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Toxicity Prediction of Selected Phytochemicals of Hatisur Weed (*Heliotropium indicum* Linnaeus) and Synthetic Medicines: An *In Silico* Approach by Using ProTox-II Tool

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Abstract: ***Heliotropium indicum Linnaeus is a common medicinal weed known as Hatisur in Bengali and many parts of the Asian country. This plant contains several phytochemicals, which are used for many disorders and also anti-inflammatory agent. This study predicted different types of toxicity parameters viz. acute toxicity (rat oral LD_{50}), organ toxicity, toxicity and genotoxicity endpoints of selected phytochemicals of H. indicum and synthetic medicines (Indomethacin and Ibuprofen) by using ProTox-II online tool. The phytocompounds Heliotridine, Retronecine and β-Linalool were predicted class V, may be harmful if swallowed. All the phytochemicals were predicted inactive, while both the synthetic medicines were found active for hepatoxicity. All compounds were obtained non-neurotoxic except Trachelanthamidine and Indomethacin. There were 5 compounds viz. Heleurine, Heliotridine, Trachelanthamidine, β-Linalool and Ibuprofen non-nephrotoxic. Only 2 compounds viz. β-Linalool and Ibuprofen were found inactive for respiratory toxicity. All compounds were obtained clinical toxicity inactive while only 1 compound (Indomethacin) was obtained immunotoxic. Regarding toxicity endpoints, only 1 compound (β-Linalool) was obtained ecotoxic. Only 5 compounds viz. Indicine N-oxide, Trachelanthamidine, Supinine, β-Linalool and Ibuprofen were obtained inactive for clinical toxicity while. only 3 compounds viz. Trachelanthamidine, β-Linalool and Ibuprofen were found inactive for nutritional toxicity. All compounds were found non-cytotoxic and non-mutagenic while only 3 compounds viz. were β-Linalool, Indomethacin and Ibuprofen were obtained non-carcinogenic. In conclusion, β-Linalool can be used as non-toxic phytocompound and substituted as synthetic medicines viz. Indomethacin and Ibuprofen. Moreover, experimental assay is suggested to validate the present prediction with this phytocompound.***

Keywords: Anti-inflammatory agent, *Heliotropium indicum*, Predictive toxicity, *In silico*, Toxicity endpoints

1. Introduction

The medicinal weed in Bengali "Hatisur" and the scientific name *Heliotropium indicum* Linnaeus under the Family: Boraginaceae, this name as is derived from the Greek words "Helios" meaning "sun" and "Tropein" meaning "to turn," which is indicating that the flowers and leaves turn toward the sun. [1,2] This weed is commonly found throughout Bangladesh, Nepal, Sri Lanka, Thailand, India, and other areas of tropical Asia and in few sites of Africa.[3]

Interestingly, this medicinal weed has potential phytochemicals to prevent many disorders such as inflammation, pain, bone fracture, nociceptive activity, wound healing, etc. [2,4-6] But some studies reported that plant extract may contain allelochemicals cause allelopathy and toxicity to animals. [7,8]

With regards, prior to using crude extract, toxicity screening is extreme concern based on *in vitro* and *in vivo* assay, but these studies may take long-duration, huge laboratory cost and animal sacrifice. Whereas *in silico* prediction through computational tools helps faster screening, no animal harming and inexpensive method.[9,10] In recent research, many investigators reported *in silico* toxicity prediction of natural and synthetic compounds for new drug design. [9-12]

This study was focused to predict toxicity of selected phytochemicals of Hatisur weed (*Heliotropium indicum* Linnaeus) and synthetic medicines used for anti-inflammation by using ProTox II online tool.

2. Materials and Methods

All established phytochemicals of *Heliotropium indicum* Linnaeus and synthetic medicines viz. Indomethacin and Ibuprofen were considered from available literature. [2,4]

As per the protocol by Banerjee et al.[10] in the ProTox-II online tool (version, 3.0) in which different types of toxicity parameters viz. acute toxicity (rat oral LD_{50}),[9] organ toxicity, toxicity and genotoxicity endpoints of selected phytochemicals of *H. indicum* and synthetic medicines (Indomethacin and Ibuprofen) were evaluated.

The pictorial representation of medicinal weed specimen is exhibited in Fig 1.

Fig 1: Medicinal weed specimen (*Heliotropium indicum* Linnaeus)

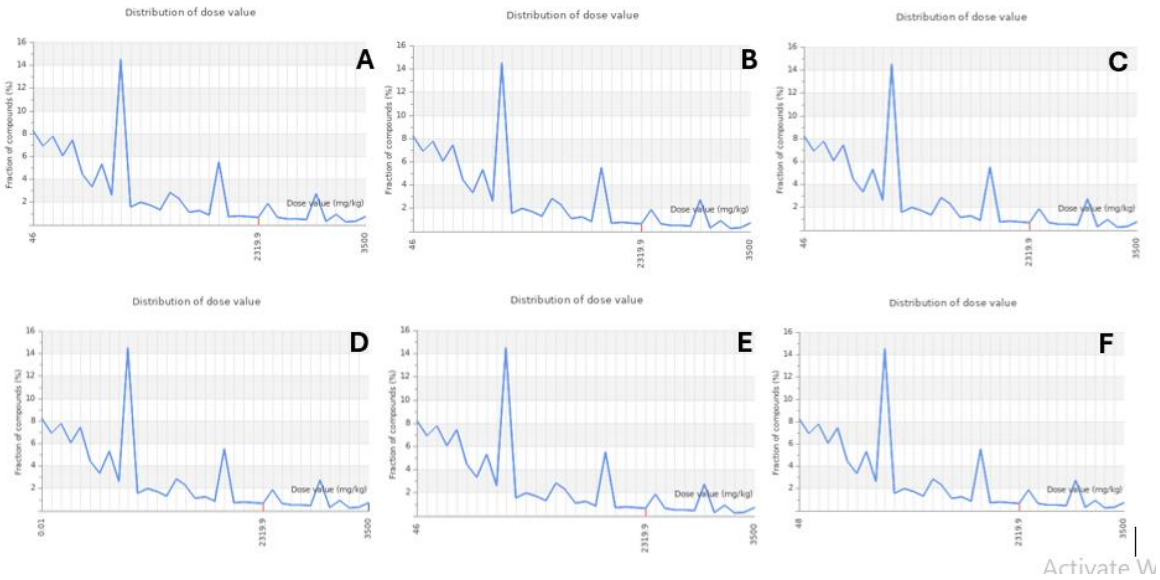
3. Results

Table 1 summarizes the values of predictive oral acute (LD_{50}) toxicity (mg/Kg), class and predictive accuracy of phytochemicals and synthetic medicines. The phytocompounds Heliotridine, Retronecine and β-Linalool were predicted class V, may be harmful if swallowed. Fig 2 depicts the dose response curves of studied compounds.

Table 1: Prediction of acute toxicity of phytochemicals of areal parts of *H. indicum* and synthetic medicines

Sl. No.	Compounds name	Rat oral LD_{50} value (mg/Kg)	Predicted toxicity class	Probability
Phytochemicals				
1.	Heleurine	46.0	II	69.26
2.	Echinatine	46.0	II	70.97
3.	Heliotrine	46.0	II	70.97
4.	Heliotridine	3500.0	V	67.38
5.	Indicine	46.0	II	70.97
6.	Indicine N-oxide	48.0	II	69.26
7.	Lasiocarpine	110.0	III	100.00
8.	Trachelanthamidine	227.0	III	70.97
9.	Retronecine	3500.0	V	67.38
10.	Supinine	46.0	II	69.26
11.	β-Linalool	2200	V	100.00
Synthetic medicines				
1.	Indomethacin	12.0	II	100.00
2.	Ibuprofen	299.0	III	100.00

Class II: fatal if swallowed ($5 < LD_{50} \leq 50$); Class III: toxic if swallowed ($50 < LD_{50} \leq 300$); Class V: may be harmful if swallowed ($2000 < LD_{50} \leq 5000$)



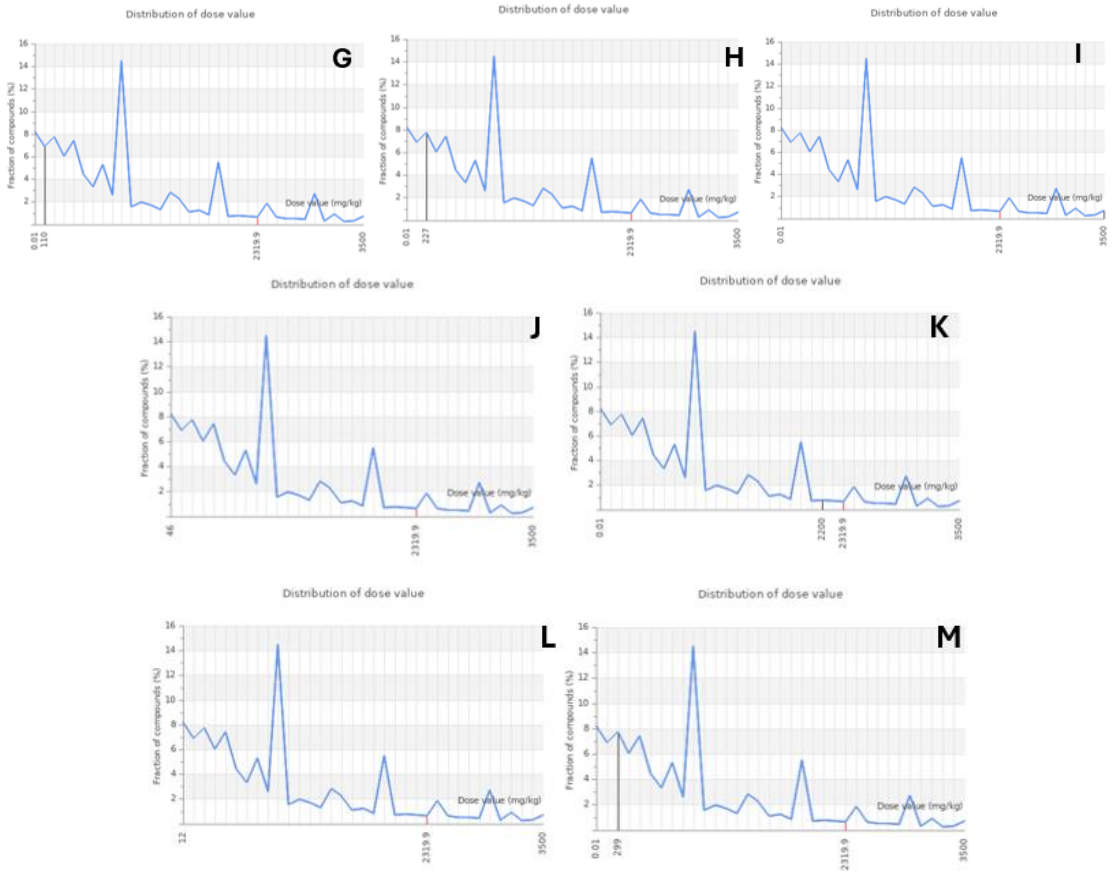


Figure 2: Dose response curve for phytochemicals of areal parts of *H. indicum* and synthetic medicines (A = Heleurine; B = Echinatine; C = Heliotrine; D = Heliotridine; E = Indicine; F = Indicine N-oxide; G = Lasiocarpine; H = Trachelanthamidine; I = Retronecine; J = Supinine; K = Indomethacin; L = Ibuprofen

Table 2 predicts organ toxicity of phytochemicals of areal parts of *H. indicum* and synthetic medicines. For hepatotoxicity, all phytochemicals were obtained inactive while both the synthetic medicines were found active for hepatoxicity. For neurotoxicity, all compounds were obtained non-neurotoxic except Trachelanthamidine and Indomethacin. For nephrotoxicity, 5 compounds viz. Heleurine, Heliotridine, Trachelanthamidine, β-Linalool and Ibuprofen were non-nephrotoxic. For respiratory toxicity, 2 compounds viz. β-Linalool and Ibuprofen were found inactive. All compounds were obtained clinical toxicity inactive while only 1 compound (Indomethacin) was obtained immunotoxic.

Table 2: Prediction of organ toxicity of phytochemicals of areal parts of *H. indicum* and synthetic medicines

Sl. No.	Compounds name	HT	P	NT	P	NPT	P
Phytochemicals							
1.	Heleurine	I	0.92	I	0.65	I	0.51
2.	Echinatine	I	0.90	I	0.66	A	0.53
3.	Heliotrine	I	0.91	I	0.66	A	0.53
4.	Heliotridine	I	0.91	I	0.52	I	0.58
5.	Indicine	I	0.90	I	0.66	A	0.79
6.	Indicine N-oxide	I	0.77	I	0.75	A	0.52
7.	Lasiocarpine	I	0.89	I	0.67	A	0.54
8.	Trachelanthamidine	I	0.92	A	0.54	I	0.60
9.	Retronecine	I	0.91	I	0.52	I	0.58
10.	Supinine	I	0.90	I	0.66	A	0.50
11.	β-Linalool	I	0.76	I	0.62	I	0.87
Synthetic medicines							
1.	Indomethacin	A	0.86	A	0.59	A	0.59
2.	Ibuprofen	A	0.66	I	0.89	I	0.52
		RT	**P**	**CT**	**P**	**IT**	**P**
Phytochemicals							
1.	Heleurine	A	0.79	I	0.78	I	0.97
2.	Echinatine	A	0.79	I	0.74	I	0.99
3.	Heliotrine	A	0.81	I	0.76	I	0.97
4.	Heliotridine	A	0.74	I	0.74	I	0.89
5.	Indicine	A	0.79	I	0.74	I	0.99
6.	Indicine N-oxide	A	0.76	I	0.75	I	0.98
7.	Lasiocarpine	A	0.80	I	0.78	I	0.87
8.	Trachelanthamidine	A	0.69	I	0.72	I	0.99
9.	Retronecine	A	0.74	I	0.74	I	0.99
10.	Supinine	A	0.80	I	0.77	I	0.99
11.	β-Linalool	I	0.99	I	0.75	I	0.99
Synthetic medicines							
1.	Indomethacin	A	0.87	I	0.91	A	0.62
2.	Ibuprofen	I	0.77	I	0.71	I	0.99

HT = Hepatotoxicity; NT = Neurotoxicity; NPT = Nephrotoxicity; RT = Respiratory toxicity; CT = Cardiotoxicity; IT = Immunotoxicity, I= Inactive; A = Active; P = Probability

Table 3 predicts toxicity endpoints such as ecotoxicity, clinical toxicity and nutritional toxicity of phytochemicals of areal parts of *H. indicum* and synthetic medicines. For ecotoxicity, only 1 compound (β-Linalool) was obtained ecotoxic. For clinical toxicity, 5 compounds viz. Indicine N-oxide, Trachelanthamidine, Supinine, β-Linalool and Ibuprofen were obtained inactive. For nutritional toxicity, 3 compounds viz. Trachelanthamidine, β-Linalool and Ibuprofen were found inactive.

Table 3: Prediction of toxicity endpoints of phytochemicals of areal parts of *H. indicum* and synthetic medicines

S. No.	Compounds name	ET	P	CLT	P	NUT	P
Phytochemicals							
1.	Heleurine	I	0.52	A	0.50	A	0.53
2.	Echinatine	I	0.64	A	0.59	A	0.71
3.	Heliotrine	I	0.64	A	0.61	A	0.69
4.	Heliotridine	I	0.63	A	0.58	A	0.57
5.	Indicine	I	0.64	A	0.59	A	0.71
6.	Indicine N-oxide	I	0.59	I	0.67	A	0.71
7.	Lasiocarpine	I	0.68	A	0.60	A	0.69
8.	Trachelanthamidine	I	0.51	I	0.57	I	0.63
9.	Retronecine	I	0.63	A	0.58	A	0.57
10.	Supinine	I	0.58	I	0.50	A	0.60
11.	β-Linalool	A	0.56	I	0.63	I	0.70
Synthetic medicines							
1.	Indomethacin	I	0.69	A	0.70	A	0.77
2.	Ibuprofen	I	0.59	I	0.71	I	0.96

ET = Ecotoxicity; CLT = Clinical toxicity; NUT = Nutritional toxicity; I= Inactive; A = Active; P = Probability

Table 4 predicts genotoxicity endpoints such as cytotoxicity, mutagenicity and carcinogenicity of phytochemicals of areal parts of *H. indicum* and synthetic medicines. All compounds were found non-cytotoxic and non-mutagenic while only 3 compounds viz. were β-Linalool, Indomethacin and Ibuprofen were obtained non-carcinogenic.

Table 4: Prediction of genotoxicity endpoints of phytochemicals of areal parts of *H. indicum* and synthetic medicines

S. No.	Compounds name	CYT	P	MUT	P	CARC	P
Phytochemicals							
1.	Heleurine	I	0.67	I	0.80	A	0.76
2.	Echinatine	I	0.66	I	0.91	A	0.83
3.	Heliotrine	I	0.66	I	0.86	A	0.79
4.	Heliotridine	I	0.74	I	0.89	A	0.59
5.	Indicine	I	0.66	I	0.91	A	0.83
6.	Indicine N-oxide	I	0.66	I	0.80	A	0.71
7.	Lasiocarpine	I	0.65	I	0.83	A	0.92
8.	Trachelanthamidine	I	0.73	I	0.75	A	0.54
9.	Retronecine	I	0.74	I	0.99	A	0.59
10.	Supinine	I	0.66	I	0.87	A	0.79
11.	β-Linalool	I	0.82	I	0.95	I	0.64
Synthetic medicines							
1.	Indomethacin	I	0.79	I	0.86	I	0.63
2.	Ibuprofen	I	0.85	I	0.99	I	0.74

CYT = Cytotoxicity; MUT = Mutagenicity; CARC = Carcinogenicity; I = Inactive; A = Active; P = Probability

4. Discussion

The phytocompounds Heliotridine, Retronecine and β-Linalool were predicted class V, may be harmful if swallowed but rest compounds were obtained class II (fatal if swallowed) and III (toxic if swallowed). Earlier toxicity studies reported that 14-day oral administration of 1–2 gm/kg of *H. indicum* aqueous extracts induced pathological impacts on the liver, kidney, heart, and lungs, [2,13] which is supported the present study that most of the phytocompounds obtained nephrotoxic and respiratory toxic but did not show hepatoxicity and cardiotoxicity.

Moreover, pain relief was observed by ethanolic and aqueous extracts of the aerial parts of *H. indicum* (30-300 mg/kg) in a mouse model after formalin-induced pain. [2,13] In another study, the chloroform extract of leaf of *H. indicum* extract (150 mg/kg of body weight) observed a significant anti-inflammatory effect (80.0%) on carrageenan-induced paw edema in albino Wistar rats.[14]

All compounds were found non-cytotoxic and non-mutagenic while only 3 compounds viz. were β-Linalool, Indomethacin and Ibuprofen were obtained non-carcinogenic. Azeez et al. [15] observed chromosomal aberrations for the cytotoxic and genotoxic activities of *H. indicum*.

In the present study, β-Linalool observed inactive for the studied parameters except ecotoxicity. An agreement with earlier study that Linalool has anti-inflammatory effects.[16] On the other hand, for genotoxicity end points, β-Linalool did not observe cytotoxicity, mutagenicity and carcinogenicity, which is an agreement with other experimental study that Linalool was found non-genotoxic. [17]

5. Conclusion

In conclusion, among these phytocompounds and synthetic medicines, β-Linalool was found to be better efficacious for all toxicity parameters like Indomethacin and Ibuprofen like drugs. Moreover, experimental assay is suggested to validate the present prediction with this phytocompound.

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Conflict of interest
It is declared none.

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