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Research Article

Molecular Docking Studies to Identify Inhibitors of Norepinephrine Reuptake from Marine Algae

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ABSTRACT

Depression is one of the major mental health problems at prevalence nowdays. It can be characterized by poor concentration, low self-esteem, losing interest in family or social life, feeling tired or fatigued, suicidal thoughts and similar symptoms. There are treatments like psychotherapies, anti-depressants and electroconvulsive therapies available, but there is need to identify more effective treatments with lesser side effects. Marine organisms like algae, sponges or corals have been investigated to explore their potential as anti-depressants. This article aims to explore the potential of some compounds from marine algae by molecular docking and assessment of pharmacokinetics. Human norepinephrine transporter (hNET) was used as target for this study as this transporter is responsible to reuptake norepinephrine and disturbs the chemical balance of the brain, which can be a cause for depression. These compounds' Binding affinity was compared with the binding affinity of prescribed levomilnacipran. From 14 selected compounds, 13 showed higher binding affinity towards hNET. Among all compounds, saringosterone has the highest binding score. Pharmacokinetics properties were constructive for most compounds. Compounds showed weaker druglikeness and drugs score but can be optimized to enhance it. Compounds identified as inhibitors of NET can be developed as drug molecules in the future or algal sources for it can be taken as a food supplement.

Introduction

Naturally occurring compounds are used dominantly as medicines for different diseases as plenty of structurally and chemically distinct molecules are available. Almost all terrestrial organisms have been investigated by researchers in search of crucial compounds that possess some bioactivity. Available resources are being overburdened by a growing population. Low bioavailability, higher cost of synthetic drugs, time consumed for a single drug to come into market are some reasons for the shortage of pocket-friendly medicines. Hence, researchers are always in search of new and effective drugs.[1] Large portion of earth is covered with oceans. The biodiversity of ocean is much greater than the biodiversity on land. Out of 36 animal phyla identified, 34 are from marine domain.[2] Ocean is a treasure of useful compounds like steroids, anti-neoplastic agents, anti-cancerous agents, enzymes, nutritional supplements and other secondary metabolites. Marine bacteria, fungi, dinoflagellates, algae, sponges, cnidarans are sources of beneficial therapeutic compounds.[3] Traditional Chinese medicines and Japanese herbs of marine origin have been used since long for their therapeutic effects. One of the first drugs used was toxin (nereistoxin) and was obtained from flatworm *Lumbriconeris heteropoda*.[4] The first commercial marine compounds used as drugs are Ara A and Ara C. FDA approved Ara-C as anti-cancer agent and Ara-A as an anti-viral agent in 1969 and 1976, respectively.[5] Various challenges are faced while isolating the compounds from marine origin. Variable metabolites are produced by same organism due to different environmental conditions.[1]

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Organisms from which compounds isolated are not always the producers but microbes residing on them are producing the metabolite.[6] To enhance the research, derivatives of isolated compounds are being developed synthetically.[7] There are lots of articles throwing light into the area of marine pharmaceutics. Very few neuroprotective compounds were identified from marine origin. Indian seaweed *Ulva reticulata* is efficient in the treatment of Alzheimer's disease.[8] Compounds derived from marine algae *S. siliquastrum* have been reported for their neuroprotective activities.[9] *Sargassum polycystum* and *Laurencia obtusa* are some algae that possess antioxidant activity.[10] Fucoxanthin[11] fucosterol[12] fucoidan[13] sargaquinoic acid[14] and omega-3 fatty acids[15] are some therapeutic compounds of marine origin. Marine compounds are potent in exhibiting properties like anti-aging, neuroprotective, anti-cholinesterase, monoamine oxidase inhibition, anti-fungal activities and lot more. A lot of research has been made on marine compounds for Alzheimer's disease, Parkinson's disease, anti-inflammatory, anti-cancerous, anti-fungal properties and other neurological disorders. Compounds from marine origin can also be effective in treating depression and anxiety disorders. Depression can be characterized by continuous feeling of sadness, anxiety or, feeling worthless and losing interest in daily activities. It can be due to stress, emotional loss, genetic vulnerability or faulty mood regulations by some chemicals in brain. If not treated, it can lead to suicide. Different mechanisms have been proposed to indicate the cause of depression, but none of them completely justifies the interaction between external environment and biological characteristics that lead towards depression.[16] A single reason cannot be given behind the cause of depression. Diet plays an important role in the prevention of diseases. A healthy diet leads to a healthy body and healthy mind. Diet, including long-chain omega-3 fatty acids is helpful in preventing neurological disorders. Intake of diet including fishes and other seafood, nuts, whole grains can lower the risk of developing depression.[17,18] Deficiencies of omega-3 fatty acids, vitamin B12 and folate cause depression.[19,20] In a study in Japan, 1745 pregnant women were observed for diet they consumed and onset of depression. High consumption of seaweed was inversely related with lower levels of depression in subjects.[21]

Marine compounds have also been evaluated for their anti-depressant activity, but number of clinical trials as compared to other neurodegenerative diseases is less. Some clinical studies carried out on algae and sterols of marine origin showed positive results in rodents. Marine compounds like β-sitosterol and other sterols show anti-depressant effects in forced swim test and tail suspension test.[22] Brown seaweed (algae) has benefits like cancer prevention, reducing inflammation, anti-diabetic properties, and antioxidant properties.[23]

Eicosapentanoic acid (EPA) and docosahexaenoic acid (DHA) are omega-fatty acids obtained from fishes and marine algae.[24] A study carried out on omega-3 fatty acids combined with anti-depressants resulted in reduction of depressive symptoms.[25] *Padina gymnospora, S. angustifolium, S. fusiforme, P. australis*, *Nannochloropsis salina*, *Undaria pinnatifida* are some of the algae that contain pharmacologically active compounds.[23,26] Increase in neurotransmitters were observed by treatment of fucosterol from *S. fusiforme* in mouse brain.[27] Disturbed sleep pattern is associated with depression. It is seen that fucoxanthin improves sleep pattern in animals.[28] Extracts of *Padina australis* Hauck reduced depression like effects by targeting mitochondrial oxidation.[29] *S. ilicifolium* extract showed efficacy parallel to fluoxetine and imipramine.[30] Methanol extracts of macroalgae like *Nizamuddinia zanardinii* (Shiffner) P. C. Silva, *Stoechospermum marginatum* (C. Agardh) Kützing and *Sargassum swarzii* C. Agardh extracted from Ormara in Pakistan showed anti-depressant activity. Depression-like behavior was improved in rat models after their administration for 28 days.[31]

At present, there are different types of anti-depressants depending on different targets. Anti-depressants can be inhibiting serotonin reuptake, serotonin-noradrenaline reuptake, noradrenaline-dopamine reuptake, monoamine oxidase or some may be atypical. These anti-depressants may have severe side effects and all of them are not in generic form. Cost of these anti-depressants is very high. Metabolic profile may vary person to person. So, same medication can affect differently in individuals. Duloxetine, vanlaflexine, levomilnacipran, milnacipran are anti-depressants that target serotonin and norepinephrine reuptake. Duloxetine and venlaflexine have higher selectivity towards inhibiting serotonin reuptake than inhibition of norepinephrine reuptake. Milnacipran have almost an affinity towards both levomilnacipran has higher selectivity towards inhibiting norepinephrine reuptake than inhibiting serotonin reuptake.[32] But these drugs have side effects like nausea, headaches, loss of apetite, and diarrhoea. Also all of them are not generic. Some anti-depressants are very costly and are not affordable by patients with low income. There is need to identify natural compounds that are effective against depression and other such neurological disorders so the medication must be easily available with lesser side effects and must have sufficient bioavailability.

The current study evaluates docking score and absorption, distribution, metabolism, excretion and toxicity (ADMET) properties of some selected compounds from marine algae to be potent anti-depressants. Algal sources are used as supplements from ancient times, but the mechanism of interaction of these compounds in the body is not very well known. We need to explore the targets for these compounds to enhance their use as drugs. These natural

compounds mitigate the risk of side effects and will be less toxic as compared to currently prescribed synthetic drugs. The anti-depressant effect of marine algae is studied in different experiments but actual process underlying this anti-depressant effect needs to be investigated. This study aims to identify compounds from marine algae using *an in-silico* approach by analyzing selected compounds' binding affinity for NET. Levomilnacipran is inhibitor for NET. Hence, was chosen as a reference molecule. ADMET properties of selected compounds were also predicted to evaluate their potency as a drug. As NET is an important target for depression, compounds efficiently interacting with it can be selected as better anti-depressants.

Materials and Methods

Selection of Target

X-ray crystal structure of human norepinephrine transporter (hNET) was not available in Research Collaboratory for Structural Bioinformatics (RCSB) Protein Data Bank (PDB). So, Alphafold (https://alphafold.ebi.ac.uk/)[33,34] was used to retrieve the structure (https://alphafold.ebi.ac.uk/entry/P23975). Structure was downloaded in PDB format (Alphafold: AF-P23975-F1). Alphafold predicts protein structure on the basis of its sequence. Alphafold help researchers to use computationally predicted structures and fills the gaps of unavailability of x-ray or NMR structures.

Structure Evaluation

PROCHECK from PDBsum (http://www.ebi.ac.uk/thornton-srv/databases/pdbsum/)[35] was used to evaluate the protein structure. PDBsum can be searched by entering ID or sequence of protein or just by simple text search by compound, author or protein name. It also allows searching by UniProt, Pfam or Ensembl id. PROCHECK evaluation gives information about the number of residues that lie in the favored or disallowed region through Ramachandran plot. It also gives information about G-factors regarding dihedral angles and main chain covalent forces.

Ligand Selection

Ligands were taken from literature by searching for marine sources that had some significance in curing neurological disorders. PubChem (https://pubchem.ncbi.nlm.nih.gov/)[36] were used to download 3D SDF files for the selected ligands. Ligands whose 3D SDF files were not available in PubChem were downloaded using ChemSpider (http://www.chemspider.com/)[37,38] in .mol format. Along with selected compounds, SDF file for anti-depressants levomilnacipran was also downloaded from PubChem. Ligands must be in PDB format for further process. Hence, they were converted from SDF to PDB using PyMol (https://pymol.org/2/)[39] which is used to visualize molecules and also helps to convert them in PDB/mmCIF/PQR/Maestro and other such molecular file formats.

Binding Site Prediction

ProteinPlus server (https://proteins.plus/)[40] was used to predict the binding pocket of hNET for levomilnacipran. .pdb file for Alphafold structure of hNET and .sdf file for levomilnacipran as ligand were given as input. Pocket can be modified by adding or removing amino acids in it.

Virtual Screening

Virtual screening was performed with the help of PyRx (https://pyrx.sourceforge.io/)[41] for hNET and selected compounds. hNET was defined as macromolecule and compounds were defined as ligands. This will turn your ligands and receptors into .pdbqt format. Residues in the binding pocket were selected and grid box was adjusted according to it. Binding affinity in kcal/mol was obtained and saved in CSV format.

Preparing Protein-ligand Complex

PyMol is used to prepare protein-ligand complex files. Once the docking is performed the out files are generated. For saving ligand-receptor complex, output files for receptor and ligand were opened in PyMol and exported as a single file.

Visualization of Interactions

LigPlot+ (https://www.ebi.ac.uk/thornton-srv/software/LigPlus/)[42] is a used for generating 2D diagrams for ligand-receptor interaction. If your file contains more than one ligand, you must choose ligand whose interactions must be viewed. LigPlot+ is also used for showing interactions across the interface with the help of DIMPLOT and antibody plot for plotting antigen-antibody interactions.

ADMET Property

ADMET properties were predicted using AdmetSAR server (http://lmmd.ecust.edu.cn/admetsar2).[43,44] It is freely available tool to evaluate ADMET properties like $CaCo_2$ cell permeability, blood brain barrier, human intestinal absorption, human oral bioavailability, etc.

Druglikeness Prediction

ORISIS property explorer (https://www.organic-chemistry.org/prog/peo/)[45] is used to predict properties like ClogP, molecular weight, TPSA, druglikeness, drug-score as well as indicates toxicity risk. Input can be given in SMILES or molecule can be drawn.

Results

Selection of Target

Target retrieved from Alphafold database (ID: AF-P23975-F1) is shown in Fig. 1. 3D Protein structure is shown using predicted local distance difference test

(pLDDT). The structure is represented with different colours to indicate the higher and lower pLDDT score of the particular residue. Regions with very high pLDDT i.e. pLDDT>90 are shown by dark blue color and have very high model confidence. Region with pLDDT scores between 90 and 70 are shown in light blue and are classified as confident. pLDDT between 70 to 50 is low confidence region and is shown in yellow, whereas regions with pLDDT <50 are very low confidence and shown in orange color.

Structure Evaluation

Structure was evaluated using PROCHECK tool available at PDBsum. PROCHECK statistics showed that 93% of residues were in favored region and only 0.4% was in disallowed region. A good quality structure must have more than 90% residues in favoured region. Ramachandran plot for AF-P23975-F1 is shown in Fig. 2.

Retrieving Ligands

Ligands are downloaded from PubChem and ChemSpider databases in. sdf and .mol format and then converted to PDB using PyMol visualizer. Compound Id for selected ligands are given in Table 1.

Binding Site Prediction

ProteinPlus Server is used to predict binding pocket for levomilnacipran. Fig. 3 shows the detected pocket for levomilnacipran.

Residues detected within binding pocket are: PHE_72, ALA_73, VAL_74, ASP_75, LEU_76, ALA_77, ASN_78, TRP_80, ARG_81, ILE_144, ALA_145, LEU_146, TYR_147, VAL_148, GLY_149, TYR_151, TYR_152, THR_313, PHE_316, PHE_317, SER_318, GLY_320, PHE_323, VAL_325, LEU_415, GLY_416, SER_419, SER_420, GLY_422, GLY_423, ASP_473, ALA_477, GLY_478, ILE_481.

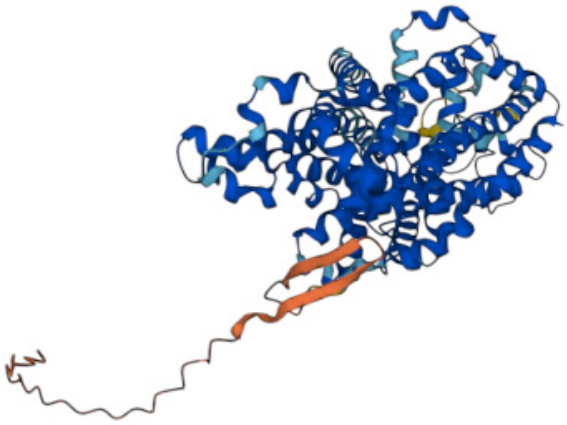

Fig. 1: 3D structure of hNET retrieved from Alphafold Database (Alphafold id: AF-P23975-F1)

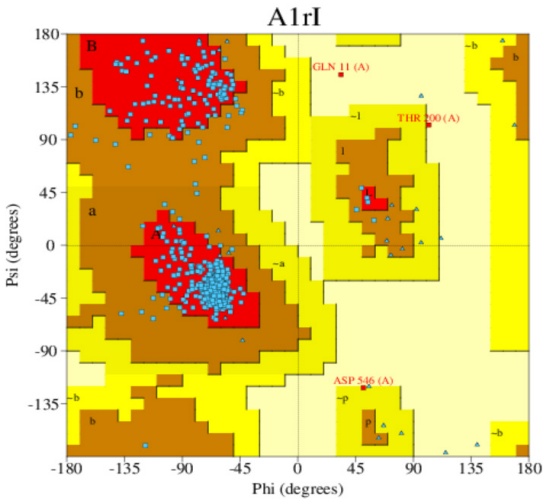

Fig. 2: Ramachandran plot for hNET from Alphafold (AF-P23975-F1)

Virtual Screening

Binding affinities were predicted using PyRx. PyRx generates 9 poses for each ligand. Higher the negative value, more efficiently the ligand binds. Ligands having higher binding affinity with respect to levomilnacipran are shown in Table 2. Levomilnacipran showed binding affinity of -6.4 kcal/mol. Brassicasterol, campesterol, cholesterol, DHA, EPA, fucosterol, fucoxanthin, pheophytin a, phytol, saringosterol, saringosterone, stigmatosterol and β-sitosterol showed higher binding affinity as compared to marketed anti-depressant levomilnacipran. Among all molecules highest binding affinity was shown by saringosterone.

Table 1: Molecules and their compound ID

S. No.	*Compounds*	*Compound ID*
1	Levomilnacipran	CID 6917779
2	Brassicasterol	CID 5281327
3	Campesterol	CID 173183
4	Cholesterol	CID 5997
5	Docosahexaenoic acid	CID 445580
6	Eicosapentaenoic acid	CID 446284
7	Fucoidan	CID 92023653
8	Fucosterol	CID 5281328
9	Fucoxanthin	CID 5281239
10	Pheophytin A	CSID 4573175
11	Phytol	CID 5280435
12	Saringosterol	CID 14161394
13	Saringosterone	CSID 29213222
14	Stigmasterol	CID 5280794
15	β-sitosterol	CID 222284

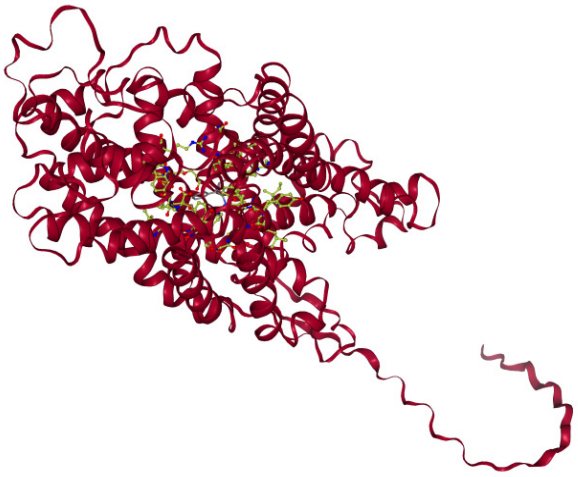

Fig. 3: Pocket detection of levomilnacipran within hNET (AF-P23957-F1)

Visualization of Interactions

LigPlot+ gives interactions in the form of hydrogen bonds and its bond length, hydrophobic interaction, covalent bonds and elastic bonds. 2D representation of interactions for all ligands having higher binding score. Table 3 summarizes the hydrophobic and hydrogen bond forming residues. Levomilnacipran do not have any hydrogen bond with target but hydrophobic interactions can be observed. Like levomilnacipran, fucoxanthin, pheophytin A and phytol do not show any hydrogen bond formation with receptor but only hydrophobic interactions. Campesterol, cholesterol, fucosterol, saringosterol, saringosterone and β –sitosterol showed hydrogen bonding with Ala384. Eicosapentanoic acid, brassicasterol and stigmasterol have hydrogen bonding with Ala145, Ser536 and Phe317, respectively. DHA forms hydrogen bonds with Thr470 and Ala384. Saringosterol forms hydrogen bond with Thr479 and Ser536. Amino acid residues forming hydrophobic interactions are similar for most compounds. Ile155, Trp80, Tyr151, Ala77, Val148, Tyr152, Ser419, Phe72, Phe323, Asp75, Arg81, Asp473, Gly383, Leu469, Phe317 are some residues forming hydrophobic interactions with ligands.

ADMET Property

AdmetSAR predicts properties like Human intestinal absorption, blood brain barrier, $CaCo_2$ cell permeability, acute oral toxicity, etc. Results are presented by "-" or "+" indicating a no or yes value for the predicted property. Predicted properties are shown in Table 4. Human intestinal absorption refers to the absorption of drug from gastrointestinal system into blood stream. All ligands showed positive result for human intestinal absorption. $CaCo^{-2}$ cells depict human intestinal absorption of drug molecule. DHA, EPA, fucoxanthin, pheophytin a, saringosterol and saringosterone showed negative value for $CaCo^{-2}$ cell permeability, rest were positive for it. Blood brain barrier refers to the regulation of molecules between blood and brain. Anti-depressants must cross blood brain barrier to function. Leaving brassicasterol, fucoxanthin and saringosterone rest all showed positive blood brain barrier permeability. Human oral bioavailability indicates the amount of drug that enters circulation after intake. It is positive for cholesterol, DHA, EPA and β- sitosterol. p-glycoprotein inhibition can improves the ADME (absorption, distribution, metabolism and elimination) pathway of drug. P-glycoprotein inhibition was positive for fucosterol, fucoxanthin, pheophytin a and saringosterone. CYP3A4 inhibition and carcinogenicity were negative for all. Ames mutagenesis was positive only for fucoxanthin. Acute oral toxicity category I is said to be highly toxic, category II is moderately toxic, category III is slightly toxic and category IV is practically non-toxic. DHA and EPA were under category IV, phytol and pheophytin a were under category III and remaining molecules were under category I according to admetSAR results.

Table 2: Binding affinities of selected molecules compared to levomilnacipran

S. No.	*Compounds*	*Binding affinity (kcal/mol)*
1	Levomilnacipran	-6.4
2	Brassicasterol	-7.6
3	Campesterol	-9.5
4	Cholesterol	-8.8
5	Docosahexaenoic acid	-7.7
6	Eicosapentaenoic acid	-7.8
7	Fucoidan	-5.6
8	Fucosterol	-9.7
9	Fucoxanthin	-7.8
10	Pheophytin A	-7.7
11	Phytol	-6.8
12	Saringosterol	-7.6
13	Saringosterone	-11
14	Stigmasterol	-7.9
15	β-sitosterol	-9.4

Druglikeness Prediction

With the help of OSIRIS property explorer druglikeness, clogP, topological polar surface area (TPSA) were predicted. Table 5 shows predicted druglikeness properties. Almost all compounds leaving fucosterol, pheophytin A and saringosterone showed no toxicity risk. cLogP is higher for all molecules, which may result in low hydrophobicity. Solubility of these molecules is also not very high. TPSA for all molecules leaving fucoxanthin and pheophytin A is below 60 $Å^2$ that specifies their permeability. Druglikeness is higher for fucoxanthin than rest of the compounds. Overall, among all properties toxicity risk,

Table 3: Molecular interactions of ligands with hNET

S. No.	*Molecules*	*Hydrogen Bonds with bond length*	*Hydrophobic interactions*
1	Levomilnacipran		Leu469, Ala384, Asp473, Arg81, Gly383, Leu386, Tyr84, Trp80
2	Brassicasterol	Ser536 (3.16, 2.89)	Leu386, Gly383, Trp80, Leu85, Tyr84, Arg81, Asp473, Thr474, Gly478, Thr479, Phe540
3	Campesterol	Ala384(2.91)	Gly383, Trp80, Tyr151, Leu469, Ile155, Asp473, Tyr152, Val148, Ser419, Phe323, Phe72, Asp75, Ala77, Phe317, Arg81
4	Cholesterol	Ala384(2.89)	Phe72, Ser419, Val148, Asp75, Phe317, Tyr152, Phe323, Arg81, Ala77, Gly383, Trp80, Leu469, Tyr151, Asp473, Ile155
5	Docosahexaenoic acid	Thr470(2.75), Ala384(2.89)	Asp75, Phe323, Tyr152, Phe72, Phe317, Asp473, Gly383, Tyr545, Ile466, Leu469, Trp80, Arg81, Tyr151, Ser429, Val148, Ser420
6	Eicosapentaenoic acid	Ala145(2.79)	Trp80, Ile155, Asp473, Ala77, Arg81, Val148, Gly149, Ser419, Tyr151, Met424, Ser420, Gly423, Phe323, Tyr152, Phe72, Asp75, Phe317
7	Fucosterol	Ala384(2.94)	Phe323, Ser419, Asp75, Phe72, Val148, Ile155, Tyr152, Tyr151, Leu469, Gly383, Arg81, Asp473, Ala77, Phe317
8	Fucoxanthin		Glu382, Asp546, Thr318, Thr544, Gly383, Asp473, Phe323, Gly423, Tyr152, Val148, Ser420, Phe72, Gly149, Ser419, Phe317, Asp75, Ala77, Arg81, Trp80
9	Pheophytin A		Lys541, Pro542, Thr474, Thr544, Gly478, Ala384, Gly383, Ala77, Leu469, Ile155, Try152, Phe72, Ser419, Val148, Ala145, Asp75, Gly149, Ser420, Gly423, Phe323, Phe317, Trp80, Met424, Asr81, Asp473
10	Phytol		Phe323, Phe72, Ser419, Gly149, Asp75, Ser420, Gly423, Phe317, Ala383, Ala77, Arg81, Ile155, Trp80, Asp473, Tyr151, Val148, Tyr152
11	Saringosterol	Ala384(3.08), Thr479(3.26), Ser536(2.80)	Gly383, Tyr545, Asp473, Thr474, Gly478, Lys541, Phe475, Phe540, Asn539
12	Saringosterone	Ala384(2.81)	Ile155, Gly383, Asp473, Trp80, Arg81, Tyr151, Ser419, Gly423, Phe323, Phe72, Val148, Tyr152, Phe317, Asp75, Ala77
13	Stigmasterol	Phe317(2.70)	Thr470, Tyr545, Gly383, Asp473, Tyr151, Tyr152, Val148, Phe323, Asp75, Ala77, Ala384, Arg81, Ile155, Trp80
14	β-sitosterol	Ala384(2.90)	Gly383, Ile155, Leu469, Asp473, Tyr152, Tyr151, Val148, Asp75, Ser419, Phe317, Ser318, Phe323, Gly320, Phe72, Ala77, Arg81, Trp80

Table 4: ADMET profile of compounds having higher binding affinity

S. No.	*Compound*	*HIA*	*Caco-2*	*BBB*	*HOB*	*p-gp inhibitor*	*CYP3A4 inhibition*	*Ames mutagenesis*	*Carcino-genecity (binary)*	*AOT (c)*
1	Levomilnacipran	+	+	+	+	-	-	-	-	III
2	Brassicasterol	+	+	-	-	-	-	-	-	I
3	Campesterol	+	+	+	-	-	-	-	-	I
4	Cholesterol	+	+	+	+	-	-	-	-	I
5	Docosahexaenoic acid	+	-	+	+	-	-	-	-	IV
6	Eicosapentaenoic acid	+	-	+	+	-	-	-	-	IV
7	Fucosterol	+	+	+	-	+	-	-	-	I
8	Fucoxanthin	+	-	-	-	+	-	+	-	I
9	Pheophytin A	+	-	+	-	+	-	-	-	III
10	Phytol	+	+	+	-	-	-	-	-	III
11	Saringosterol	+	-	+	-	-	-	-	-	I
12	Saringosterone	+	-	-	-	+	-	-	-	I
13	Stigmasterol	+	+	+	-	-	-	-	-	I
14	β-sitosterol	+	+	+	+	-	-	-	-	I

HIA: Human intestinal absorption, BBB: Blood brain barrier, HOB: Human oral bioavailability, p-gp: p-glycoprotein, AOT: Acute oral toxicity.

Table 5: Druglikeness prediction using osiris property explorer

S. No.	*Compounds*	*Toxicity risk*	*cLogP*	*solubility*	*Mol. Weight*	*TPSA*	*Drug-likeness*	*Drug-score*
1	Levomilnacipran	No	1.24	-2.3	246	46.33	8.08	0.94
2	Brassicasterol	No	7.15	-6.17	398	20.23	-1.2	0.19
3	Campesterol	No	7.4	-6.4	400	20.23	-8.19	0.14
4	Cholesterol	No	7.18	-6.24	386	20.23	-2.13	0.17
5	Docosahexaenoic acid	No	7.28	-4.49	328	37.3	-10.83	0.21
6	Eicosapentaenoic acid	No	6.62	-4.18	302	37.3	-14.26	0.24
7	Fucosterol	irritant	7.99	-6.41	412	20.23	-6.28	0.08
8	Fucoxanthin	no	9.51	-6.25	658.0	96.36	3.46	0.17
9	Pheophytin A	irritant	12.56	-10.3	870	121.9	-7.03	0.04
10	Phytol	No	7.42	-4.63	296	20.23	-3.77	0.21
11	Saringosterol	No	6.81	-6.61	428	40.46	-9.62	0.15
12	Saringosterone	tumorigenic, reproductive effective	6.95	-6.21	426	37.3	-9.06	0.07
13	Stigmasterol	No	7.6	-6.44	412	20.23	1.22	0.25
14	β-sitosterol	No	7.86	-6.67	414	20.23	-4.48	0.13

molecular weight and TPSA is good for all compounds. But optimization of molecules must be performed to enhance properties like druglikeness and drug-score.

Discussion

In this study, binding affinity of some compounds from marine algae was evaluated for human norepinephrine transporter. All of the selected compounds except fucoidan showed higher binding affinity to the receptor suggesting the reliability of study and can be initial step for selecting specific compounds for treatment of depression. Pharmacokinetic evaluation was also performed for these compounds. Selected compounds were better at ADMET properties but showed lower druglikeness and drug-score. Most of these algae derived compounds can cross blood brain barrier and are effectively interacting with the target. Compounds having lower druglikeness can be optimized to elevate these properties. Prior researches carried on marine algae were dose dependent studies and most of them were not bound to particular receptor. This type of *in-silico* work can be helpful in identifying the efficiency of compounds for the target and speed up the process of drug discovery. With the help of network pharmacology mechanism of these interactions can be studied more effectively. As well as these type of studies must be supported by animal and cellular experimental data to confirm the *in-vivo* behaviour of compounds. Marine algae have great variety of biologically active compounds. It can also be studied for other neurological disorders related to depression. Natural compounds have tendency to interact with more than one ligand. Hence, are effective in multiple target inhibition. Also, there is need to identify and study other targets so that multi-target drugs can be developed.

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