Antioxidant and α-glucosidase inhibitory activities of *Murraya koenigii* leaf extracts

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Submission Date: 1-6-2012; Review Completed: 19-7-2012

**ABSTRACT**

Medicinal plants are source for a wide variety of natural antioxidants. Dietary antioxidant consumption may be an important approach for inhibiting or delaying the oxidation of susceptible cellular substrates and thus is relevant to disease prevention in many paradigms. *Murraya koenigii* is one of the important medicinal herb which is used as a food ingredient across India. In this study, we demonstrated the reducing power, total antioxidant potential, radical scavenging capabilities and α-glycosidase inhibitory property of different crude extracts of curry leaves (*Murraya koenigii* L.). The extracts were evaluated for their radical scavenging activities by means of DPPH, NO, •OH, O2− and anti-lipid peroxidation assays. In addition, extract of *M. koenigii* were tested for α-glucosidase inhibitory property. The extracts of *M. koenigii* scavenged radicals effectively in varied degree. Similarly, the total reducing power of alcohol extract was found higher in both phosphomolybednum and FRAP methods. In *vitro* assay of α-glucosidase activity of MKA and MKW showed an IC50 of 174.74 and 287.00 μg/ml respectively, while other two extracts did not show any significant effect. Simultaneously, total phenolic and total flavonoid contents of extracts were studied, where values of MKW were found to be higher than that of other extracts. In present study, we found that MKA and MKW extracts contain effective antioxidant and radical scavenging activities as compared to other extracts. Our study provides a proof for the ethno-medical claims and reported biological activities that curry leaves have significant therapeutic potential.

**Keywords:** *Murraya koenigii*, antioxidant, free radical, α-glucosidase.

**INTRODUCTION**

Reactive oxygen species (ROS) such as singlet oxygen (O2•), hydrogen peroxide (H2O2), superoxide anion (O2−) and hydroxyl (OH) radical are often generated as byproducts of biological reactions or from exogenous. These ROS create homeostatic imbalance which generate oxidative stress and cause cell death and tissue injury.[1] Free radicals and ROS are well known inducers of cellular and pathological processes including diabetes, cell proliferation, inflammatory conditions and many neurodegenerative disorders apart from aging processes.[2,3,4] A potent scavenger of these species may serve as a possible preventive intervention for free radical-mediated diseases.[5] It has been established that antioxidants provide protection to living organisms from damage triggered by uncontrolled production of ROS and associated lipid peroxidation, protein damage and DNA strand breaking.[6] The health promoting effect of antioxidants from plants is thought to arise from their potential effects on the reactive oxygen/nitrogen species. Plant-derived antioxidants could function as singlet and triplet oxygen quenchers, peroxide decomposers, enzyme inhibitors or synergists.[7]
Most plant species possess tremendous medicinal properties because they are used both to sustain health and to cure illness. Dietary antioxidant intake may be an important approach for inhibiting or delaying the oxidation of susceptible cellular substrates and is thus relevant to disease prevention in many models. Phenolic compounds such as flavonoids, phenolic acids, diterpenes etc. have received attention for their high antioxidative activity. In vitro bioassay systems have been extensively used to monitor biological activities of medicinal plant extracts used in traditional medicines.

Numerous plants used in traditional medicine are effective in treating various ailments caused by oxidative stress, bacterial and/or viral infections. Since ancient times, spices and herbs have been added to different types of food to improve flavours as well as for their antioxidant capacity. Converging evidence from both experimental and epidemiological studies have demonstrated that medicinal plants in particular contain a myriad of phenolic compounds and still present a large source of natural antioxidants that might serve as leads for the development of novel drugs. Therefore, the exploration for natural antioxidants and other preparations of plant origin to achieve this objective has been gained importance over the years.

*Murraya koenigii* L. Spreng (Rutaceae) is a small and strong smelling perennial shrub or small tree commonly found in forests as undergrowth, cultivated in India for its characteristic flavour and aroma. Leaves of this plant are used as a condiment in the preparation of curry powder, pickle, chutney, sausages and seasonings. The flavour and fragrance of leaves is retained even after drying. Leaves relieve nausea, indigestion, vomiting and used as a cure for diarrhoea and dysentery.

In this study, we have demonstrated the antioxidant efficiency of different *M. koenigii* using series of in vitro assays. In addition, we also evaluated its α-glucosidase inhibitory activity apart from determination of phytochemical constituents.

**MATERIALS AND METHODS**

**Materials**

All solvents used in this study were of analytical grade. Methanol, ethyl acetate, hexane, and Folin-Ciocalteu reagent obtained from Merck (Merck, India) while other chemicals were procured from HiMedia chemicals, India.

**Plant Material**

Curry leaves (*M. koenigii* L.) were purchased from local market in Hyderabad, India in the month of July, 2011. The identity of the plant was confirmed by Dr. VC Gupta, Taxonomist, Central Research Institute of Unani Medicine, Hyderabad. The voucher specimen of the plant was kept for future reference. One hundred grams of curry leaves were dried at ambient temperature for 10–15 days. After drying completely, leaves were grounded to a coarse powder using domestic electric grinder.

**Preparation of Extracts**

The coarse powder was subjected to successive extraction in a soxhlet apparatus using different solvents such as hexane, ethyl acetate, ethanol and water. Each time before extracting with the next solvent, the material was dried in hot air oven at 40°C. Furthermore, extracts were filtered through Whatman No.1 paper filter and concentrated to the dry mass with the aid of rotary evaporator. The yield of each extract was measured and residues were stored in dark for further analysis. Different extracts were designated as MKH (for hexane extract), MKE (for ethyl acetate extract), MKA (for ethanol extract) and MKW (for water extract). Dried extracts of 20 mg/ml stock solution were prepared and different concentrations were used in various experiments.

**DETERMINATION OF PHYTOCHEMICAL CONSTITUENTS**

**Total Phenolic Content**

Total phenolic content of extracts were determined by Folin-Ciocalteu method with little modification. Briefly, 10 μl of the extracts were taken to which 500 μl of double distilled water was added, followed by 100 μl of FolinCiocalteu’s reagent. After incubating the mixture for 10 min at room temperature, 300 μl of 20% Na₂CO₃ was added, thoroughly vortexed and the volume of the reaction mixture was adjusted to one ml with double distilled water. The mixture was then incubated for 2 hrs in dark and the absorbance was measured at 765 nm against blank. Results were expressed as mg of gallic acid equivalents (GAE)/g dry weight (dw).

**Total Flavonoid Content**

Total flavonoid content was quantified by following the method of Barreira with minor modifications. Briefly, 20 μl of each extract were mixed with 500 μl double
distilled water and 30 µl of 5% NaNO<sub>2</sub> solution. After 5 min of incubation at room temperature, 60 µl of 10% AlCl<sub>3</sub> solution was added. Subsequently, 350 µl of 1 M NaOH and 40 µl of double distilled water were added to make the final volume to one ml. Samples were further incubated for 15 min at room temperature and the absorbance of samples was measured at 510 nm. The total flavonoids were determined as quercetin equivalents (mg QE)/g of dry weight.

**Determination of Total Antioxidant Activity**

Total antioxidant activities of extracts were evaluated by phosphomolybdenum method.<sup>[19]</sup> The assay is based on the reduction of Mo<sup>6+</sup>–Mo<sup>5+</sup> by the antioxidant compounds and subsequent formation of a green phosphate/Mo<sup>5+</sup> complex at acidic pH. The reagent solution contains ammonium molybdate (4 mM), disodium hydrogen phosphate (28 mM) and sulfuric acid (0.6 M) mixed with the extracts. Samples were incubated for 60 min at 90°C and the absorbance of the green phosphomolybdenum complex was recorded at 695 nm. Ascorbic acid was used as reference and reducing capacity of the extracts was expressed as the mg ascorbic acid equivalents per gram dry weight.

**Determination of Reducing Antioxidant Power (FRAP)**

The reducing antioxidant power of the plant extracts was determined according to the method described by Oyaizu<sup>[20]</sup> with slight modifications. Briefly, 10 µl of each extract was taken and the volumes were made to 250 µl with double distilled water. Further, 250 µl of potassium ferricyanide (1%) was added to the tubes and incubated for 20 min at 50°C. Then 250 µl of trichloroacetic acid (10%) was added to the incubated mixture. Upper part of the mixtures (500 µl) were taken and mixed with 400 µl of double distilled water and 100 µl of ferric chloride (0.1%). The absorbance of the mixture was measured at 700 nm and reducing power of extracts was expressed as mg ascorbic acid equivalents (AAE) per gram (g) of dry weight (dw).

**DPPH Radical Scavenging Activity**

The DPPH radical scavenging activity of the plant extracts was determined according to the method described by Braca.<sup>[21]</sup> The activity was assessed using stable 1, 1-diphenyl-2-picrylhydrazyl (DPPH). DPPH solution (0.004% w/v) was prepared in 95% methanol, mixed with different dilutions of plant extracts and thoroughly vortexed. The reaction mixture was then incubated in dark at room temperature for 45 min and absorbance solution was measured at 517 nm against the blank. Methanol (95%) and ascorbic acid were used as blank and reference compound respectively.

**Nitric Oxide Radical Scavenging Activity**

The activity was measured according to the modified method of Sreejayan and Rao.<sup>[22]</sup> To 100 µl of the extract having different concentrations (40–400 µg/ml), 20 µl of sodium nitroprusside (SNP) solution (10 mM) was added and incubated for 15 min under light conditions. After incubation, the mixture was diluted with 300 µl of Griess reagent (1% sulfanilamide in 2% H<sub>3</sub>PO<sub>4</sub>). The reaction mixture was further incubated for 45 min under light conditions at 30°C followed by addition of 10 µl of 0.1% naphthylethylene diamine dihydrochloride in 2.5% H<sub>3</sub>PO<sub>4</sub>. Final volume was made to 1 ml with double distilled water. The absorbance of the chromophore was taken immediately at 546 nm and compared to the standard (ascorbic acid).

**Hydroxyl Radical Scavenging Activity**

Hydroxyl radical scavenging activity was measured as per the protocol of Kunchandy and Rao<sup>[23]</sup> with minor changes by studying the competition between deoxyribose and extracts for hydroxyl radicals generated by Fenton’s reaction. Briefly, solution of Fenton’s reagent [Fe (III) chloride, ascorbic acid and H<sub>2</sub>O<sub>2</sub>] was prepared in distilled water just prior to use. To 0.1 mL of Fenton’s reagent, thiobarbituric acid (1% w/v) in 25 mM NaOH (1 ml) and trichloroacetic acid (1 ml, 2.8% w/v) were added and volume was made to 3 ml with distilled water. The mixture was then incubated at 80°C for 90 min and amount of pink chromogen produced was considered as control and was measured spectrophotometrically at 532 nm. The protection of oxidation of D-ribose has been conducted by pre-incubation with extracts in different concentrations and decrease in the formation of pink colour was considered as antioxidant property which was compared to standard (ascorbic acid).

**Superoxide Radical Scavenging Activity**

The activity was evaluated using nitro blue tetrazolium (NBT) reduction method.<sup>[24]</sup> The reaction mixture consisted of 0.5 ml of NBT solution (156 µM, 0.5 ml nicotinamide adenine dinucleotide (468 µM, NADH), and extracts of different concentrations (40–400 µg/ml). The reaction was initiated by adding 50 µl of phenazine methosulfate solution (60 µM, PMS) in phosphate buffer.
(pH 7.4). The reaction was incubated at 25°C for 20 min and then absorbance was measured at 560 nm against blank. Ascorbic acid was used as the reference.

**Determination of Inhibition of Lipid Peroxidation**

Lipid peroxidation inhibitory activity of *M. koenigii* extracts and standard (ascorbic acid) were performed as per the protocol given by Halliwell and Gutteridge. The rat liver homogenate was used for induction of lipid peroxidation, mediated by FeCl₃ as pro-oxidant. Healthy albino rats of the wister strain (250 g) were sacrificed and perfused the liver with 0.15 M KCl followed by centrifugation of homogenate at 800 g for 15 min at 4°C and supernatant was used for thiobarbituric acid assay. The extracts at different concentrations (40–400 μg/ml) were mixed with the liver microsome preparation and the mixtures were incubated in presence and absence of fenton’s reagent (50 μl of 10 mM FeCl₃; 10 μl of 2.5 mM H₂O₂) in phosphate buffer (0.2 M, pH 7.4) and the final volume was made to 1 ml. The reaction mixtures were incubated at 37°C for 30 min. After incubation, 2 ml of ice-cold HCl (0.25 N) containing 15% trichloroacetic acid, 0.5% thiobarbituric acid, and 0.5% butylated hydroxytoluene (BHT) was added to the reaction mixture followed by heating at 100°C for 10 min. The reaction mixture was put in an ice bath for 5 min for cooling. After that the mixture was centrifuged at 1000 g for 10 min and the extent of lipid peroxidation was subsequently monitored by the formation of thiobarbituric acid reactive substances (TBARS) as pink chromogen in the presence or absence of extracts and standard (ascorbic acid). The absorbance of supernatant was measured spectrophotometrically at 532 nm and decline in the formation of pink chromogen in the pre-treated reactions was considered as inhibition of lipid peroxidation.

**Effects of Extracts on α-glucosidase Activity In Vitro**

The α-glucosidase inhibitory activity was determined according to the method given by Matsui with slight modifications by measuring the release of 4-nitrophenol from 4-nitrophenyl α-D-glucopyranoside (4-NPGP).

Briefly, the enzyme reaction was performed using p-nitrophenyl-α-D-glucoside (PNP-glycoside) as a substrate in 0.1 M phosphate buffer (pH 6.8). PNP-glycoside (10 mM) and 10 μl of GSH (3 mM) was pre-mixed with samples at various concentrations. Each mixture was added to an enzyme solution (0.01 units) to make 1 ml of final volume. The reaction was terminated by adding 5 μl of 100 mM sodium carbonate solution. Enzymatic activity was quantified by measuring the p-nitrophenol released from PNP-glycoside at 400 nm. All reactions were carried out at 37°C for 30 min with three replications. Acarbose was used as a positive control and IC₅₀ values were calculated by the graphic method.

**Calculations and statistical analysis**

The percentage inhibitions of radicals, lipid peroxidation and α-glucosidase inhibitory activities of the extracts were calculated using the formula:

\[
\text{Percentage inhibition} = \frac{A_{\text{control}} - A_{\text{sample}}}{A_{\text{control}}} \times 100, 
\]

All the experiments were performed in triplicates and experimental results were expressed as mean ± standard deviation of mean (SEM) of three replicates. IC₅₀ value (the concentration of the extracts required to scavenge 50% of radicals) was calculated for different extracts of *M. koenigii*. Graphpad prism 5 software was used for statistical analysis and to prepare the graphical representation of results.

**RESULTS AND DISCUSSION**

**Determination of Phytochemical Constituents**

The results of total phenolic content of different leaf extracts of *M. koenigii* were significant and found in the range of 2.37 to 28.84 mg GAE/g dw (Table 1). Among the tested extracts, the highest amount of total phenolics was observed in MKW (28.84 mg GAE/g dw) whereas MKH showed least (2.37 mg GAE/g dw) content of phenolics. The flavonoid contents of the extracts in terms of quercetin equivalent were between 8.28 and 39.90 mg QE/g dw (Table 1), highest being in MKW (39.90 mg QE/g dw) and lowest in MKH (8.28 mg QE/g dw). It is well known that plant polyphenols are widely distributed in the plant kingdom and are very important plant constituents. It has been recognised that phenolic compounds such as flavonoids, phenolic acid and tannins possess diverse biological activities like anti-diabetic anti-

**Table 1. Total Polyphenol, Flavonoid, Antioxidants and Ferric Reducing Antioxidant Power of *M. koenigii* Extracts.**

<table>
<thead>
<tr>
<th>Extract</th>
<th>Total polyphenol content</th>
<th>Total flavonoid content</th>
<th>Total antioxidant activity</th>
<th>Ferric reducing power</th>
</tr>
</thead>
<tbody>
<tr>
<td>MKH</td>
<td>2.37±0.36</td>
<td>8.28±0.18</td>
<td>6.04±0.25</td>
<td>3.31±0.012</td>
</tr>
<tr>
<td>MKE</td>
<td>12.41±1.60</td>
<td>14.93±1.19</td>
<td>21.86±1.04</td>
<td>9.91±0.02</td>
</tr>
<tr>
<td>MKA</td>
<td>21.18±1.47</td>
<td>23.52±0.81</td>
<td>25.54±0.72</td>
<td>17.12±0.47</td>
</tr>
<tr>
<td>MKW</td>
<td>28.84±0.49</td>
<td>39.90±1.23</td>
<td>-</td>
<td>2.69±0.08</td>
</tr>
</tbody>
</table>

a: gallic acid; b: quercetin; c: ascorbic acid equivalents mg/g dw plant material respectively; Results represented in means ± standard deviation (n = 3).
inflammatory, anti-carcinogenic and anti-atherosclerotic activities and their effects on human nutrition and health are considerable.\(^{[28]}\) It is interesting to note that both the polyphenol and flavonoid contents of MKW are higher than other extracts. This may be due to the better solubility of the polyphenol and flavonoid type of constituents in the aqueous solvent than the other medium. According to our study, the high contents of these phytochemicals in \(M.\ koenigii\) can explain its high radical scavenging activity.

**Determination of Total Antioxidant Activity**

Total antioxidant activity of plant extracts was determined by phosphomolybdenum method which is based on the reduction of \(\text{Mo (VI)}\)–\(\text{Mo (V)}\) by the antioxidant compounds and subsequent formation of green phosphate/\(\text{Mo (V)}\), is quantitative since the antioxidant activity is expressed as the number of equivalents of ascorbic acid (AA) per gram dry weight of extract. All the four extracts possess antioxidant potential (Table 1), but discrepancies were noticed in the extracts of different polarity. The antioxidant activity was in the range of 6.04 to 25.54 mg AAE/g dw in the leaf extracts. MK showed the greatest value of 25.54 mg AAE/g dw, as was the case of total phenols and flavonoids, whereas the lowest value of 6.04 mg AAE/g dw was found in MKH. In our study, total polyphenol, flavonoid content and antioxidant activity of the plant extracts correlated significantly and could also contribute to the overall antioxidant potential.

**Determination of Reducing Antioxidant Power (FRAP)**

The reducing ability to convert \(\text{Fe}^{3+}\) to \(\text{Fe}^{2+}\) is also an indirect evidence for the antioxidant activity of an extract or a compound.\(^{[29]}\) In this assay system, the antioxidants present in the extract causes the reduction of the \(\text{Fe}^{3+}/\text{ferricyanide}\) complex to form \(\text{Fe}^{2+}\) ions, which was monitored spectrophotometrically by recording the absorbance of the reaction mixture at 700 nm.\(^{[30]}\) All the tested samples showed some degree of electron donation capacity (Table 1). The reducing power of MKA was the highest amongst all the tested samples, with 17.12±0.47 mg AAE/g dw, followed by MKE (9.91±0.02 mg AAE/g dw), whereas those of the others were much lower, with a varied range from 2.69±0.08 to 3.31±0.012 mg AAE/g dw. The data presented here indicate that the marked reducing power of \(M.\ koenigii\) extracts seem to be attributed to their antioxidant activity.

**DPPH Radical Scavenging Activity**

Substances that are capable of donating hydrogen or an electron to DPPH, nitrogen centered free-radical are considered as antioxidants and therefore, radical scavengers.

The degree of discoloration of violet colour of DPPH, as it gets reduced, indicates the radical scavenging potential of the antioxidant.\(^{[31]}\) In this assay, all extracts showed significant dose-dependent DPPH radical scavenging capacity (Figure 1). The \(\text{IC}_{50}\) values ranged from 22.12±2.97 to 32.55±1.42 and the DPPH radical-scavenging efficiency increased as follows: MKH < MKA < MKW < MKE. MKE was most efficient, with the lowest \(\text{IC}_{50}\) value, 22.12 µg/ml. The antioxidant(s) in crude extracts neutralized the free radical character of DPPH by transferring either electrons or hydrogen atoms to DPPH\(^{[32]}\), thereby changing the colour from purple to the yellow-coloured diphenyl picrylhydrazine. This experiment suggests that the plant extracts could contain more bioactive compounds that may attribute the antioxidant properties of \(M.\ koenigii\).

**Nitric Oxide Radical Scavenging Activity**

Abnormally high level of NO has been linked with chronic inflammation and may be associated with the etiology and pathology of a number of chronic diseases.\(^{[33]}\) Besides its own toxicity, this radical can further react with other species instigating even more toxic radicals, such as peroxynitrite, which results from its reaction with superoxide. We tested \(M.\ koenigii\) extracts for their inhibitory effect on nitric oxide production and nitric oxide radical generated from sodium nitroprusside at physiological pH was found to be inhibited by the extracts. The extracts at varied concentrations (40–400 µg/ml) showed significant inhibitory effect of nitric oxide radical scavenging activity. The percentage scavenging activity increased with increasing concentration. Among the different extracts, the lower \(\text{IC}_{50}\) value was observed for MKE (162.27±2.26 µg/ml) followed by MKW (163.06±1.86 µg/ml) and highest \(\text{IC}_{50}\) was observed for MKA (Table 2; Figure 2).
Superoxide Radical Scavenging Activity

Superoxide anion is a free radical generated from the normal energy process of energy generation in the human body. It is toxic to cells and tissues and can act as precursors to other reactive oxygen species. It was found that the superoxide-scavenging activities of different extracts of *M. koenigii* were increased markedly with increasing concentrations (Figure 3). The inhibitory activity of MKH was significantly higher than that of other extracts (Figure 3). The IC$_{50}$ values of extracts was found to be in the order of MKW > MKA > MKE > MKH (Table 2). These results imply that water extract is better superoxide scavenger and its capacity to scavenge superoxide may contribute to its antioxidant activity.

Hydroxyl Radical Scavenging Activity

Hydroxyl radicals are the major reactive oxygen species that are responsible for oxidation of lipids and massive biological damage. The evaluation of radical scavenging activity was based on the generation of 'OH by Fenton reaction. The percentage inhibitions against the hydroxyl radical of different extracts of *M. koenigii* are presented in Figure 4. MKW was found to be efficient scavengers of hydroxyl radicals with least IC$_{50}$ value (82.23±0.33 μg/ml), while other extracts were found to be less efficient scavengers. By comparing the IC$_{50}$ values of all extracts, we can point out that the aqueous extracts of this plant (MKW) was more efficient hydroxyl radical scavengers than its other counterparts.

Determination of Inhibition of Lipid Peroxidation

In biological systems, lipid peroxidation generates a number of degradation products such as malondialdehyde which is one of the causes of cell membrane destruction.

<table>
<thead>
<tr>
<th>Extract</th>
<th>DPPH</th>
<th>Nitric oxide</th>
<th>Superoxide</th>
<th>Hydroxyl radical</th>
<th>Lipid peroxidation</th>
<th>α-glucosidase inhibition</th>
</tr>
</thead>
<tbody>
<tr>
<td>MKH</td>
<td>32.55±1.42</td>
<td>188.82±0.78</td>
<td>90.38±0.64</td>
<td>380.44±0.38</td>
<td>281.12±0.47</td>
<td>ND</td>
</tr>
<tr>
<td>MKE</td>
<td>22.12±2.97</td>
<td>162.27±2.26</td>
<td>129.73±0.42</td>
<td>535.90±0.36</td>
<td>31.67±1.65</td>
<td>ND</td>
</tr>
<tr>
<td>MKA</td>
<td>28.88±2.76</td>
<td>267.20±2.035</td>
<td>141.76±0.53</td>
<td>177.52±0.28</td>
<td>72.25±1.9</td>
<td>287.00±0.49</td>
</tr>
<tr>
<td>MKW</td>
<td>28.00±0.72</td>
<td>163.06±1.86</td>
<td>203.56±1.13</td>
<td>82.23±0.33</td>
<td>260.41±0.53</td>
<td>174.74±0.72</td>
</tr>
</tbody>
</table>

(Values expressed in μg/ml; Results represented in means ± standard deviation (n = 3); NA: No activity.)
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and cell damage. In order to determine if the *M. koenigii* extracts were capable of reducing *in vitro* oxidative stress, lipid peroxidation was assessed by means of an assay that determines the production of malondialdehyde and related compounds in rat liver homogenates. TBARS are produced as by-products of lipid peroxidation that occurs in the hydrophobic core of biomembranes. Our results clearly indicate that extracts in rat liver homogenate were capable of quenching the extent of lipid peroxidation caused by a Fe²⁺/ascorbate system (Figure 5). The IC₅₀ value was found to be least for MKE (31.67±1.65 μg/ml) followed by MKA (72.25±1.9 μg/ml). The IC₅₀ of other two extracts MKH and MKW were much higher (Table 2). This inhibition of lipid peroxidation may have been either due to chelation of Fe²⁺ ions or by trapping of free radicals produced by Fe²⁺/ascorbate in the reaction system.

**Effects of the Extracts on α-glucosidase Activity In Vitro**

Natural products are still the most readily available source of α-glucosidase inhibitors.[37] In recent reports from other traditional plants, polyphenols were observed to contribute to strong α-glucosidase inhibition.[38] Therefore, we investigated the activity in different extracts. It was found that out of four extracts tested for α-glucosidase inhibitory activity, only MKA and MKW showed significant inhibition property (Figure 6), whereas other extracts (MKH and MKE) did not inhibit α-glucosidase at all. The percentage inhibition of α-glucosidase by MKA and MKW exhibited significant inhibitory activity at dose-dependent acceleration suggesting a competitive type of inhibition. MKW (IC₅₀=174.74 μg/ml) exerted the most powerful inhibitory activity. Acarbose, an antidiabetic drug exerts almost similar effects (IC₅₀=128 μg/ml) under our assay conditions. The IC₅₀ value for MKA was found to be 287.00 μg/ml. Based on our results presented here, we can say that *M. koenigii* exert inhibitory effect on α-glucosidases, with MKW being the most effective. With these results, we can further support the traditional use of the plants for its wide medicinal applications.

**CONCLUSION**

In conclusion, the results of this study clearly indicate that *M. Koennigii* has powerful antioxidant activity against various oxidative systems *in vitro*. Various antioxidant properties of this potent medicinal plant may be attributed to its components effectiveness as scavengers of free radicals, reductive capacity, and metal chelating ability, as well as lipid peroxidation inhibition. The free radical-scavenging property may be one of the mechanisms by which this plant is attributed as useful for foodstuff as well as traditional medicine. Thus, our results support *M. koenigii* as an accessible source of natural antioxidants and a food supplement.

**ACKNOWLEDGMENTS**

The author MZG acknowledges the financial support from University Grants Commission, New Delhi, India in the form of Research fellowship (RFSMS). We are thankful to DBT-CREBB, DBT-FIST and UGC-SAP-CAS, UGC-XI plan seed money for supporting infra structural facilities of Department of Plant Sciences and School of Life Sciences.
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