ISSN 2581 – 4346

Available Online at www.ijms.co.in
Innovative Journal of Medical Sciences 2023; 7(1):16-25

ORIGINAL ARTICLE

Computational approaches highlighting conotoxins as potential drug against breast and pancreatic cancer treatment

Manisha Nag[1,2], Sweta Rani Chaurasia[1,3], Md. Mahfooz Khan[1,4], Pramod Kumar[1,4], Dipanjali Sharma[1,4], Sneha Priya[1,5], Shruti Kumari[1,6], Priyangulta Beck[1], Ganesh Chandra Baskey[2], Mukesh Nitin[1]*

[1]Department of Tech Biosciences, Digianalix, South Samaj Street, Tharphakna Ranchi, Jharkhand, India, [2]Department of Zoology, Dr. Shyama Prasad Mukherjee University, Ranchi, Jharkhand, India, [3]Department of Biotechnology, M.S. Ramaiah University of Applied Sciences, Bengaluru, Karnataka, India, [4]Department of Biotechnology, Marwari College Ranchi, Jharkhand, India, [5]Department of Biotechnology, Ranchi Women's College Ranchi, Jharkhand, India, [6]Department of Biotechnology, Birla institute of Technology, Mesra, Jharkhand, India

Received on: 14 Jan 2023; Revised on: 08 Feb 2023; Accepted: 08 Mar 2023

ABSTRACT

Introduction: The two most common cancer-related causes of mortality, breast and pancreatic cancer, account for a significant portion of deaths worldwide. Our research included genomic and docking investigations using a variety of cancer illness datasets, for the identification of potential hub genes as target receptors. Based on literature reviews, we focused on conotoxins, a marine animal-derived substance secreted by cone snails, which serves as a ligand protein in the treatment of many disorders like cancer. **Methods:** The Gene Expression Omnibus (GEO) dataset of NCBI was used to compile the information on breast and pancreatic cancer. In this study, two microarray datasets (GSE36775 and GSE113865) were evaluated and screened out significantly differentially expressed genes (DEGs) that had $P<0.05$, using NCBI GEO2R. Furthermore, Cytoscape is used to examine the highly conserved genes across various cancer types. A protein-protein docking study was conducted using the H-Dock server. **Results:** Gene conservancy studies revealed that 963 genes in total were preserved in two significant cases. To identify their metabolic pathways, a system biology approach was used, and docking studies of receptor proteins against diverse conotoxins were also conducted. The two compounds with the highest docking scores are CCNB1 + conotoxinPVIIA (−257.64 Kcal/mol) and CDK1 + conotoxinGeXIVA (−246.66 Kcal/mol), respectively. **Conclusion:** In the fight against cancer, our study has shown that conotoxins-derived peptides have enormous promise for selectively targeting cancer cells. We concluded that animal-based compounds may unveil new areas of study for researchers.

Keywords: Breast cancer, CDK1, Conotoxin-GeXIVA, Pancreatic cancer, Protein-Protein docking study

INTRODUCTION

Breast cancer roughly (10–20%) and pancreatic cancer are two devastating malignancies that continue to pose significant challenges to global health.[1,2] Breast cancer is a malignant tumor, that affects about 1.4 million people with the second-highest mortality rate among women, while pancreatic cancer is the most prevalent lethal tumor type worldwide and the fourth-leading cause of death.[3,4] At present, the number of people with pancreatic and breast cancer is still rising. In 2020, there were around 2.3 million new instances of breast cancer diagnosed globally.[5] Despite significant improvements in research and therapy, there is still a substantial cause of morbidity and mortality. Therefore, it is crucial to investigate novel therapeutic approaches for cancer therapy that can enhance the treatment.

***Corresponding Author:**
Dr. Mukesh Nitin
E-mail: digianalix@gmail.com

Recently, there have been advancements in the field of toxicology research; conotoxins are a diverse class of naturally occurring peptides derived from marine cone snails that have shed light on the potential of these small peptide toxins as valuable therapeutic agents. A cone snail is a marine gastropod belonging to the phylum Mollusca, family Conidae and genus conus,[6] contains a venom gland that secretes neurotoxins generally referred to as conopeptide or conotoxins, and has emerged as a promising class of natural product for cancer therapy.[7,8] Approximately 100,000 natural conotoxins have been identified in various cone snails around the world.[9-12] Therefore, the cone snail can exhibit the largest collection of natural marine drugs. The most common commercial conotoxin is ω-MVIIA (ziconotide), derived from the venom of conus magus species and approved by the U.S. Food and Drug Administration to treat chronic pain in serious cancer and AIDS patients.[13] Conotoxins are classified into different families depending on the types of their molecular targets and corresponding pharmacological activity.[14,15] Conotoxin has a wide range of structural and functional diversity and mainly targets membrane protein receptors, especially ion channel and receptor proteins. Membrane ion channels and receptor proteins have a significant role in cell proliferation and play an essential role in the development of cancer.[16] In recent years, one of the most prominent areas of research has been the potential use of natural products, including compounds derived from marine organisms as a source of new cancer treatments.[17] Ultimately proliferation studies demonstrate that inhibition of protein expression or channel blockade by any specific inhibitor reduces cell proliferation. Thus, selectively targeting and blocking receptors or ion channels would be a blessing for cancer therapy.

In this research, hub genes were identified based on genomic expression contributing crucially in the cell cycle of cancer patients by causing inflammation and proliferation in the essential organs, which results in discomfort and death. The capacity of conotoxins to target particular cell cycle-related genes like CDK1 and CCNB1 has been investigated. This article provides an overview of the potential use of conotoxins as a novel therapeutic approach for pancreatic and breast cancer, highlighting their diverse modes of action and the promising results observed.

MATERIALS AND METHODS

Data collection and identification

Data samples were screened from the NCBI's Gene Expression Omnibus (GEO) dataset,[18] a public functional genomic database containing high-throughput gene expression data, chips, and microarrays, to explore potential therapeutic targets for pancreatic and breast cancer. Two microarray datasets (GSE36775 and GSE113865) were investigated in this research. In case 1 (GSE36775), pancreatic cancer patients were taken under two different conditions: The first involved exposure of the pancreatic stellate cell line (stellate cells are resident cells of the pancreas) to the pancreatic cancer cell line (capan1) and the second involved taking only cancer cells. The second case (GSE113865) involved breast cancer under two different conditions: The first is triple-negative breast cancer (TNBC) which has low or no expression of, estrogen receptor, progesterone receptor, and human epidermal growth factor receptor 2 and the second is normal breast tissue.

Using an internal shell script, the GEO2R program was used to reanalyze the data and resulting in the generation of box plot, volcano plot, and MA plot along with gene expression table using the Benjamin-Hedge Hoch test, limma package-R software and further screening out significantly differentially expressed genes (DEGs) based on $P \leq 0.05$, which is statistically significant. We identify the conserved genes across two distinct cancer case studies using Venn Ghent software.[19]

System biology approaches

The metabolic pathways of the frequently expressed genes were then investigated utilizing gene ontology resources. Cytoscape was used to assess the highly conserved genes across various cancer conditions to highlight hub genes network.

To check for protein-protein interactions, we use string analysis in cytoscape. We take conserved genes that are frequently expressed and place them in a string protein network to analyze the annotation of the genes. Next, we use Cytohubba to provide the top 10 hub genes by activating the Hubba nodes and selecting the degree of genes to 10 as option.[20]

Preparation of receptor and ligands

After the cross-referencing studies using numerous research articles related to breast and various other cancers, target receptors for the highly expressed hub genes CDK1 and CCNB1 were taken. CDK1 dysregulation increases cell proliferation in a variety of cancers, including breast cancer,[21] and CCBN1, a regulatory protein involved in cell cycle progression, can result in tumor development in a variety of cancers.[22] In addition, we chose numerous conotoxin variants that function as ligands, namely, the alpha O-conotoxin GeXIVA,[23] alpha conotoxin MII,[24] kapa-conotoxin PVIIA,[25] and alpha-conotoxin RgIA.[26] These proteins were modeled using the Swiss model to find the three-dimensional structure in pdb format.[27]

Protein docking study

We conduct a protein-protein docking study with receptors and ligands that are both proteins using an H-dock server to conduct further investigation. Here, we choose receptors and substances released by marine snails to serve as ligands for a protein docking study to understand how these proteins bind to each other and predict the complex's spatial arrangement.[28]

Molecular dynamics simulation (MD)

The stability and movement of the resulting docked compounds were defined by the iMODS server, which was used to examine the compounds for MD modeling. The pdb file of the final docked compounds was uploaded to the iMODS server while maintaining the parameters, coarse-grained as 'CA (Calcium ions), and JSmol as "HTML" were submitted for the examination of their MD.[29]

RESULTS

We carried out a meta-analysis of genomic information obtained from NCBI's GEO datasets to find genes related to the development and prognosis of pancreatic cancer and breast cancer. The NCBI GEO-analyzer server performed a reanalysis of the data that had been obtained. In case1 (GSE36775), a total of 47230 DEGs were discovered between two pancreatic cancer circumstances, one in which pancreatic cancer cell line (capan1) was exposed to pancreatic stellate cell line and the other in which cancer cell alone was the condition. A total of 47320 differently expressed genes were discovered in Case 2 (GSE113865) between two types of breast cancer, one of which was TNBC and the other was normal breast tissue. The mean arithmetic (MA) plot, the volcano plot, and the raw read normalization box plot were then performed, as illustrated in Figure 1.

Additional significant differential gene expression investigations were conducted and based on p-values ≤ 0.05, a total of 4807 and 6414 highly significant DEGs were found in the two separate cases.

For further gene conservancy analysis, the Venn Ghent server was used to identify the potential genes that are common in these two diseased cases, highlighting conserved genes screened based on significant differential expression of genes. Out of which, 963 genes in total were discovered to be common in both, as shown in Figure 2.

The total top 10 genes were finally identified as the hub genes, and they were CDK1, CCNB1, CCNA2, AURKB, PLKI, BUB1, CDC20, TOP2A, KIF11, and CHEK1 in Figure 3.

These genes were highly expressed in our target disorders and were obtained with the aid of the Cytoscape software. As indicated in Table 1, the role of these genes in the control of carcinogenesis and inflammation was also investigated. Among these genes, the biological pathways of CDK1 and CCNB1 were examined as prominent gene as receptor, revealing positive regulation in the cell cycle process in cancer.

Physiochemical analysis of CDK1 and CCNB1, SOPMA was used to highlight secondary structures such as alpha helix, extended strand, and random coil for both receptors and ligands in Table 2.

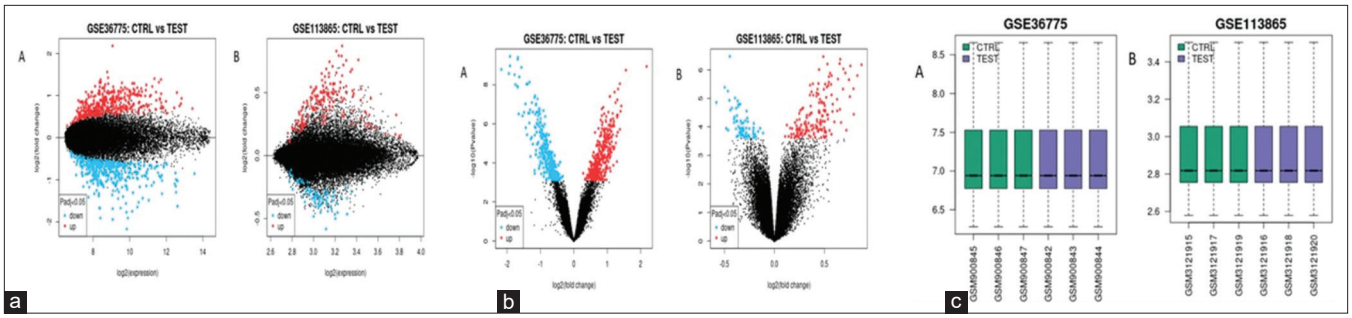

Figure 1: Results of meta-analysis (a) MA plot, (b) Box plot, (c) read normalization plot

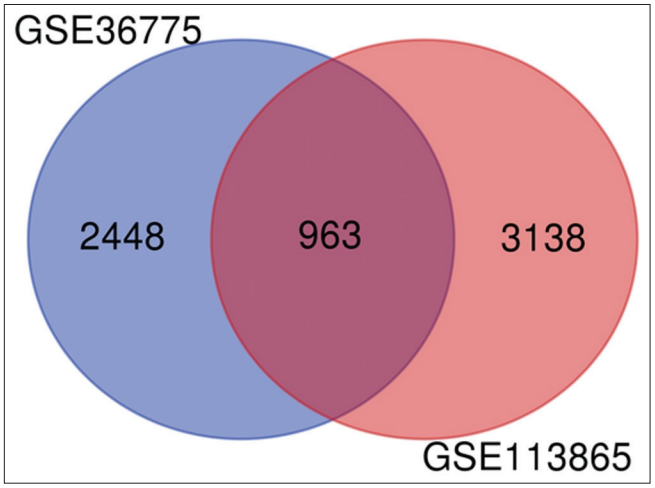

Figure 2: Gene conservancy chart analysis of target diseases illustrating intersections between cases GSE36775 and GSE113865

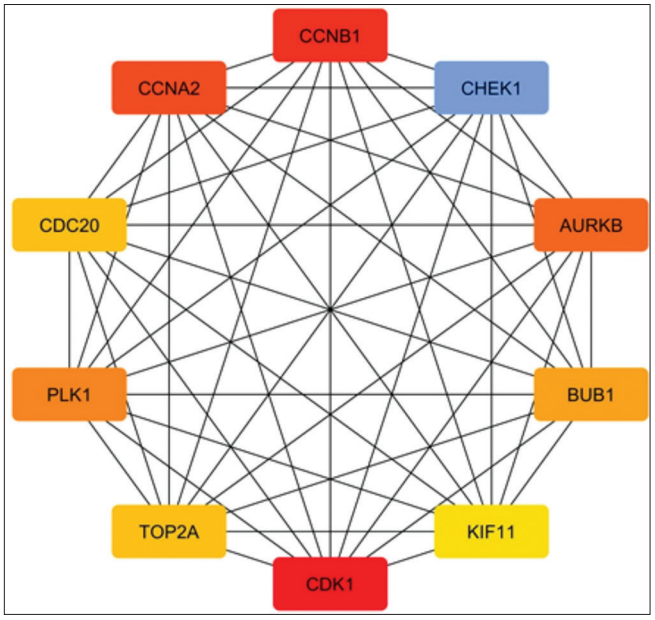

Figure 3: Network and pathway analyses screening from gene pool top 10 Hub genes

Sosui was used to highlight transmembrane structure in Figure 4. Protparam was used to highlight the number of amino acids. Theoretical pi, instability index, and gravy index are also highlighted in Table 3.

Swiss modeling was used to determine the 3D structure of the receptor in Figure 5a and ligand proteins in Figure 5b.

Table 1: Functioning of the top 10 highly expressed hub genes

Genes	Functions	References
CDK1	Dysfunctioning of CDKs which leads to increased cell proliferation has been identified in different cancers.	[21]
CCNB1	CCNB1 is involved in checkpoint control, it is highly expressed in various primary tumors, and its deregulated expression is observed in a number of different human cancers including breast cancer, cervical cancer, lung cancer, etc.	[22]
CCNA2	CCNA2 is overexpressed in various cancer types, which indicates its potential role in cancer transformation and progression.	[30]
AURKB	AURKB is frequently observed as highly expressed in tissues from some tumors, such as lung cancer and breast cancer, and AURKB overexpression is associated with poor prognosis.	[31]
PLK1	Human polo-like kinase 1 (Plk1), a key regulator of mitosis, is overexpressed in various human tumors.	[32]
BUB1	BUB1 is a mitotic checkpoint serine/threonine kinase that has been reported as an oncogene or tumor suppressor gene in various types of cancer.	[33]
CDC20	CDC20 may function as an oncoprotein to promote the development and progression of human cancers.	[34]
TOP2A	TOP2A is overexpressed in several malignancies including Breast cancer.	[35]
KIF11	The expression of KIF11 increased significantly in high-stage and malignant tumor cells	[36]
CHEK1	DNA damage induces activation of CHEk1, promotes tumor growth, and may contribute to anticancer therapy resistance	[37]

Table 2: Sopma analysis of receptor and ligand proteins

Secondary structure of proteins	Receptors proteins (%)		Ligands proteins (%)			
	CDK1	CCNB1	Conotoxin-GeXIVA	Conotoxin-MII	Conotoxin-PVIIA	Conotoxin-RgIA
Alpha helix	43.77	56.81	50.00	45.59	44.44	40.62
3_{10} helix	0.00	0.00	0.00	0.00	0.00	0.00
Pi helix	0.00	0.00	0.00	0.00	0.00	0.00
Beta bridge	0.00	0.00	0.00	0.00	0.00	0.00
Extended strand	14.48	3.23	6.76	17.65	8.33	0.00
Beta turn	7.07	2.31	5.41	2.94	6.94	0.00
Bend region	0.00	0.00	0.00	0.00	0.00	0.00
Random coil	34.68	37.64	37.84	33.82	40.28	59.38
Ambiguous	0.00	0.00	0.00	0.00	0.00	0.00

Table 3: Protparam is used to calculate physicochemical parameters of receptor and ligand proteins

Targets	Protein	Mol. Weight	pI	Instability index	Aliphatic index	Gravy
Receptors	CDK1	34095.45	8.38	39.26	97.78	−0.281
	CCNB1	48337.43	7.09	50.59	90.09	0-239
Ligands	Conotoxin-Gexiva	8621.14	10.25	72.46	73.78	−0.349
	Conotoxin-MII	7356.60	8.93	33.89	86.03	0.206
	Conotoxin-PVIIA	8316.89	9.75	33.66	87.92	−0.139
	Conotoxin-RgIA	3725.38	9.93	14.62	49.06	−0.831

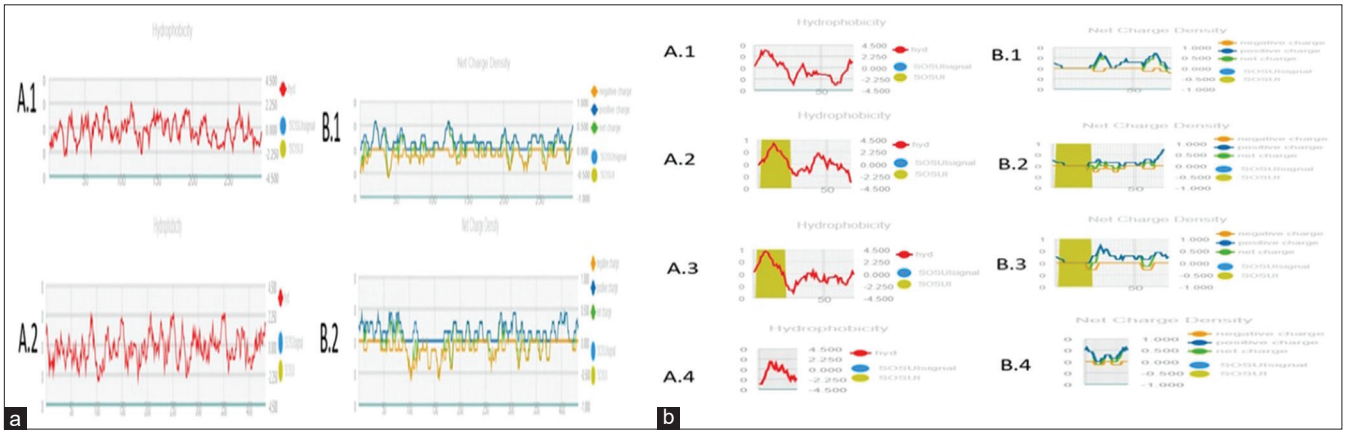

Figure 4: Sousi-secondary structure prediction of membrane protein of (a) Receptors (A.1) and (A.2) are hydrophobicity and (B.1), (B.2) are net charges (b) ligands (A.1), (A.2), (A.3), and (A.4) are hydrophobicity (B.1), (B.2), (B.3), and (B.4) are net charges, respectively.

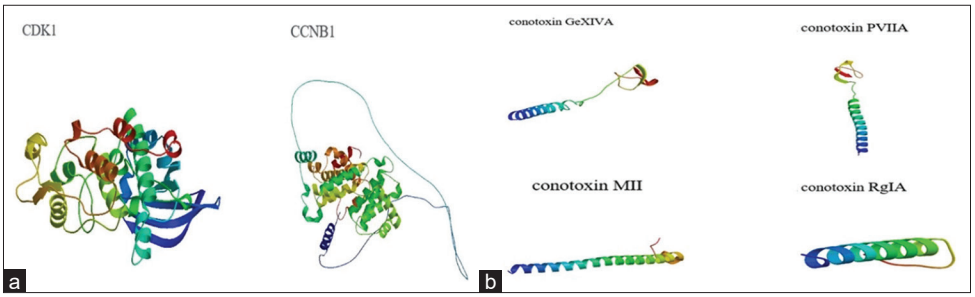

Figure 5: Swiss model is used to determine the 3D structure of receptor and ligand proteins

Ramachandran is utilized to check the favorable region of the receptor proteins, CDK1 in Figure 6a and CCNB1 in Figure 6b, and the percentage of residues is mentioned in Table 4. CDK1 and

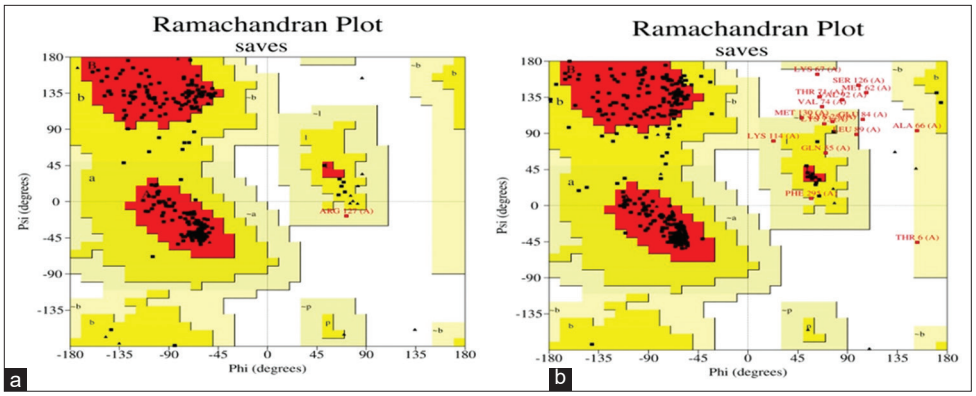

Figure 6: Ramachandran plot showing the favorable region of receptor proteins

Table 4: Percentage residue of Ramachandran plot of receptor proteins

Ramachandran plot analysis	CDK1 (%)	CCNB1 (%)
Residues in the most favorable regions (A, B, L)	93.1	86.5
Residues in additional allowed regions (a, b, l, p)	6.5	9.2
Residues in generously allowed regions (~a, ~b, ~l, ~p)	0.4	2.4
Residue in disallowed regions	0.0	1.8

CCNB1 were chosen as receptors based on their physicochemical features, and conotoxin GeXIVA, conotoxin MII, conotoxin PVIIA, and conotoxin RgIA were chosen as ligands.

In a protein docking investigation, CDK1 was combined with the toxins GeXIVA, MII, PVIIA, and RgIA. These compounds displayed the docking score, confidence score, and ligand rmsd() in Table 5, as well as the best-docked complex in Figure 7a and CCNB1 demonstrated a docking score, confidence score, and ligand rmsd() in Table 5 and the best-docked complex in Figure 7b along with the toxin GeXIVA, MII, PVIIA, and RgIA.

With the help of MD simulation, we examined the stability and movement of the docked complex. It was carried out to verify the docked complexes' stability and movement. Deformability and B-factor reveal that there was little distortion in the complex represented by the hinges, which are indicators of the stability and movement of the docked protein. The B-factor plot displays the stability of the docked complex and is proportional to the root mean square (RMS). The eigenvalue of CDK1 + conotoxinGeXIVA, CDK1 + conotoxinMII, CDKI + conotoxinPVIIA, CDK1 + conotoxinRgIA, and the eigenvalue of CCNB1 + conotoxinGeXIV, CCNB1 + conotoxinMII, CCNB1 + conotoxinPVIIA, and CCNB1 + conotoxinRgIA represents in Table 6.

The stability of the docked complex is indicated by its greater eigenvalue, which displays the energy needed to deform it. The covariance matrix graph shows correlated, uncorrelated, and anti-correlated motions represented by the colors red, white, and blue, respectively. The stiffness of the docked complex is investigated using an elastic network model. In Figure 8 darker grey indicates increased protein stiffness in specific locations which shows the best MD results of our docked complex. Hence, our docked complexes indicated a strong likelihood of a molecule with drug-potent stability.

DISCUSSION

In several malignancies, protein-associated cell cycle dysregulation has been well-documented.[38] Despite the fact that their roles in the development of cancer and prognosis have only been partially proven.[39-41]. There is a need for more bioinformatics analysis on breast and pancreatic cancer samples. In our study, meta-analysis data were carried out to find conserved hub genes, including CDK1, CCNB1, CCNA2, TOP2A, CHEK1 BUB1, AURKB, PLKI, CDC20, and KIF11. Here, we examine the expression levels with CDK1 and CCNB1, as well as their prognostic value in pancreatic and breast cancer.

The research advances our knowledge of pancreatic and breast cancer which aids in the creation of more effective treatment regimens and prognoses for these diseases. According to bibliographical investigations, CDK1 is overexpressed in breast

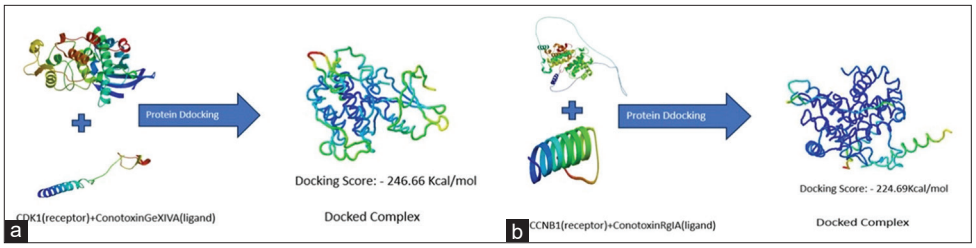

Figure 7: Docking result (a) CDK1+ Conotoxin GeXIVA and (b) CCNB1+ Conotoxin PVIIA

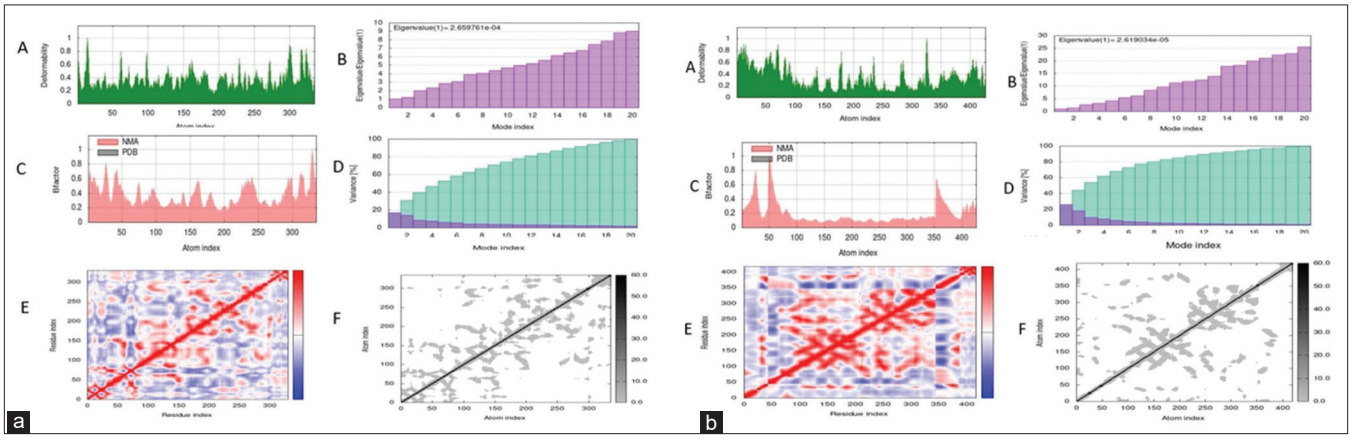

Figure 8: Visual representation of molecular dynamics (a) CDK1+GeXIVA and (b) CCNB1 + PVIIA in the graph represents (A) deformability, (B) eigenvalue, (C) b-factor, (D) variance plot, (E) elastic variance model, and (F) covariance map

Table 5: Docking result of receptors with ligands molecule

Receptor molecule	CDK1			CCNB1		
Ligand molecule	**D.S**	**C.S**	**L.rmsd(Å)**	**D.S**	**C.S**	**L.rmsd(Å)**
ConotoxinGeXIVA	−246.66	0.8736	46.32	−253.33	0.8876	56.63
Conotoxin MII	−218.62	0.7978	57.66	−243.38	0.8662	61.59
Conotoxin PVIIA	−236.02	0.8482	34.92	−257.64	0.8959	53.64
Conotoxin RgIA	−219.31	0.8000	30.96	−224.69	0.8166	36.02

Table 6: Eigenvalue of the docked protein-protein complex

Compounds	Eigenvalue
CDK1+conotoxinGeXIVA	2.659761e-04
CDK1+conotoxinMII	4.405886e-06
CDKI+conotoxinPVIIA	2.833001e-05
CDK1+conotoxinRgIA	1.758071e-04
CCNB1+conotoxinGeXIV	3.134842e-05
CCNB1+conotoxinMII	9.355029e-06
CCNB1+conotoxinPVIIA	2.619034e-05
CCNB1 + conotoxinRgIA	5.996605e-05

and pancreatic cancer.[42] In TNBC, CDK1 inhibition significantly decreased tumor formation and increased tumor cell apoptosis,[43] and it also regulates the spread of pancreatic cancer.[44] CCNB1 overexpression has been seen in numerous malignancies, including pancreatic carcinoma, according to several studies.[45] According to a study by Agarwal *et al.*, CCNB1 can be used to assess a breast cancer prognostic predictor.

In this study, we investigate the extraordinary potential of conotoxin as a cutting-edge therapeutic approach for the treatment of cancer. Conotoxins are renowned for their diversity and selectivity because of their capacity to precisely target a variety of ion channels and receptors. Certain conotoxins have shown the ability to interfere with signaling pathways necessary for tumor growth, angiogenesis, and metastasis. Several conotoxins have been found to inhibit ion channels overexpressed in cancer cells, resulting in the decreased proliferation and induction of apoptosis as well. Based on various research literatures, we selected four distinct types of conotoxins (conotoxin-GeXIVA, PVIIA, MII,

and RgIA) that act as ligand proteins generated from poisonous marine cone snails of the conus species, which have emerged as nature's special weapons against cancer. The protein-protein docking of CDK1 and CCNB1 interacting with these four conotoxin compounds was carried out to obtain the best reasonable conformations.

Many drug-potent molecules were docked with CDK1 and CCNB1. Furthermore, it was discovered that four conotoxins, namely, GeXIVA, PVIIA, MII, and RgIA, had a high affinity for CDK1 and CCNB1 demonstrated a higher docking score of CCNB1 + conotoxinPVIIA −257.64 Kcal/mol, CCNB1 + conotoxinGeXIV −253.33 Kcal/mol, CDK1 + conotoxinGeXIVA −246.66 Kcal/mol, CCNB1 + conotoxinMII −243.38 Kcal/mol, CDKI + conotoxinPVIIA −236.02 Kcal/mol, CCNB1 + conotoxinRgIA −224.69 Kcal/mol, CDK1 + conotoxinRgIA −219.31 Kcal/mol and CDK1 + conotoxinMII −218.62 Kcal/mol, respectively, representing the minimum energy values of the docked complex has higher stability between receptors and ligands molecules and therefore potential inhibitors of CDK1 and CCNB1. Similar research has been carried out in which natural marine components used to make animal-based medicinal compounds were examined for their anti-cancerous capabilities against various malignancies. In dry laboratories, our experimental values are validated computationally through MD simulation to prove our hypothesis representing the eigenvalue of the docked complex; the higher eigenvalue indicates the energy required to deform the docked complex hence representing its stability.

CONCLUSION

Our study has demonstrated immense potential in targeting cancer cells with conotoxins-derived peptides in the war against cancer. Our findings imply that the biomarkers CDK1 and CCNB1 may serve as prospective therapeutic targets and biomarkers for detecting high-risk subgroups among breast and pancreatic cancer patients and also we have been able to identify various conotoxins as anticancerous agents. Conotoxins like GeXIVA, MII, PVIIA, and RgIA were identified as promising avenues for the development of targeted cancer therapies. We are trying to find the genomic interaction between breast and pancreatic cancer along with a drug discovery approach to find animal-based drug compounds that may have therapeutic potential against our disease. The study of animal-based drug compounds may open a wide area of study for researchers to identify a drug compound that may act against multiple cancerous diseases with lesser side effects. Further, these findings call for more research into the possible therapeutic uses of various cell cycle inhibitors.

ACKNOWLEDGMENT

I would like to express my sincere gratitude to Head Dept. of Tech. Biosciences, Digianalix for allowing me to use research facilities to complete my study.

REFERENCES

1. Reis-Filho JS, Tutt AN. Triple negative tumours: A critical review. Histopathology 2008;52:108-18.
2. Bauer KR, Brown M, Cress RD, Parise CA, Caggiano V. Descriptive analysis of estrogen receptor (ER)-negative, progesterone receptor (PR)-negative, and HER2-negative invasive breast cancer, the so-called triple-negative phenotype: A population-based study from the California cancer Registry. Cancer 2007;109:1721-8.
3. Fan L, Strasser-Weippl K, Li JJ, St Louis J, Finkelstein DM, Yu KD, *et al.* Breast cancer in China. Lancet Oncol 2014;15:e279-89.
4. Martínez-Bosch N, Guerrero PE, Moreno M, José A, Iglesias M, Munné-Collado J, *et al.* The pancreatic niche inhibits the effectiveness of sunitinib treatment of pancreatic cancer. Oncotarget 2016;7:48265-79.
5. Bashar MD, Begam N. Breast cancer surpasses lung cancer as the most commonly diagnosed cancer worldwide. Indian J Cancer 2022;59:438-9.
6. Lebbe EK, Peigneur S, Wijesekara I, Tytgat J. Conotoxins targeting nicotinic acetylcholine receptors: An overview. Mar Drugs 2014;12:2970-3004.
7. Prashanth JR, Brust A, Jin AH, Alewood PF, Dutertre S, Lewis RJ. Cone snail venomics: From novel biology to novel therapeutics. Future Med Chem 2014;6:1659-75.
8. Vetter I, Lewis RJ. Therapeutic potential of cone snail venom peptides (conopeptides). Curr Top Med Chem 2012;12:1546-52.
9. Olivera BM, Rivier J, Clark C, Ramilo CA, Corpuz GP, Abogadie FC, *et al.* Diversity of Conus neuropeptides. Science 1990;249:257-63.

10. Aman JW, Imperial JS, Ueberheide B, Zhang MM, Aguilar M, Taylor D, *et al.* Insights into the origins of fish hunting in venomous cone snails from studies of Conus tessulatus. Proc Natl Acad Sci U S A 2015;112:5087-92.
11. Espiritu DJ, Watkins M, Dia-Monje V, Cartier GE, Cruz LJ, Olivera BM**.** Venomous cone snails: Molecular phylogeny and the generation of toxin diversity. Toxicon 2001;39:1899-916.
12. Kaas Q, Westermann JC, Craik DJ**.** Conopeptide characterization and classifications: An analysis using ConoServer. Toxicon 2010;55:1491-509.
13. Molinski TF, Dalisay DS, Lievens SL, Saludes JP**.** Drug development from marine natural products. Nat Rev Drug Discov 2009;8:69-85.
14. Dutertre S, Jin AH, Kaas Q, Jones A, Alewood PF, Lewis RJ**.** Deep venomics reveals the mechanism for expanded peptide diversity in cone snail venom. Mol Cell Proteomics 2013;12:312-29.
15. Robinson SD, Norton RS. Conotoxin gene superfamilies. Mar Drugs 2014;12:6058-101.
16. Ekberg J, Craik DJ, Adams DJ**.** Conotoxin modulation of voltage-gated sodium channels. Int J Biochem Cell Biol 2008;40:2363-8.
17. Khalifa SA, Elias N, Farag MA, Chen L, Saeed A, Hegazy MF, *et al.* Marine natural products: A source of novel anticancer drugs. Mar Drugs 2019;17:491.
18. Barrett T, Wilhite SE, Ledoux P, Evangelista C, Kim IF, Tomashevsky M, *et al.* NCBI GEO: Archive for functional genomics data sets-update. Nucleic Acids Res 2012**;**41:D991-5.
19. COVIDSurg Collaborative, Adamina M, Ademuyiwa A, Adisa A, Bhangu AA, Bravo AM, *et al***.** The impact of surgical delay on resectability of colorectal cancer: An international prospective cohort study. Colorectal Dis 2022;24:708-26.
20. Smoot ME, Ono K, Ruscheinski J, Wang PL, Ideker T. Cytoscape 2.8: New features for data integration and network visualization. Bioinformatics 2011;27:431-2.
21. Izadi S, Nikkhoo A, Hojjat-Farsangi M, Namdar A, Azizi G, Mohammadi H, *et al.* CDK1 in breast cancer: Implications for theranostic potential. Anticancer Agents Med Chem 2020;20:758-67.
22. Kreis NN, Sanhaji M, Krämer A, Sommer K, Rödel F, Strebhardt K, *et al*. Restoration of the tumor suppressor p53 by downregulating cyclin B1 in human papillomavirus 16/18-infected cancer cells. Oncogene 2010;29:5591-603.
23. Luo S, Zhangsun D, Harvey PJ, Kaas Q, Wu Y, Zhu X, *et al.* Cloning, synthesis, and characterization of αO-conotoxin GeXIVA, a potent α9α10 nicotinic acetylcholine receptor antagonist. Proc Natl Acad Sci U S A 2015;112:E4026-35.
24. McIntosh JM, Azam L, Staheli S, Dowell C, Lindstrom JM, Kuryatov A, *et al*. Analogs of α-conotoxin MII are selective for α6-containing nicotinic acetylcholine receptors. Mol Pharmacol 2004;65:944-52.
25. Scanlon MJ, Naranjo D, Thomas L, Alewood PF, Lewis RJ, Craik DJ. Solution structure and proposed binding mechanism of a novel potassium channel toxin κ-conotoxin PVIIA. Structure 1997;5:1585-97.
26. Ellison M, Haberlandt C, Gomez-Casati ME, Watkins M, Elgoyhen AB, McIntosh JM, *et al*. α-RgIA: A novel conotoxin that specifically and potently blocks the α9α10 nAChR. Biochemistry 2006;45:1511-7.
27. Schwede T, Kopp J, Guex N, Peitsch MC. SWISS-MODEL: An automated protein homology-modeling server. Nucleic Acids Res 2003;31:3381-5.
28. Luo W, Pei J, Zhu Y. A fast protein-ligand docking algorithm based on hydrogen bond matching and surface shape complementarity. J Mol Model 2010;16:903-13.
29. Kremer JR, Mastronarde DN, McIntosh JR. Computer visualization of three-dimensional image data using IMOD. J Struct Biol 1996;116:71-6.
30. Dong S, Huang F, Zhang H, Chen Q. Overexpression of BUB1B, CCNA2, CDC20, and CDK1 in tumor tissues predicts poor survival in pancreatic ductal adenocarcinoma. Biosci Rep 2019;39:BSR20182306.
31. Goldenson B, Crispino JD. The aurora kinases in cell cycle and leukemia. Oncogene 2015;34:537-45.
32. Wianny F, Tavares A, Evans MJ, Glover DM, Zernicka-Goetz M. Mouse polo-like kinase 1 associates with the acentriolar spindle poles, meiotic chromosomes and spindle midzone during oocyte maturation. Chromosoma 1998;107:430-9.
33. Zhou W, Guan W, Zhou Y, Rao Y, Ji X, Li J. Weighted genes associated with the progression of retinoblastoma: Evidence from bioinformatic analysis. Exp Eye Res 2021;211:108730.
34. Wang Z, Wan L, Zhong J, Inuzuka H, Liu P, Sarkar FH, *et al.* Cdc20: A potential novel therapeutic target for cancer treatment. Curr Pharm Des 2013;19:3210-4.
35. Zhou J, Chen WR, Yang LC, Wang J, Sun JY, Zhang WW, *et al.* KIF11 Functions as an oncogene and is associated with poor outcomes from breast cancer. Cancer Res Treat 2019;51:1207-21.
36. Xie Y, Wei RR, Huang GL, Zhang MY, Yuan YF, Wang HY. Checkpoint kinase 1 is negatively regulated by miR-497 in hepatocellular carcinoma. Med Oncol 2014;31:844.
37. García-Gutiérrez L, Bretones G, Molina E, Arechaga I, Symonds C, Acosta JC, *et al.* Myc stimulates cell cycle progression through the activation of Cdk1 and phosphorylation of p27. Sci Rep 2019;9:18693.
38. Zhang P, Kawakami H, Liu W, Zeng X, Strebhardt K, Tao K, *et al.* Targeting CDK1 and MEK/ERK overcomes apoptotic resistance in BRAF-mutant human colorectal cancer. Mol Cancer Res 2018;16:378-89.
39. Guo W, Yu H, Zhang L, Chen X, Liu Y, Wang Y, *et al.* Effect of hyperoside on cervical cancer cells and transcriptome analysis of differentially expressed genes. Cancer Cell Int 2019;19:235.
40. Chen EB, Qin X, Peng K, Li Q, Tang C, Wei YC, *et al.* HnRNPR-CCNB1/CENPF axis contributes to gastric cancer proliferation and metastasis. Aging (Albany NY) 2019;11:7473-91.

41. Kang J, Sergio CM, Sutherland RL, Musgrove EA. Targeting cyclin-dependent kinase 1 (CDK1) but not CDK4/6 or CDK2 is selectively lethal to MYC-dependent human breast cancer cells. BMC Cancer 2014;14:32.
42. Xia Q, Cai Y, Peng R, Wu G, Shi Y, Jiang W. The CDK1 inhibitor RO3306 improves the response of BRCA-proficient breast cancer cells to PARP inhibition. Int J Oncol 2014;44:735-44.
43. Huang J, Chen P, Liu K, Liu J, Zhou B, Wu R, *et al.* CDK1/2/5 inhibition overcomes IFNG-mediated adaptive immune resistance in pancreatic cancer. Gut 2021;70:890-9.
44. Zhou L, Li J, Zhao YP, Cui QC, Zhou WX, Guo JC, *et al.* The prognostic value of Cyclin B1 in pancreatic cancer. Med Oncol 2014;31:107.
45. Agarwal R, Gonzalez-Angulo AM, Myhre S, Carey M, Lee JS, Overgaard J, *et al.* Integrative analysis of cyclin protein levels identifies cyclin b1 as a classifier and predictor of outcomes in breast cancer. Clin Cancer Res 2009;15:3654-62.