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Original Research

Determination of Nutritive Value and Mineral Elements of Some Species of Genus *Memecylon* Linn. from Central Western Ghats

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Abstract

Five species of *Memecylon* were investigated for proximate analysis and elemental composition. These species include *M. umbellatum* *M. malabaricum*, *M. talbotianum, M. wightii* and *M. lushingtonii*. Among the proximates *M. wightii* showed highest percentage of ash, crude fiber and carbohydrate. *M. umbellatum* showed high percentage of moisture and crude fat. Crude protein was more in *M. malabaricum*. Nutritive value was highest in *M. lushingtonii* among all the species. Among the macronutrients Na and K, Ca and Mg P and N were highest in *M. malabaricum, M. talbotianum* and *M. umbellatum* respectively. *M. umbellatum* recorded highest values of Cu and Mn where as *M. lushingtonii* and *M. wightii* recorded highest values of Zn and Fe. Among the heavy metals Pb and Cd *M. umbellatum* recorded highest values of Pb and *M. talbotianum* recorded highest values of Cd but both the heavy metals are absent in *M. malabaricum*.

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INTRODUCTION

The most important nutrients present in plants are carbohydrates, oil, proteins, minerals, ascorbic acid and the antioxidant phenols (Ekanayake and Nair, 1998). The trace elements together with other essential nutrients are necessary for growth, normal physiological functioning and maintaining of life. These trace elements must be supplied by food because the body cannot synthesis them. They required by the body in amounts less than 100 mg daily and deficiency of some of these trace elements can lead to diseases (Janab and Thompson, 2002).

Plants have great importance due to their nutritive value and a major source of medicines as they have been found throughout human history (Balick *et al.,* 1996). Human body comprises chemical compounds such as water proteins, fatty acids, nucleic acids and carbohydrates these in turn consists of elements such as carbon, hydrogen, oxygen, nitrogen and phosphorus and may or may not contains minerals such as calcium, iron, magnesium and zinc (Katzmaezyk and Waist, 2004).

Each medicinal plant species has its own nutrient composition besides having pharmacologically important phytochemicals. These nutrients are essential for the physiological functions of human body and they play an important role in satisfying human needs for energy and life processes (Hoffman *et al.,* 1998; Mathews *et al.,* 1999; Dingman, 2002).

Memecylon Linn. belongs to the family Melastomaceae / Memecylaceae and there are about 150 species are reported from different regions of the world. The different floras, taxonomic literatures and other publications were referred and the identified species are recorded.

There are a few reports regarding the medicinal uses of a few species of *Memecylon* Linn. Nadakarni (1976) mentioned the medicinal use of *M. amplexicaule*, *M. angustifolium* and *M. edule*. Kirtikar and Basu (1991) reported the uses of leaves of *M. umbellatum* for the treatment of leucorrhoea and gonorrhea; they also mentioned the application of flowers and shoots of *M. amplexicaule* for the treatment of skin diseases. In addition the root of *M. amplexicaule* acts as ecobolic. The bark of *M. angustifolium* is used as a tonic and refrigerant. Maruthi *et al.* (2000) reported the application of leaves of *M. umbellatum* for the treatment of herpes. It is also reported that *M. edule* is used for contusion and the roots of *M. harmandi* in the treatment of diabetes (Ayyanara and Ignacimuthu, 2005). The crushed leaves extracts of *M malabaricum* is used in the treatment of skin problems. In addition to medicinal uses the wood of *M. umbellatum* is an excellent source of firewood and charcoal. The pulp of the fruit is edible, the leaves and flowers contain dyes and these dyes are employed in the dyeing industry to dye the mats and cotton fabrics (Subhash, 1988). The fruit of *M. edule* has been being used by the tribal community of

Tamil Nadu. Hullihatti *et al.* (2004) studied the antimicrobial activity of leaves of *M. malabaricum*. Amalraj and Ignacimuthu (1998) evaluated the hypoglycemic effect of *M. umbellatum* in normal & alloxan diabetic mice.

MATERIALS AND METHODS

Study Area

Karnataka state is situated between 11°40' and 18°27 north latitude and 74°5' and 78°33' east longitude in the center of western peninsular India. The area of Karnataka comprises of about 1, 92,204 square kilometers. The state is broadly divided into two climatic regions namely southern and northern Karnataka. The southern Karnataka is rich in forests and receives maximum rain during the south west monsoon. The northern Karnataka has comparatively dry region.

Sample Collection

The leaves of *M. umbellatum* Burm. were collected from two different regions of Karnataka. The species was identified with the help of some taxonomic literature [Flora of British India (Hooker, 1894); flora of presidency of Madras (Gamble, 1928), flora of Davanagere District (Manjunath*et al.,* 2002), (Yoganarasimhan *et al.,* 1982), Compendium of Indian Medicinal plants (Rastogi and Mehrohra, 1991), flora of Presidency of Bombay (Cooke, 1903), The forest trees of Travancore (Bourdillon, 1908), flora of south Indian hill stations (Fyson, 1932; Flora of Coorg (Kodagu); (Keshava Murthy and Yoganarasimhan, 1990); Flora of Hassan District; Saldanha, 1976)]. The voucher specimens are deposited as herbarium in the department of Applied Botany, Kuvempu University.

Sample Preparation

The collected leaves were washed thoroughly 2-3 times with running tap water and once with sterile water, shade dried, powdered and stored in airtight bottles for further investigation.

Nutritive Value and Elemental Composition

The components of nutritive value and elemental composition were estimated by following the standard procedures (Indrayana *et al.,* 2005; Deepak Dhyani, 2007 and Gali *et al.,* 1999) at the department of Applied Botany, Kuvempu University and Central Coffee Research Institute, Balehonnur by using Atomic Absorption Spectroscopy (AAS) and Flame Photometer.

Statistical Analysis

Each experiment was repeated for three times. The results were represented with their means, standard deviation and standard error. The correlation matrix has been calculated.

RESULTS

Components of Nutritive Value

The average values of proximate of five species of *Memecylon* Linn represented in table 1. The percentage of ash was highest in *M. wightii* (5.32%) it was followed by *M. lushingtonii* (5.16%), *M. malabaricum* (4.82%), *M. talbotianum* (3.99%) and *M. umbellatum* (3.16%).

The *M. umbellatum* contain highest of 53.99% of moisture and *M. wightii* contain lowest of 46.83% of moisture *M. malabaricum*, *M. talbotianum* and *M. lushingtonii* recorded 48.99, 48.91 and 48.16 % moisture respectively.

The crude fat percentage was highest in *M. umbellatum* (6.08%) and it was followed by *M. lushingtonii* (5.99%), *M. malabaricum* (5.61%), *M. talbotianum* (5.53%) and *M. wightii* (4.95%).

The crude fiber percentage was highest in *M. wightii* (6.08%) and lowest in *M. umbellatum* (3.62%) and *M. lushingtonii*, *M. malabaricum*, and *M. talbotianum* recorded 4.86, 4.32 and 4.16 % of crude fiber respectively.

The percentage of crude protein was highest in *M. malabaricum* (1.87%), and it was followed by *M. wightii*, *M. talbotianum, M. lushingtonii* and *M. umbellatum* recorded 1.86, 1.8, 1.78 and 1.68 % of crude protein respectively.

M. wightii recorded highest of 40.99% of carbohydrate and *M. umbellatum* recorded lowest of 35.05% of carbohydrate *M. talbotianum, M. lushingtonii* and *M. malabaricum* recorded 39.76, 38.87 and 38.65 % respectively.

Table 1: Average values of proximates [moisture, ash, crude fat, crude fiber, crude protein and carbohydrate (in %) and nutritive values (in cal/100gm)] of five species of *Memecylon* Linn

Samples	Ash	Moisture	Crude fat	Crude fiber	Crude Protein	Carbohydrate	Nutritive Value
M. umbellatum	3.166±0.709	54.000±2.268	6.083±0.222	3.625±0.231	1.683±0.100	35.054±1.586	201.750±6.195
M. malabaricum	4.833±0.843	49.000±2.670	5.625±0.386	4.333±0.518	1.872±0.137	38.665±2.015	212.792±8.578
M. talbotianum	4.000±0.716	48.917±1.584	5.542±0.566	4.167±0.544	1.803±0.109	39.692±1.209	216.050±8.229
M. wightii	5.167±0.527	46.833±2.038	4.958±0.804	4.917±0.667	1.870±0.116	40.989±1.277	216.125±12.35
M. lushingtonii	5.167±0.500	48.167±1.167	6.000±0.333	4.833±0.500	1.787±0.163	38.877±1.540	216.667±7.500

± Shows mean and standard error

Nutritive Value

Among the five species three species of *Memecylon* namely *M, lushingtonii* (216.66), *M. wightii* (216.12) *M. talbotianum* (216.03) recorded more or less equal values of nutritive value but *M. umbellatum* recorded lowest of 201.74 and *M. malabaricum* recorded 213.03 cal/100 gm of nutritive value.

Elemental Composition

Table 2 shows Average values of elemental composition of five species of *Memecylon* Linn.

Macronutrients

Among the five macronutrients analyzed all the five species recorded highest percentage of N, Ca and K and these were followed by Na, Mg and P in *M. umbellatum, M. malabaricum, M. talbotianum* and *M. lushingtonii* but in *M. wightii* N, Ca and K were followed by Mg, P and Na.

Table 2: Average values of elemental composition [macro (in %), micro (in ppm) and heavy metals (in ppm) of five species of *Memecylon*

	Macro elements						Micro elements				Heavy metals	
Samples	**Na**	**K**	**P**	**Ca**	**Mg**	**N**	**Zn**	**Cu**	**Mn**	**Fe**	**Pb**	**Cd**
M. umbellatum	0.36± 0.170	0.655± 0.196	0.133± 0.020	0.646± 0.249	0.158± 0.058	8.648± 0.274	17.519± 6.311	74.358± 58.929	233.347± 123.628	284.289± 122.138	30.45± 17.762	0.48± 0.279
M. malabaricum	0.66± 0.125	0.747± 0.230	0.071± 0.022	1.779± 0.614	0.224± 0.075	3.758± 1.528	18.926± 4.454	28.717± 18.366	47.379± 17.544	304.523± 87.221	0	0
M. talbotianum	0.37± 0.159	0.603± 0.203	0.086± 0.032	1.722± 0.869	0.227± 0.106	7.582± 0.469	17.262± 6.630	10.681± 5.017	142.030± 79.420	312.233± 130.141	12.961± 12.961	0.328± 0.328
M. wightii	0.11± 0.014	0.801± 0.080	0.148± 0.020	1.399± 0.155	0.299± 0.062	8.519± 0.308	25.579± 6.279	75.454± 56.141	55.801± 4.694	804.283± 159.168	51.163± 2.868	1.278± 0.166
M. lushingtonii	0.11± 0.084	0.214± 0.181	0.043± 0.030	0.251± 0.206	0.072± 0.072	3.872± 2.243	9.010± 7.356	14.998± 13.251	45.4033± 38.8780	191.945± 149.226	11.044± 11.044	0.283± 0.283

±shows mean and standard error

The highest percentage of N was recorded in *M. malabaricum* (0.642%) and it was followed by *M. umbellatum* (0.505%), *M. talbotianum* (0.485%), *M, lushingtonii* (0.306%) and *M. wightii* (0.114%).

The percentage of P was highest in *M. umbellatum* (0.176%) and lowest in *M. malabaricum* (0.083%) and *M. wightii, M. talbotianum* and *M, lushingtonii* recorded 0.147, 0.115 and 0.098 % of P respectively.

The highest percentage of Ca was highest in *M. talbotianum* (2.133%) and it was followed by *M. malabaricum* (1.828%), *M. wightii* (1.32%), *M. umbellatum* (1.164%) and *M. lushingtonii* (0.666%).

M. talbotianum recorded highest of 0.342% of Mg and *M. lushingtonii* recorded lowest of 0.187% of Mg and *M. malabaricum, M. wightii* and *M. umbellatum* recorded 0.328, 0.298 and 0.248 % of Mg respectively.

N was the most dominant among the five macronutrient analyzed. The highest value of N 25.94% was recorded in *M. umbellatum* and it was followed by *M. wightii* (25.55%), *M. lushingtonii* (23.23%), *M. talbotianum* (23.17%) and *M. malabaricum* (11.27%).

Micronutrients

M, lushingtonii recorded highest value of 36.97 ppm of Zn and *M. malabaricum* recorded lowest of 24.49 ppm of Zn. *M. talbotianum, M. umbellatum* and *M. wightii* recorded 33.43 ppm, 30.17 and 25.57 ppm of Zn respectively.

Cu was highest in *M. umbellatum* (78.93ppm) and lowest in *M. talbotianum* (15.15ppm) and *M. wightii, M. lushingtonii* and *M. malabaricum* recorded moderate values of 74.03ppm, 35.91ppm and 26.72 ppm of Cu respectively.

M. umbellatum recorded highest of 460.95ppm of Mn which is followed by *M. talbotianum, M. lushingtonii*, *M. malabaricum* and *M. wightii* and the values recorded were 263.81, 210.42, 58.97 and 55.79 ppm respectively.

Fe was the dominant among the four micronutrients analyzed. *M. wightii* contain highest of 878.44ppm and *M. malabaricum* recorded lowest of 352.06ppm of Fe and *M. lushingtonii*, *M. talbotianum* and *M. umbellatum* recorded 658.80, 560.46 and 449.23 ppm of Fe respectively.

Heavy metals

Among the two heavy metals analyzed Pb was dominant in all the five species. *M. umbellatum* recorded highest of 60.9ppm and *M, lushingtonii* recorded lowest of 44.17ppm of Pb and *M. talbotianum* and *M. wightii* recorded 51.84 and 51.07ppm of Pb respectively.

Cd was highest (1.13ppm) in *M. talbotianum* and lowest (0.961ppm) in *M. umbellatum* and *M. wightii* and *M, lushingtonii* recorded 1.27 and 1.13 ppm of Cd respectively. *M. malabaricum* did not recorded both the heavy metals.

Statistical Analysis Proximate Parameters

Moisture and ash, fiber and moisture are negatively significantly correlated at 0.05 level, nutritive value and carbohydrate are positively significantly correlated at 0.05 level. Carbohydrate and moisture, nutritive value and moisture are negatively significantly correlated at 0.01 level where as fiber and ash are positively significantly correlated at 0.01 level (Table 3).

Table 3: Correlation matrix of proximate parameters

	Ash	**Moisture**	**Fat**	**Fiber**	**Protein**	**Carbohydrate**	**Nutritive value**
Ash	1						
Moisture	-0.911*	1					
Fat	-0.542	0.701	1				
Fiber	0.967**	-0.920*	-0.546	1			
Protein	0.795	-0.854	-0.755	0.682	1		
Carbohydrate	0.788	-0.966**	-0.814	0.818	0.839	1	
Nutritive value	0.813	-0.959**	-0.546	0.841	0.762	0.930*	1

* Correlation is significant at the 0.05 level (2-tailed) and ** Correlation is significant at the 0.01 level (2-tailed)

Elemental Composition

Zinc and potassium are negatively significantly correlated at 0.05 level, cadmium and nitrogen, cadmium and lead are positively significantly correlated at 0.05 level. Magnesium and calcium, lead and nitrogen are positively significantly correlated at 0.01 level where as iron and sodium are negatively significantly correlated at 0.01 level (Table 4).

Table 4: Correlation matrix of elemental composition

	Na	K	P	Ca	Mg	N	Zn	Cu	Mn	Fe	Pb	Cd
Na	1											
K	0.156	1										
P	-0.304	0.356	1									
Ca	0.494	0.385	-0.221	1								
Mg	0.329	0.565	-0.142	0.962**	1							
N	-0.649	-0.269	0.752	-0.420	-0.403	1						
Zn	-0.098	-0.952*	-0.100	-0.386	-0.589	0.447	1					
Cu	-0.465	0.464	0.853	-0.508	-0.336	0.559	-0.313	1				
Mn	0.281	-0.215	0.602	-0.188	-0.346	0.524	0.496	0.270	1			
Fe	-0.985**	-0.162	0.247	-0.349	-0.192	0.625	0.092	0.355	-0.334	1		
Pb	-0.516	-0.273	0.766	-0.317	-0.336	0.985**	0.483	0.494	0.635	0.502	1	
Cd	-0.693	-0.454	0.479	-0.243	-0.252	0.920*	0.557	0.238	0.321	0.725	0.905*	1

* Correlation is significant at the 0.05 level (2-tailed) and ** Correlation is significant at the 0.01 level (2-tailed)

DISCUSSION

The leaves of five species of *Memecylon* were analyzed for their moisture, ash, crude fat, crude fiber, crude protein, carbohydrate, nutritive value and mineral contents. The five species *M. umbellatum*, *M. malabaricum, M. talbotianum, M. wightii* and *M. lushingtonii* were selected for the present study.

The leaves have recorded ash content ranging from 3.16% for *M. umbellatum*, to 5.17% for *M. wightii*. The high content of ash is useful in assessing the quality of grading the plant and it also gives an idea of the amount of minerals present in the sample (Michael and David, 2002). Moisture content ranging from 46.83% for *M. wightii* to 54% for *M. umbellatum*, crude fat content ranging from 4.95% for *M. wightii* to 6.08% for *M. umbellatum*, The fat are primarily used to produce hormone like substances that regulate a wide range of functions, including blood pressure, blood clotting, blood lipid levels, the immune response and the inflammation response to injury infection. It play an important role in the life and death of cardiac cells because they are essential fuels for mechanical and electrical activities of heart (Reiffel and Donold, 2006; Landmark and Alm, 2006; Herbant, 2006). The fat content can be used for storage and transport forms of metabolic fuel (Michael and David, 2002). Crude fiber content ranging for 3.62% for *M. umbellatum* to 4.91% for *M. wightii* which plays an important role in decreasing the risks of many disorders such as constipation, diabetes, cardiovascular disease (CVD), diverticulosis and obesity (Spiller, 2001). Food fiber helps in absorption of trace elements in the gut (Kelsay, 1981) and reduces absorption of cholesterol (Le Veille and Sanberlich, 1966). The fiber also increases the bowel movement and promotes abortion. (Michael and David, 2002). Crude protein content ranging from 1.68% for *M. umbellatum* to 1.87% for *M. malabaricum* and *M. wightii.* Proteins contain amino acids which are utilized by the cells of the body to synthesize all the numerous proteins required for the function of the cell and also to furnish energy (Robinson, 1978). The low content of the protein can contribute to the formation of hormones which controls a variety of body functions such as growth repair and maintance of body protein (Mau *et al*, 1999). All the samples were rich in carbohydrate which is ranging from 35.05% for *M. umbellatum* to 40.98% for *M. wightii,* Carbohydrate constitutes a major class of naturally occurring organic compounds that are essential for the maintenance of plant and animal life and also provide raw materials for many industries (Ebun-Oluwa and Alade, 2007). The high carbohydrate content is used as energy source and also it is necessary in the digestion and assimilation of other foods (Michael and David, 2002). And nutritive value was ranging from 201.75% for *M. umbellatum* to 216.66% for *M. lushingtonii*.

Among the macro nutrients the Na and K were highest in *M. malabaricum.* Na and K take part in ionic balance of the human body and maintain tissue excitability, carrying of normal muscle contraction help in the formation of gastric juice in stomach (Brody Tom, 1998). K helps in releasing of chemicals which are acts as nerve impulses, regulating heart rhythms. Its deficiency causes nervous irritability, mental disorientation, low blood sugar, insomnia and coma (Underwood and Suttle, 1999).

Ca and Mg were highest in *M. talbotianum*. Ca plays an important role in building and maintaining of strong bones and teeth and also constitutes a large part of human blood and extra cellular fluids and it is also necessary for normal functioning of cardiac muscle, blood coagulation, milk clotting and regulation of cell permeability (Heaney, 1994). Its deficiency causes rickets, back pain, osteoporosis, indigestion, irritability, premenstrual tension and cramping of the uterus (Hasling *et al.,* 1991). Mg plays an important role in the formation of bones, muscles and prevents high disorders, high blood pressure and depression (Smith and Hammarstam, 1958) along with these it also plays important role in enzyme activity, deficiency interfere with transmission of nerve and muscle, impulse, causing irritability and nervousness and in the prevention of heart diseases (Scelig, 1989).

P and N were highest in *M. umbellatum*. P helps in maintaining of blood sugar level, normal heart contraction dependent on P (Linder and Manria, 1991). Also important for normal cell growth and repair, which are needed for bone growth, kidney functions and cell growth. It also plays an important role in maintaining body's acid-alkaline balance (Johns and Dequette, 1991). N plays an important role in digestion of food and growth (Cooper, 1984) but in excess it is harmful for living body.

The micronutrients Zn and Fe were highest in *M. lushingtonii* and *M. wightii* respectively. In enzyme metabolism Fe, Mn and Zn are the three essential elements. Fe is important in maintaining the good health of human being (Vaughan and Judd, 2003).

Whereas Cu and Mn were recorded high in *M. umbellatum.* Mn is a component of several enzymes including manganese-specific glycosyl transferase and phosphoenol pyruvate carboxykinase and essential for normal bone structure. Mn deficiency can cause manifest as transient dermatics, hypocholesterolemia and increased ALP level. Cu is a universally important cofactor for hundreds of enzymes. It functions as a cofactor and numerous enzymes which are involved in development and maintance of the cardiovascular system. The deficiency of Cu can result in a decrease in the tinsel strength of arterial walls, leading to aneurysm formation and skeletal mal development (Tilson, 1982). Mn is an important modulator of cells function and plays a vital role in the control of diabetes (Kore, 1988).

The toxic elements such as Pb and Cd were recorded highest in *M. umbellatum* and *M. talbotianum* respectively. Trace elements are essential for normal growth of plants, their protection against viruses and completion of their life cycle (Bennet *et al*., 2000).

Nutritional deficiency may lead to diseases e.g.; Kwashiorkor, goiter. The increasing use of highly refined foods which have low percentage of minerals, vitamins etc contributes to the problems of health. Dietary supplements which increase the total dietary intake of one or more essential vitamin or mineral are very common. USA alone spent about $ 3 billion per year on vitamins, elements and nutritional products. The possible adverse effects of long term ingestion of high dose mineral supplements are unknown (Ivey and Elmen, 1986). The traditional healers prescribe mixture of medicinal plants for curing diseases ranging from common cold malaria, arthritis, ulcers etc (Obiajunwa *et al*., 2002). To prevent imminent development of diseases or for treatment of vitamin or mineral deficiencies nonprescription drugs such as minerals or vitamins are used (Hay, 1984).

The differences in the concentrations of various elements may be due to the differences in botanical structures of the plants and also due to the mineral composition of the soil. Moreover the differences may be due to the ability of plants to accumulate the elements from the surrounding aerial or aquatic environment either for their physiological requirement or as a precautionary measure (Viksna and Selin, 2001). Some inorganic mineral elements (potassium, zinc, calcium, traces of chromium, etc.) play important roles in the maintenance of normal glucose tolerance and in the release of insulin from beta cells of islets of Langerhans (Choudhary and Bandyopadhyay, 1999). All the plant parts have nutritional qualities which when used in the right proportions could be of tremendous benefit to the body (Abolaji *et al.,* 2007).

The result of the present study reveals that *M. umbellatum* recorded highest values of moisture, crude fat, P, N, Cu, Mn and Pb but low in nutritive value. Whereas *M. malabaricum* recorded lowest values of crude protein, P, N, Zn and Fe and it is very interesting to note that both the heavy metals Pb and Cd were almost absent in the sample and *M. lushingtonii* recorded highest amount of nutritive value. So these plants can be used as fodder for animals. Many researchers studied different plants for their nutritive value and element composition. Ekpa, (1996) studied nutrient composition of three Nigerian medicinal plants and recommended that the leaves of *Ipomea aserifolia* with high percentage of protein and low level of toxicity could be serve as a good source of food for form animals. Javid *et al*., (2010) clarifies the use of four medicinal plants from Pakistan with very lesser level of Pb as food supplement. Indrayana *et al*. (2005) analyzed some medicinally valued plant from Uttarachal and reported that *Artocarpus heterophyllus* leaves are not rich in desired mineral elemental and nutritive value. So, these leaves are good for those who require high sodium and also good as fodder.

CONCLUSIONS

In the present study the five species of *Memecylon* were investigated for proximate analysis and elemental composition. *M. umbellatum* recorded highest values of moisture, crude fat, phosphorus, nitrogen, copper, manganese and lead, *M, malabaricum* recorded highest values of crude protein, sodium, potassium. In this species both the heavy metals lead and cadmium were absent. *M. talbotianum* recorded highest values of calcium, magnesium and cadmium, where as *M, wightii* recorded highest values of ash, crude fiber, carbohydrate and iron and *M. lushingtonii* recorded highest values of nutritive value and zinc. From the obtained result it can be concluded that all these species of *Memecylon* may be further explored for their phytochemical profile to identify the presence of active constituents for medicinal uses.

Conflict of Interest

Conflict of interest none declared.

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