Life Science Informatics Publications

Research Journal of Life Sciences, Bioinformatics, Pharmaceutical and Chemical Sciences

Journal Home page http://www.rjlbpcs.com/

RJLBPCS

ISSN 2454-6348

Original Review Article **DOI: 10.26479/2023.0903.02**

CONOTOXINS: AN EMERGING DRUG-BASED SOURCE AGAINST VARIOUS CANCERS AND LUNG DISEASES

Manisha Nag[1,2], Sweta Rani Chaurasia[1,3], Sana Tasneem[1,4], Pramod Kumar[1,5], Subhashini Singh Thakur[1,5], Sonia Choubey[1,6], Annie Jessica Toppo[1], Priyangulta Beck[1], Ganesh Chandr Baskey[2], Mukesh Nitin[1]*

1. Department of Tech Biosciences, Digianalix, South Samaj Street Tharphakna Ranchi, -834001, Jharkhand, India.
2. Department of Zoology, Dr. Shyama Prasad Mukherjee University, Ranchi, -834008, Jharkhand, India.
3. Department of Biotechnology, M.S. Ramaiah University of Applied Sciences, Bengaluru - 560054, Karnataka, India.
4. Department of Zoology, Ranchi Women's College Ranchi, -834001, Jharkhand, India.
5. Department of Biotechnology, Marwari College Ranchi, -834008, Jharkhand, India.
6. Department of Botany (Biotechnology), Ranchi University, Ranchi, -834008, Jharkhand, India.

ABSTRACT: Conotoxins are proteinaceous compounds extracted from cone snail venom and have pharmacological effects against a variety of illnesses, including several types of cancer (including breast, lung, pancreatic cancer etc.) and numerous lung ailments (including asthma and tuberculosis). The property of conotoxins inhibit pro inflammatory conditions and tumor suppression thereby providing us a new idea of a potent drug target component for treating the diseases mentioned above. Based on the genomics literature review, selective proteins and biological compounds like conotoxins played very important role in curing variable diseases like cancer, asthma, tuberculosis etc, which was predicted through advanced computational biology and docking analysis.

Keywords: Conotoxin, Conus, Cancer, Lung diseases, Molecular docking.

Article History: Received: May 22, 2023; Revised: June 14, 2023; Accepted: June 20, 2023.

Corresponding Author: Dr. Mukesh Nitin***** Ph.D.
Scientist and Head, Dept. of Tech Biosciences, Digianalix, INDIA
Genomic Scientific Consultant, Gentan, Izmir, TURKEY
Email- digianalix@gmail.com

1. INTRODUCTION

Cancer is a life-threatening and devastating disease that affects millions of people worldwide. In 2020, the world health organization (WHO) anticipated that there will be more than 13 million deaths and 21 million new cases of cancer by 2030. Despite significant advances in research and treatment, cancer still accounts for a major cause of morbidity and fatality. As a result, there is a critical need to develop new methods and approaches to cancer therapy that can improve outcomes for patients [1-4]. Cancer is defined as the uncontrolled growth and division of abnormal cells in the body due to some fault in the machinery of the cell cycle that controls cell proliferation [5,6]. Chemotherapy is one of the existing and effective methods to treat various types of cancerous cells. Although chemotherapy is quite effective in the treatment of cancer, it has many side effects and limited efficacy, especially in the advanced stages of the disease. Additionally, chemotherapy drugs often target both cancerous and healthy cells, leading to significant toxicity and adverse effects [7-9]. The potential utilization of natural products, particularly substances originating from the marine life, as a source of fresh cancer treatments has recently emerged as one of the most important fields of research. [10]. In particular, marine gastropods (conus snails) belonging to the Conidae family [11], contains a venom gland that secretes neurotoxin generally referred to as conopeptide or conotoxins, have emerged as a promising class of natural product for cancer therapy [12,13]. Conotoxins are classified into different families depending on the types of their molecular target 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 [15,16]. Membrane ion channels have a significant role in cell proliferation and play an important role in the development of cancer [17-25]. Ultimately, proliferation studies show that inhibition of K+ channel expression or channel blockade by any specific inhibitor reduces cell proliferation. Thus, selectively targeting and blocking ion channels would be a significant therapeutic approach for cancer therapy. Hence in this review, we will focus on the development of anticancer drugs using conotoxins for cancer therapy, along with the mechanism of action of conopeptide and the opportunities & obstacles involved in creating conotoxins-based cancer therapies. By analyzing and synthesizing the available data, we aim to provide a comprehensive and up-to-date perspective on the potential of conotoxins as cancer therapeutics and to identify key

areas for future research and development in this exciting and rapidly evolving field.

2. APPLICATIONS AND USAGE OF MARINE PRODUCTS

Marine habitats are an excellent source of bio-active natural compounds due to their unique and diverse chemical structures [26]. Biomedical exploration of marine natural compounds that exhibit various pharmacological activities, including anticancer, antimicrobial, antiviral, anti-inflammatory, and analgesic properties [27]. Applications of these compounds have shown potential for the treatment of various diseases, including cancer, infection, and inflammation [28,29]. For anti-cancer treatment, two marine natural products, ziconotide and brentuximab vedotin, have been commercialized over the past few years. Also, 3 out of 4 novel medications produced for the treatment of cancer have been derived from both marine and terrestrial natural products [30]. In 2011, another marine product synthesized as a drug (Adcetris) developed from sea hare for the treatment of cancer. Further, several other marine-derived products are under different phases of clinical trials in various countries for significant cancer treatment such as plitidepsin glembatumumab vedotin [31]. Toxins secreted from animals are peptides that interact with specific target molecules such as ion channels (e.g., sodium and/or potassium ion channels), thus affecting the neuromuscular, cardiovascular and immune systems [32,33].

3. CONE SNAILS (CONUS)

Cone snails are invertebrates belonging to the phylum Mollusca, class gastropods and genus conus, these marine gastropods consist of 700 species from the genus conus [34]. These are found in various marine habitats across the world including coral reefs and shallow sandy waters in Western Atlantic, Indian and Pacific oceans. However, they cannot survive in freshwater [35]. Predator cone snails have been a matter of growing interest due to their greatly evolved hunting approach that makes use of conotoxins to paralyze prey. Cone snails are usually slowly moving creatures surrounded by fast-moving prey, which presents a major existential challenge to these predators. However, they successfully deal with it and control it by developing a venomous apparatus, which is accountable for the synthesis, storage, and delivery of large amounts of the most sophisticated conotoxins peptides [36]. These toxins are effective in treating various diseases. Other factors include the fact that some toxins are extremely selective for particular receptors in the body and also marine snails are abundant & easily accessible source of venom toxins. They have a specialized radula tooth that is modified into a hollow, harpoon-like structure called a radula tooth. When the snail hunts for its prey, it extends this tooth and injects the venom into the prey. The venom of cone snails contains a complex mixture of toxins that can paralyze or kill their prey [37,38]. The venom gland is a modified salivary gland of marine cone snails that can secrete neurotoxic peptides in the large amount commonly knowns as conopeptide or conotoxins reached in disulphide bridges with pharmacological activities [39,40].

4. PROPERTIES OF CONOTOXIN

Conotoxins typically consist of 8- 35 amino acids and a high number of modifications such as being rich in disulphide bonds, hydroxylation and glycosylation. These modifications are responsible for the structural stability and specificity of conotoxins [41-44]. Cone snail venom conopeptide are encoded by gene superfamilies, and a single species consist of 100-400 venom peptides [45-47]. The nomenclature used for classifying conopeptides was proposed by Cruz et al. in 1985. It categorizes them into superfamilies based on sequence and framework homology, and then into pharmacological families based on the targets they interact with. Conopeptide with no disulfide comes under six groups, viz., the ntulakins (which target the neurotensin receptor), the conantokins (which target the N-methyl-D-aspartic acid receptor) [48], the conorfamides (thought to target the Rfamide receptor) [49], the conolysins (thought to target cellular membranes) [50], the conophans (target unknown) [51], and the conomarphins (target unknown) [52]. Peptides containing one disulfide bond are classified as either the contryphans (target undefined) [53] or the conopressins (vasopressin homologs) [54].

5. VARIANCE IN THE STRUCTURAL PROPERTIES

The disulfide-rich peptides are referred to as conotoxins and are classified into six different classes based on their structures. They are α conotoxins, ω conotoxins, δ conotoxins, μ conotoxins, κ conotoxins and contulakin[55].

(a) **α conotoxins-** These conotoxins have a compact globular structure stabilized by two disulfide bonds. They typically consist of 12- 19 amino acid residues [56,57]. α conotoxins are known to block nicotinic acetylcholine receptors in both neuronal and muscle types [58,59]. These peptides are used to treat various diseases, anxiety, Parkinson's disease, pain, hypertension, cancer, and also muscle relaxants [60].

(b) **ω conotoxins**- These conotoxins are typically 24-30 amino acid residues in length and contain three disulfide bonds. The first isolated ω conotoxins were GVIA from Conus geographus [61,62]. They block voltage-gated calcium channels in the nervous system [63,64]. ω conotoxins are the most selective inhibitor among the other conotoxins because of their therapeutic potential in the management of severe pain.

(c) **δ conotoxins-** These conotoxins are approximately 30 amino acid residues in length and contain three disulfide bonds [65]. They block voltage-gated sodium channels.

(d) **μ conotoxins**-These conotoxins have a compact globular structure stabilized by three disulfide bonds. They typically consist of 16-25 amino acid residues and are known to block voltage-gated sodium channels in the nervous system and caused paralyzing [65].

(e) **K conotoxins**-The first κ conotoxin was PVIIA, isolated from the fish hunting cone C. purpurascens, it is a27 amino acid residue [66]. They block voltage-gated potassium channels.

6. MODULATION OF IMMUNE SYSTEM WITH RELATED TO CONOTOXIN

Conotoxins can highly regulate the immune system due to their therapeutic potential [67-71]. Conotoxins target ion channels that have vital roles in immune cells and immune-related diseases. One such ion channel immunomodulatory is Voltage-gated potassium channels that suppress immunomodulatory activation through blockade [72-76]. Voltage-gated potassium channels are involved in various immunological processes such as leukocyte, macrophage activation, lymphocytes, and proliferation [77,78]. K- conotoxins, PVIIA blocks the shaker potassium channels by binding to the specific binding site(triethanolamine) and this channel which is expressed in immuno-cells such as macrophages and T-lymphocytes, is a potent immunomodulatory target [79]. Other conotoxins are α-conotoxins, two PnlA and ImI α-conotoxins isolated from the venoms of Conus pennaceus and Conus imperialis respectively. PnlA block the molluscan neuronal acetylcholine receptors [80]. Later found that it was also an inhibitor of mammalian nAChRs. In the year 1994, another mammalian nAChR-specific conotoxin ImI was discovered [81]. ImI has IC50 values of 220nM and 1800nM for the homomeric nAChRs consist two subunits α7 and α9 respectively [82]. α7 have a particular interest as a pharmacological target in the immune system. α7 nAChR involve in the production of antibody [83] and T-cell proliferation [84]. Literature suggests that anti-inflammatory effects were predominately produced by the activation of the α7 nAChR [85].

7. EFFECTS OF CONOTOXIN ON CANCER CELLS

The mechanism of action of conotoxins in cancer cells varies depending on the specific conotoxins and the type of cancer being targeted. Conotoxins in particular target the ion channel that communicates with cancer cells. One common mechanism by which conotoxins can affect cancer cells is by binding to and blocking ion channels that are overexpressed in cancer cells. The mechanism of conotoxins in cancer cells with nAChRs involves binding to a specific site on the receptor. The nicotinic acetylcholine receptors (nAChRs) are known to have a role in inflammation [86] and oncogenesis [87]. one of the frequently utilised techniques in the study of nAChRs are α conotoxins which specifically and efficiently inhibit different subtypes of nAChRs [88]. Several nAChR subunits, including α3, α7, α9 and β4, are expressed in a variety of tumour cells and are involved in the control of cell proliferation, apoptosis, invasion, migration, and angiogenesis [89-92]. the α-conotoxin AuIB may prevent small-cell lung carcinomas (SCLC) cells from surviving by binding to nAChRs with the subunits α3/α5/β4 [93]. In response to tumour microenvironments, α9 nAChRs play a crucial role in driving cancer cell proliferation, angiogenesis, cancer metastasis, and apoptosis suppression during carcinogenesis [94-96]. For examples, Conus generalis produced the αO-conotoxin GeXIVA, which effectively inhibits nAChRs α9 and α10 subunits [97]. In vivo, it also demonstrated significant neuropathic pain relief in one rat model [98,99]. One recent research also demonstrated that GeXIVA played a crucial role

in the prevention of the growth of cervical cancer cells [100].

8. INVESTIGATION

Few tumour causing genes were identified through literature review on different cancers conditions and a meta-analysis was carried out in three other lung disease conditions (TB, Asthama and Lung Cancer). The hub genes were identified from genomic data set of variable above mentioned lung diseases through NCBI GEO datasets-GSE54712, GSE43696, GSE20050. Further, effective significant differential expressed genes qualifying p-value < 0.05 were screened for hub genes studies through gene conservancy analysis. The selected and screened genes were further prepared as receptors, and then docked with the conotoxin variant taken (CONOTOXIN MII, CONOTOXIN GeXIVA, 3 alpha subunit CONOTOXIN). Analyzation of the effective ligand-receptor docked complex results given in **(Table 1-2) and (Figure 1-2)** shows that more effective docking score obtained is from conotoxin variant GeXIVA with receptor CSF1R is -287.13 and conotoxin variant GeXIVAwith receptor NAChRsβ2 is -326.20. Moreover, MD simulations were carried out with best docked complex highlighting its eigenvalue of docked complex of conotoxin variant GeXIVA with receptor CSF1R is 1.715798e-05 and conotoxin variant GeXIVAwith receptor NAChRsβ2 is 1.231755e-05 which shows better stability of docked complex as shown in **(Figure 3-4).**

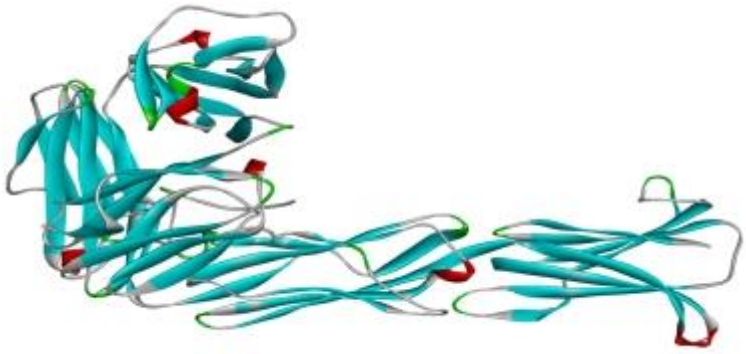

Figure 1: Docked complex Conotoxin GeXIVA with CSF1R receptor

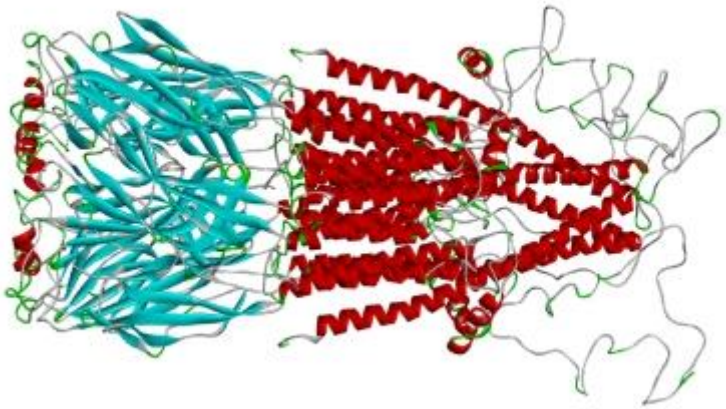

Figure 2: Docked complex Conotoxin GeXIVA with NAChRsβ2 receptor

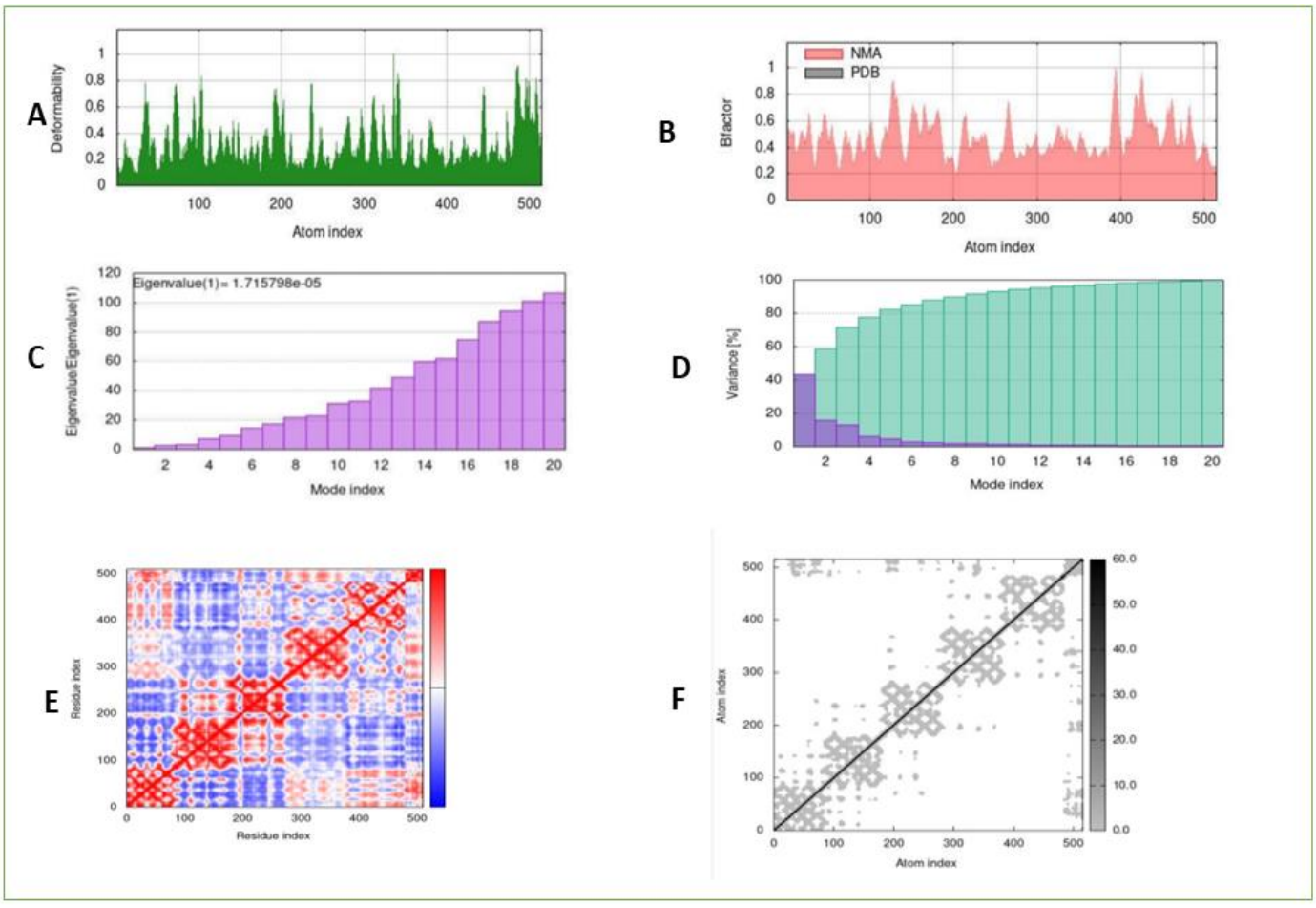

Figure 3. MD simulation result; A: deformabiloty, B: b-factor, C: eigenvalue, D: variance, E: residue index, F: atom index of receptor CSF1R with ligand conotoxin GeXIVA

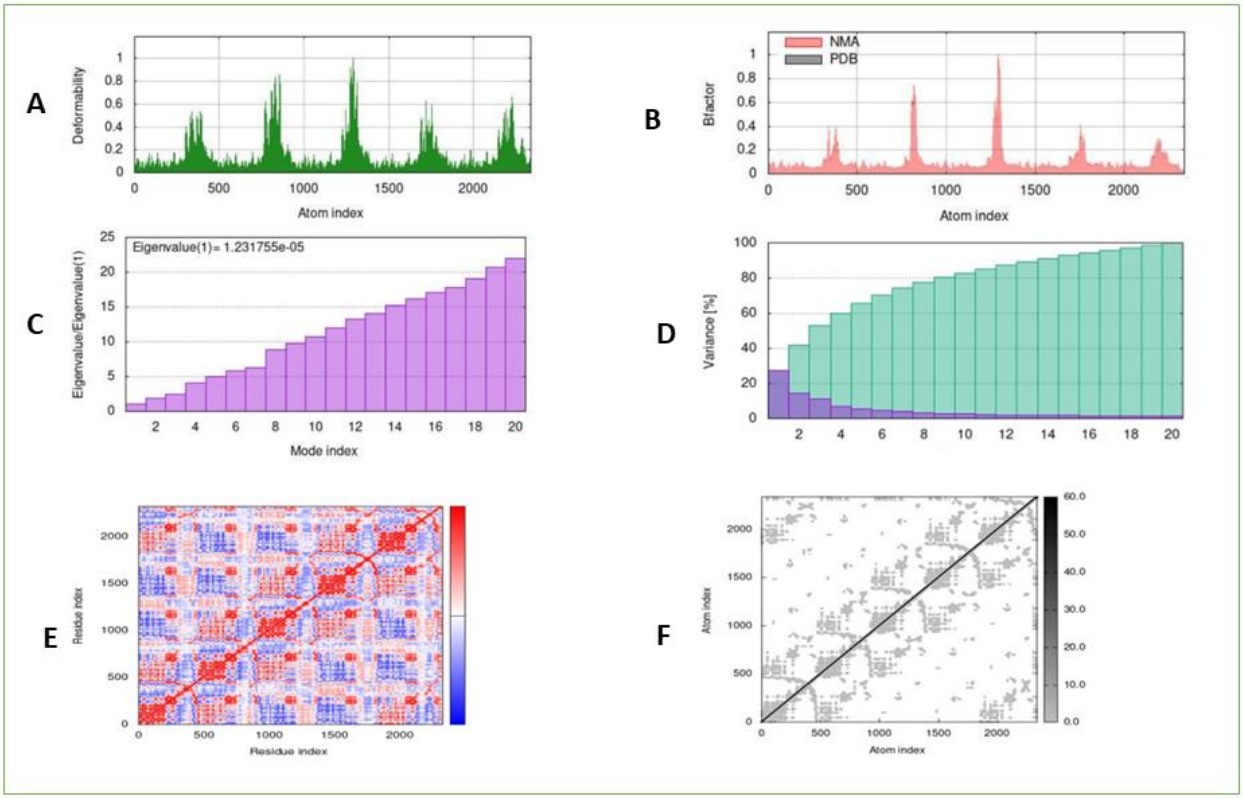

Figure 4. MD simulation result; A: deformabiloty, B: b-factor, C: eigenvalue, D: variance, E: residue index, F: atom index of receptor NAChRsβ2 with ligand conotoxin GeXIVA

B-Factor: The experimental B-factor is taken from the corresponding PDB field and the calculated from NMA is obtained by multiplying the NMA mobility by (8pi^2). Be aware that many PDB files of averaged NMR models contain no B-factors (actually, the B-factor column gives an averaged RMS).

Eigenvalues: The eigenvalue associated to each normal mode represents the motion stiffness. Its value is directly related to the energy required to deform the structure. The lower the eigenvalue, the easier the deformation.

Variance: The variance associated to each normal mode is inversely related to the eigenvalue. Colored bars show the individual (red) and cummulative (green) variances.

Covariance map: Covariance matrix indicates coupling between pairs of residues, i.e. whether they experience correlated (red), uncorrelated (white) or anti-correlated (blue) motions.

Elastic network: The elastic network model defines which pairs of atoms are connected by springs. Each dot in the graph represents one spring between the corresponding pair of atoms. Dots are colored according to their stiffness, the darker grays indicate stiffer springs and vice versa.

9. DISCUSSION

The clinical investigation of these conotoxins has been hardly reported. So far, only a few studies have published the anticancer activity of conotoxins [101]. To date, several conotoxins have proceeded to the clinical research stage, including ω-MVIIA (chronic pain, marketed), ω-CVID (analgesia, phase IIa), contulakin-G (analgesia, phase I), conantokin-G (analgesia/anti-epileptic, phase Ib), χMrIA (analgesia, phase IIa), α-Vc1.1 (analgesia, phase II, terminated), μO-MrVIB (analgesia, phase II), etc. [102,103]. ω-conotoxin MVIIA (ziconotide is a synthetic version of the peptide) is a commercial conotoxin derived from the venom of C. magus to treat chronic pain in serious cancer and AIDS patients since its launch in 2004 approved by the U.S. Food and Drug Administration (FDA) [104,105]. Conotoxins are also rapidly undergoing development for the treatment of various health conditions like pain, Alzheimer's disease, Parkinson's disease, cardiac infarction, hypertension, and various other neurological diseases [106-108]. Conopeptide is a vast drug resource; cone snails can develop up to one million different bio-active compounds [109]. But less than 0.1% of these effective compounds have been structurally and functionally identified [110-112]. Hence, conotoxins can be a significant therapeutic approach for the development of drugs treating cancer disease.

10. CONCLUSION

In this study, we identified a special category of compound conotoxins, obtained from invertebrate sources like, molluscs which cover wide aspects of medicinal properties providing cure against various diseases with the use of computational biology and metanalysis. We successfully identified potential target receptors which plays vital role in various metabolic pathways for different kind of cancers, asthma and tuberculosis. Further, in our study we identified particular class of conotoxins (GeXIVA, MII, 3Alpha subunit) which can inhibit proliferation of cancerous cells and acts as tumor suppressors. Future studies on these variants can build a potential path for developing drugs against various diseases.

ACKNOWLEDGEMENT

I am very thankful to Head, Dept. of Tech Biosciences, Digianalix, Ranchi for providing research facility for successful completion of this work.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Not applicable.

HUMAN AND ANIMAL RIGHTS

No Animals/Humans were used for studies that are base of this research.

CONSENT FOR PUBLICATION

Not applicable.

FUNDING

None.

CONFLICT OF INTEREST

The authors have no conflict of interest.

REFERENCES

1. Bohlin L, Göransson U, Alsmark C, Wedén C, Backlund A. Natural products in modern life science. Phytochem. Rev. 2010; 9:279-301.
2. Fattorusso, E., Gerwick, W. H., & Taglialatela-Scafati. Handbook of marine natural products*:* NY: Springer. 2012, (pp. 191-293).
3. Fenical W, Jensen PR- Developing a new resource for drug discovery: marine actinomycete bacteria. Nat. chem. Biol. 2006; 2:666-73.
4. Gerwick WH, Moore BS-Lessons from the past and charting the future of marine natural products drug discovery and chemical biology. Chem. biol. 2012; 19:85-98.
5. Balmain A, Barrett JC, Moses H, Renan MJ- How many mutations are required for tumorigenesis? Implications from human cancer data. Mol. carcinog. 1993; 7:139-46.
6. Wang Z-Roles of K+ channels in regulating tumour cell proliferation and apoptosis. Pflug. Arch. 2004; 448:274–286
7. Espinosa E, Zamora P, Feliu J, Barón MG- Classification of anticancer drugs – a new system based on therapeutic targets. Cancer Treat Rev 2003; 29:515–523.
8. Cassidy J, Misset JL-Oxaliplatin-related side-effects: characteristics and management. Semin Oncol 2002; 29:11–20.
9. Kalyanaraman B, Joseph J, Kalivendi S, Wang S, Konorev E, Kotamraju S- Doxorubicin-induced apoptosis: implications in cardiotoxicity. Mol Cell Biochem 2002; 234–235:119–s124.
10. Khalifa SA, Elias N, Farag MA, Chen L, Saeed A, Hegazy ME, Moustafa MS, Abd El-Wahed A, Al-Mousawi SM, Musharraf SG, Chang FR-Marine natural products: A source of novel anticancer drugs. Mar. drugs. 2019; 17:491.
11. Lebbe EK, Peigneur S, Wijesekara I, Tytgat J-Conotoxins targeting nicotinic acetylcholine receptors: an overview. Mar. Drugs. 2014; 12:2970–3004
12. 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
13. Vetter I, J Lewis R- Therapeutic potential of cone snail venom peptides (conopeptides). Curr. Top. Med. Chem. 2012; 12:1546–1552.
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. Proteom. 2013; 12: 312–329.
15. Robinson SD, Safavi-Hemami H, McIntosh LD, Purcell AW, Norton RS, Papenfuss AT-Diversity of conotoxin gene superfamilies in the venomous snail, Conus victoriae. PLoS one. 2014; 9:123-127.

16. Lewis RJ, Garcia ML -Therapeutic potential of venom peptides. Nat. Rev. Drug Discov. 2003; 2:790–802.
17. Prevarskaya N, Skryma R, Shuba Y- Ion channels and the hallmarks of cancer. Trends Mol. Med. 2010; 16:107-121.
18. Arcangeli A, Crociani O, Lastraioli E, Masi A,Pillozzi S, Becchetti A- Targeting ion channels in cancer: a novel frontier in antineoplastic therapy. Curr. Med. Chem. 2009; 16: 66-93.
19. Conti M- Targeting ion channels for new strategies in cancer diagnosis and therapy. Curr. Clin. Pharmacol., 2007; 2:135-144.
20. Prevarskaya N, Skryma R, Bidaux G, Flourakis M, Shuba Y- Ion channels in death and differentiation of prostate cancer cells. Cell Death Differ. 2007; 14: 1295-1304.
21. Fiske J. L, Fomin VP, Brown ML, Duncan RL, Sikes RA- Voltage-sensitive ion channels and cancer. Cancer Metastasis Rev. 2006; 25: 493-500.
22. Schonherr R- Clinical relevance of ion channels for diagnosis and therapy of cancer. J. Membr. Biol. 2005; 205: 175-184.
23. Kunzelmann K- Ion channels and cancer. J. Membr. Biol. 2005; 205 :159-173.
24. Trepel JB- Ion channels as molecular targets in prostate cancer. Clin. Prostate. Cancer. 2003; 2: 188-189.
25. Abdul M, Hoosein N- Expression and activity of potassium ion channels in human prostate cancer. Cancer Lett. 2002; 186: 99- 105.
26. Ireland CM- Uniqueness of the marine chemical environment: categories of marine natural product from invertebrates. Mem Calif Acad Sci. 1988; 13:41-57.
27. Ireland CM, Copp BR, Foster MP, McDonald LA, Radisky DC, Swersey JC- Biomedical potential of marine natural products. Pharmaceutical and bioactive natural products. 1993; 1-43.
28. Blunt JW, Copp BR, Hu WP, Munro MH, Northcote PT, Prinsep MR- Marine natural products. Nat. Prod. Rep. 2009; 26: 170–244.
29. Maggon K- Best-selling human medicines 2002-2004. Drug Discov. 2005;10: 739–742.
30. Newman DJ, Cragg GM. Natural products as sources of new drugs over the last 25 years. J. nat. prod. 2007; 70:461-77.
31. Leal M, Sapra P, Hurvitz SA, Senter P, Wahl A, Schutten M, Shah DK, Haddish-Berhane N, Kabbarah O. Antibody–drug conjugates: an emerging modality for the treatment of cancer.Ann. N. Y.Acad. Sci. 2014; 1321: 41–54.
32. Zhang Y. Why do we study animal toxins? Zool. Res.2015; 36:183–222.
33. Ponte G, Modica MV- Salivary glands in predatory mollusks: Evolutionary considerations. Front. Physiol.2017; 8:580.

34. Olivera BM, Gray WR, Zeikus R, McIntosh JM, Varga J, Rivier J, De Santos V, Cruz LJ- Peptide neurotoxins from fish-hunting cone snails. Science. 1985; 23:1338-43.
35. Nelson L- Venomous snails: One slip, and you're dead... Nature. 2004; 429:798-800.
36. Olivera BM, Seger J, Horvath MP, Fedosov AE- Prey-capture strategies of fish-hunting cone snails: behavior, neurobiology and evolution. Brain Behav. Evol. 2015; 86: 58–74.
37. Kohn AJ, Saunders PT, Wiener S- Preliminary studies on the venom of the marine snail Conus. Ann N Y Acad Sci. 1960; 90: 706–725.
38. Terlau H, Shon KJ, Grilley M, Stocker M, Stühmer W, Olivera BM- Strategy for rapid immobilization of prey by a fish-hunting marine snail. Nature. 1996; 381:148-51.
39. 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: 1659–1675.
40. Vetter I, J Lewis R. Therapeutic potential of cone snail venom peptides (conopeptides). Curr. Top. Med. Chem. 2012; 12: 1546–1552.
41. Nasiripourdori A, Taly V, Grutter T, Taly A- From toxins targeting ligand-gated ion channels to therapeutic molecules. Toxins. 2011; 3:260-93.
42. Hart SL- Beyond greening: strategies for a sustainable world. Harvard business review. 1997; 75:66-77.
43. Bingham JP, Broxton NM, Livett BG, Down JG, Jones A, Moczydlowski EG- Optimizing the connectivity in disulfide-rich peptides: α-conotoxin SII as a case study. Analytical biochemistry. 2005 ;338:48-61.
44. Monteiro MC, Romao PR, Soares AM- Pharmacological perspectives of wasp venom. Protein Pept. Lett. 2009; 16:944-52.
45. Li Q, Barghi N, Lu A, Fedosov AE, Bandyopadhyay PK, Lluisma AO, Concepcion GP, Yandell M, Olivera BM, Safavi-Hemami -Divergence of the Venom Exogene Repertoire in Two Sister Species of 500 Turriconus. Genome biol and evol. 2017; 9:2211-2225.
46. Phuong MA, Mahardika GN, Alfaro ME- Dietary breadth is positively correlated with venom complexity in cone snails. BMC genomics. 2016; 17:1-5.
47. Robinson SD, Li Q, Lu A, Bandyopadhyay PK, Yandell M, Olivera BM, Safavi-Hemami H - The venom repertoire of Conus gloriamaris (Chemnitz, 1777), the glory of the sea. Marine drugs. 2017; 15:145.
48. Mena EE, Gullak MF, Pagnozzi MJ, Richter KE, Rivier J, Cruz LJ, Olivera BM. Conantokin-G - a novel peptide antagonist to the N-methyl-D-aspartic acid (NMDA) receptor. Neuroscience letters. 1990; 118:241-4.
49. Maillo M, Aguilar MB, Lopez-Vera E, Craig AG, Bulaj G, Olivera BM, De La Cotera EH- Conorfamide, a Conus venom peptide belonging to the RFamide family of neuropeptides. Toxicon. 2002; 40:401-7.

50. Biggs JS, Rosenfeld Y, Shai Y, Olivera BM. Conolysin-Mt- a conus peptide that disrupts cellular membranes. Biochemistry. 2007; 46:12586-93.
51. Pisarewicz K, Mora D, Pflueger FC, Fields GB, Marí F. Polypeptide chains containing d-γ-hydroxyvaline. J. Am. Chem. Soc. 2005; 127: 6207–6215.
52. Han Y, Huang F, Jiang H, Liu L, Wang Q, Wang Y, Shao X, Chi C, Du W, Wang C- Purification and structural characterization of ad-amino acid-containing conopeptide, conomarphin, from Conus marmoreus. The FEBS Journal. 2008; 275:1976-87.
53. Jacobsen R, Jimenez EC, Grilley M, Watkins M, Hillyard D, Cruz LJ, Olivera BM. The contryphans, ad-trvptophan-containing family of Conus peptides: interconversion between conformers. J Pept Res, 1998; 51: 173–179.
54. Cruz LJ, De Santos V, Zafaralla GC, Ramilo CA, Zeikus R, Gray WR, Olivera BM. Invertebrate vasopressin/oxytocin homologs. Characterization of peptides from Conus geographus and Conus straitus venoms. J Biol Chem,.1987; 262: 15821–15824
55. McIntosh JM, Olivera BM, Cruz LJ- Conus peptides as probes for ion channels. In Methods in enzymology Academic Press. 1999; 294: 605-624.
56. McIntosh JM, Santos AD, Olivera BM- Conus peptides targeted to specific nicotinic acetylcholine receptor subtypes. Annu. Rev. Biochem., 1999; 68: 59–88.
57. Zhang R, Snyder GH- Factors governing selective formation of specific disulfides in synthetic variants of. alpha-conotoxin. Biochemistry. 1991; 30:11343-8.
58. Dutton JL, Craik DJ. Alpha conotoxins nicotinic acetylcholine receptor antagonists as pharmacological tools and potential drug leads. Curr. Med. Chem. 2001; 8: 327– 344.
59. Olivera BM, Quik M, Vincler M, McIntosh JM- Subtype-selective conopeptides targeted to nicotinic receptors: Concerted discovery and biomedical applications. Channels. 2008; 2:143-52.
60. Arias HR, Blanton MP- α-Conotoxins. *Int. J. Biochem. Cell Biol*. 2000; 32:1017-28.
61. McIntosh JM, Ghomashchi F, Gelb MH, Dooley DJ, Stoehr SJ, Giordani AB, Naisbitt SR, Olivera BM-Conodipine-M, a Novel Phospholipase A2 Isolated from the Venom of the Marine Snail Conus magus . J. Biol. Chem. 1995; 270:3518-26.
62. McIntosh JM, Olivera BM, Cruz LJ, Gray WR- Gamma-carboxyglutamate in a neuroactive toxin. J. Biol. Chem. 1984; 259:14343-6.
63. Adams DJ, Smith AB, Schroeder CI, Yasuda T, Lewis RJ- ω-Conotoxin CVID inhibits a pharmacologically distinct voltage-sensitive calcium channel associated with transmitter release from preganglionic nerve terminals. J. Biol. Chem. 2003; 278:4057-62.
64. Feng ZP, Doering CJ, Winkfein RJ, Beedle AM, Spafford JD, Zamponi GW. Determinants of inhibition of transiently expressed voltage-gated calcium channels by ω-conotoxins GVIA and MVIIA. J. Biol. Chem. 2003; 278:20171-8.

65. Xia Z, Chen Y, Zhu Y, Wang F, Xu X, Zhan J- Recombinant ω-conotoxin MVIIA possesses strong analgesic activity. Bio. Drugs. 2006; 20:275-81.
66. Mirshafiey A- Venom therapy in multiple sclerosis. Neuropharmacology. 2007; 53:353-61.
67. Callaghan B, Haythornthwaite A, Berecki G, Clark RJ, Craik DJ, Adams DJ- Analgesic α-conotoxins Vc1. 1 and Rg1A inhibit N-type calcium channels in rat sensory neurons via GABAB receptor activation. J. Neurosci. 2008; 28: 10943-10951.
68. Park SP, Kim BM, Koo JY, Cho H, Lee CH, Kim M, Na HS, Oh U- A tarantula spider toxin, GsMTx4, reduces mechanical and neuropathic pain. PAIN. 2008; 137:208-17.
69. Satkunanathan N, Livett B, Gayler K, Sandall D, Down J, Khalil Z. Alpha-conotoxin Vc1. 1 alleviates neuropathic pain and accelerates functional recovery of injured neurons. Brain Res. 2005; 1059: 149-158.
70. Beeton C, Smith BJ, Sabo JK, Crossley G, Nugent D, Khaytin I, Chi V, Chandy KG, Pennington MW, Norton RS- The D-diastereomer of ShK toxin selectively blocks voltage-gated K+ channels and inhibits T lymphocyte proliferation. J. Biol. Chem.2008; 28: 988-997.
71. Beraud E, Viola A, Regaya I, Confort-Gouny S, Siaud P, Ibarrola D, Le Fur Y, Barbaria J, Pellissier JF, Sabatier JM, Medina I. Block of neural Kv1. 1 potassium channels for neuroinflammatory disease therapy. Ann. Neurol. 2006; 60: 586-596.
72. Meuth SG, Melzer N, Kleinschnitz C, Budde T, Wiendl H- Multiple sclerosis–a channelopathy? Targeting ion channels and transporters in inflammatory neurodegeneration. Der nervenarzt. 2009; 80:422-9.
73. Norton RS, Pennington MW, Wulff H- Potassium channel blockade by the sea anemone toxin ShK for the treatment of multiple sclerosis and other autoimmune diseases. Curr. Med. Chem. 2004; 11 : 3041- 3052.
74. Becchetti A, Arcangeli A. A comment on ion channels as pharmacological targets in oncology. J. Gen. Physiol. 2008; 132: 313-314.
75. Gao B, Peng C, Lin B, Chen Q, Zhang J, Shi Q- Screening and validation of highly-efficient insecticidal conotoxins from a transcriptome-based dataset of chinese tubular cone snail. Toxins. 2017; 9:214.
76. Dhiman V, Pant D- Human health and snails. J. Immunoass. and Immunochem. 2021; 42: 211–235.
77. Cahalan MD, Wulff H, Chandy KG- Molecular properties and physiological roles of ion channels in the immune system. J. Clin. Immunol. 2001; 21: 235-252.
78. Vicente R, Escalada A, Soler C, Grande M, Celada A, Tamkun MM, Solsona C, Felipe A- Pattern of Kvβ subunit expression in macrophages depends upon proliferation and the mode of activation. J. Immunol. 2005; 174: 4736-4744.
79. Vicente R, Escalada A, Villalonga N, Texido L, Roura-Ferrer M, Martín-Satué M, Lopez-

Iglesias C, Soler C, Solsona C, Tamkun MM, Felipe A - Association of Kv1. 5 and Kv1. 3 contributes to the major voltage-dependent K+ channel in macrophages. J. Biol. Chem. 2006; 281: 37675-37685.

80. Fainzilber M, Hasson A, Oren R, Burlingame AL, Gordon D, Spira ME, Zlotkin E- New mollusk-specific. alpha-conotoxins block Aplysia neuronal acetylcholine receptors.Biochem. 1994; 33: 9523-9529.
81. McIntosh JM, Yoshikami D, Mahe E, Nielsen DB, Rivier JE, Gray WR, Olivera BM- A nicotinic acetylcholine receptor ligand of unique specificity, alpha-conotoxin ImI. J. Biol. Chem. 1994; 269: 16733-16739.
82. Johnson DS, Martinez J, Elgoyhen AB, Heinemann SF, McIntosh JM- alpha-Conotoxin ImI exhibits subtype-specific nicotinic acetylcholine receptor blockade: preferential inhibition of homomeric alpha 7 and alpha 9 receptors. Mol. Pharmacol. 1995; 48: 194-199.
83. Fujii YX, Fujigaya H, Moriwaki Y, Misawa H, Kasahara T, Grando SA, Kawashima K- Enhanced serum antigen-specific IgG1 and proinflammatory cytokine production in nicotinic acetylcholine receptor α7 subunit gene knockout mice. J. Neuroimmunol. 2007; 189: 69-74.
84. De Rosa MJ, Dionisio L, Agriello E, Bouzat C, del Carmen Esandi M- Alpha 7 nicotinic acetylcholine receptor modulates lymphocyte activation. Life Sci. 2009; 85: 444-449.
85. Rosas-Ballina M, Tracey KJ- The neurology of the immune system: neural reflexes regulate immunity. Neuron. 2009; 64: 28-32.
86. Kawashima K, Fujii T, Moriwaki Y, Misawa H- Critical roles of acetylcholine and the muscarinic and nicotinic acetylcholine receptors in the regulation of immune function. Life Sci. 2012; 91: 1027–1032.
87. Linnoila RI- From nicotine to breast cancer, implications of cholinergic receptor pathway. J. Natl. Cancer Inst. 2010; 102: 1298–1299.
88. Lebbe EK, Peigneur S, Wijesekara I, Tytgat J- Conotoxins targeting nicotinic acetylcholine receptors: an overview. Mar. Drugs. 2014; 12: 2970–3004.
89. Zdanowski R, Krzyżowska M, Ujazdowska D, Lewicka A, Lewicki S- Role of α7 nicotinic receptor in the immune system and intracellular signaling pathways. Cent. Eur. J. Immunol. 2015; 40: 373–379.
90. Sun H, Ma X- α5-nAChR modulates nicotine-induced cell migration and invasion in A549 lung cancer cells. Exp. Toxicol. Pathol. 2015; 67: 477–482.
91. Ho YS, Lee CH, Wu CH- The alpha 9-nicotinic acetylcholine receptor serves as a molecular target for breast cancer therapy. J. Exp. Clin. Med. 2011; 3: 246–251.
92. Calleja-Macias IE, Kalantari M, Bernard HU- Cholinergic signaling through nicotinic acetylcholine receptors stimulates the proliferation of cervical cancer cells: an explanation for

the molecular role of tobacco smoking in cervical carcinogenesis? Int. J. Cancer 2009; 124: 1090–1096.

93. Improgo MR, Soll LG, Tapper AR, Gardner PD- Nicotinic acetylcholine receptors mediate lung cancer growth. Front. Physiol. 2013; 4: 251.
94. Grando SA- Connections of nicotine to cancer. Nat. Rev. Cancer 2014; 14: 419–429.
95. Lee CH, Huang CS, Chen CS, Tu SH, Wang YJ, Chang YJ, Tam KW, Wei PL, Cheng TC, Chu JS, Chen LC- Overexpression and activation of the α9-nicotinic receptor during tumorigenesis in human breast epithelial cells. J. Natl. Cancer Inst. 2010; 102: 1322–1335.
96. Huang LC, Lin CL, Qiu JZ, Lin CY, Hsu KW, Tam KW, et al. Nicotinic acetylcholine receptor subtype alpha-9 mediates triple-negative breast cancers based on a spontaneous pulmonary metastasis mouse model. Front. Cell. Neurosci. 2017; 11: 336.
97. Luo S, Zhangsun D, Harvey PJ, Kaas Q, Wu Y, Zhu X, et al. Cloning, synthesis, and characterization of αO-conotoxinGeXIVA, a potent α9α10 nicotinic acetylcholine receptor antagonist. Proc. Natl. Acad. Sci. 2015; 112: 4026–4035.
98. Wang H, Li X, Zhangsun D, Yu G, Su R, Luo S. The α9α10 nicotinic acetylcholine receptor antagonist αO-conotoxinGeXIVA [1, 2] alleviates and reverses chemotherapy-induced neuropathic pain. Mar. Drugs 2019; 17: 265.
99. Li X, Hu Y, Wu Y, Huang Y, Yu S, Ding Q, et al. Anti-hypersensitive effect of intramuscular administration of αO-conotoxinGeXIVA [1, 2] and GeXIVA [1, 4] in rats of neuropathic pain. Prog. Neuropsychopharmacol. Biol. Psychiatry 2016; 66: 112–119.
100. Liu Y, Qian J, Sun Z, Zhangsun D, Luo S. Cervical cancer correlates with the differential expression of nicotinic acetylcholine receptors and reveals therapeutic targets. Mar. Drugs 2019; 17: 256.
101. Gao B, Peng C, Lin B, Chen Q, Zhang J, Shi Q. Screening and validation of highly- efficient insecticidal conotoxins from a transcriptome-based dataset of chinese tubular cone snail. Toxins. 2017; 9:214
102. Dhiman V, Pant D. Human health and snails. J. Immunoass. Immunochem. 2021; 42: .211–235.
103. Fu Y, Li C, Dong S, Wu Y, Zhangsun D, Luo S. Discovery methodology of novel conotoxins from Conus species. Mar. Drugs 2018; 16: 417.
104. Rigo FK, Dalmolin GD, Trevisan G, Tonello R, Silva MA, Rossato MF, et al. Effect of ω-conotoxin MVIIA and Phα1β on paclitaxel-induced acute and chronic pain. Pharmacol. Biochem. Behav. 2013; 114: 16–22.
105. Eisapoor SS, Jamili S, Shahbazzadeh D, GhavamMostafavi P, PooshangBagheri K. A new, high yield, rapid, and cost-effective protocol to deprotection of cysteine-rich conopeptide, omega-conotoxinMVIIA. Chem. Biol. Drug Des. 2016; 87: 687–693.

106. Han TS, Teichert RW, Olivera BM, Bulaj G. Conus venoms-a rich source of peptide-based therapeutics. Curr. Pharm. Des. 2008; 14: 2462–2479.
107. Olivera BM, Teichert RW. Diversity of the neurotoxic Conus peptides: A model for concerted pharmacological discovery. Mol. Interv. 2007; 7: 251–260.
108. Fedosov AE, Moshkovskii SA, Kuznetsova KG, Olivera BM. Conotoxins: From the biodiversity of gastropods to new drugs. Biomed. Chem. 2013; 59: 267–294.
109. Jin AH, Muttenthaler M, Dutertre S, Himaya SW, Kaas Q, Craik DJ, et al. Conotoxins: Chemistry and biology. Chem. Rev. 2019; 119: 11510–11549.
110. Vetter I, J Lewis R. Therapeutic potential of cone snail venom peptide (conopeptides).Curr. Top Med. Chem. 2012; 12: 1546–1552.
111. Lewis RJ, Dutertre S, Vetter I, Christie MJ. Conus venom peptide pharmacology. Pharmacol. Rev. 2012; 64: 259–298.
112. Fu Y, Li C, Dong S, Wu Y, Zhangsun D, Luo S. Discovery methodology of novel conotoxins from Conus species. Mar. Drugs 2018; 16: 417.