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Phytochemical Screening of *Murraya koenigii* (L.) Spreng

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Abstract: *Murraya koenigii* (L.) Spreng., commonly known as curry leaf tree is well known in Ayurvedic medicinal system for its varied pharmacological activities like anticancer activity, antioxidant activity, anti-inflammatory activity, anthelmintic activity, antidiabetic and antimicrobial activity. The leaves of this tree had been commonly used to enhance the flavour in culinary purposes for its specific taste and aroma. It's Stem part is commonly used as datun for oral health care purposes.The secondary metabolites present in the plant are also known for its different pharmacological activities. The present study focuses on preliminary qualitative phytochemical Screening and analysis from the hydroalcoholic extracts of root, stem and leaves part which detected the presence of alkaloids, phenols, flavonoids, terpenoids, steroids and tannins in the prepared hydroalcoholic extracts using Soxhlet extraction method.

Keywords: *Murraya koenigii* (L.) Spreng; Soxhlet extraction; Secondary metabolites; Phytochemical Screening.

1. INTRODUCTION AND PHARMACOLOGICAL PROPERTIES OF *MURRAYA KOENIGII* (L.) SPRENG.

It had been noted by WHO that 80% of the population in the developing countries rely on plant based natural products for their primary health care needs (Gahlawat DK, *et al*, 2014). *Murraya koenigii* (L.) Spreng. plant, a medicinal herb, which is well known in Ayurvedic system of medicine, had been widely used in India and other Asian countries as a spice and condiment in culinary purposes. It had been known to possess a varied range of biological activities too (Bhandari PR, 2012). *Murraya koenigii* (L.) Spreng. belongs to the plant family Rutaceae and is native to India and also found in various regions of Asian subcontinent (Verma, S, 2018). In the traditional medicinal system, it is widely as a cure in various lifestyle diseases and disorders. It is used as a febrifuge, tonic, antidiabetic, antidiarrheal, anti-obesity and as a flavor enhancer in varied culineries (Kumari B, 2018). Varied mineral elements such as iron, calcium, magnesium, Zinc, Sodium had been found to be present along with varied vitamins such as Vitamin A, Vitamin B1, Vitamin B2, Vitamin B3 and Vitamin E which suggests that the leaves can be used as a supplement for nutrient scarcity (Igara CE *et a*l., 2016).

Murraya koenigii (L.) Spreng. possess varied pharmacological properties which are as follows:

Antioxidant property

Maryam Zahin et al. demonstrated that the benzene fraction extract showed the highest content of phenolic compounds as well as showed maximal antioxidant activity in the three experimental assays like DPPH assay, FRAP assay and CUPRAC assays which shows that the exogenous supply of antioxidants in the form of *Murraya koenigii* (L.) Spreng. bio-actives would further help in quenching the endogenous reactive oxygen species generated in the body (Zahin M. *et al*, 2013). It had also been found that in the methylene chloride extract of curry leaves the presence of various antioxidant carbazole alkaloids had been found of which the DPPH assay showed the efficacy of the phytochemicals bismurrayafoline E, euchrestine B, mahanine were found to be the highest (Tachibana Y*et al*, 2001)The essential oils in *Murraya koenigii* (L.) Spreng leaves and stem had also been found to posses moderate antioxidant potential when studied through DPPH assay experiment (Iqbal Z*et al*., 2017). A carbazole alkaloid Girinimbine present in the stem bark of *Murraya koenigii* (L.) Spreng. had also been found to posses antioxidant capacity which could be compared to the antioxidant capacity of alpha tocopherol. It had been also found to exhibit anti tumor activity by the inhibition of the expression of EA-EBV virus in the Raji cells (Kok YY*et al*., 2012).

Anti-inflammatory potential

The methanolic extracts of *Murraya koenigii* (L.) Spreng. leaves had been found to posses analgesic and anti-inflammatory activity in acute inflammation in dose dependent manner when experimented on male albino rats (Gupta S*et al*, 2010). The carbazole alkaloid, girinimbine had been found to possess anti-cancer and anti-inflammatory potential by apoptosis induction in HT-29 colon cancer cells and a dose dependent anti-inflammatory activity by inhibiting the nitric oxide production along with reduction in pro-inflammatory cytokine levels (Iman V*et al*, 2017).

Antimicrobial activity

The compound Murrayamine-J and koeninmbine isolated from the crude CHCl3 : MeOH extract of stem and leaves had been found to posses antimicrobial activity against the microorganism *Staphylococcus aureus* whereas the compounds like girinimbine and 1-hydroxy-7-methoxy-8-(3-methylbut-2-en-1-yl)-9*H*-carbazole-3-carbaldehyde had been found to posses potential anti-microbial activity against the *Bacillus cerus* (Nalli Y*et al*., 2018)The chloroform extracts of the roots of plant *Murraya koenigii* (L.) Spreng. shows

inhibitory potential against the microorganisms *A. niger*, *P.aeruginosa and C. albicans* at lower concentrations as well (Vats M*et al*., 2011). *Murraya koenigii* (L.) Spreng. bark's ethanolic extract had been found to possess ant-microbial activity against the microorganisms *S.aureu*s, whereas the leaf ethanolic extracts showed highest anti-microbial activity against the microorganism *P. vulgaris* (Akula P *et al*., 2016)

Antidiabetic activity

The chloroform extract of the leaves of *Murraya koenigii* (L.) Spreng. had been found to exhibit hypogycemic effect of the alloxan induced diabetes in the rats, which had been due to the protective effects such as preservation of pancreatic cell integrity and showing a decreased oxidative stress in rats (Vijayanand S., 2015). The ethanolic extract of the leaves had been also found to exhibit decreased blood sugar levels in dose dependent manner which also showed a decrease in depression levels in the diabetic patients (Tembhurne SV *et al*., 2017). The ethanolic extract and the nano-particles found in *Murraya koenigii* (L.) Spreng. leaves also showed the reduced blood glucose levels in rats (Handayani R *et al*, 2017). The carbazole alkaloid mahanimbine, isolated from *Murraya koenigii* (L.) Spreng. leaves exhibited hypolipidemic and anti-diabetic effect in the diabetic rats which shows that the compound can be useful in management of diabetes (Dineshkumar B *et al*., 2010).

Anticancer activity

The hydroalcoholic extract of *Murraya koenigii* (L.) Spreng. leaves had been found to exhibit anti-cancer activity against the breast cancer cells MDA-MB-231 through the proteasome inhibitory activity and induction of apoptosis in vitro. Reduced tumor size had also been observed in mice (Noolu B *et al*., 2016). Leaf extracts from different study locations had also been found to possess inhibitory potential against the breast cancer cells proving its chemopreventive potentiality (Ghasemzadeh A *et al*., 2014). The carbazole alkaloids mahanine, murrafoline-I and pyrafoline-D had been found to exhibit cytotoxic activity against the human lekemia cell line HL-60 by the caspase-3 pathway actvation and mitochondrial dysfunction (Ito C*et al*., 2006).

Anthelmintic activity

The aqueous and ethanolic extracts of *Murraya koenigii* (L.) Spreng. had been found to exhibit anthelmintic activity against the organism *Pheretima posthuma*at the highest concentration of 100 mg/ml (Sharma US et al., 2010). The methanolic extract of *Murraya koenigii* (L.) Spreng. leaves had been found to posses anthelmintic activty against the *Haemonchus contortus* worms in the larval motility assay and egg hatch assay (Sujith S *et al*., 2018).

2. MATERIALS AND METHODOLOGY

2.1Collection and Preparation of Plant Material for Preparation of Extract

The root, stem and leaves part of the plant was collected. The collected plant material was washed with distilled water thoroughly and air dried at room temperature. The collected, washed and dried plant materials was ground into powder form using mixture grinder and stored in air tight containers for further experimentation.

2.2Extract Preparation Using Soxhlet Apparatus

10 grams of each sample in powder form was taken for its extract preparation using Soxhlet extraction method (Rajhans S*et al*., 2019). The sample had been finely packed in the filter paper and kept in the thimble part of the extractor. Solvent system of 70:30, methanol: water, was chosen for extraction and 100 ml of the solvent system was taken in the round bottom flask. The heating mantle, round bottom flask, Soxhlet apparatus and condenser were connected to the running tap water and was assembled for the further extraction. The heat from the heating mantle rose up to the Soxhlet apparatus in form of vapor wherein, after condensation, droplets of the solvent system dripped down from the Soxhlet apparatus where the sample had been kept for extraction from the sample. The siphon tube of apparatus was then filled with the extract which further dripped down to the round bottom flask. This process was repeated again and again until all the necessary materials were extracted from the solid part of the sample into the organic solvent in the round bottom flask (Rajhans S *et al*., 2019). The extract accumulated in the round bottom flask was brought to normal room temperature and were then filtered using Whatman filter paper number-1 and then transferred and poured into clean petriplates. The prepared hydroalcoholic extracts of roots, stem and leaves were air dried at room temperature in the petriplates.

2.3Phytochemical Screening of the extracts

Phytochemical screening of the hydroalcoholic extract of Root, Stem and Leaves parts of *Murraya koenigii* (L.) Spreng. had been done which revealed the results in Table 1.In the screening of root extracts of *Murraya koenigii* (L.) Spreng. by Vats M., the presence of steroids, alkaloids and flavonoids had been observed with different solvent systems (Vats M*et al*., 2011)

	Root	Stem	Leaves
Test for Alkaloids			
Mayer's Test	+	+	+
Wagner's Test	-	-	-
Hager's test	+	+	-
Dragendroff's test	+	+	+
Test for phenols			
$FeCl_3$ test	+	+	+
Lead acetate test	+	+	+
Tests for Flavonoids			
Alkaline Reagent Test	+	+	+
Ammonia test	+	+	+
Lead acetate test	+	+	+
Test for Saponin			
Foam Test	-	+	+
Test for Terpenoids			
Salkowaski's test	+	+	+
Test for Triterpenoids			
Salkowaski's test	+	+	+
Test for Anthraquinone			
Borntrager's test	-	-	-
Test for Phlobatannin			
Precipitate test	-	-	-
Test for Glycoside			
Bromine water test	+	+	+
Fehling Test	+	+	+
Test for Coumarin			
NaOH + chloroform test	+	+	+
Test for Steroids			
Salkowaski's test	+	+	+
Test for Tannin			
Lead acetate test	+	+	+

Table: 1 Preliminary Phytochemical Screening of Root, stem and Leaves extracts of *Murraya koenigii* (L.) Spreng. (+ indicates positive results & - indicates negative results)

The phytochemical screening for varied phytoconstituents present in the root , stem and leaves part of the plant had been carried out on the basis of reviewed papers by different authors related to their publications on phytochemical screening. The protocols followed for certain tests were, from the research articles by, Verma, D. & Shrivastava, K. 2018,L Sophiyamole. *et al*.,2017, Sasikala M. and Sundaraganapathy R., 2017, A kumar *et al* 2016, RK Bargah *et al* 2015, K.Sahira Banu & L. Cathrine, 2015, S. Pendli *et al* 2014, S C ugochukwa *et al.* 2013, S. Sharmila *et al*., 2013, ME Halilu *et al*. 2012 and KP Salna *et al*., 2011.

3. RESULTS AND DISCUSSION

***3.1*Extract Preparation for phytochemical analysis**

For phytochemical analysis 50 mg of air dried extracts of root, stem and leaves was taken and dissolved in 50 ml of the hydroalcoholic solvent system

***3.2*Preliminary phytochemical screening**

In a study by Sophiyamole L. it had been observed that in MK leaves, the phenols,flavonoids,tannins, saponins, terpenoids and alkaloids had been found to be present [25]. Vijayvargia P. and Vijayvargia R. carried out the preliminary phytochemical analysis of *Murraya koenigii* (L.) Spreng. in different solvent systems. Alkaloids, triterpenoids and phytosterols had been found to be present in the stem and leaves part of the *Murraya koenigii* (L.) Spreng (Vijayvargia P. & Vijayvergia R., 2016).

Presence of Tannins, alkaloids, steroids, tritepenoids and flavonoids had been found to be present in the *Murraya koenigii* (L.) Spreng. leaves and twings part's crude ethanolic extract in the phytochemical investigations by Kusuma IW (Kusuma IW *et al*., 2011).

For the detection of alkaloids, Mayer test, Wagner test, Hager test and Dragendroff test had been performed for the root stem and leaves extract. The Mayer test and

Dragendroff test were found to be positive for all the three extracts whereas, the Wagner test had been found to be negative for all extracts. Furthermore Hager test showed negative results for leaves but positive for root and stem extracts. For the detection of phenols $FeCl_3$ test and Lead acetate test showed positive results for all extracts. Alkaline reagent test, ammonia test and lead acetate test for detection of flavonoids showed positive results for all the three extracts. Foam test for saponins showed negative results for roots whereas it showed positive results for stem and leaves. Salkowaski test performed for the detection of steroids, terpenoids and triterpenoids showed positive results for all the three extracts. Test for anthraquinone and Phlobatannin showed negative results for all the three extracts. Bromine water test and Fehling test for glycosides, showed positive results for all the three extracts. The test for coumarin, NaOH + chloroform test showed positive results for all three sample extracts. The lead acetate test for detection of tannins also showed positive results for all the three sample extracts.

4. CONCLUSION

The present study reveals the results of preliminary phytochemical screening from the hydroalcoholic extracts of root, stem and leaves of the *Murraya koenigii* (L.) Spreng. plant had been found to be rich in phyto-constituents such as alkaloids, phenols, flavonoids, terpenoids, steroids, glycosides, coumarins and tannins. Further *In vitro* and *In silico* research on the individual phytochemicals and its extraction can provide more insights into the varied pharmacological roles of different phytochemicals. Computational methods such as machine learning approaches using different algorithms could help in providing the insights into the different activities of diverse phytochemicals.

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