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Research Article

Artificial Intelligence and Machine Learning approach based *in-silico* ADME-Tox and Pharmacokinetic Profile of α-Linolenic acid from *Catharanthus roseus* (L.) G. Don.

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Abstract

Current craze and concomitant rise of Artificial Intelligence and Machine Learning (AI&ML) in the post-COVID-era holds significant contribution to Drug Design and Development. Along with IoT, AI&ML has reduced human interface and improved the Quality of Life though Quality-Health-Care products. AI&ML approaches driven Rational Drug Design along with customised molecular modelling techniques such as *in-silico* simulation, pharmacophore modelling, molecular dynamics, virtual screening, and molecular docking aims to elucidate unforeseen bioactivity of natural products confined to limited timeframe with at-most perfection. Besides, it also defines the molecular determinants that partake in the interface with in the drug and the target to design more proficient drug leads. α-Linolenic acid (ALA), a carboxylic acid with 18 carbons and three cis double bonds, is an essential fatty acid required for normal human health and can be acquired through regular dietary supplementation of food. During the metabolic process, ALA is bio-transformed into EPA and DHA. ALA decreases the risk of heart disease by maintaining normal heart rhythm and pumping. Studies suggest that ALA is associated with reduced risk of fatal ischemic heart disease further higher intake may reduce the risk of sudden death among prevalent myocardial infarction patients consistent with induced antiarrhythmic effect. It reduces blood clots, besides, cardiovascular-protective, anti-cancer, neuro-protective, anti-osteoporotic, anti-inflammatory, and anti-oxidative effects. However, data on pharmacological and toxicological aspects of ALA is limited; on the other hand, no serious adverse effects of ALA have been reported yet. In the present study AI&ML approach based *in-silico* ADME-Tox and pharmacokinetic profile of ALA from *Catharanthus roseus* is envisaged.

Keywords: IoT; AI&ML; ADME-Tox; α-Linolenic Acid (ALA); EPA; DHA Pharmacokinetics; *Catharanthus roseus*.

INTRODUCTION

The ripple effect of COVID-19 outbreak has brought major changes and challenges to worldwide healthcare systems[1]. Since the outbreak of novel coronavirus disease – COVID – 19, celebrity of pharmaceutical drug development as intractable hot area of research and development (R&D) in the environs of pharma-industries is on the raise. Drug development in the post COVID era is a high-risk - high-return business that guarantees huge profit as returns upon success, paradoxically the success rate is extremely low[2]. Drug development requires extensive R&D program that includes clinical trials and investment-cost-returns approach for the successful development of a single drug from the industry into the market[3]. For these reasons, pharmaceutical companies sought for alternative strategies to increase the probability of success in their drug development project[4]. One of the possible solutions is to exploit AI&ML approach that has undergone tremendous *ad-interim* exponential growth[5]. As of now, multi-national pharmaceutical companies are adopting data mining and AI&ML technologies to reduce time and cost required for R&D program[6].

Since antiquity, medicinal plants have been a valuable source of therapeutic agents, and even today most of the drugs available in the market are either obtained from plant based natural products or their derivative[7] biomolecules with therapeutic potential. Medicinal plants remain a vital storehouse for the discovery of novel drug leads[8-11]. Plant Based Natural Products (PBNPs) offer unique features in

comparison with their synthetic counterparts. PBNPs confer both advantages and challenges for the drug discovery process as they are characterized by structural scaffold diversity, functional specificity and molecular complexity[8]. In the past, pharmaceutical industry focused on libraries of synthetic compounds as a source for drug discovery[9] however, this cumbersome process has been given up for their unwanted side effects. On the other hand, PBNPs leads with GRAS standards are easy to produce for resupply with good compatibility on high throughput screening (HTS) platforms[8-11].

Catharanthus roseus (L) G. Don (Family – Apocynaceae), is native to Madagascar, but grown elsewhere as an ornamental plant in gardens, farms and landscape. In India, in Ayurvedic system of medicine, different parts of *C. roseus* have been reported for their use in the treatment of cancer, diabetes, stomach disorders, kidney, liver and cardiovascular diseases. Apart from India, this plant is used in traditional system of medicine in South Africa, China, Mexico and Malaysia, as remedy for diabetics[12,13].

Significance of *C. roseus* in modern system of medicine has gained prominence after the characterization of anticancer indole alkaloids - vincristine and vinblastine. As isolation and purification process of vincristine and vinblastine from the leaves is a time-consuming and costly affair due to the low content of these compounds, Mekky et al.[14] potentiated the biosynthesis of anticancer alkaloids vincristine and vinblastine in callus cultures of *C. roseus.* As of now, advanced and high-throughput separation/ analytical techniques have been used for isolation, purification, identification, characterization and quantitation of other alkaloids from the crude extract prepared from *C. roseus*[13,15].

Alkaloids of *C. roseus* possess hypotensive, sedative, tranquilizing, and anticancer properties[12]. *C. roseus* is recommended for the treatment of nose bleeding, gum bleeding, mouth ulcers, and sore throats, hypertension, cystitis, gastritis, enteritis, and diarrhea and memory loss[13]. Alkaloids including vinblastine, vincristine, vinorelbine, and vinflunine isolated from this plant have proven antitumor activity[15,16]. Apart from biomedical application *C. roseus* is exploited for its antibacterial, biopesticidal activities[17,18]. Recent phytochemical investigation has revealed a total of 344 compounds including monoterpene indole alkaloids (MIAs) (110), bis-indole alkaloids (35), flavonoids (34), phenolic acids (9) and volatile constituents (156) have been reported in the various extracts and fractions of different plant parts of *C. roseus*[13]. Similarly aerial parts of *C. roseus* contain vindoline, vindolidine, vindolicine, roseadine, leurosine-N′b-oxide, leurocolombine, catharanthamine, pleuroside, dimethylvinblastin, 5′-oxoleurosine, leurosidineN′b-oxide, vinorelbine, vinzolidine, vineamine, raubasin, 16-epi-19S-vindolinine, and vindolinine[16].

ALA (18:3n-3) is essential ω-3 fatty acid found in nuts[19]. It is necessary for normal growth and development thus an aspect of the human diet, probably because it is the main substrate for the synthesis of longer-chain fatty acids. ALA is the precursor of two long chain ω-3 fatty acids viz., EPA (eicosapentaenoic acid, 20:3n-5) and DHA (docosahexaenoic acid, 22:3n-6), both of them have vital roles in brain development, cardio-vascular health, inflammatory response, etc.[20]

The metabolic pathways of ALA have been reported by Fekete and Decsi, 2010[21]. During the metabolic process, ALA is bio-transformed into EPA and DHA. ALA is readily converted to EPA has been reported to be 8%, while conversion rates of ALA to DHA has been reported to be 4%, no direct link between DHA concentration and increase in rate of intake has been reported yet. Burdge et al.[22] reported that in humans, enzymes desaturase and elongase are involved in the bioconversion of ALA to EPA and DHA[22,23] respectively. Pawlosky et al.[24] reported that coefficient constant of EPA to DHA was about 4-fold higher in women than in men. Invariably, it has been reported that women have a higher concentration of DHA than men[25]. Further, high conversion rate of ALA to EPA/ DHA in women has been related to the level of estrogen[26]. In human system, ALA possesses hypo-cholesterolemic, nematicide, anti-arthritic, hepatoprotective anti-androgenic, hypo-cholesterolemic, 5-α reductase inhibitor, antihistaminic, anti-coronary, anti-eczemic, anti-acne properties[25]. Pharmacological studies show that ALA has the anti-metabolic syndrome, anticancer, anti-inflammatory, anti-oxidant, anti-obesity, neuro-protection properties[25-28]. Recently, it has been proved that ALA plays a major role in the functional regulation of gut microflora[28].

MATERIALS AND METHODS

Botanical Description of the plant

***Catharanthus roseus* (L) G. Don (Family – Apocynaceae)**

Habit: Suffrutex up to 1 m high, perennial, woody at the base, herbaceous above; **Stem:** glabrous or thinly pubescent; **Leaves:** opposite, obovate, oblong or oblanceolate, apex rounded or, rarely, sub-acute, apiculate, base cuneate, 4-8 cm long 1-3 cm broad, membranous to thinly coriaceous, glabrous or finely pubescent; **Petiole** 2-5 mm long; axillary glands forming a fringe, outer longer than inner; **Stipules:** absent; **Flowers:** axillary, solitary/ paired, subsessile, pink or white, or white with pink centre; **Calyx:** divided at base; sepals - 5, linear-subulate, 4-6 mm long, glabrous or pubescent; **Corolla:** salver-shaped; tube slender cylindrical, 2.3-2.6 cm long and 2-2.5 mm in dia, mouth constricted, thickened, pubescent; lobes broadly obovate, apiculate, 1.6-2.0 cm long; **Stamens:** 5, pentamerous, inserted near the mouth; anthers 2 mm long, subsessile. Disc replaced by 2 linear-subulate glands 2 mm long alternating with the carpels; **Ovary:** bicarpelary, 2 carpels, free; style filiform, stigma at the level of the anthers, capitate with a reflexed hyaline frill at the base; **Ovules:** numerous, 2-seriate. **Fruit:** two follicular mericarps erect, slightly spreading; follicles 2.5-3.5 cm long, cylindric, striate. **Seeds:** numerous, oblong, 2 mm long, black, rugose, grooved on one face; **Cotyledons:** flat, slightly shorter than the radicle; **Endosperm:** scanty; **Fl & Fr:** round the year.

Plant materials were collected from the College Campus, Identified and authenticated by Department of Botany, Government Arts College, Melur, Madurai, TamilNadu using Flora[29.30]. The plant material was shade dried, pulverized at room temperature, sieved and stored in vials until used. Dried

powder was dissolved in double distilled water and the aqueous leaf extract of *C. roseus* was analyzed for phytochemicals using standard protocols. GCMS analysis was performed as described previously[31-33].

ADMET predications

Phyto-components of AqLE of *C. roseus* were subjected to ADME prediction using QikProp (Schrödinger, LLC, NY) and toxicity prediction using TOPKAT (Accelrys, Inc., USA, 2015). QikProp develops and employs QSAR/QSPR models using partial least squares, principal component analysis and multiple linear regression to predict physicochemical significant descriptors and pharmaco-kinetically relevant properties that are essential for rational drug design. The computational toxicity was assessed using TOPKAT (TOxicity Prediction by Komputer Assisted Technology). TOPKAT calculates the toxicity on the basis of the quantitative structure-toxicity relationship (QSTR) model using linear regression on the structural descriptor and it considers 4 nearest neighbours with a similarity distance of <0.25 to assign probabilities of the toxicity such as carcinogenicity, mutagenicity, rat oral LD_{50}, skin irritation, aerobic biodegradability[34-38].

RESULTS

Isolation, purification and characterization of PBNPs (secondary metabolites) remains the key aspect of phytochemical screening[39-49]. In the present study ALA among the phyto-components of **AqLE** of *Catharanthus roseus* were ADMET predicted *in-silico.*

Chemical kingdom	:	Organic compounds
Super class	:	Lipids and lipid-like molecules
Class	:	Fatty Acyls
Subclass	:	Lineolic acids derivatives
PubChem Identifier	:	5280934
ChEBI Identifier	:	25048
CAS Identifier	:	28290-79-1
Synonyms	:	α-LINOLENIC ACID;
Canonical SMILES	:	CC/C=CC/C=CC/C=CCCCCCCCC(=O)O
InChI Key	:	DTOSIQBPPRVQHS-PDBXOOCHSA-N

Physicochemical Properties

Molecular weight of ALA was calculated as 278.44 g/mol; LogP value was predicted as 5.66; LogD value was predicted as 3.68; LogSw value was predicted as -4.78. Number of stereo-centers was predicted as 0; Stereo-chemical complexity was predicted as 0.000; Fsp3 was predicted as 0.611; Topological polar surface area was calculated as 37.30Å^2; Number of hydrogen bond donors was calculated as 1; Number of hydrogen bond acceptors was calculated as 1; Number of smallest set of smallest rings (SSSR) was calculated as 0; Size of the biggest system ring was calculated as 0; Number of rotatable bond was calculated as 13; Number of rigid bond was calculated as 4; Number of charged group was calculated as 1; Total charge of the compound was calculated as -1; Number of carbon atoms was ascertained as 18; Number of heteroatoms was ascertained as 2; Number of heavy atoms was ascertained as 20; Ratio between the number of non-carbon atoms and the number of carbon atoms was ascertained as 0.11 (Table 1). TPSA of ALA was calculated as 37.30; natoms in ALA was 20; MW of ALA was calculated as 278.44; nON was calculated as 2; nOHNH value was calculated as 1; nviolations value for ALA was calculated as 1; number of rotatable bonds in ALA was 13; and the theoretical volume of ALA was calculated as 306.47. The 3D structure of ALA is illustrated in Fig. 1.

Druggability Properties

In-silico studies are expected to reduce the risk of late-stage attrition of drug development and to optimize screening/ testing by looking at Druggability Properties, Lipinski's rule of 5 violations was predicted as 1; Veber rule was predicted as Good; Egan rule was predicted as Good; Oral PhysChem score (Traffic Lights) was predicted as 4; GSK's 4/400 score was predicted as Good; Pfizer's 3/75 score was predicted as BAD; Weighted quantitative estimate of drug-likeness (QEDw) score was predicted as 0.31; Solubility of EA was predicted as 2342.23; Solubility Forecast Index of EA was predicted as Good (Table 2).

ADMET Properties

Experimental evaluation of small-molecule is both time-consuming and expensive. On the other hand evolution of computational approaches to optimize pharmacokinetic and toxicity properties is said to drive the progression of drug discovery. Prediction of ADMET-associated properties of new chemicals, however, is a challenging task with only tenuous links between many physicochemical characteristics and pharmacokinetic and toxicity properties. This has led to a need for novel approaches to understand, explore, and predict ADMET properties of small molecules as a way to improve compound quality and success rate. ADMET prediction models, including performance measures for the selected candidate molecule ALA were performed online (Table 3).

Human Intestinal Absorption (HIA+) for ALA had a calculated probability value of 0.990; Blood Brain Barrier (BBB+) had a calculated probability value of 0.931; Caco-2 permeable (Caco2+) for ALA had a calculated probability value of 0.774; P-glycoprotein substrate that served as Non-substrate for ALA had a calculated probability value of 0.677; P-glycoprotein inhibitor I that served as Non-inhibitor for ALA had a calculated probability value of 0.950; P-glycoprotein inhibitor II for ALA served as Non-inhibitor and had a predicted probability value of 0.903 (Table 3).

CYP450 2C9 substrate for ALA served as Non-substrate with a predicted probability value of 0.774; CYP450 2D6 substrate for ALA served as Non-substrate with a predicted probability value of 0.908; CYP450 3A4 substrate for ALA served as Non-substrate with a predicted probability value of 0.688; CYP450 1A2 inhibitor for ALA worked as Inhibitor with a predicted probability value of 0.692; CYP450 2C9 inhibitor for ALA functioned as Non-inhibitor with a predicted probability value of 0.880; CYP450 2D6 inhibitor for ALA served as Non-inhibitor with a predicted probability value of 0.963; CYP450 2C19 inhibitor for ALA remained as Non-inhibitor with a predicted probability value of 0.964; CYP450 3A4 inhibitor for ALA was as Non-inhibitor with a predicted probability value of 0.947; CYP450 inhibitory promiscuity for ALA had Low CYP Inhibitory Promiscuity with a predicted probability value of 0.943 respectively (Table 3).

ADMET Ames test for ALA served as Non AMES toxic with a predicted probability value of 0.913; Carcinogenicity for ALA served as Non-carcinogens with a predicted probability value of 0.650; Biodegradation potential for ALA served as Ready biodegradable with a predicted probability value of 0.781; Rat acute toxicity 1.450 LD_{50}, mol/kg for ALA was Not applicable; hERG inhibition (predictor I) for ALA served as Weak inhibitor with a predicted probability value of 0.882; hERG inhibition

(predictor II) for ALA served as Non-inhibitor with a predicted probability value of 0.932 respectively (Table 3). In the present study 15 models covered a diverse set of ADMET endpoints including Maximum Recommended Therapeutic Dose (MRTD), chemical mutagenicity, human liver microsomal (HLM), Pgp inhibitor/ substrates. ADMET data for performance measures of vNN models in 10-fold cross validation using a restricted/ unrestricted applicability domain is given in Table 4a,b.

Liver Toxicity - DILI

Drug-induced liver injury (DILI) is one of the most commonly cited reasons for drug withdrawals from the market. This application predicts whether a compound could cause DILI. A dataset of 1,431 compounds was obtained from online sources. The dataset contained both pharmaceuticals and non-pharmaceuticals compounds; compounds were classified as **causing DILI** if it was associated with a high risk and **non DILI** if there was no such risk with the compound.

Cytotoxicity (HepG2)

Cytotoxicity is the degree to which a chemical causes damage to cells. A cytotoxicity prediction model was developed using *in-vitro* data on toxicity against HepG2 cells for 6,000 structurally diverse compounds, collected from ChEMBL. In developing model, compounds with $IC_{50} \leq 10$ µM in *in-vitro* assay as cytotoxic was considered.

Metabolism - HLM

Human Liver Microsomal (HLM) stability assay is commonly used to identify and exclude compounds that are too rapidly metabolized. For a drug to achieve effective therapeutic concentrations in the body, it cannot be metabolized too rapidly by the liver. Compounds with a half-life of 30 min or longer in an HLM assay were considered as stable; otherwise considered unstable. HLM data was retrieved from ChEMBL database, manually curated and classified as stable or unstable based on the reported half-life ($T_{1/2} > 30$ min was considered stable, and $T_{1/2} < 30$ min unstable). The final dataset contained 3,654 compounds. Of these, 2,313 compounds were stable and 1,341 were unstable.

Metabolism - Cytochrome P450 enzyme (CYP) inhibition

CYPs constitute a superfamily of proteins that play an important role in the metabolism and detoxification of xenobiotics. *In-vitro* data was derived from five main drug-metabolizing CYPs - 1A2, 3A4, 2D6, 2C9, and 2C19 was used to develop CYP inhibition models. CYP inhibitors were retrieved from PubChem and classified, a compound with an $IC_{50} \leq 10$ µM for an enzyme as an inhibitor of the enzyme. Prediction values for CYP1A2, CYP3A4, CYP2D6, CYP2C9, and CYP2C19 is given in Table 4.

Membrane Transporters - BBB

The blood-brain barrier (BBB) is a highly selective barrier that separates the circulating blood from the central nervous system. A vNN-based BBB model was developed, using 352 compounds, their corresponding BBB permeability values (logBB) were obtained from online sources. The compounds were further classified with logBB values of less than –0.3 and greater than +0.3 as BBB non-permeable and permeable respectively.

Membrane Transporters - Pgp Substrates and Inhibitors

P-glycoprotein (Pgp) is an essential cell membrane protein that extracts many foreign substances from the cell. Cancer cells often overexpress Pgp, which increases the efflux of chemotherapeutic agents from the cell and prevents treatment by reducing the effective intracellular concentrations of such agents through a phenomenon known as multidrug resistance. For this reason, identifying compounds that can either be transported out of the cell by Pgp (substrates) or impair Pgp function (inhibitors) is of great interest. In the present study, models were developed to predict both Pgp substrates and Pgp inhibitors. The dataset contained 422 substrates and 400 non-substrates. To generate a large Pgp inhibitor dataset, two datasets were combined and duplicates were removed to form a combined dataset consisting of a training set of 1,319 inhibitors and 937 non-inhibitors. Results are given in Table 4.

hERG (Cardiotoxicity)

Human ether-à-go-go-related gene (hERG) codes for a potassium ion channel involved in the normal cardiac repolarization activity of the heart. Drug-induced blockade of hERG function can cause long term QT syndrome, which may result in arrhythmia which may ultimately lead to death. A data set of 282 known hERG blockers were retrieved from the literature and classified compounds with an IC_{50} cut-off value of 10 µM or less as blockers. A set of 404 compounds with IC_{50} values greater than 10 µM were collected from ChEMBL and classified as non-blockers. Results are provided in Table 4.

MMP (Mitochondrial Toxicity)

Given the fundamental role of mitochondria in cellular energetics and oxidative stress, mitochondrial dysfunction has been implicated in cancer, diabetes, neurodegenerative disorders, and cardiovascular diseases. In the present study a largest dataset of chemical-induced changes in mitochondrial membrane potential (MMP) were used based on the assumption that a compound that causes mitochondrial dysfunction is also likely to reduce the MMP. vNN based MMP prediction model was developed using 6,261 compounds collected from a screened library of 10,000 compounds at 15 concentrations, each in triplicate, to measure changes in the MMP in HepG2 cells. Data indicate that 913 compounds decreased the MMP, whereas 5,395 compounds had no or insignificant effect (Table 4).

Mutagenicity (AMES Test)

Mutagens are chemicals that cause abnormal genetic mutations leading to cancer. A common way to assess a chemical's mutagenicity is the Ames test. In the present study, a prediction model was developed using a dataset of 6,512 compounds. Data indicate that 3,503 compounds were Ames-positive.

Maximum Recommended Therapeutic Dose (MRTD)

MRTD is an estimated upper daily dose that is safe. In the present study a prediction model was built based on a dataset of MRTD values publically disclosed by the FDA, mostly of single-day oral doses for an average adult with a body weight of 60 kg, for 1,220 compounds (small organic drugs) however organometallics, high-molecular weight polymers (>5,000 Da), nonorganic chemicals, mixtures of chemicals, and very small molecules (<100 Da) were excluded. An external test set of 160 compounds were used that were collected by the FDA for validation. The total dataset for the model contained 1,185 compounds. The predicted MRTD value is reported in mg/day unit based upon an average adult weighing 60 kg (Table 4).

Biological properties - G-PCRs (GPCRs)

GPCRs are the largest family of signalling proteins. Structurally, GPCRs are similar: extracellular N-terminus, seven membrane-spanning α-helices (TM), and intracellular C-terminus, with variable extracellular and intracellular

elements. These cell surface receptors act like an inbox for messages in the form of light energy, peptides, lipids, sugars, and proteins. Calculated distribution of activity scores (version 2011.06) for GPCR ligands for the molecule was 0.33; kinase inhibitors, ion channel modulators, nuclear receptor ligands, protease inhibitors and other enzyme targets compared with scores for about 100'000 average drug-like molecules. The calculated value for Ion channel modulator (0.23); Kinase inhibitor (-0.19); Nuclear receptor ligand (0.35); Protease inhibitor (0.13); Enzyme inhibitor (0.42) respectively, the score allows efficient separation of active and inactive molecules. Further, cytoscape network of predicted human targets of ALA- human target proteins were predicted using STITCH (26590256), a database of chemical-protein interaction networks is provided in Fig 2. Further, predicted bioactivity target classes for ALA from *Catharanthus roseus* with probability score provided in Fig. 3.

pkCSM - pharmacokinetic properties of ALA using graph-based signatures

Drug development is a fine balance of optimizing drug like properties to maximize efficacy, safety, and pharmacokinetics. Many early stage drug discovery programs focus on identifying molecules that bind to a target of interest. While potency is a driving factor in these early stages, ultimately the pharmacokinetic and toxicity properties dictate whether it will ever advance its effectiveness and success therapeutically. Mathematical calculation and graph-theory based Graph modelling is an intuitive and well established mathematical representation of chemical entities, from where the descriptors encompassing both molecule structure and chemistry can be extracted for rational drug design. The pharmacokinetic properties of ALA predicted using graph-based signatures is given in Table 5.

DISCUSSION

Absorption: There is very limited information on the absorption of ALA in the human gut. However, absorption of ALA in humans is assumed to be efficient. Absorption can be determined through the difference between intake levels of ALA in foods and excretion in the feces. Absorption efficiency of ALA through the human gut and carrier-mediated transporters involved in the absorption is assumed to be quite high[50].

Distribution: Available information on the distribution of ALA is limited. Lin and Salem[51], reported whole body distribution of ALA in rats. Through inter conversion of EPA and DHA, DHA was deposited in brain, spinal cord, heart, testes, and eye over time. About 16–18% of ALA was deposited in adipose tissue, skin, and muscle. About 6% of ALA was elongated and desaturated, and stored, in muscle, adipose tissue, and carcass. Remaining 78% of ALA was eventually excreted.

Metabolism: Metabolic conversion of ALA to EPA and DHA is relatively poor because ALA absorbed in the system undergoes b-oxidation[52]. Although 67% of ALA undergoes b-oxidation in brain, only 30% of fatty acids, such as arachidonic acid, undergo b-oxidation in brain[53]. In the brain, small percentage of fatty acids undergoes b-oxidation. However, the rapidity of the process is difficult to measure owing to the pace of lipid metabolism occurs within seconds/ minutes, rather than hours.

Toxicity: It has been suggested that EDF containing enriched ALA is safe when orally administered to rats. Furthermore, EDF could reduce the increase of triglyceride levels in plasma. Prospective meta-analysis studies concluded that there exists no association between dietary intake of ALA and prostate cancer risk[54]. Therefore, overall evidence of prostate cancer risk with ALA remains inconclusive. Association of ALA with the risk of macular degeneration has been reported[55], however, more research is required before any conclusion is drawn. Flaxseed/ oil is rich dietary sources of ALA is prospected to induce adverse gastrointestinal effects, such as flatulence, bloating and stomach aches/cramps[56]. Further it has been pointed out that ALA can induce lipid peroxidation when exposed to UV radiation, which may produce have adverse effects if not monitored[57]. ALA is well-known for its anti-inflammatory activity. Recently, it has been pointed out that ALA rich diet influences microbiota composition and villus morphology of the mouse small intestine.

CONCLUSION

ALA from AqLE of *C. roseus* was screened and ADMET predicted for the functional properties. It has been well established that in the human body, ALA is converted to EPA and DHA, which is protective against cardiovascular, neuronal, osteoporotic inflammatory diseases. In addition, EPA and DHA lower the blood cholesterol level that reduces the risk of heart disease. However, the conversion rates of ALA to EPA/ DHA is very low. With limited toxicological data, it is concluded that ALA is safe as a dietary ingredient because it doesn't produce serious health problems, this essential fatty acid could be used as nutraceutical and pharmacological food ingredient. However, overall evidence on the association of ALA with risks remains inconclusive at this point of time. The data and mathematical calculation based *in-silico* predication models presented in the paper is hopefully is expected to facilitate the drug development process by enabling the rapid design, evaluation, and prioritization of ALA owing to its overwhelming biomedical applications.

REFERENCES

1. Ayati N, Saiyarsarai P, Nikfar S. Short and long term impacts of COVID-19 on the pharmaceutical sector. DARU Journal of Pharmaceutical Sciences 2020; 28(2):799-805. https://doi.org/10.1007/s40199-020-00358-5

2. Won JH, Lee H. Can the COVID-19 pandemic disrupt the current drug development practices? International Journal of Molecular Sciences 2021; 22(11):5457. https://doi.org/10.3390/ijms22115457

3. Tamimi NA, Ellis P. Drug development: from concept to marketing!. Nephron Clinical Practice 2009; 113(3):c125-31. https://doi.org/10.1159/000232592

4. Kiriiri GK, Njogu PM, Mwangi AN. Exploring different approaches to improve the success of drug discovery and development projects: a review. Future Journal of Pharmaceutical Sciences 2020; 6(1):1-2. https://doi.org/10.1186/s43094-020-00047-9

5. Kim H, Kim E, Lee I, Bae B, Park M, Nam H. Artificial intelligence in drug discovery: a comprehensive review of data-driven and machine learning approaches. Biotechnology and Bioprocess Engineering 2020; 25(6):895-930. https://doi.org/10.1007/s12257-020-0049-y

6. Gupta R, Srivastava D, Sahu M, Tiwari S, Ambasta RK, Kumar P. Artificial intelligence to deep learning: machine intelligence approach for drug discovery Molecular diversity 2021; 25(3):1315-60. https://doi.org/10.1007/s11030-021-10217-3

7. Balunas MJ, Kinghorn AD. Drug discovery from medicinal plants Life sciences 2005; 78(5):431-41. https://doi.org/10.1016/j.lfs.2005.09.012

8. Atanasov AG, Zotchev SB, Dirsch VM, Supuran CT. Natural products in drug discovery: advances and opportunities. Nature Reviews Drug Discovery 2021; 20(3):200-16. https://doi.org/10.1038/s41573-020-00114-z

9. Atanasov AG, Waltenberger B, Pferschy-Wenzig EM, Linder T, Wawrosch C, Uhrin P, Temml V, Wang L, Schwaiger S, Heiss EH, Rollinger JM. Discovery and resupply of pharmacologically active plant-derived natural products: A review. Biotechnology advances 2015; 33(8):1582-614. https://doi.org/10.1016/j.biotechadv.2015.08.001

10. Newman DJ, Cragg GM. Natural products as sources of new drugs over the nearly four decades from 01/1981 to 09/2019. Journal of Natural Products. 2020; 83(3):770-803. https://doi.org/10.1021/acs.jnatprod.9b01285

11. Dzobo K. The Role of Natural Products as Sources of Therapeutic Agents for Innovative Drug Discovery. Reference Module in Biomedical Sciences. 2021; B978-0-12-820472-6.00041-4.

12. Das S, Sharangi AB. Madagascar periwinkle (Catharanthus roseus L.): Diverse medicinal and therapeutic benefits to humankind. Journal of Pharmacognosy and Phytochemistry. 2017; 6(5):1695-701.

13. Kumar S, Singh B, Singh R. Catharanthus roseus (L.) G. Don: A review of its ethnobotany, phytochemistry, ethnopharmacology and toxicities. Journal of Ethnopharmacology. 2022; 284:114647. https://doi.org/10.1016/j.jep.2021.114647

14. Mekky H, Al-Sabahi J, Abdel-Kreem MF. Potentiating biosynthesis of the anticancer alkaloids vincristine and vinblastine in callus cultures of Catharanthus roseus. South African Journal of Botany. 2018; 114:29-31. https://doi.org/10.1016/j.sajb.2017.10.008

15. Tiong SH, Looi CY, Hazni H, Arya A, Paydar M, Wong WF, Cheah SC, Mustafa MR, Awang K. Antidiabetic and antioxidant properties of alkaloids from Catharanthus roseus (L.) G. Don. Molecules. 2013; 18(8):9770-84. https://doi.org/10.3390/molecules18089770

16. Singh B, Sharma RA. Secondary Metabolites of Medicinal Plants, 4 Vol Set: Ethnopharmacological Properties, Biological Activity and Production Strategies. John Wiley & Sons; 2020. https://doi.org/10.1002/9783527825578

17. Ramya S. In Vitro Evaluation of Antibacterial Activity Using Crude Extracts of Catharanthus roseus L. (G.) Don. Ethnobotanical Leaflets. 2008; 2008(1):140.

18. Ramya S. Biopesticidal effect of leaf extracts of Catharanthus roseus L (G) Don. on the larvae of gram pod borer-Helicoverpa armígera (Hübner). Ethnobotanical Leaflets. 2008; 2008(1):145.

19. Yuan Q, Xie F, Huang W, Hu M, Yan Q, Chen Z, Zheng Y, Liu L. The review of alpha-linolenic acid: Sources, metabolism, and pharmacology. Phytotherapy Research. 2022; 36(1):164-88. https://doi.org/10.1002/ptr.7295

20. Allaire J, Couture P, Leclerc M, Charest A, Marin J, Lépine MC, Talbot D, Tchernof A, Lamarche B. A randomized, crossover, head-to-head comparison of eicosapentaenoic acid and docosahexaenoic acid supplementation to reduce inflammation markers in men and women: the Comparing EPA to DHA (ComparED) Study. The American journal of clinical nutrition. 2016; 104(2):280-7. https://doi.org/10.3945/ajcn.116.131896

21. Fekete K, Decsi T. Long-chain polyunsaturated fatty acids in inborn errors of metabolism. Nutrients. 2010; 2(9):965-74. https://doi.org/10.3390/nu2090965

22. Burdge GC, Calder PC. Conversion of α-linolenic acid to longer-chain polyunsaturated fatty acids in human adults. Reproduction Nutrition Development. 2005; 45(5):581-97. https://doi.org/10.1051/rnd:2005047

23. Zhang JY, Kothapalli KS, Brenna JT. Desaturase and elongase limiting endogenous long chain polyunsaturated fatty acid biosynthesis. Current opinion in clinical nutrition and metabolic care. 2016; 19(2):103. https://doi.org/10.1097/MCO.0000000000000254

24. Pawlosky R, Hibbeln J, Lin Y, Salem N. n-3 fatty acid metabolism in women. British Journal of Nutrition. 2003; 90(5):993-4. https://doi.org/10.1079/BJN2003985

25. Domenichiello AF, Kitson AP, Bazinet RP. Is docosahexaenoic acid synthesis from α-linolenic acid sufficient to supply the adult brain? Progress in lipid research. 2015; 59:54-66. https://doi.org/10.1016/j.plipres.2015.04.002

26. Kitson AP, Stroud CK, Stark KD. Elevated production of docosahexaenoic acid in females: potential molecular mechanisms. Lipids. 2010; 45(3):209-24. https://doi.org/10.1007/s11745-010-3391-6

27. Samanta S. Potential bioactive components and health promotional benefits of tea (Camellia sinensis) Journal of American College of Nutrition. 2020:1-29. https://doi.org/10.1080/07315724.2020.1827082

28. Todorov H, Kollar B, Bayer F, Brandão I, Mann A, Mohr J, Pontarollo G, Formes H, Stauber R, Kittner JM, Endres K. α-Linolenic acid-rich diet influences microbiota composition and villus morphology of the mouse small intestine. Nutrients. 2020; 12(3):732. https://doi.org/10.3390/nu12030732

29. Matthew KM. Flora of the Tamilnadu Carnatic. The Rapinat Herbarium, St. Joseph's College, Tiruchirapalli, India; 198117.

30. Gamble JS, Fischer CE. Flora of the Presidency of Madras. London, UK: West, Newman and Adlard; 1915. https://doi.org/10.5962/bhl.title.21628

31. Kandeepan C, Sabitha M, Parvathi K, Senthilkumar N, Ramya S, Boopathi NM, Jayakumararaj R. Phytochemical Screening, GCMS Profile, and In-silico properties of Bioactive Compounds in Methanolic Leaf Extracts of Moringa oleifera. Journal of Drug Delivery and Therapeutics. 2022; 12(2):87-99. https://doi.org/10.22270/jddt.v12i2.5250

32. Soorya C, Balamurugan S, Basha AN, Kandeepan C, Ramya S, Jayakumararaj R. Profile of Bioactive Phyto-compounds in Essential Oil of Cymbopogon martinii from Palani Hills, Western Ghats, India. JDDT 2021; 11(4):60-65 https://doi.org/10.22270/jddt.v11i4.4887

33. Loganathan T, Barathinivas A, Soorya C, Balamurugan S, Nagajothi TG, Jayakumararaj R. GCMS Profile of Bioactive Secondary Metabolites with Therapeutic Potential in the Ethanolic Leaf Extracts of Azadirachta indica: A Sacred Traditional Medicinal Plant of India. JDDT 2021; 11(4-S):119-126. https://doi.org/10.22270/jddt.v11i4-S.4967

34. Soorya C, Balamurugan S, Ramya S, Neethirajan K, Kandeepan C, Jayakumararaj R. Physicochemical, ADMET and Druggable properties of Myricetin: A Key Flavonoid in Syzygium cumini that regulates metabolic inflammations. Journal of Drug Delivery and Therapeutics. 2021; 11(4):66-73. https://doi.org/10.22270/jddt.v11i4.4890

35. Loganathan T, Barathinivas A, Soorya C, Balamurugan S, Nagajothi TG, Ramya S, Jayakumararaj R. Physicochemical, Druggable, ADMET Pharmacoinformatics and Therapeutic Potentials of Azadirachtin-a Prenol Lipid (Triterpenoid) from Seed Oil Extracts of Azadirachta indica A. Juss. Journal of Drug Delivery and Therapeutics. 2021; 11(5):33-46. https://doi.org/10.22270/jddt.v11i5.4981

36. Kandeepan C, Kalaimathi RV, Jeevalatha A, Basha AN, Ramya S, Jayakumararaj R. In-silico ADMET Pharmacoinformatics of Geraniol (3, 7-dimethylocta-trans-2, 6-dien-1-ol)-acyclic monoterpene alcohol drug from Leaf Essential Oil of Cymbopogon martinii from Sirumalai Hills (Eastern Ghats), India. Journal of Drug Delivery and Therapeutics. 2021; 11(4):109-118. https://doi.org/10.22270/jddt.v11i4-S.4965

37. Krishnaveni K, Sabitha M, Murugan M, Kandeepan C, Ramya S, Loganathan T, Jayakumararaj R. vNN model cross validation towards Accuracy, Sensitivity, Specificity and kappa performance measures of β-caryophyllene using a restricted-unrestricted applicability domain on Artificial Intelligence & Machine Learning approach based in-silico prediction. Journal of Drug Delivery and Therapeutics. 2022; 12(1-S):123-131. https://doi.org/10.22270/jddt.v12i1-S.5222

38. Kalaimathi RV, Jeevalatha A, Basha AN, Kandeepan C, Ramya S, Loganathan T, Jayakumararaj R. In-silico Absorption, Distribution, Metabolism, Elimination and Toxicity profile of Isopulegol from

Rosmarinus officinalis. Journal of Drug Delivery and Therapeutics. 2022; 12(1):102-8. https://doi.org/10.22270/jddt.v12i1.5188

39. Jeevalatha A, Kalaimathi RV, Basha AN, Kandeepan C, Ramya S, Loganathan T, Jayakumararaj R. Profile of bioactive compounds in Rosmarinus officinalis. Journal of Drug Delivery and Therapeutics. 2022; 12(1):114-122. https://doi.org/10.22270/jddt.v12i1.5189

40. Rajasekaran C, Meignanam E, Vijayakumar V, Kalaivani T, Ramya S, Premkumar N, Siva R, Jayakumararaj R Investigations on antibacterial activity of leaf extracts of Azadirachta indica A. Juss (Meliaceae): a traditional medicinal plant of India. Ethnobotanical leaflets. 2008; 2008(1):161

41. Ramya S, Alaguchamy N, Maruthappan VM, Sivaperumal R, Sivalingam M, Krishnan A, Govindaraji V, Kannan K, Jayakumararaj R. Wound healing ethnomedicinal plants popular among the Malayali tribes in Vattal Hills, Dharmapuri, TN, India. Ethnobotanical Leaflets. 2009; 2009(10):6.

42. Ramya S, Jepachanderamohan PJ, Kalayanasundaram M, Jayakumararaj R. In vitro antibacterial prospective of crude leaf extracts of Melia azedarach Linn. against selected bacterial strains. Ethnobotanical Leaflets. 2009; 2009(1):32

43. Ramya S, Krishnasamy G, Jayakumararaj R, Periathambi N, Devaraj A. Bioprospecting Solanum nigrum Linn.(Solanaceae) as a potential source of Anti-Microbial agents against selected Bacterial strains. Asian Journal of Biomedical and Pharmaceutical Sciences. 2012; 2(12):65

44. Ramya S, Murugan M, Krishnaveni K, Sabitha M, Kandeepan C, Jayakumararaj R. In-silico ADMET profile of Ellagic Acid from Syzygium cumini: A Natural Biaryl Polyphenol with Therapeutic Potential to Overcome Diabetic Associated Vascular Complications. Journal of Drug Delivery and Therapeutics. 2022; 12(1):91-101 https://doi.org/10.22270/jddt.v12i1.5179

45. Ramya S, Neethirajan K, Jayakumararaj R. Profile of bioactive compounds in Syzygium cumini-a review. Journal of Pharmacy research. 2012; 5(8):4548-53

46. Sabitha M, Krishnaveni K, Murugan M, Basha AN, Pallan GA, Kandeepan C, Ramya S, Jayakumararaj R. In-silico ADMET predicated Pharmacoinformatics of Quercetin-3-Galactoside, polyphenolic compound from Azadirachta indica, a sacred tree from Hill Temple in Alagarkovil Reserve Forest, Eastern Ghats, INDIA. Journal of Drug Delivery and Therapeutics. 2021; 11(5-S):77-84 https://doi.org/10.22270/jddt.v11i5-S.5026

47. Shanmugam S, Sundari A, Muneeswaran S, Vasanth C, Jayakumararaj R, Rajendran K. Ethnobotanical Indices on medicinal plants used to treat poisonous bites in Thiruppuvanam region of Sivagangai district in Tamil Nadu, India. Journal of Drug Delivery and Therapeutics. 2020; 10(6-s):31-6 https://doi.org/10.22270/jddt.v10i6-s.4432

48. Sundari A, Jayakumararaj R. Herbal remedies used to treat skin disorders in Arasankulam region of Thoothukudi District in Tamil Nadu, India. Journal of Drug Delivery and Therapeutics. 2020; 10(5):33-8 https://doi.org/10.22270/jddt.v10i5.4277

49. Sundari A, Jayakumararaj R. Medicinal plants used to cure cuts and wounds in Athur region of Thoothukudi district in Tamil Nadu, India. Journal of Drug Delivery and Therapeutics. 2020; 10(6-s):26-30. https://doi.org/10.22270/jddt.v10i6-s.4429

50. Kim KB, Nam YA, Kim HS, Hayes AW, Lee BM. α-Linolenic acid: Nutraceutical, pharmacological and toxicological evaluation. Food and chemical toxicology. 2014; 70:163-78. https://doi.org/10.1016/j.fct.2014.05.009

51. Lin YH, Salem N. Whole body distribution of deuterated linoleic and α-linolenic acids and their metabolites in the rat. Journal of Lipid Research. 2007; 48(12):2709-24. https://doi.org/10.1194/jlr.M700369-JLR200

52. Poumès-Ballihaut C, Langelier B, Houlier F, Alessandri JM, Durand G, Latge C, Guesnet P. Comparative bioavailability of dietary α-linolenic and docosahexaenoic acids in the growing rat. Lipids. 2001; 36(8):793-800. https://doi.org/10.1007/s11745-001-0786-5

53. Barceló-Coblijn G, Murphy EJ. Alpha-linolenic acid and its conversion to longer chain n− 3 fatty acids: Benefits for human health and a role in maintaining tissue n− 3 fatty acid levels. Progress in lipid research. 2009; 48(6):355-74. https://doi.org/10.1016/j.plipres.2009.07.002

54. Carleton AJ, Sievenpiper JL, de Souza R, McKeown-Eyssen G, Jenkins DJ. Case-control and prospective studies of dietary α-linolenic acid intake and prostate cancer risk: a meta-analysis. BMJ open. 2013; 3(5):e002280. https://doi.org/10.1136/bmjopen-2012-002280

55. Seddon JM, Rosner B, Sperduto RD, Yannuzzi L, Haller JA, Blair NP, Willett W. Dietary fat and risk for advanced age-related macular degeneration. Archives of ophthalmology. 2001; 119(8):1191-9. https://doi.org/10.1001/archopht.119.8.1191

56. Austria JA, Richard MN, Chahine MN, Edel AL, Malcolmson LJ, Dupasquier CM, Pierce GN. Bioavailability of alpha-linolenic acid in subjects after ingestion of three different forms of flaxseed. Journal of the American College of Nutrition. 2008; 27(2):214-21. https://doi.org/10.1080/07315724.2008.10719693

57. Regensburger J, Knak A, Maisch T, Landthaler M, Bäumler W. Fatty acids and vitamins generate singlet oxygen under UVB irradiation. Experimental dermatology. 2012; 21(2):135-9. https://doi.org/10.1111/j.1600-0625.2011.01414.x

Table 1: Physicochemical Properties of Linolenic acid from *Catharanthus roseus*

PROPERTY	VALUE
Molecular weight	278.44 g/mol
LogP	5.66
LogD	3.68
LogSw	-4.78
Number of stereocenters	0
Stereochemical complexity	0.000
Fsp3	0.611
Topological polar surface area	37.30 $Å^2$
Number of hydrogen bond donors	1
Number of hydrogen bond acceptors	1
Number of smallest set of smallest rings (SSSR)	0
Size of the biggest system ring	0
Number of rotatable bonds	13
Number of rigid bonds	4
Number of charged groups	1
Total charge of the compound	-1
Number of carbon atoms	18
Number of heteroatoms	2
Number of heavy atoms	20
Ratio between the number of non-carbon atoms and the number of carbon atoms	0.11

Physicochemical properties were computed using FAF-Drugs4 (28961788) and RDKit open-source cheminformatics platform

Table 2: Druggability Properties of Linolenic acid from *Catharanthus roseus*

PROPERTY	VALUE
Lipinski's rule of 5 violations	1
Veber rule	Good
Egan rule	Good
Oral PhysChem score (Traffic Lights)	4
GSK's 4/400 score	Good
Pfizer's 3/75 score	Bad
Weighted quantitative estimate of drug-likeness (QEDw) score	0.31
Solubility	2342.23
Solubility Forecast Index	Good Solubility

Druggabiity scoring schemes were computed using FAF-Drugs4 (28961788) and FAF-QED (28961788) open-source cheminformatics platform

Table 3: ADMET Properties of Linolenic acid from *Catharanthus roseus*

PROPERTY	VALUE	PROBABILITY
Human Intestinal Absorption	HIA+	0.990
Blood Brain Barrier	BBB+	0.931
Caco-2 permeable	Caco2+	0.774
P-glycoprotein substrate	Non-substrate	0.677
P-glycoprotein inhibitor I	Non-inhibitor	0.950
P-glycoprotein inhibitor II	Non-inhibitor	0.903
CYP450 2C9 substrate	Non-substrate	0.774
CYP450 2D6 substrate	Non-substrate	0.908
CYP450 3A4 substrate	Non-substrate	0.688
CYP450 1A2 inhibitor	Inhibitor	0.692
CYP450 2C9 inhibitor	Non-inhibitor	0.880
CYP450 2D6 inhibitor	Non-inhibitor	0.963
CYP450 2C19 inhibitor	Non-inhibitor	0.964
CYP450 3A4 inhibitor	Non-inhibitor	0.947
CYP450 inhibitory promiscuity	Low CYP Inhibitory Promiscuity	0.943
Ames test	Non AMES toxic	0.913
Carcinogenicity	Non-carcinogens	0.650
Biodegradation	Ready biodegradable	0.781
Rat acute toxicity	1.450 LD50, mol/kg	NA
hERG inhibition (predictor I)	Weak inhibitor	0.882
hERG inhibition (predictor II)	Non-inhibitor	0.932

ADMET features were predicted using admetSAR (23092397) open-source tool.

Table 4a: ADMET Predictions for Linolenic acid from *Catharanthus roseus* results based on restricted/ unrestricted applicability domain

Query	Liver Toxicity		Metabolism						Membrane Transporters			Others			
			Cyp Inhibitors for												
	DILI	Cyto-toxicity	HLM	1A2	3A4	2D6	2C9	2C19	BBB	P-gp Inhibitor	P-gp Substrate	hERG Blocker	MMP	AMES	MRTD (mg/day)
	No	Yes	Yes	No	No	No	No	No	Yes	No	No	No	No	No	0.16

Table 4b: Performance measures of vNN models in 10-fold cross validation using a restricted or unrestricted applicability domain

Model	Data[a]	d_0[b]	h[c]	Accuracy	Sensitivity	Specificity	*kappa*	R[d]	Coverage
DILI	1427	0.60	0.50	0.71	0.70	0.73	0.42	0.00	0.66
		1.00	0.20	0.67	0.62	0.72	0.34	0.00	1.00
Cytotox (hep2g)	6097	0.40	0.20	0.84	0.88	0.76	0.64	0.00	0.89
		1.00	0.20	0.84	0.73	0.89	0.62	0.00	1.00
HLM	3219	0.40	0.20	0.81	0.72	0.87	0.59	0.00	0.91
		1.00	0.20	0.81	0.70	0.87	0.57	0.00	1.00
CYP1A2	7558	0.50	0.20	0.90	0.70	0.95	0.66	0.00	0.75
		1.00	0.20	0.89	0.61	0.95	0.60	0.00	1.00
CYP2C9	8072	0.50	0.20	0.91	0.55	0.96	0.54	0.00	0.76
		1.00	0.20	0.90	0.44	0.96	0.46	0.00	1.00
CYP2C19	8155	0.55	0.20	0.87	0.64	0.93	0.58	0.00	0.76
		1.00	0.20	0.86	0.52	0.94	0.50	0.00	1.00
CYP2D6	7805	0.50	0.20	0.89	0.61	0.94	0.57	0.00	0.75
		1.00	0.20	0.88	0.52	0.95	0.51	0.00	1.00
CYP3A4	10373	0.50	0.20	0.88	0.76	0.92	0.68	0.00	0.78
		1.00	0.20	0.88	0.69	0.93	0.64	0.00	1.00
BBB	353	0.60	0.20	0.90	0.94	0.86	0.80	0.00	0.61
		1.00	0.10	0.82	0.88	0.75	0.64	0.00	1.00
Pgp Substrate	822	0.60	0.20	0.79	0.80	0.79	0.58	0.00	0.66
		1.00	0.20	0.73	0.73	0.74	0.47	0.00	1.00
Pgp Inhibitor	2304	0.50	0.20	0.85	0.91	0.73	0.66	0.00	0.76
		1.00	0.10	0.81	0.86	0.74	0.61	0.00	1.00
hERG	685	0.70	0.70	0.84	0.84	0.83	0.68	0.00	0.80
		1.00	0.20	0.82	0.82	0.83	0.64	0.00	1.00
MMP	6261	0.50	0.40	0.89	0.64	0.94	0.61	0.00	0.69
		1.00	0.20	0.87	0.52	0.94	0.50	0.00	1.00
AMES	6512	0.50	0.40	0.82	0.86	0.75	0.62	0.00	0.79
		1.00	0.20	0.79	0.82	0.75	0.57	0.00	1.00
MRTD	1184	0.60	0.20	0.00	0.00	0.00	0.00	0.79	0.69
		1.00	0.20	0.00	0.00	0.00	0.00	0.74	1.00

[a]Number of compounds in the dataset; [b]Tanimoto-distance threshold value; [c]Smoothing factor; [d]Pearson's correlation coefficient; [e]Regression model.

Table 5: Pharmacokinetic properties of ALA

PROPERTY	MODEL NAME	PREDICTED VALUE	UNIT
Absorption	Water solubility	-5.787	(log mol/L)
Absorption	CACO2 permeability	1.577	(log Papp in 10^{-6} cm/s)
Absorption	Intestinal absorption (human)	92.836	Numeric (% Absorbed)
Absorption	Skin Permeability	-2.722	Numeric (log Kp)
Absorption	P-glycoprotein substrate	No	Categorical (Yes/No)
Absorption	P-glycoprotein I inhibitor	No	Categorical (Yes/No)
Absorption	P-glycoprotein II inhibitor	No	Categorical (Yes/No)
Distribution	VDss (human)	-0.617	Numeric (log L/kg)
Distribution	Fraction unbound (human)	0.056	Numeric (Fu)
Distribution	BBB permeability	-0.115	Numeric (log BB)
Distribution	CNS permeability	-1.547	Numeric (log PS)
Metabolism	CYP2D6 substrate	No	Categorical (Yes/No)
Metabolism	CYP3A4 substrate	Yes	Categorical (Yes/No)
Metabolism	CYP1A2 inhibitior	Yes	Categorical (Yes/No)
Metabolism	CYP2C19 inhibitior	No	Categorical (Yes/No)
Metabolism	CYP2C9 inhibitior	No	Categorical (Yes/No)
Metabolism	CYP2D6 inhibitior	No	Categorical (Yes/No)
Metabolism	CYP3A4 inhibitior	Yes	Categorical (Yes/No)
Excretion	Total Clearance	1.991	Numeric (log ml/min/kg)
Excretion	Renal OCT2 substrate	No	Categorical (Yes/No)
Toxicity	AMES toxicity	No	Categorical (Yes/No)
Toxicity	Max. tolerated dose (human)	-0.84	Numeric (log mg/kg/day)
Toxicity	hERG I inhibitor	No	Categorical (Yes/No)
Toxicity	hERG II inhibitor	No	Categorical (Yes/No)
Toxicity	Oral Rat Acute Toxicity (LD_{50})	1.441	Numeric (mol/kg)
Toxicity	Oral Rat Toxicity (LOAEL)	3.115	(log mg/kg_bw/day)
Toxicity	Hepatotoxicity	Yes	Categorical (Yes/No)
Toxicity	Skin Sensitisation	Yes	Categorical (Yes/No)
Toxicity	*T.pyriformis* toxicity	0.722	Numeric (log ug/L)
Toxicity	Minnow toxicity	-1.183	Numeric (log mM)

Table 5: Prospected target for α-linolenic acid with predicted probability

TARGET	COMMON NAME	TARGET CLASS	PROBABILITY
Peroxisome proliferator-activated receptor γ	PPARG	Nuclear receptor	0.976
Peroxisome proliferator-activated receptor α	PPARA	Nuclear receptor	0.976
Peroxisome proliferator-activated receptor δ	PPARD	Nuclear receptor	0.976
Fatty acid binding protein adipocyte	FABP4	FABPF	0.723
Free fatty acid receptor 1	FFAR1	Family A G-PCR	0.690
Fatty acid binding protein muscle	FABP3	FABPF	0.682
Cyclooxygenase-1	PTGS1	Oxidoreductase	0.658
Fatty acid binding protein epidermal	FABP5	FABPF	0.281
Acyl-CoA desaturase	SCD	Enzyme	0.207
Anandamide amidohydrolase	FAAH	Enzyme	0.199
Telomerase reverse transcriptase	TERT	Enzyme	0.199
Fatty acid-binding protein, liver	FABP1	FABPF	0.199
Cannabinoid receptor 1	CNR1	Family A G-PCR	0.166
Protein-tyrosine phosphatase 1B	PTPN1	Phosphatase	0.133
Arachidonate 5-lipoxygenase	ALOX5	Oxidoreductase	0.133
T-cell protein-tyrosine phosphatase	PTPN2	Phosphatase	0.133
Prostaglandin E synthase	PTGES	Enzyme	0.117
Leukotriene B4 receptor 1	LTB4R	Family A G-PCR	0.109
DNA polymerase β	POLB	Enzyme	0.109
Estrogen receptor β	ESR2	Nuclear receptor	0.109
Protein-tyrosine phosphatase 1C	PTPN6	Phosphatase	0.109
11-β-hydroxysteroid dehydrogenase 1	HSD11B1	Enzyme	0.101
Carboxylesterase 2	CES2	Enzyme	0.101
Nuclear receptor ROR-γ	RORC	Nuclear receptor	0.101
DNA topoisomerase I	TOP1	Isomerase	0.101
Prostanoid EP2 receptor	PTGER2	Family A G-PCR	0.101
Arachidonate 12-lipoxygenase	ALOX12	Enzyme	0.101

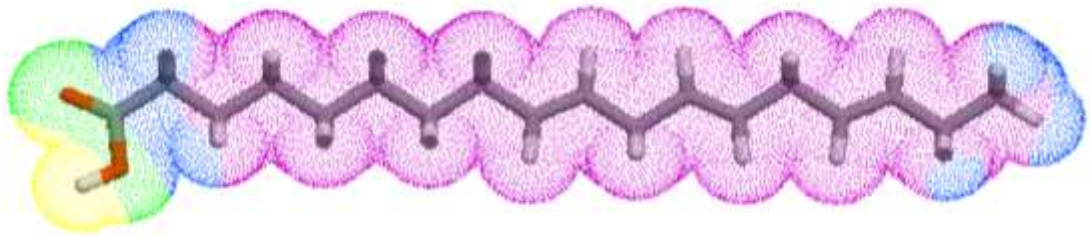

Figure 1: 3D structure of α-Linolenic Acid

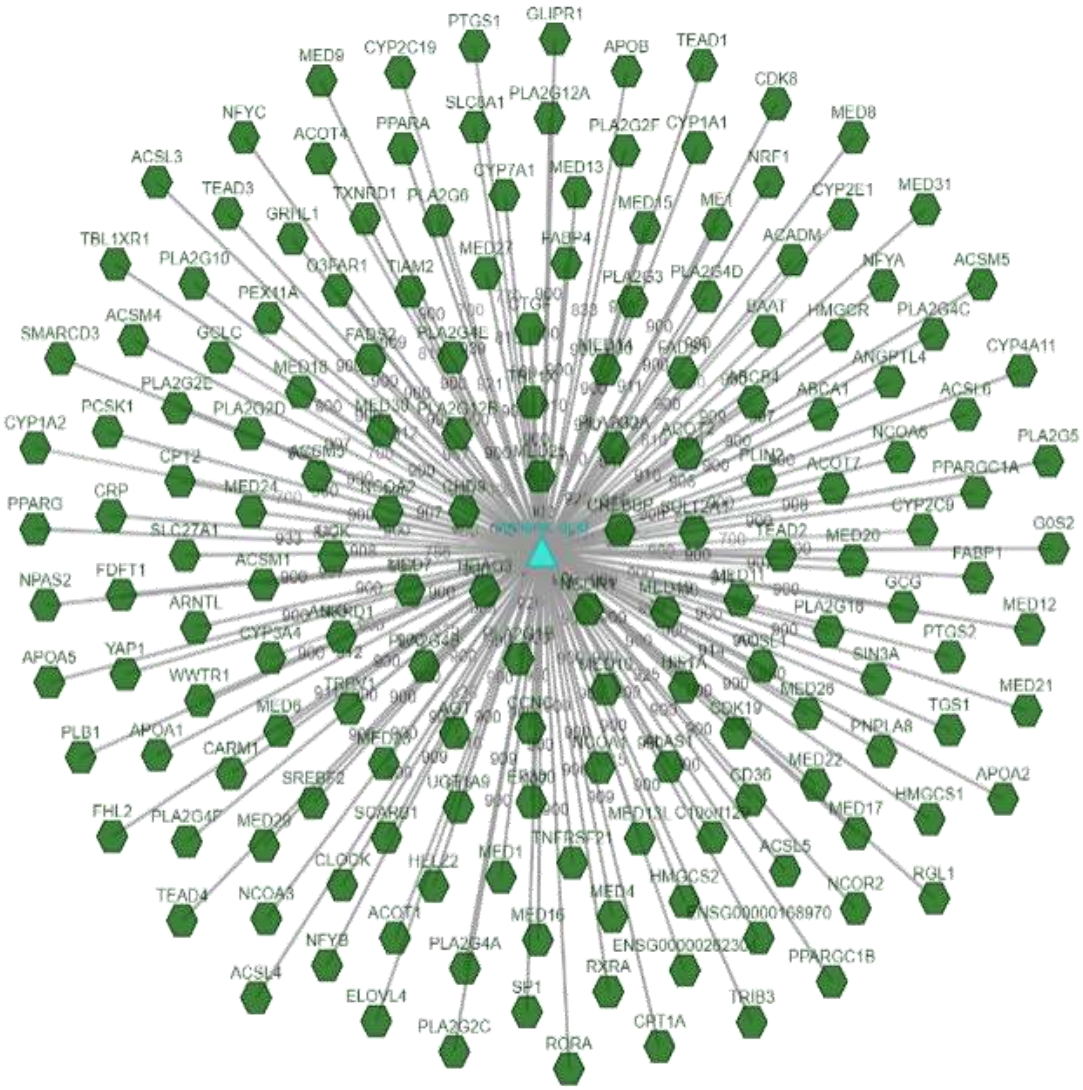

Figure 2: Cytoscape network of predicted human targets of ALA- Human Target Proteins were predicted using STITCH (26590256), Database of Chemical-Protein Interaction Networks.

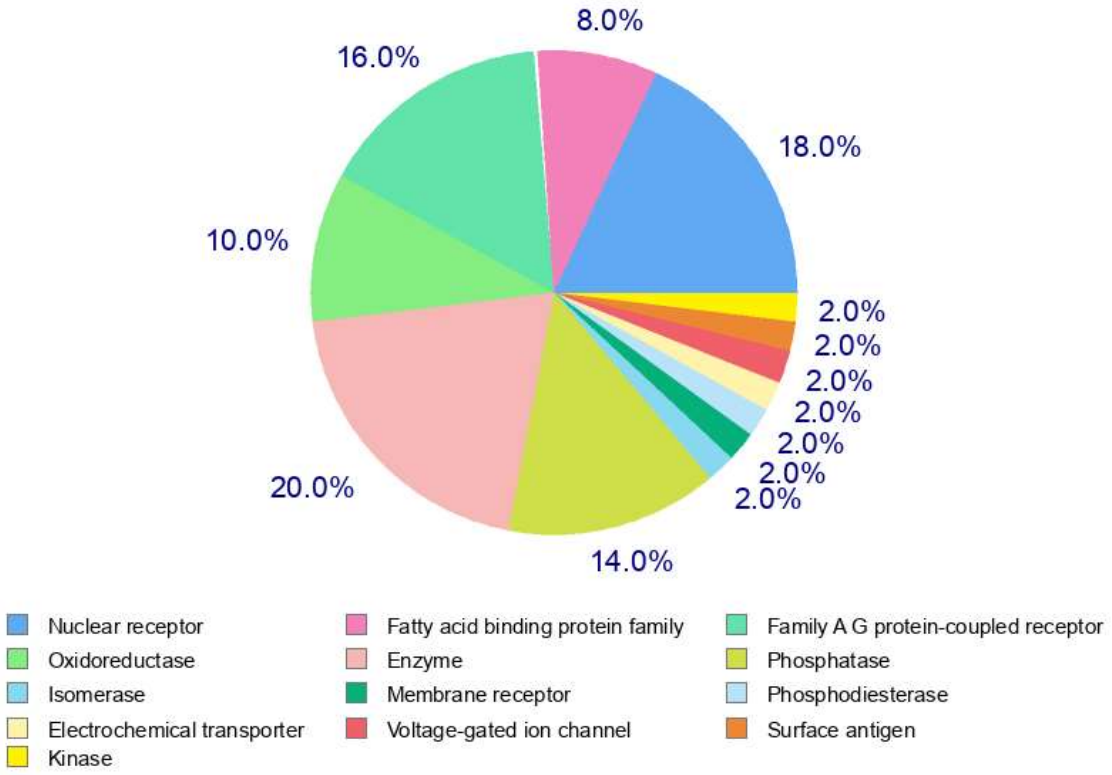

Figure 3: Predicted bioactivity target classes for α-linolenic acid from *C. roseus* with percentage probability