ISSN: 2348-8948
Vol: 2; Issue: 11

RESEARCH ARTICLE

INSILICO RELATIVE COMPOUND TOXICITY OF *PEDILANTHUS TITHYMALOIDES* AGAINST ORAL RAT

N.K. UDAYA PRAKASH.,* R. PUJITHA LAKSHMI., N. ASHWIN KARTHICK., A. BALAMURUGAN.,N. SRIPRIYA AND S. BHUVANESWARI

Research and Development, Marina Labs, Nerkundram, Chennai 600107
E-mail: nkudayaprakash@gmail.com

Date Received:	Date of Accepted:	Date Published:
06-Nov-2014	15-Nov-2014	18-Nov-2014

Abstract:
Pedilanthus tithymaloides, a common plant of the tropical countries, is widely used in ethno medicine. However, the toxicity profile of the plant is not much studied. The present study focuses on the prediction of toxicity in Oral rats of the chemicals detected through GC-MS. The compounds of *Pedilanthus tithymaloides* were subjected to toxicity analysis using Toxicity Estimation Software Tool (T.E.S.T). This *in-silico* QSAR model was used to predict the $LD_{50}$and the predicted values of the compounds 10-Octadecenoic acid, methyl ester; Cyclopropane butanoic acid, 2-[[2-[[2-[(2-pentyl cyclo propyl) methyl] cyclopropyl] methyl] cyclo propyl] methyl]-, methyl ester; Pentadecanoic acid, 14-methyl-, methyl ester; (4,4-Diphenyl-butyl)-(3-phenyl-piperidin-4-yl-)-amine and Rescinnamine against Oral rat are 15263.11, 4111.09, 16739.33, 754.43 and 603.44mg/kg respectively, concluding that these compounds were less or non-toxic to oral consumption.

Keywords: *Pedilanthustithymaloides, In-silico toxicity,* TEST, Oral rat.

Introduction

Pedilanthus tithymaloides, a common inhabitant of the tropical countries, is widely studied for its medicinal uses[1,2] and also for its antioxidant properties[3]. But, the plant is also reported to be toxic. The plant leaves and stems contain beta-sitosterol, cycloartenone, octacosanol, and oxime, all of which have known medicinal as well as toxic properties[4]. They also contain euphorbol (a complex terpene) and other diterpene esters in its root, stem and leaves in form of milky white latex. These are also known carcinogens[5,6]. The toxic effects of the plant include irritation of mucosal membranes, causing nausea and vomiting, skin irritation, inflammation, blisters, eye pain, kerato conjunctivitis, reduced visual activity and extreme diarrhoea[7].Despite this nature, the plant is highly used in ethnomedicine[8,9], which arises a need to study its toxicity profile. Previous studies in lower forms of animal models reported the plant compounds to be super toxic[10,11]and a need for studies on higher/equivalent forms has been realized.

Higher animal model in testing toxicityis expensive and time-consuming with subsequent ethical issues. This scenario creates a need for an alternate method to predict the toxicity in Animal models.

Quantitative structure-activity relationship (QSAR), a highly reliable *in-silico* model in toxicology[12-14], fits the rationale. QSAR models are models linking a property or effect such as boiling point or toxicity, predicting parameters associated with chemical parameters such as molecular descriptors[15].Toxicity Estimation Software Tool (TEST) is a highly reliable QSAR model used in assessing toxicity of compounds which is highly advantageous and predicts the toxicity in lesser time and cost with a data that is highly reliable.
Thus, TEST is used in this study to predict the toxicity of compounds detected by GC-MS analysis of the methanolic extract of plant, *Pedilanthus tithymaloides* against Oral Rat.

Materials and methods:
The plant has been subjected to GC-MS analysis and has been reported[16]. The detected compounds 10-Octadecenoic acid, methyl ester; Cyclopropane butanoic acid, 2-[[2-[[2-[(2-pentyl cyclo propyl) methyl] cyclopropyl] methyl] cyclo propyl] methyl]-, methyl ester; Pentadecanoic acid, 14-methyl-, methyl ester; (4,4-Diphenyl-butyl)-(3-phenyl-piperidin-4-yl-)-amine and Rescinnamine, their structural information, Molecular weight and representation are tabulated in table 1.

The *in-silico* toxicity prediction was carried out using QSAR model Toxicity Estimation Software Tool (T.E.S.T). TEST provides multiple prediction methodologies (Hierarchical clustering, Food and Drug Administration (FDA), nearest neighbor and Consensus method) so that one can have greater confidence in the predicted toxicities (assuming the predicted toxicities are fairly similar from different methods)[17,18]. Oral rat LD_{50} (amount of chemical in mg/kg body weight that causes 50% of rats to die after oral ingestion) was chosen as the toxicity end point for these compounds and the prediction was carried out.

Results:
The compounds detected by GC-MS analysis of the plant extract were subjected to toxicity prediction using various TEST models. The data obtained showed that the compounds 10-Octadecenoic acid, methyl ester; Cyclopropanebutanoic acid, 2-[[2-[[2-[(2-pentyl cyclo propyl) methyl] cyclopropyl] methyl] cyclo propyl] methyl]-, methyl ester; Pentadecanoic acid, 14-methyl-, methyl ester and (4,4-Diphenyl-butyl)-(3-phenyl-piperidin-4-yl-)-amine lacked experimental data while Rescinnamine had data on toxicity in Oral rat.

Table 2 corresponds to Consensus method (an average of Hierarchical, FDA and nearest neighbor method), while table 3 gives the prediction results given by modeling the compounds using Hierarchical, FDA and nearest neighbor methods.

The results showed that Rescinnamine has previous data on toxicity in Oral rats[19] and also TEST predictions inferred that the compound had the highest toxicity among the detected GC-MS compounds, which is followed by (4,4-Diphenyl-butyl)-(3-phenyl-piperidin-4-yl-)-amine; Cyclopropane butanoic acid, 2-[[2-[[2-[(2-pentyl cyclo propyl) methyl] cyclopropyl] methyl] cyclo propyl] methyl]-, methyl ester; 10-Octadecenoic acid, methyl ester and Pentadecanoic acid, 14-methyl-, methyl ester.

Discussion:
The prediction results using TEST for Rescinnamine could explain the reliability of data on toxicity. The experimental data on toxicity of Rescinnamine against Oral rat has a closer association with predicted data, whereas a ten-fold value is acceptable in chemical risk assessments. The data also complies with previous works[20]implying TEST as a highly reliable tool in toxicity predictions, though reliability is an outcome of user's confidence on the tool.

Using acceptable toxicity scales, the chemicals are assigned to various groups. One of the most common Hedge and Sterner scale was used to classify the compounds. The scale assigned three of the five compounds, Rescinnamine; (4, 4-Diphenyl-butyl)-(3-phenyl-piperidin-4-yl-)-amine and Cyclopropane butanoic acid, 2-[[2-[[2-[(2-pentyl cyclo propyl) methyl] cyclopropyl] methyl] cyclo propyl] methyl]-, methyl ester as slightly toxic while 10-Octadecenoic acid, methyl ester and Pentadecanoic acid, 14-methyl-, methyl ester as practically non-toxic compounds.

Though the milky latex of the plant is highly toxic to sensitive parts of human, the *in-silico* testing of these compounds in Oral rat showed that the plant compounds were practically not toxic or slightly toxic. The results could imply that the plant, highly used as ethno-medicine is not/less toxic when ingested orally. However, data on toxicity of these compounds in sensitive parts such as eyes and skin is necessary to realize the extent to use *Pedilanthus tithymaloides* in drug discovery.

Conclusion:
The data shows that the compounds of *Pedilanthus tithymaloides* are practically nontoxic or slightly toxic and can be used as an Oral medicine. The study could also serve as a reference for further *in-silico* toxicity studies using TEST.

References:

1. Ghosh, S., Samanta, A., Mandal, N.B., Bannerjee, S., Chattopadhyay, D., 2012. Evaluation of the wound healing activity of methanol extract of *Pedilanthus tithymaloides* (L.) Poit leaf and its isolated active constituents in topical formulation. J Ethnopharmacol. 142(3), 714-722.
2. *Vidottia, G.J., Zimmermanna, A., Sarragiottoa, M.H., Nakamurab, C.V., Dias Filho, B.P.,* 2006. Antimicrobial and phytochemical studies on *Pedilanthus tithymaloides.* Fitoterapia77(1), 43–46.
3. Abreu, P., Matthew, S., Gonza´lez, T., Costa, D., Segundo, M.A., Fernandes, E., 2006. Anti-inflammatory and antioxidant activity of a medicinal tincture from *Pedilanthus tithymaloides,* Life Sci. 78(14), 1578-1585.
4. Khare, C.P., Indian medicinal plants-An illustrated dictionary, Springer Science. (2007)

Table 1: List of compounds detected by GC-MS analysis of *Pedilanthus tithymaloides*

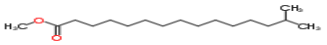
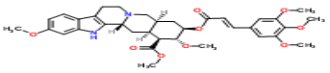
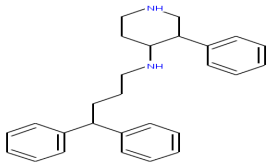
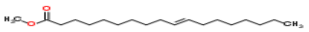
S.No	Chemical name	Structure	Representation	Molecular weight (Da)
1	10-Octadecenoic acid, methyl ester		$C_{19}H_{36}O_2$	296.49
2	Cyclopropanebutanoic acid, 2-[[2-[[2-[(2-pentylcyclopropyl)methyl] cyclopropyl] methyl] cyclo propyl] methyl]-, methyl ester		$C_{25}H_{42}O_2$	374.60
3	Pentadecanoic acid, 14-methyl-, methyl ester		$C_{17}H_{34}O_2$	270.45
4	(4,4-Diphenyl-butyl)-(3-phenyl-piperidin-4-yl-)-amine		$C_{27}H_{32}N_2$	384.61
5	Rescinnamine		$C_{35}H_{42}N_2O_9$	634.72

Table 2: Experimental and predicted values of the compounds predicted by Consensus method

S.no	Compound	Experimental value(48 hr) $-Log_{10}$ (mol/L)	Experimental value(48 hr) (mg/L)	Predicted value(48hr) $-Log_{10}$ (mol/L)	Predicted value(48hr) (mg/kg)
1	$C_{19}H_{36}O_2$	N/A	N/A	1.29	15263.11
2	$C_{25}H_{42}O_2$	N/A	N/A	1.96	4111.09
3	$C_{17}H_{34}O_2$	N/A	N/A	1.21	16739.33
4	$C_{27}H_{32}N_2$	N/A	N/A	3.	711.11
5	$C_{35}H_{42}N_2O_9$	2.80	999.15	3.02	603.44

Table 3: Prediction values of the compounds by Hierarchical, FDA and nearest neighbor methods

S.No	Compound	Heirarchical clustering	FDA	Nearest Neighbor
1	$C_{19}H_{36}O_2$	1.32	1.36	1.19
2	$C_{25}H_{42}O_2$	2.67	1.34	1.88
3	$C_{17}H_{34}O_2$	1.30	1.11	1.21
4	$C_{27}H_{32}N_2$	2.67	2.71	2.74
5	$C_{35}H_{42}N_2O_9$	3.19	2.69	3.19

5. Spoerke and Susan C Smolinske, Toxicity of Houseplants, 1st edition, CRC press, 181 (1990)
6. David W Nellis, Poisonous Plants and Animals of Florida and the Caribbean, Pineapple Pr Inc., 182 (1997).
7. Kenneth Lampe, Mary Ann McCann, American Medical Association, AMA Handbook of Poisonous and Injurious Plants, American Medical Association, 1st edition, Amer Medical Assn, 128 (1985).
8. Rashtra Vardhana, Direct Uses of Medicinal Plants and Their Identification, Sarup & Sons, 261 (2008).
9. J. K. Maheshwari, Ethnobotany in South Asia, Scientific Publishers (1996).
10. Udaya Prakash N.K., Balamurugan, A., Sripriya, N., AshwinKarthick, N., Pujitha Lakshmi, R and S. Bhuvaneswari. 2014, *In-silico* prediction of relative compound toxicity of *Pedilanthus tithymaloides* against *Daphnia magna*, Int J PharmTech Res. 6(6), 1908-1913.
11. Udaya Prakash N.K., Balamurugan, A., Sripriya, N., Ashwin Karthick, N.,Pujitha Lakshmi, R and S. Bhuvaneswari, 2014, *In-silico* evaluation of relative compound toxicity of *Pedilanthus tithymaloides* against *Pimephales promelas*, Der Pharma Chemica, 6(5), 105-110.
12. Zhu, H., Martin, T.M., Ye, L., Sedykh, A., Young, D.M., Tropsha, A., 2009, Quantitative structure activity relationship modeling of rat acute toxicity by oral exposure, Chem. Res. Toxicol.22(12), 1913–1921.
13. Demchuk, E., Ruiz, P., Chou, S., Fowler, B.A., 2011, SAR/QSAR methods in public health practice, Toxicol. Appl. Pharmacol. 254(2), 192–197.
14. Enslein, K., 1993, The future of toxicity prediction with QSAR, In Vitro Toxicol., 6, 163–169.
15. Theory, guidance and applications on QSAR and REACH. http://www.antares-life.eu
16. Udayaprakash, N.K., Ranjithkumar, M., Sripriya, N., Pujitha Lakshmi, Deepa, S., Bhuvaneswari, S., 2014, Antioxidant, free radical scavenging activity and GC-MS studies on *Pedilanthus tithymaloides* (L.) Poit. Int. J. Pharma and Pharmaceutical Sci. (Accepted).
17. User Manual Version 5.5, Simulation Plus Inc, S. P., Lancaster, CA, USA, (2011).
18. T.E.S.T. Tool, User's Guide for T.E.S.T, Version 4.0., US EPA, Cincinatti, OH, USA, (2011).
19. Drugs in Japan 6, 898, (1982).
20. Ruiz, P., Begluitti, G., Tincher, T., Wheeler, J., Mumtaz, M., 2012, Prediction of Acute Mammalian Toxicity Using QSAR Methods: A Case Study of Sulfur Mustard and Its Breakdown Products, Molecules, 17(8), 8982-9001.