ORIGINAL ARTICLE

Prediction of Prospective Anti-Parkinson Phytochemicals using Prediction of Activity Spectra of Substances Software to Justify 3R's Ethics of *In Vivo* Evaluation

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Abstract

Background: Parkinson's disease (PD) is a chronic progressive devastative disorder of neurons characterized by a muscle rigidity, tremors, bradykinesia etc. In present scenario, it is affecting more than 1% population above 50 years of age and hence is an important concern in society. Advancement in research field in recent decades has led to upsurge the use of animals for evaluation of new drugs. **Objective:** In contemplation of upward trend in use of animals, PASS (prediction of activity spectra of substances), a web tool, provides an informative prediction data for different pharmacological activity of compounds without using the animals which justifies the 3R's ethics (Reduction, Replacement, and refinement) to be followed for *in vivo* evaluation. **Methods:** For prediction of pharmacological activities of anti-parkinson compounds, canonical smiles of phytochemicals were obtained from Pubmed and used in the software for prediction of relevant pharmacological activity so that phytochemicals, showing best results can be further explored for *in vivo* evaluation against PD. Using PASS online software, biological activity spectra for nine different activities related to Parkinson's disease for selected phytochemicals was predicted and compared with marketed compounds. **Result:** Out of selected phytochemicals, scopolamine and atropine have shown highest antiparkinsonian activities. Piperine was also found to have antiparkinsonian activity. Elaeocarpine, harmine and oxyresveratrol have found to have comparable activity for this condition. **Conclusion:** This article describes the utility of PASS to justify the 3R's concept which is to be followed for the further *in vivo* exploration of compounds.

Key words: Anti-Parkinson phytochemicals, PASS software, 3R's ethics

INTRODUCTION

Parkinson's disease (PD) is named after Dr. James Parkinson, who was the first to identify conditions and record symptoms of PD. PD is a progressive neurodegenerative disorder which affects the substantia nigra pars compacta region of the brain, characterized by decrease in dopamine level in the brain. It is reported that 80% of dopamine level lost during the Parkinson's. It is the second most devastative neurological disease, primarily affects the persons with age 50 and above. Currently, some of the reported clinical symptoms are muscle rigidity, stoop posture, bradykinesia, and resting tremors of extremities.[1-4] The underlying pathology of PD is increased oxidative stress, mitochondrial dysfunction, and deposition of alpha-synuclein (termed as Lewy bodies and dystrophic neurites), a misfold protein in the brain causing microglia activation followed by neuroinflammation and neurotoxicity.[5-7]

The etiology of PD is still unknown, but some factors such as age, environment, genetics, and medication have been reported. A large number of synthetic drugs are available for drug therapy [Figure 1] but have a wide number of side effects, such as nausea, vomiting, hallucinations,

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Figure 1: Available drug therapy of Parkinson's disease

and convulsions. Moreover, they are known to provide symptomatic relief only, i.e., not able to cure the underline cause of PD.[8-10]

Because of concern about side effects of synthetic medicines, there is serious consideration and research for the natural medicines in the last few decades. Antioxidant and neuroprotective actions of natural medicines are utilized in the treatment of PD.[11] A large number of phytochemicals have been reported to be effective in *in vivo* and *in vitro* models of PD but fail to enter in the mainstream of drug development due to lack of information.

PASS, a computer-based software program, provides information regarding different biological activities of chemical compounds on the basis of their chemical structures. The current version of PASS can predict more than 3750 biological effects, biochemical modes of action, specific toxicities, and metabolic terms based on two-dimensional structures or canonical simplified molecular-input line-entry system (SMILES), with a mean accuracy of almost 95%. It predicts activity in terms of probabilities; probable activity (Pa) and probable inactivity (Pi). The values vary from 0.000 to 1.000. The activity of compound is considered only if Pa > Pi; moreover, compounds having Pa activities >0.7 are considered to have high pharmacological actions. Similarly, compounds having Pa values <0.7 have less probability of observing the activity.[12-15] The present study incorporates the use of PASS for exploration of the pharmacological potential of selected phytochemicals in the treatment of PD, with respect to various disease-associated targets.

MATERIALS AND METHODS

Materials

Several phytoconstituents were selected on the basis of existing literature, suggesting their applicability in the treatment of PD [references mentioned in Table 1]. One marketed drug to treat PD was also selected to predict the biological activity spectra. The canonical SMILES, which work as a formula of these phytochemicals and marketed drugs, were obtained from PubChem (www.pubchem.ncbi.nlm.nih.gov), as shown in Table 2.

Methods

An extravagant search of existing literature was conducted to collect the information pertaining to previously reported activities (*in vivo* and *in vitro*) of phytochemicals. The canonical SMILES of different compounds were copied into the PASS online software (www.pharmaexpert.ru/passonline/predict.php/), and the biological activity spectra were obtained.

RESULTS AND DISCUSSIONS

Using PASS online software, biological activity spectra for nine activities of selected phytochemicals and marketed compound were predicted. These activities are as follows:

- Dopamine precursors
- Caspase 3 inhibitors
- Central anticholinergics
- Free radical scavengers
- Nootropic activity
- Dopamine-release stimulants
- Monoamine oxidase (MAO) inhibitors
- N-methyl D-aspartate (NMDA) receptor antagonists
- Antiparkinsonian.

The biological activity spectra for different activities for phytochemicals and marketed compound, i.e., safinamide, are represented in Table 2.

PD is a progressive neurodegenerative disorder, which to date has no effective treatment. Search is generally still going on around the world in this stream. Several compounds have passed pre-clinical trials but are able to enter the clinical trials stage, due to lack of sufficient informative data. PASS is an online program that can be used by anyone after completing the free registration, which predicts the biological activity of a chemical compound on the basis of chemical composition and interaction with different targets. This program provides information that a particular compound can be useful in a particular disease.

One marketed standard drug (safinamide) for PD was selected to compare with various phytochemicals. Pharmacological

Table 1: Canonical simplified molecular-input line-entry system of different phytochemicals

Name of compound	Canonical SMILES
Safinamide	CC(C(=O)N)NCC1=CC=C(C=C1)OCC2=CC(=CC=C2)F
Atropine	CN1C2CCC1CC(C2)OC(=O)C(CO)C3=CC=CC=C3
Harmine	CC1=NC=CC2=C1NC3=C2C=CC(=C3)OC
Curcumin	COC1=C(C=CC(=C1)C=CC(=O)CC(=O)C=CC2=CC(=C(C=C2)O)OC)O
Scopolamine	CN1C2CC(CC1C3C2O3)OC(=O)C(CO)C4=CC=CC=C4
Oxyresveratrol	C1=CC(=C(C=C1O)O)C=CC2=CC(=CC(=C2)O)O
Oleuropein	CC=C1C(C(=COC1OC2C(C(C(C(O2)CO)O)O)O)O)C(=O)OC)CC(=O)OCCC3 =CC(=C(C=C3)O)O
Tenuigenin	CC1(CCC2(CCC3=C(C2C1)C(CC4C3(CCC5C4(CC(C(C5(C)C (=O)O)O)O)O)C)C)CCl)C(=O)O)C
Quercetin	C1=CC(=C(C=C1C2=C(C(=O)C3=C(C=C(C=C3O2)O)O)O)O)O
Glutathione	C(CC(=O)NC(CS)C(=O)NCC(=O)O)C(C(=O)O)N
Rudrakine	O=C1C2C(CCN3C2CCC3)OC4=C1C(C)CC(O)C4
Huperzine A	CC=C1C2CC3=C(C1(CC(=C2)C)N)C=CC(=O)N3
Nobiletin	COC1=C(C=C(C=C1)C2=CC(=O)C3=C(O2)C(=C(C(=C3OC)OC)OC)OC)OC
Iso rhynchophylline	CCC1CN2CCC3(C2CC1C(=COC)C(=O)OC)C4=CC=CC=C4NC3=O
Elaeocarpidine	C1CC2N(C1)CCC3N2CCC4=C3NC5=CC=CC=C45
Creatine	CN(CC(=O)O)C(=N)N
Morin	C1=CC(=C(C=C1O)O)C2=C(C(=O)C3=C(C=C(C=C3O2)O)O)O
Piperin	C1CCN(CC1)C(=O)C=CC=CC2=CC3=C(C=C2)OCO3
Asiaticoside	CC1CCC2(CCC3(C(=CCC4C3(CCC5C4(CC(C(C5(C)CO)O)O)C)C)C2C1C)C)C(=O)O C6C(C(C(C(O6)COC7C(C(C(C(O7)CO)OC8C(C(C(C(O8)C)O)O)O)O)O)O)O)O)O
Apigenin	C1=CC(=CC=C1C2=CC(=O)C3=C(C=C(C=C3O2)O)O)O

SMILES: Simplified molecular-input line-entry system

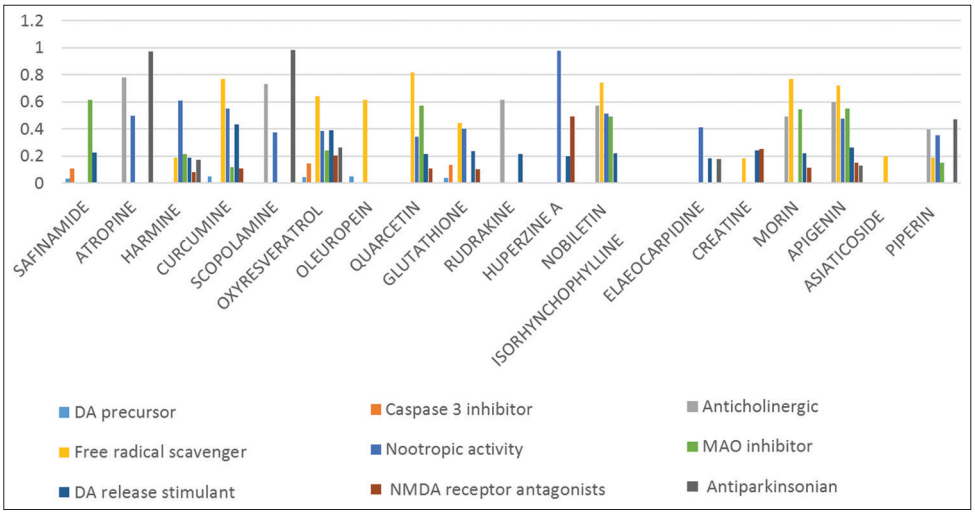

Figure 2: All activities for phytochemicals with respect to safinamide

activity for all phytochemicals was tested for nine activities, as shown in Figure 2.

It was found that safinamide has less dopaminergic precursor activity as compared to phytochemicals [Figure 3]. Of the phytochemicals, oleuropein has high value for dopamine (DA) precursor activity while phytochemicals such as rudrakine, huperzine A, nobiletin, iso rhynchophylline, elaeocarpidine, creatine, morin, apigenin, asiaticoside, and piperin were zero DA precursor activity [Figure 3].

The DA precursor activity for various phytochemicals in decreasing sequence was oleuropein >curcumin >oxyresveratrol >glutathione.

Table 2: Probable activity predicted by prediction of activity spectra of substance software

Compounds	Dopamine precursor Pa	Caspase 3 inhibitor Pa	Anticholinergic Pa	Free radical scavenger Pa	Nootropic activity Pa	MAO inhibitor Pa	Dopamine release stimulant Pa	NMDA receptor antagonists Pa	Antiparkinsonian Pa
Safinamide[16]	0.032	0.109	0	0	0	0.614	0.223	0	0
Atropine[17]	0	0	0.777	0	0.494	0	0	0	0.973
Harmine[18]	0	0	0	0.187	0.608	0.215	0.188	0.079	0.173
Curcumin[19]	0.047	0	0	0.771	0.552	0.118	0.433	0.107	0
Scopolamine[20]	0	0	0.733	0	0.373	0	0	0	0.983
Oxyresveratrol[19]	0.045	0.145	0	0.640	0.383	0.240	0.388	0.202	0.262
Oleuropein[19]	0.050	0	0	0.614	0	0	0	N	0
Quercetin[21]	0	0	0	0.816	0.341	0.572	0.216	0.109	0
Glutathione[22]	0.035	0.132	0	0.442	0.399	0	0.234	0.101	0
Rudrakine[23]	0	0	0.616	0	0	0	0.211	0	0
Huperzine A[24]	0	0	0	0	0.977	0	0.196	0.493	0
Nobiletin[25]	0	0	0.569	0.742	0.513	0.489	0.221	0	0
Iso rhynchophylline[26]	0	0	0	0	0	0	0	0	0
Elaeocarpidine[27]	0	0	0	0	0.409	0	0.183	0	0.177
Creatine[28]	0	0	0	0.181	0	0	0.239	0.250	0
Morin[29]	0	0	0.491	0.766	0	0.544	0.217	0.113	0
Apigenin[30]	0	0	0.598	0.719	0.475	0.549	0.259	0.148	0.129
Asiaticoside[31]	0	0	0	0.197	0	0	0	0	0
Piperin[11]	0	0	0.393	0.185	0.350	0.148	0	0	0.469

NMDA: N-methyl D-aspartate, MAO: Monoamine oxidase, Pa: Probable activity

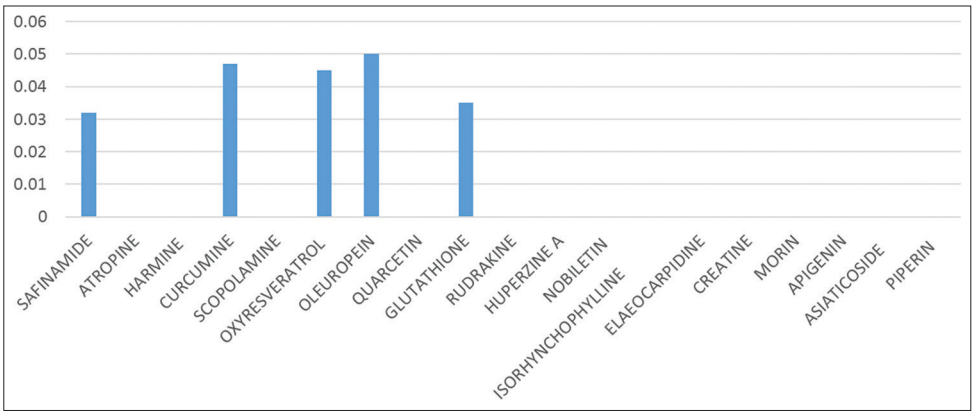

Figure 3: Dopamine precursor activity with respect to safinamide

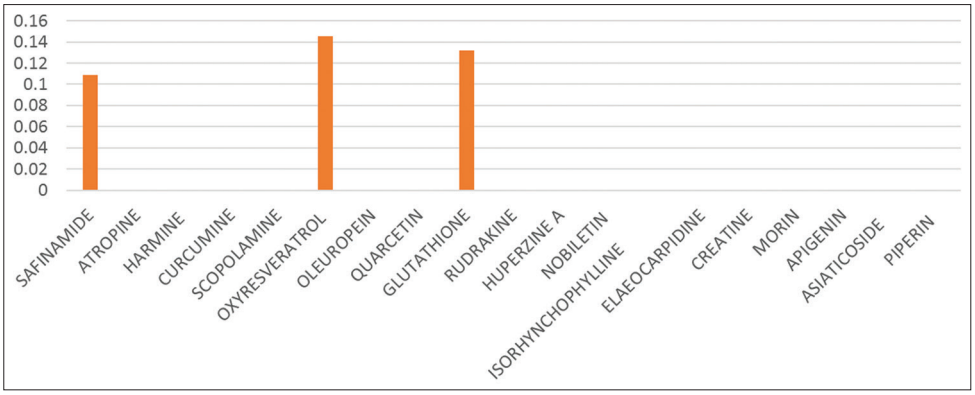

Figure 4: Caspase 3 inhibitor activity with respect to safinamide

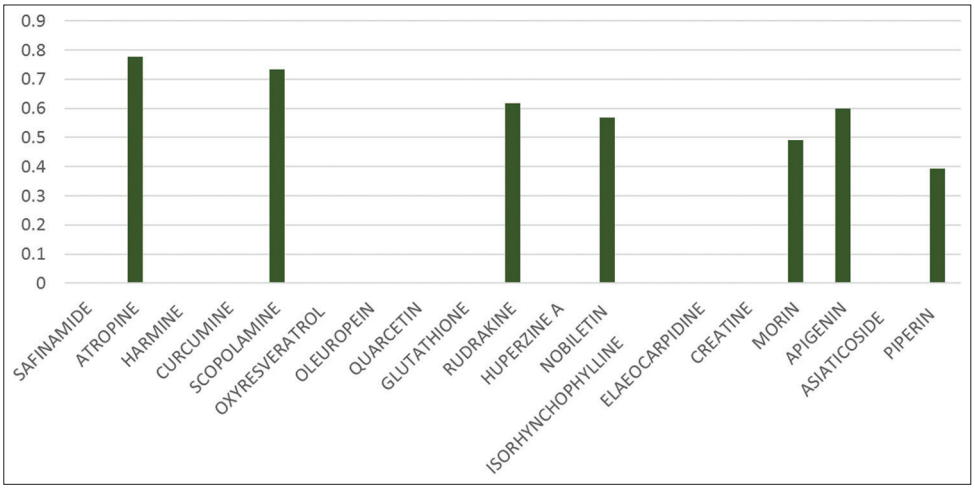

Figure 5: Anticholinergic activity with respect to safinamide

Safinamide was found to have less caspase 3 inhibitory activity. Of the phytochemicals, only two were showing caspase 3 inhibitory activity. Oxyresveratrol has high caspase 3 inhibitory activity and glutathione with the least caspase 3 inhibitor activity [Figure 4]. Similarly, anticholinergic activity for all phytochemicals was determined. Safinamide was found to have maximum anticholinergic activity as compared to phytochemicals. The pattern for anticholinergic activity for phytochemicals is scopolamine >rudrakine = apigenin >nobiletin >morin >piperin [Figure 5].

Safinamide was found to have no free radical scavenging activity. Phytochemicals for free radical activity follow the pattern quercetin >curcumin =morin >nobiletin >apigenin >asiaticoside >harmine =creatine =piperin as shown in Figure 6. In the same manner, safinamide has no nootropic activity, when compared with phytochemicals. Huperzine A has the highest nootropic activity. Other phytochemicals having nootropic activity as follows: huperzine A >harmine >curcumin >nobiletin =apigenin >glutathione = elaeocarpidine >scopolamine >quercetin >piperin [Figure 7].

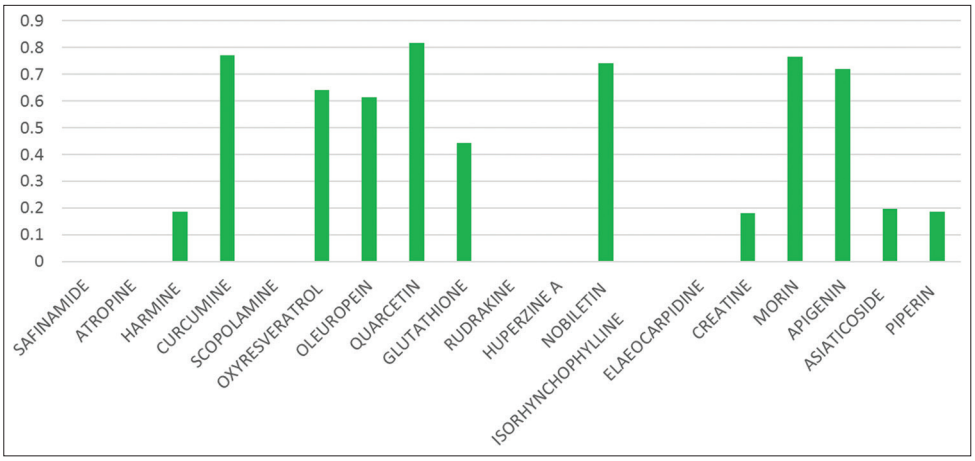

Figure 6: Free radical scavenging activity with respect to safinamide

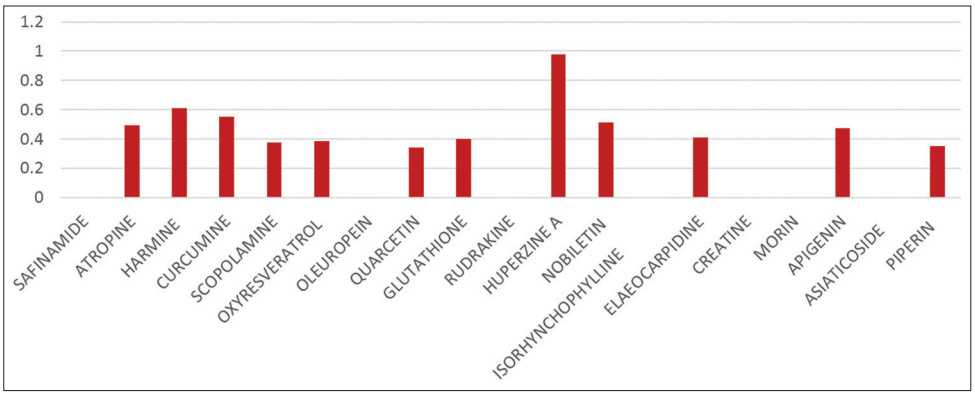

Figure 7: Nootropic activity with respect to safinamide

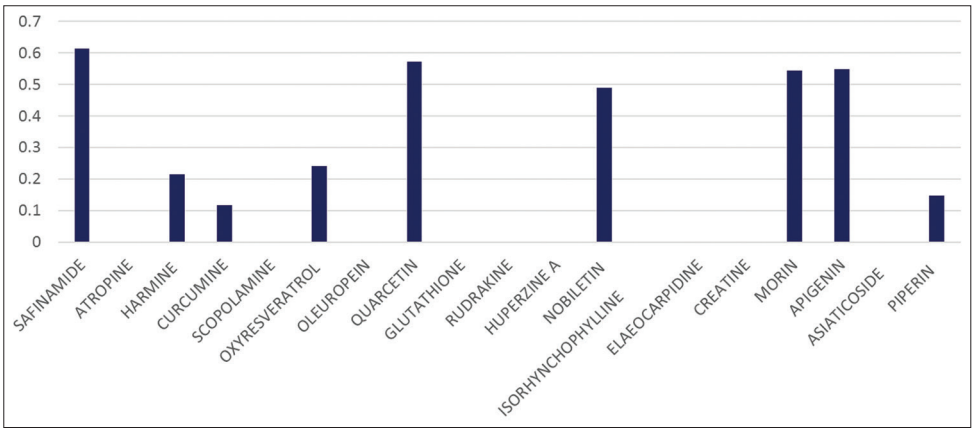

Figure 8: Monoamine oxidase inhibitor activity with respect to safinamide

In MAO activity, safinamide has the highest activity than phytochemicals. Quercetin, morin, and apigenin were found to have equivalent MAO inhibitory activity followed by nobiletin, oxyresveratrol, harmine, piperin, and curcumin, respectively [Figure 8]. In contrast, safinamide has less DA-releasing stimulant activity when compared with phytochemicals. The pattern followed by phytochemicals for DA-stimulant activity is curcumin >oxyresveratrol >apigenin >glutathione =creatine >quercetin =nobiletin =morin [Figure 9].

In case of NMDA receptor antagonist activity, both safinamide and phytochemicals have no significance activity except huperzine A, which is found to have 0.5 Pa value. Some of phytochemicals have activities as follows: creatine >oxyresveratrol >apigenin >curcumin =glutathione =quercetin =morin >harmine [Figure 10].

Standard drug safinamide was found to have no antiparkinsonian activity. Of phytochemicals, scopolamine and atropine have the highest antiparkinsonian activities.

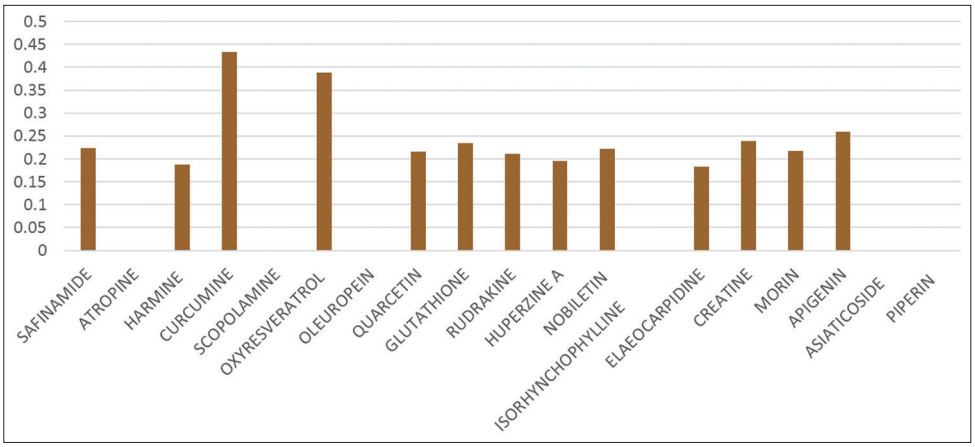

Figure 9: Dopamine-releasing stimulant with respect to safinamide

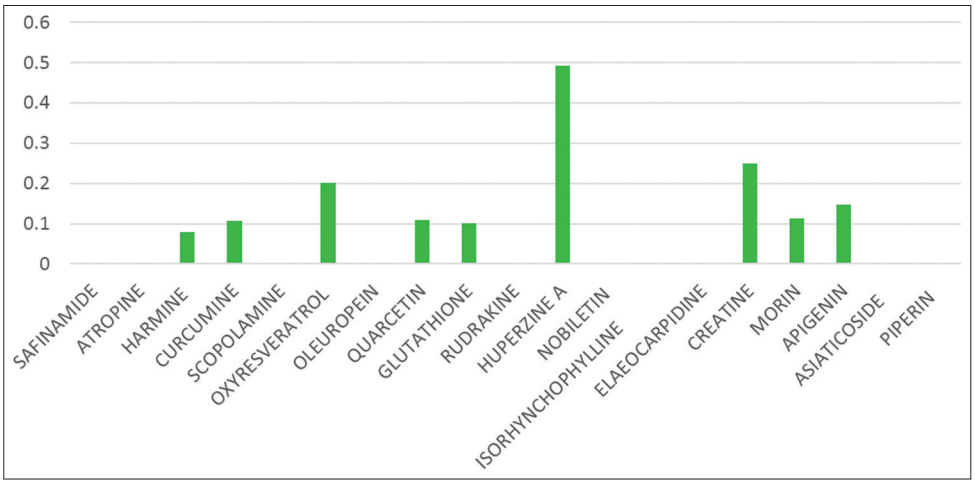

Figure 10: N-methyl D-aspartate receptor antagonists with respect to safinamide

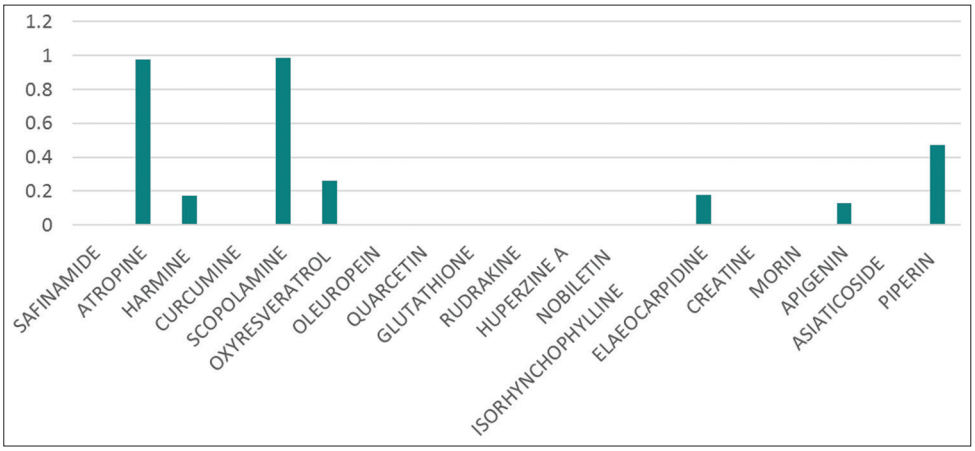

Figure 11: Antiparkinsonian activity with respect to safinamide

Piperin was also found to have antiparkinsonian activity up to some extent. Elaeocarpidine, harmine, and oxyresveratrol have values closer to each other [Figure 11].

CONCLUSION

From the above study, PASS, an online software, helps in the prediction of pharmacological action of the phytochemicals. This software also aids 3R's ethics for animal usage before *in vivo* evaluation, which reduces money, time, as well as number of animals.

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