam]	2074 [O3]
Doxorubicin	1	7A	SER	2.32	3.11	140.04	66 [O3]	2101 [O3]
	2	8A	GLY	2.17	3.06	147.68	69 [Nam]	2101 [O3]
	3	53A	LEU	2.30	3.28	164.15	516 [Nam]	2087 [O3]
	4	54A	LYS	2.29	3.25	158.32	533 [N3+]	2099 [O2]
	5	54A	LYS	2.79	3.76	166.05	525 [Nam]	2103 [O3]
	6	126A	ASN	2.79	3.74	162.12	2105 [O_3]	1273 [O2]
	7	126A	ASN	2.65	3.27	119.31	2095 [N3]	1273 [O2]
Vinorelbine	1	8A	GLY	2.69	3.66	163.46	69 [Nam]	2110 [O3]
	2	54A	LYS	3.56	4.07	113.91	533 [N3+]	2097 [Nar]

doxorubicin only showed an interaction with SER-7A and both the drugs shown an interaction with GLY-8A (Fig. 6). Among them 4-nitroimidazoles, the **3f** only has a hydrogen bonding interaction with LEU-53A (Fig. 3). Four 4-nitroimidazolium cations [**3a**-H]$^+$, [**3d**-H]$^+$-[**3f**-H]$^+$, two anions FA and NMP and the conventional medicine doxorubicin, on the other hand, have only exhibited an interaction with the protein's LEU-53A. The 4-nitroimidazoles (**3a-c**, **3e**, **3h** and **3i**), as well as 4-nitroimidazolium cations [**3g**-H]$^+$ and [**3i**-H]$^+$ and organic anions DAB and DNB, have exhibited hydrogen bonding interactions with the protein residue LYS-54A (Figs. 3-5). Doxorubicin has two interactions with LYS-54A and vinorelbine has one as well (Fig. 6). 4-Nitroimidazolium cations [**3a**-H]$^+$, [**3e**-H]$^+$ and [**3f**-H]$^+$, as well as the organic anion FA, only showed a hydrogen

TABLE-7
π-CATION BINDING INTERACTIONS OF 4-NITROIMIDAZOLES

Imidazole(s)	Binding index	Protein residue	Amino acid	Distance (Å)	Offset	Ligand group	Ligand atom(s)
3a	1	54A	LYS	3.82	1.27	Aromatic	2066, 2067, 2068, 2069, 2070
3b	1	54A	LYS	3.82	1.96	Aromatic	2066, 2067, 2068, 2069, 2070
3c	1	54A	LYS	3.61	0.85	Aromatic	2066, 2067, 2068, 2069, 2070
	2	54A	LYS	3.95	1.60	Aromatic	2072, 2073, 2074, 2075, 2076, 2078
3d	1	187A	ARG	5.74	1.30	Aromatic	2072, 2073, 2074, 2075, 2076, 2078
	2	187A	ARG	4.61	1.91	Aromatic	2066, 2067, 2068, 2069, 2070
3e	1	54A	LYS	3.68	1.86	Aromatic	2066, 2067, 2068, 2069, 2070
3h	1	54A	LYS	3.61	1.93	Aromatic	2066, 2067, 2068, 2069, 2070
Vinorelbine	1	54A	LYS	3.60	0.98	Aromatic	2079, 2080, 2081, 2082, 2083, 2084

TABLE 8
SALT BRIDGE BINDING INTERACTIONS OF NMP AND *m*CPBA

Organic anion(s)	Binding index	Protein residue	Amino acid	Distance (Å)	Ligand group	Ligand atom(s)
NMP	1	51A	ARG	5.00	Carboxylate	2072, 2073
	2	187A	ARG	4.92	Carboxylate	2076, 2077
	3	187A	ARG	4.52	Carboxylate	2072, 2073
*m*CPBA	1	54A	LYS	4.29	Carboxylate	2074, 2073

bond interaction with the protein residue ASN-126A. There are no hydrogen bonding contacts between ASM-126A and any of ten 4-nitroimidazole derivatives (**1a**, **3a-i**) and the medication doxorubicin has two hydrogen bonding interactions with ASM-126A (Figs. 3-6). The cations [**3g**-H]$^+$ and [**3i**-H]$^+$ showed two contacts with LEU-9A, as did the imidazole **3c**, anion DNB and *m*CPBA. Imidazole **1a**, **3g**, cations [**3b**-H]$^+$, [**3c**-H]$^+$ & FA anion demonstrated an interaction and **3f** & [**3h**-H]$^+$ shown three interactions with ARG-51A of the protein (Figs. 3-5). Imidazole **3d** has shown an interaction with ASP-165A and GLU-188A, two interactions with ARG-187A and **3g** has an interaction with ARG-187A & GLU-188A. The anion DAB interacts with the residues VAL-6A, THR-10A and ASN-30A, while the DNB anion interacts with ASN-30A and the NMP anion interacts with THR-125A (Fig. 5).

π-Cation binding interactions: Among the simulated 4-nitroimidazoles, cations and anions, the six 4-nitroimidazoles **3a-e** & **3h** had a high π-cation interaction with the breast cancer protein *via* the aromatic imidazolyl group (Table-7, Fig. 3). 4-nitroimidazole **3a**, **3b**, **3e** & **3h** showed a π-cation contact while **3c** showed two π-cation interactions with the residue LYS-54A of the breast cancer protein (Fig. 3). Only the residue LYS-54A (3.60) has a π-cation interaction with the typical medication vinorelbine (Fig. 6). With ARG-187A, 4-nitroimidazole **3d** has shown new two π-cation interactions (Fig. 3).

Salt bridge binding interactions: Only two organic anions NMP and *m*CPBA have shown additional salt bridge interactions with the breast cancer protein through the carboxylate group among docked 4-nitroimidazoles, 4-nitroimidazoilum cations and organic anions (Table-8). The breast cancer protein residues ARG-51A (5.00) and ARG-187A interact with the anion NMP (4.52 & 4.92) (Fig. 5). Through a salt bridge, LYS-54A (4.29) interacts with the anion *m*CPBA (Fig. 5).

Conclusion

As anticancer agents, ionic salts or liquids are gaining popularity. As a result, ten 4-nitroimidazole derivatives (**1a**, **3a-i**), nine protonated 4-nitroimidazolium cations ([**3a**-H]$^+$-[**3i**-H]$^+$) and five aromatic carboxylate counter anions (DAB, DNB, FA, NMP, & *m*CPBA) were successfully docked and executed in this study to examine the binding affinity, binding modes and binding interactions with the (PDB code: 3K0K). Three 4-nitroimidazoles (**3c**, **3e** and **3f**), four 4-nitroimidazolium cations ([**3b**-H]$^+$, [**3c**-H], [**3d**-H]$^+$ and [**3f**-H]$^+$) and two organic anions (DNB and NMP) were shown to have a higher binding affinity (-5.0 to -5.2 kcal/mol) with the breast cancer protein. These classes of 4-nitroimidazoles and their 4-nitroimidazolium cations, as well as organic anions, have greater interactions with the breast cancer protein, according to the findings. Finally, those 4-nitroimidazolium salts were found to be effective anticancer competitors in the treatment of human breast cancer.

REFERENCES

1. C. DeSantis, J. Ma, L. Bryan and A. Jemal, Breast Cancer Statistics, 2013, *CA Cancer J. Clin.*, **64**, 52 (2014); https://doi.org/10.3322/caac.21203
2. H. Varmus, The New Era in Cancer Research, *Science*, **312**, 1162 (2006); https://doi.org/10.1126/science.1126758
3. World Health Organization (WHO), Cancer (2020).
4. Statistics at a glance: The burden of Cancer in the United Sates. https://www.cancer.gov/about-cancer/understanding/statistics
5. C.E. DeSantis, S.A. Fedewa, A. Goding Sauer, J.L. Kramer, R.A. Smith and A. Jemal, Breast Cancer Statistics, 2015: Convergence of Incidence Rates between Black and White Women, *CA Cancer J. Clin.*, **66**, 31 (2016); https://doi.org/10.3322/caac.21320
6. M. Ghoncheh, Z. Pournamdar and H. Salehiniya, Incidence and Mortality and Epidemiology of Breast Cancer in the World, *Asian Pac. J. Cancer Prev.*, **17(sup3)**, 43 (2016); https://doi.org/10.7314/APJCP.2016.17.S3.43
7. Cancer Fact and Figure American Cancer Society (2017); https://www.cancer.org/research/cancer-facts-statistics/all-cancer-facts-figures/cancer-factsfigures-2017.html (2017) pp 1–71.
8. Health Care Global Enterprises Ltd, Annual Report On Cancer 2016-2017; http://icmr.nic.in/icmrsql/archive/2016/7.pdf
9. L.A. Haldosén, C. Zhao and K. Dahlman-Wright, Estrogen Receptor Beta in Breast Cancer, *Mol. Cell. Endocrinol.*, **382**, 665 (2014); https://doi.org/10.1016/j.mce.2013.08.005

10. B.J. Deroo and K.S. Korach, Estrogen Receptors and Human Disease, *J. Clin. Invest.*, **116**, 561 (2006); https://doi.org/10.1172/JCI27987
11. R.A. Magarian, L.B. Overacre, S. Singh and K.L. Meyer, The Medicinal Chemistry of Nonsteroidal Antiestrogens: A Review, *Curr. Med. Chem.*, **1**, 61 (1994); https://doi.org/10.2174/0929867301666220210212635
12. B.H. Mitlak and F.J. Cohen, Selective Estrogen Receptor Modulators, *Drugs*, **57**, 653 (1999); https://doi.org/10.2165/00003495-199957050-00001
13. C. Descôteaux, V. Leblanc, G. Bélanger, S. Parent, E. Asselin and G. Bérubé, Improved Synthesis of Unique Estradiol-Linked Platinum(II) Complexes Showing Potent Cytocidal Activity and Affinity for the Estrogen Receptor Alpha and Beta, *Steroids*, **73**, 1077 (2008); https://doi.org/10.1016/j.steroids.2008.04.009
14. S. Adsule, S. Banerjee, F. Ahmed, S. Padhye and F.H. Sarkar, Hybrid Anticancer Agents: Isothiocyanate–Progesterone Conjugates as Chemotherapeutic Agents and Insights into their Cytotoxicities, *Bioorg. Med. Chem. Lett.*, **20**, 1247 (2010); https://doi.org/10.1016/j.bmcl.2009.11.128
15. J. Ellmén, P. Hakulinen, A. Partanen and D.F. Hayes, Estrogenic Effects of Toremifene and Tamoxifen in Postmenopausal Breast Cancer Patients, *Breast Cancer Res. Treat.*, **82**, 103 (2003); https://doi.org/10.1023/B:BREA.0000003957.54851.11
16. T.L. Taras, G.T. Wurz and M.W. DeGregorio, *In vitro* and *in vivo* Biologic Effects of Ospemifene (FC-1271a) in Breast Cancer, *J. Steroid Biochem. Mol. Biol.*, **77**, 271 (2001); https://doi.org/10.1016/S0960-0760(01)00066-8
17. G. Kaur, M.P. Mahajan, M.K. Pandey, P. Singh, S.R. Ramisetti and A.K. Sharma, Design, Synthesis and Evaluation of Ospemifene Analogs as Anti-breast Cancer Agents, *Eur. J. Med. Chem.*, **86**, 211 (2014); https://doi.org/10.1016/j.ejmech.2014.08.050
18. B.F. Abdel-Wahab, G.E. Awad and F.A. Badria, Synthesis, Antimicrobial, Antioxidant, Anti-hemolytic and Cytotoxic Evaluation of New Imidazole based Heterocycles, *Eur. J. Med. Chem.*, **46**, 1505 (2011); https://doi.org/10.1016/j.ejmech.2011.01.062
19. L. Zhang, X.M. Peng, G.L.V. Damu, R.X. Geng and C.H. Zhou, Comprehensive Review in Current Developments of Imidazole-Based Medicinal Chemistry, *Med. Res. Rev.*, **34**, 340 (2014); https://doi.org/10.1002/med.21290
20. A.K. Jain, V. Ravichandran, M. Sisodiya and R.K. Agrawal, Synthesis and Antibacterial Evaluation of 2-Substituted-4,5-Diphenyl-*N*-Alkyl Imidazole Derivatives, *Asian Pac. J. Trop. Med.*, **3**, 471 (2010); https://doi.org/10.1016/S1995-7645(10)60113-7
21. X.L. Hu, Z. Xu, M.L. Liu, L.S. Feng and G.D. Zhang, Recent Developments of Coumarin Hybrids as Anti-fungal Agents, *Curr. Top. Med. Chem.*, **17**, 3219 (2017); https://doi.org/10.2174/1568026618666171215100326
22. J. Pandey, V.K. Tiwari, S.S. Verma, V. Chaturvedi, S. Bhatnagar, S. Sinha, A.N. Gaikwad and R.P. Tripathi, Synthesis and Antitubercular Screening of Imidazole Derivatives, *Eur. J. Med. Chem.*, **44**, 3350 (2009); https://doi.org/10.1016/j.ejmech.2009.02.013
23. A. Bistrovic, L. Krstulovic, A. Harej, P. Grbcic, M. Sedic, S. Kostrun, S.K. Pavelic, M. Bajic and S. Raic-Malic, Design, Synthesis and Biological Evaluation of Novel Benzimidazole Amidines as Potent Multi-Target Inhibitors for the Treatment of Non-Small Cell Lung Cancer, *Eur. J. Med. Chem.*, **143**, 1616 (2018); https://doi.org/10.1016/j.ejmech.2017.10.061
24. B.L. Cui, B.L. Zheng, K.L. He and Q.Y. Zheng, Imidazole Alkaloids from *Lepidium meyenii*, *J. Nat. Prod.*, **66**, 1101 (2003); https://doi.org/10.1021/np030031i
25. V.S. Souza, J.R. Corrêa, P.H.P.R. Carvalho, G.M. Zanotto, G.I. Matiello, B.C. Guido, C.C. Gatto, G. Ebeling, P. F.B. Gonçalves, J. Dupont and B.A.D.Neto, *Sens. Actuators, B: Chem.*, **321**, 128530 (2020); https://doi.org/10.1016/j.snb.2020.128530
26. R. Bonsignore, A. Notaro, A.M.P. Salvo, A. Spinello, G. Fiasconaro, A. Terenzi, F. Giacalone, B.K. Keppler, M. Giuliano, M. Gruttadauria and G. Barone, DNA-Binding and Anticancer Activity of Pyrene-Imidazolium Derivatives, *ChemistrySelect,* **1**, 6755 (2016); https://doi.org/10.1002/slct.201601502
27. M. Guncheva, K. Paunova, P. Ossowicz, Z. Rozwadowski, E. Janus, K. Idakieva, S. Todinova, Y. Raynova, V. Uzunova, S. Apostolova, R. Tzoneva and D. Yancheva, *Rapana thomasiana* Hemocyanin Modified with Ionic Liquids with Enhanced Anti Breast Cancer Activity, *Int. J. Biol. Macromol.*, **82**, 798 (2016); https://doi.org/10.1016/j.ijbiomac.2015.10.031
28. A. Aubé, S. Campbell, A.R. Schmitzer, A. Claing and J.F. Masson, Ultra-Low Fouling Methylimidazolium Modified Surfaces for the Detection of HER2 in Breast Cancer Cell Lysates, *Analyst*, **142**, 2343 (2017); https://doi.org/10.1039/C7AN00056A
29. S.C. Jadhvar, H.M. Kasraliker, S.V. Goswami, A.V. Chakrawar and S.R. Bhusare, One-Pot Synthesis and Evaluation of Anticancer Activity of Polyhydroquinoline Derivatives Catalyzed by [Msim]Cl, *Res. Chem. Intermed.*, **43**, 7211 (2017); https://doi.org/10.1007/s11164-017-3069-2
30. M. Galluzzi, C. Schulte, P. Milani and A. Podestà, Imidazolium-Based Ionic Liquids Affect Morphology and Rigidity of Living Cells: An Atomic Force Microscopy Study, *Langmuir*, **34**, 12452 (2018); https://doi.org/10.1021/acs.langmuir.8b01554
31. F. Al-Blewi, N. Rezki, A. Naqvi, H. Qutb Uddin, S. Al-Sodies, M. Messali, M.R. Aouad and S. Bardaweel, A Profile of the *in vitro* Anti-Tumor Activity and *in silico* ADME Predictions of Novel Benzothiazole Amide-Functionalized Imidazolium Ionic Liquids, *Int. J. Mol. Sci.*, **20**, 2865 (2019); https://doi.org/10.3390/ijms20122865
32. X.L. Xu, J. Wang, C.L. Yu, W. Chen, Y.C. Li, Y. Li, H.B. Zhang and X.D. Yang, Synthesis and Cytotoxic Activity of Novel 1-((Indol-3-yl)-methyl)-1*H*-imidazolium salts, *Bioorg. Med. Chem. Lett.*, **24**, 4926 (2014); https://doi.org/10.1016/j.bmcl.2014.09.045
33. D. Satheesh, A. Rajendran, R. Saravanan, S. Kannan and K. Chithra, An Efficient Room Temperature Synthesis of N^1-(4-Substituted benzyl)-2-methyl-4-nitro-1*H*-imidazoles and N^1-Butyl-2-methyl-4-nitro-1*H*-imidazoles, *Iran. J. Org. Chem.*, **10**, 2325 (2018).
34. D. Satheesh, A. Rajendran, K. Chithra and R. Saravanan, Synthesis of Some New Protic N^1-Benzyl/Butyl-2-methyl-4-nitro-1*H*-imidazol-3-ium salts with 3,5-Diaminobenzoate, 3,5-Dinitrobenzoate, (*E*)-3-(4-Hydroxy-3-methoxyphenyl)acrylate and 2-Carboxy-5-nitrobenzoate as Organic Anions, *Results Chem.*, **2**, 100033 (2020); https://doi.org/10.1016/j.rechem.2020.100033
35. D. Satheesh, A. Rajendran, K. Chithra and R. Saravanan, Synthesis and Antimicrobial Evaluation of N^1-Benzyl/butyl-2-methyl-4-nitro-3-imidazolium 3′-chloroperoxy Benzoates, *Chem. Data Coll.*, **28**, 100406 (2020); https://doi.org/10.1016/j.cdc.2020.100406
36. Structure of Doxorubicin, https://pubchem.ncbi.nlm.nih.gov/compound/Doxorubicin#section=Structures
37. Structure of Vinorelbine, https://pubchem.ncbi.nlm.nih.gov/compound/Vinorelbine#section=Structures.
38. S.J. Campbell, R.A. Edwards and J.N. Glover, Comparison of the Structures and Peptide Binding Specificities of the BRCT Domains of MDC1 and BRCA1, *Structure*, **18**, 167 (2010); https://doi.org/10.1016/j.str.2009.12.008
39. S. Salentin, S. Schreiber, V.J. Haupt, M.F. Adasme and M. Schroeder, PLIP: Fully Automated Protein–Ligand Interaction Profiler, *Nucleic Acids Res.*, **43(W1)**, W443 (2015); https://doi.org/10.1093/nar/gkv315
40. D. Satheesh, A. Rajendran and K. Chithra, Protein-Ligand Binding Interactions of Imidazolium Salts with SARS CoV-2, *Heliyon*, **6**, e05544 (2020); https://doi.org/10.1016/j.heliyon.2020.e05544