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"ANALYSIS OF BIOACTIVE CONSTITUENTS FROM MURRAYA KOENIGII L. BY HRLC-MS TECHNIQUE"

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ABSTRACT
The study is aimed to review all medicinally active phytoconstituents and their chemical compositions present in ethanolic leaves extract of Murraya Koenigii and to support the scientific logic behind its medicinal properties with the investigation and study of these phytoconstituents by HRLC-MS analysis which conveniently provide the presence of all the possible active constituents with their exact mass, m/z ratio, structure and relative abundance in the extract. In this study all over fifty constituents are reported but out of them only relative to the study and its therapeutic values are selected for the further judgment like trolamine, 3-guanidinopropionic acid, ethosuximide, metaraminol and vigabatrin etc.

KEYWORDS: Murraya Koenigii, HRLC-MS, trolamine, 3-guanidinopropionic acid, ethosuximide, metaraminol, vigabatrin.

INTRODUCTION
Murraya Koenigii is commonly known as curry leaf and traditionally it is known as a rich source of medicinal values. It is also known for its nuetraceutical properties and applied externally to relief from the bites of poisonous animals.[1] It is also known for hypoglycemic effects in type 2 diabetes milletus.[2] Recent researches reveals that that the extract of Murraya Koengii reveals many therapeutic activities like anti-ulcer, anti-microbial, anti-bacterial, analgesic, anti-inflammatory and chemo protective activity.[3] It is having tremendous history of using from long time in ayurvedic medicine. As per its literature survey, it becomes more interesting to find out all possible phytoconstituents responsible to impart the therapeutic properties and helping it to gain ideal position as medicinal herb in herbalism. The medicinal properties of Curry leaves are due to presence of certain constituents which are inconsistent and variable.[4] The herb is also studied for lipid profile, glaciated proteins and amino acids in non-insulin-dependent diabetic patients.[5]

Currently it is having wide scope to investigate the Murraya Koenigii because of regular uses of its leaves in Indian culinary recipies. It needs to study its proper taxonomic status and different biological activities. Since, from the ancient times in India, it is believed that the herb is having potential to combat various diseases so it is commonly used in many Indian recepies.

Materials and experimental part
The plant material was collected from the local market Kharghar and was authenticated from the expert of botanical field. The fresh plants leaves were washed under running tap water then with distilled and leaves were separated out by scissor. Then the air dried leaves were homogenized to fine powder and stored in air tight bottles. 10 gms of sample powder was dispersed in 50 ml of ethanol and the solution was left to stand at room temperature for twenty four hours, then it was filtered and filtrate was used for the HRLC-MS analysis which was carried out at SAIF, IIT, Bombay.

Table 1: List of active constituents present in the extract of Murraya Koenigii identified by HRLC-Ms anlaysis.

S/N	Name of the compound	Molecular Formula	m/z value
1	Trolamine	$C_6H_{15}NO_3$	150.1
2	3-Guanidinopropionic acid	$C_4H_9N_3O_2$	132.07
3	Ethosuximide	$C_7H_9NO_3$	138.05
4	Metaraminol	$C_9H_{13}NO_2$	150.09
5	Vigabatrin	$C_6H_{11}NO_2$	130.08

Relative compounds with their mass spectrum and peak values are represented below.

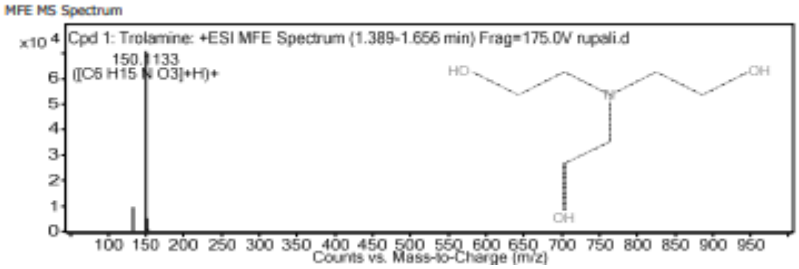

Fig. 1: Mass spectrum of Trolamine.

Table 2: Showing MS Spectrum peaklist of Trolamine.

m/z	Abundance	Formula	Ion
150.1133	70865.12	$C_6H_{15}NO_3$	$(M+H)^+$
151.1163	5014.22	$C_6H_{15}NO_3$	$(M+H)^+$
152.1178	699.95	$C_6H_{15}NO_3$	$(M+H)^+$

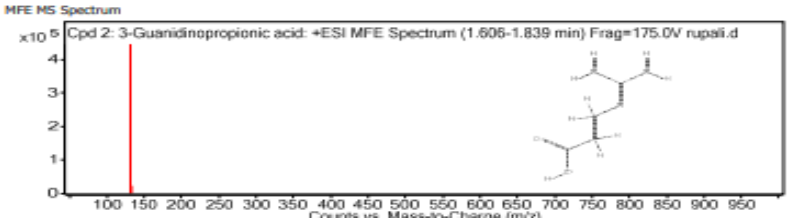

Fig. 2: Mass spectrum of 3-Guanidinopropionic acid.

Table 3: Showing MS Spectrum peaklist of 3-Guanidinopropionic acid.

m/z	Abundance	Formula	Ion
132.0779	445209.66	$C_4H_9N_3O_2$	$(M+H)^+$
133.0803	21931.4	$C_4H_9N_3O_2$	$(M+H)^+$

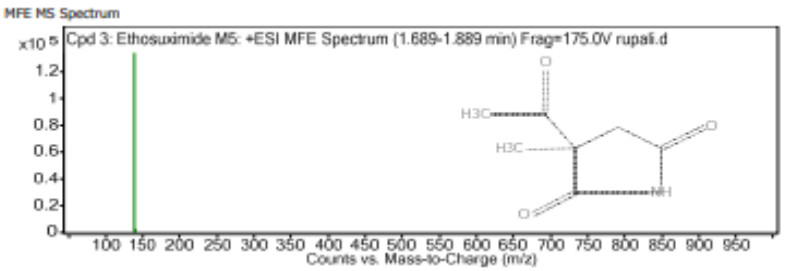

Fig. 3: Showing mass spectrum of Ethosuximide M5.

Table 4: Showing MS Spectrum peaklist of Ethosuximide.

m/z	Abundance	Formula	Ion
138.0558	133915.9	$C_7H_9NO_3$	$(M+H)^+$
139.0527	31979.92	$C_7H_9NO_3$	$(M+H)^+$
140.056	2792.15	$C_7H_9NO_3$	$(M+H)^+$

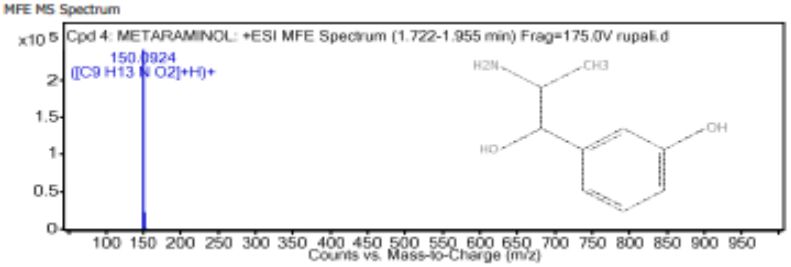

Fig. 4: Showing MS Spectrum peaklist of Metaraminol.

Table 5: Showing MS Spectrum peaklist of Metaraminol.

m/z	Abundance	Formula	Ion
150.0924	240468.17	$C_9H_{13}NO_2$	$(M+H)^+$
151.0953	21891.42	$C_9H_{13}NO_2$	$(M+H)^+$
152.0977	1464.59	$C_9H_{13}NO_2$	$(M+H)^+$

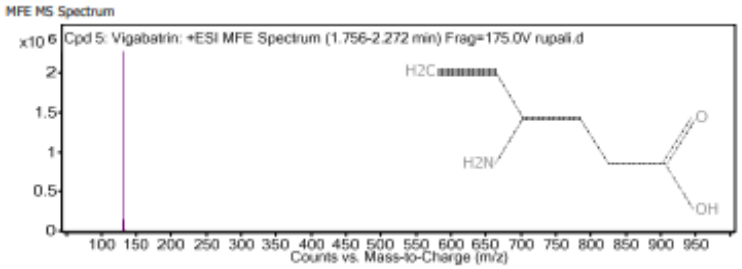

Fig. 5: Showing mass spectrum of Vigabatrin.

Table 6: Showing MS Spectrum peak list of Vigabatrin.

m/z	Abundance	Formula	Ion
130.087	2270806.5	$C_6H_{11}NO_2$	$(M+H)^+$
131.0905	139125.15	$C_6H_{11}NO_2$	$(M+H)^+$
132.0917	11763.36	$C_6H_{11}NO_2$	$(M+H)^+$

RESULT AND DISCUSSION

The study hereby reveals the presence of few medicinally active constituents which are already the backbone of existing synthetic drugs as well as are found to be present in many ointments, drugs and medicines. In this study only few of them are mentioned to support the scientific logic behind the use of Murraya Koenigii as prime medicinal herbs. HRLC-MS analysis hereby shows the presence of trolamine which is an active ingredient of anti-inflammatory and analgesic cream applied on delayed onset muscle soureness.[6] It is also reported to show physical and chemical enhancer effect on rat skin.[7] It is also found in topical analgesic in powder, capsule, cream and ointment form etc.[8] Trolamine herby is observed showing m/z value at 150.1133.

Traditionally Murraya Koenigii is also known for anti-diabetic effect and it is showing the presence of 3-Guanidinopropionic acid somewhere supports the presence of this therapeutic value as 3-Guanidinopropionic acid is well known and reported for its biological activity similar to anti-diabetic agent.[9] It can also capable to increase fatigue tolerance of skeletal muscles.[10] 3-Guanidinopropionic acid is observed at m/z value at 132.07. Ethosuximide with m/z value at 138.05; belong to the anticonvulsant type of medication. The pharmacodyanmics hence can suggest future development in the extract of Murraya Koenigii which is also reported to show the presence of some other constituents like ethosuximide M5, metaraminol with m/z value at 150.09, and vigabatrin with m/z value at 130.08 etc. Indirectly literature survey reveals that these phytoconstituetns are also present in many marketed drugs so it is interesting to study the medicinal activity of individual one. The chemical constituents present in extract vigabatrin with molecular formula $C_6H_{11}NO_2$ and relative abundance 2270806.5 is a kind of drug used in children with intractable epilepsy.[11]

Murraya Koenigii is also reported to have antioxidant properties helping for neutralizing free radicals[12] which is recently more essential for scientific and pharmaceutical purposes. The free radicals are actually occupied for holding responsible to cause many ailments like diabetes, cardiovascular disease and Alzheimer's disease.[13] Due to presence of these kinds of phytochemical, the medicinal herbs are widely found as an alternative source for the treatment of various ailments.[14] Hence it is essential to investigate all phytochemical to formulate exactly the herbal based products and to list out the medicinal values.

CONCLUSION

The current herb is rich source of medicinally active phytoconstituents and hence can be good alternative or the basis for the development and synthesis of novel drugs for the treatment of many diseases. It becomes more interesting to carry out further investigation of Murraya Koenigii in detail and it can also consider as a promising herb not only for drug synthesis but also in many health drinks, herbal tea, cuisines, many health promoting products, immunity boosters, anti-inflammatory and analgesic cream. It can also assume as promising medicinal herb for the treatment of diabetes or somewhere as a supportive medicine for diabetes as it is well known to have potential for decreasing blood sugar level if it is regularly consumed. So, there is wide scope to investigate the medicinal herb Murraya Koenigii further through scientific way.

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