

TOTAL PHENOLIC CONTENT IN AQUEOUS EXTRACTS OF *MURRAYA KOENIGII* STORED FOR SIX MONTHS AT ROOM TEMPERATURE AND AT 4°C

Thilageswary Kumutharanjan , Sivakanesan Ramaiyah and Vasanthy

Arasaratnam

* Unit of Siddha Medicine, University of Jaffna

** Department of Biochemistry, Faculty of Medicine, University of Peradeniya

*** Department of Biochemistry, Faculty of Medicine, University of Jaffna

Abstract- Oxidative stress is inevitable. Preventive actions should be taken to minimize the oxidative damages to biomolecules which may lead to health problems. *Murraya koenigii* leaves are reported to possess potent antioxidant properties. This study was initiated to evaluate the Total Phenol Content (TPC) and Antioxidant Activity of aqueous extracts of *M. koenigii*. Leaves were dried to constant weight, powdered and sieved. TPC was estimated in the cold and hot aqueous extracts of the *M. koenigii* leaf powder stored at room temperature and at 4°C in monthly interval for six months. The initial TPC of cold and hot extracts of *Murraya koenigii* leaf powder was 22.6 and 25.08 µg TAE/g dry weight respectively. When the *Murraya koenigii* leaf powder was stored at room temperature for 6 months, TPC of cold and hot water extracts was 11.02 & 11.71 µg TAE/g dry weight respectively while the TPC of cold and hot water extracts of the leaf powder stored at 4°C for six months respectively was 12.31 & 12.84 µg TAE/g dry weight. Extraction of antioxidant activity was better with hot water than with cold water. TPC of *Murraya koenigii* powder decreased when stored both at room temperature and at 4°C. Both cold as well as hot extracts exhibited antioxidant activity even after storing for 6 months. At 3 months the decline in the TPC of the powder stored at Room temperature is higher than that stored at 4°C. In the Siddha Medicine the lifespan of 'Chooranam' which is prepared from herbs is used for 3 months. Hence the leaf powder can be used for the 'Chooranam' preparation. However freshly prepared leaf powder is recommended for the preparation of the 'Chooranam'.

Index Terms- antioxidant activity, *Murraya koenigii*, SiddhaMedicine, Total Phenoic content

I. INTRODUCTION

Nature has been a potential source of a variety of plants with diversified medicinal values and herbs were used for the treatment various ailments for thousands of years and plant based drugs continue to play an essential role in the primary health care of 80% of the worlds Under Developed and Developing Countries (Grover and Vats, 2001). Plant drugs are frequently considered to be less toxic and free of side effects than synthetic ones (Wang and Ng, 1999). *Murraya koenigii* belongs to the Family of Rutaceae (Tamil- 'Kariveppilai'; Sinhala-'Curripincha'; English- 'Curry leaves'). *Murraya*

koenigii leaves are used as flavoring agents and condiments, and used in Traditional Medicine for the treatment of various metabolic and infectious diseases. The leaves, barks and the roots are used intensively in Indigenous system of medicine from ancient time as a tonic for stomach ache, stimulant and carminative (Anonymous, 1998; Murugesumuthaliyar 1998; Prajapati *et al.*, 2003). Phytochemical screening of *M. koenigii* revealed the presence of some vitamins, carbazole alkaloids, triterpenoids, phenolic compounds, and mineralss such as iron, calcium, zinc, and vanadium, etc. (Narendhirakannan *et al.*, 2005; Chakraborty *et al.*, 1965; Kong *et al.*, 1986). In addition it contains antioxidant activity (Tachibana *et al.*, 2003) and several biological activities such as antihypercholesterolemic (Khan *et al.*, 1996a & b) antiinflammatory (Ramsewak *et al.*, 1999) and antidiabetic activity (Tachibana *et al.*, 2003; Tembhurne and Sakarkar 2009c & 2010). *Murraya koenigii* also has its efficacy against colon carcinogenesis (Khan *et al.*, 1996b) and increases the gastrointestinal motility (Khan *et al.*, 1996a & b) have been reported. In Sidha Medicine 'chooranam' is prepared by the physicians which contains plant materials as ingredients. The *Murraya koenigii* leaf powder is one of the ingredients and the leaf powder is prepared and stored for some time for "Chooranam" preparation. To improve the efficacy of the 'chooranam', it is important to retain its active principles intact for efficient bioactivity. Thus in this studies the antioxidant property of *Murraya koenigii* leaf powder after storing for different period was studied, so that under emergency situations the stored *Murraya koenigii* powder can be used for 'Chooranam' preparation.

II. MATERIALS AND METHODS

Plant Material

Leaves of *Murraya koenigii* were collected from Karaveddy Village in Jaffna Peninsula.

Preparation of plant extract

Murraya koenigii leaves were cleaned, washed and dried under shade at room temperature for 10 days. The dried leaves were powered by wooden motor and pestle and sieved with muslin cloth. The powder was divided into 100 mg quantities packed in polythene packets and stored either at 4°C and room temperature. Cold and hot aqueous extract were prepared at monthly interval by suspending 100mg of the powder in 10ml distilled water, where one part was stirred at room temperature and the other part was kept in water bath at 100°C while stirring for 5 minutes. Then these were centrifuged at 10,000

rpm for 10 minutes. Supernatant was analysed for Total phenolic content (Mc Donald *et al.*, 2001). The phenolic content of the sample was expressed as µg of TAE (Tannic acid equivalent)/g of dried powder.

III. RESULTS AND DISCUSSION

Initially the Total Phenolic Content of *Murraya koenigii* leaves powder cold and hot extracts was 22.6 & 25.08 µg TAE/g dry weight respectively (Table 1). The TPC was better extracted with hot water than with cold water and hence when compared with the cold extracts, the hot extracts contained higher TPC (Table 1). Initially the extraction of TPC with hot water was 1.1 times (11%) more than with cold water.

When the powder was stored at room temperature for a month and the TPC was analysed, the cold and hot water extracts contained 20.97 & 22.53 µg TAE/g dry weight respectively. The leaf powder stored at room temperature for six months showed TPC of 11.02 & 11.71 µg TAE/g dry weight respectively in cold and hot water extracts. With time, the TPC of the leaf powder stored at room temperature decreased. After six months of storage at room temperature, TPC in cold extract and hot extract were 50% of the initial TPC content and almost same (Table 1).

When the powder was stored at 4°C for a month and the TPC was analysed, the cold and hot water extracts contained 21.01 & 24.73 µg TAE/g dry weight respectively (Table 1). When compared with the cold extracts, the hot extracts contained higher TPC than cold extract (Table 1). The leaf powder stored at 4°C for six months showed TPC of 12.31 & 12. µg TAE/g dry weight respectively in cold and hot water extracts. With time, the TPC of the leaf powder stored at 4°C decreased. After six month of storage at room temperature, TPC in cold extract and hot extract were almost same (Table 1).

The decrease in TPC of the *Murraya koenigii* leaf powder stored at room temperature and at 4°C was analysed by extracting with cold and hot water. The results indicated that the loss in TPC was not uniform with time (Table 2). When the loss of TPC in *Murraya koenigii* leaf powder stored at room temperature and 4°C were compared, the TPC loss was not significant. TPC of *Murraya koenigii* powder decreased when stored at either room temperature or at 4°C, and also after 3 month the decline percentage was higher than in the first three months.

In Siddha Medicine, the plant materials are used for the 'Chooranam' preparation. Usually the 'Chooranam' is used for 3 months. The loss of TPC from *Murraya koenigii* leaf powder stored at room temperature for 3 months decreased by 21.5 and 20.41% respectively when extracted with cold and hot water. On the other hand when the leaf powder was stored at 4°C, the loss was 12.1 and 10.2% when extracted with cold and hot water respectively.

The loss of TPC at 6 months was compared with the powder stored at room temperature and 4°C and extracted with cold and hot water was 51.2 & 53.3% and 45.53 & 48.8% respectively. Thus the loss of TPC is more when stored at 4°C than at room

temperature. However the difference is statistically not significant.

As the lifespan of 'Chooranam' is considered as 3 month, freshly prepared plant parts are recommended for the preparation. And the 'Chooranam' should be used within 3 month and can be stored at room temperature.

IV. CONCLUSION

This study showed that the *Murraya koenigii* leaf powder could be used for 'Chooranam' preparation immediately after the preparation of the leaf powder. Under emergency situations, the powder stored for three months at room temperature could be used, but not the powder preparation stored for more than 3 months.

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AUTHORS

First Author – Thilageswary Kumutharanjan, Unit of Siddha Medicine, University of Jaffna, tkumutharanjan@yahoo.com

Second Author – Ramaiyah Sivakanesan, Department of Biochemistry, Faculty of Medicine, University of Peradeniya, sivaskanda@gmail.com

Third Author – Vasanthy Arasaratnam, Department of Biochemistry, Faculty of Medicine, University of Jaffna, arva26arva@yahoo.com

Table 1: Total Phenolic Content in cold and hot water extracts of *Murraya koenigii* powder stored at Room Temperature and at 4°C.

Time (Month)	Total Phenolic Content (µg TAE/g dry weight)			
	Room Temperature		4°C	
	Cold water	Hot water	Cold water	Hot water
0	22.60	25.08	–	–
1	20.97	22.53	21.01	24.73
2	18.58	20.61	20.31	23.67
3	17.73	19.95	19.87	22.53
4	16.43	18.03	17.91	18.94
5	13.20	13.57	14.26	14.63
6	11.02	11.71	12.31	12.84

Table 2: Decrease in the Total Phenolic Content of *Murrya koenigii* leaf powder stored at Room Temperature and at 4⁰C . For analysis the TPC was extracted with cold and hot water.

Time (Months)	Decrease in TPC (µg TAE/g dry weight)							
	Stored at Room Temperature				Stored at 4 ⁰ C			
	Cold water extract		Hot water extract		Cold water extract		Hot water extract	
	Loss	Cumulative loss	Loss	Cumulative loss	Loss	Cumulative loss	Loss	Cumulative loss
1	1.63	1.63	2.55	2.55	1.59	1.95	0.35	0.35
2	2.39	4.02	1.92	4.47	0.70	2.65	1.06	1.41
3	0.85	4.87	0.66	5.13	0.44	3.09	1.14	2.55
4	1.30	5.00	1.92	7.05	1.96	5.05	3.59	6.14
5	3.23	8.23	4.46	11.51	3.65	8.70	4.31	10.45
6	2.18	10.41	1.86	13.37	1.95	10.65	1.79	12.24