

In-silico analysis of compounds characterized from *Murraya koenigii* against cancer

Satyam Ray¹, Preetha Bhadra*

Satyam Roy: 2nd Year, B.Sc, Centurion University of Technology and Management, Odisha.

Preetha Bhadra: Assistant Professor, Department of Biotechnology, M.S.Swaminathan School of Agriculture, Centurion University of Technology and Management, Odisha

Abstract

Murraya koenigii is a culinary important plant of Indian origin, and also been a component of many formulations used in the Ayurvedic system of medicine since many centuries. A scrutiny of literature reveals some notable pharmacological activities of the plant. Carbazole alkaloids which are abundantly present in the leaves, fruits, roots and bark of this plant, have been reported for their antidiabetic, anticancer, antibacterial, anti-nociceptive and antioxidant activities. Besides these activities, the plant is described to have a wide array of therapeutic activities. Phytochemistry and pharmacology of this plant necessitates a comprehensive review of its prospects as an important therapeutic agent focusing on its anti carcinogenic activities. The current review provides a detailed report of the phytochemical, pharmacological, clinical and pre-clinical works carried out on this culinary plant and also throws light on its therapeutic prospects.

Keywords: culinary, anti-nociceptive, phytochemistry, therapeutic, pharmacology, carcinogenic

Introduction

Murraya koenigii commonly known as curry leaves or curry patta in Indian dialects which represents more than 150 genera and 1600 species. Curry leaves belongs to the family rutaceae. *M. koenigii* is a highly valued plant for its aroma and medicinal value. *M. koenigii* contains a number of chemical constituents that interact to elicit their pharmacodynamic response. It grows through the Indian subcontinent and has wide culinary effect and is one of the main components of formulations in the traditional ayurvedic system^[1]. There are different forms of *Murraya koenigii* due to which they are found as the useful plant such as extract, essential oil, or directly used due to the presence of following active constituent bismahanine, murrayanine, murrayafoline-A, bi-koeniquinone-A, bismurrayquinone, mukoenine-A, mukoenine-B, mukoenine-C, murrastifoline, Murrayazolinol, murrayacine, murrayazolidine, murrayazoline, mahanimbine, girinimbine, koenioline, xynthyletin, koenigine-Quinone A and koenigine-Quinone B for therapeutic purpose by folk people^[2-5]. These natural products have been shown to have antioxidant properties and are capable of scavenging free superoxide radicals, thus providing antiaging benefits as well as reducing the risk of cancer. It has been found that flavonoids reduce the lipid and glucose levels in blood and support the human immune system^[6-7]. The health-promoting effects of flavonoids are the result of their ability to induce the activity of protective enzyme systems^[8]. Several studies have suggested that flavonoids such as catechin and quercetin are able to control the growth of cancer cells in human body.

Chemical constituent:

Murraya koenigii is very rich source of organic compounds with different chemical composition such as alkaloids, flavonoids carbohydrates, and sterol is present in the plant extract prepared in solvents such as petroleum ether, ethyl acetate, chloroform, ethanol and water^[10,11-14]. For the confirmation of the phyto-constituents in the plant extract, various numbers of tests were performed: The presence of alkaloids was confirmed by using Mayer's reagent, which shows formation of white or cream colored precipitates in the extract of *Murraya koenigii*.^[15] Phenolic compounds were confirmed by formation of white precipitate by the addition of few drops of 5% lead acetate solution to alcoholic extracts of *Murraya koenigii*.^[15] The presence of flavonoids is detected by Yellow coloration of filter paper by dipping in ammoniated alcoholic upon the extract.^[15] Presence of Saponins is considered when the extract showed honey comb like frothing formation after giving a shake with sodium bicarbonate.^[15] The presence of proteins and free amino acids is indicated by the conducting the following tests i.e., Millons, Biurets and Ninhydrins test. vi. Presence of sterol and triterpenes are indicated by alcoholic extract which was shaken with chloroform and few drops of acetic anhydride along with few drops of concentrated sulphuric acid from the side of the tube form the blue to brick red coloration.^[16] The essential oil composition of *Murraya koenigii* was studied and then presence of D-Sabinene, D- α -Terpinol, di- α -phellendrene, D- α -pinene, caryophyllene and dipentene^[17] and the property of *Murraya koenigii* oil is explained in the following table.^[15]

Sl. no	Property	Value
1.	Specific Gravity (25°C)	0.9748
2.	Refractive Index (25°C)	1.5021
3.	Optical Rotation (25°C)	+ 4.8
4.	Saponification Value	5.2
5.	Saponification after Acetylation	54.6
6.	Moisture	66.3%
7.	Protein	6.1%
8.	Fat (Ether Extract)	1.0%
9.	Carbohydrate	18.7%
10.	Fibre	6.4%
11.	Mineral Matter	4.2%
12.	Calcium	810Mg/100 G
13.	Phosphorus	600Mg/100 G
14.	Iron Of Edible Portion	3.1Mg/100 G
15.	Carotene (As Vitamin A)	12 600I μ /100 G
16.	Nicotinic Acid	2.3Mg/100 G
17.	Vitamin C	4 Mg/100 G
18.	Thiamine And Riboflavin	Absent

Determination of Anticancer Activity:

MTT (3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium Bromide) Assay

The assay was conducted as follows: cancer cells were seeded in 96-well plates at a density of 1×10^4 cells/well in 100 μ L RPMI. At 24 h after seeding, the medium was removed and the cells were incubated for 3 days with RPMI in the absence or presence of various concentrations of curry leaf extracts. Extracts concentrations used ranged between 20, 40, 80, 160, 320, and 640 μ g/mL. After incubation, 20 μ L of MTT reagent was added into each well. The plate was incubated again in a CO₂ incubator at 37°C for 4 h. The resulting MTT-products were determined by measuring the absorbance at 570 nm using ELISA reader. ^[19] Each point represents the mean of triplicate experiments. The cell viability was determined using the formula.

Statistical Analysis:

All analytical values shown represent the means of three replicates. Data were analysed using analysis of variance by Statistical Analysis System (SAS 9.0). Mean separation test between treatments was performed using Duncan multiple range test and a *P* value ≤ 0.05 was regarded as significant. ^[18]

Other studies related to anti-carcinogenic properties

Girinimbine, acarbazole isolated from the bark of *Murrya koenigii* significantly induced programmed cell death in HepG2 cells suggesting the necessity for further evaluations in preclinical human hepatocellular carcinoma models. ^[20] The results from the study conducted by Bhattacharya *et al.* 2010 provides evidence for the involvement of death receptor mediated extrinsic pathway of apoptosis in mahanine-induced anticancer activity in MOLT-3 cells, but not in K562 cells which are deficient in Fas/FasL. ^[21] Furthermore, 3 carbazole alkaloids mahanine, pyrayafoline and murrafoline, showed significant activity against HL-60 cells by inducing apoptosis through of capsase-9/capsase-3 pathway, through mitochondrial dysfunction. ^[22] Down regulation of cell survival factors by activation of capsase-3 through mitochondrial dependent pathways and disruption of cell cycle progression could be an additional mechanism. ^[23] The mean number of neoplasms in the colon and intestines were significantly low as demonstrated by morphological and histological studies in the *Murrya koenigii* treated animals. ^[24] The methanolic extract of *Murrya koenigii* leaves demonstrated a significant increase in the phagocytic index by the rapid removal of carbon particles from blood stream. It also demonstrated an increase in the antibody titer against ovalbumin and protection against cyclophosphamide-induced myelosuppression.

Some major chemicals specifically showing anti-carcinogenic effects.

Conclusion:

Murrya koenigii is a leafy medicinal as well as green leafy plant that belongs to family Rutaceae. The various pharmacological activities of the plant has been seen such as activity on Anti diabetic, cholesterol reducing property, antimicrobial activity, antiulcer activity, Antioxidative property, cytotoxic activity, antidiarrhoea activity, anti-cancer activity with many other phagocytic activity. In this study we specifically took interest over the anti-carcinogenic properties. The chemical composition of the *Murrya koenigii* consists of certain alkaloids which are proved to express certain anti-cancerous activities upon in-vitro exposure to cancer cells. Thus Curry leaves merits further phytochemical, pharmacological and clinical investigations for development of an effective natural plant.

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