

# Antioxidant potentials of Miyan Kuka (Baobab leaves)

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## Abstract

Antioxidant is any substance that directly scavenges reactive oxygen species (ROS) or indirectly acts to up-regulate antioxidant defenses or inhibit ROS production. The review is aimed at highlighting the components of baobab leaves with antioxidant capabilities. Articles were searched from the Directory of Open Access Journals, PubMed, Google Scholar, Science Direct using key words such as composition of baobab leaves, antioxidant properties of baobab leaf, reactive oxygen species, oxidative stress and antioxidant. The result showed that baobab leaves are good sources of food with a lot of antioxidant components such as vitamin C, carotenes, flavonoids, zinc etc. These antioxidant components help to regulate ROS including free radicals such as superoxide anion, hydroxyl radical, as well as non-radical molecules like hydrogen peroxide, singlet oxygen. Baobab leaves carry out the antioxidant activities through the following processes: scavenging and neutralizing free radicals, singlet oxygen quenching, superoxide radical scavenging, hydrogen donation, metal chelating, enzymes inhibition, peroxide decomposing and synergies. They also help in activating antioxidant enzymes, reduce  $\alpha$ -tocopherol radicals, reduce

nitrosative stress, prevent oxidases, and increase levels of uric acid and low molecular weight molecules which ultimately help to reduce oxidation in the long run. Baobab leaves are rich in phytochemicals with a lot of antioxidant capabilities due to the present of several components like vitamin C, carotenes, flavonoids, etc. These components work in different ways to regulate reactive oxygen species and by extension prevent oxidative stress.

## Introduction

Baobab leaves are staple food for many African populations especially the central and western regions of the continent.<sup>1-2</sup> Baobab leaves are used throughout Africa either as fresh leaves or dried in the sun.<sup>3</sup> The plant is known to survive seasonal fluctuations providing families with adequate nutrients and energy.<sup>4-5</sup> In Nigeria, “Miyan kuka” is a popular soup within the Hausas in the northern part of the country made by the used of baobab leaves.<sup>6</sup> In Zimbabwe, baobab leaves are used as vegetables substitute for the commercially grown leafy vegetables such as lettuce and cabbages.<sup>7</sup> In Mali, the leaves are used in making sauce and they are called lalo.<sup>8</sup> The Baobab leaves are rich in pro-vitamins A and vitamin C; in WHO standards, and in terms of protein content, leaves of baobab can be rated *good* in that they score well for 5 of the 8 essential amino acids.<sup>5</sup> Phytochemical screening shows that it contains glycosides, saponins, steroids, and flavonoids which are very crucial in antioxidant activities, maintenance of electrolyte level, protection against infections, antimicrobial, anti-inflammatory, and immune-stimulating properties.<sup>9,10</sup>

In nature, reactive oxygen species (ROS) is produced as a result of aerobic metabolism. These reactive oxygen species play major roles in the maintenance and regulation of cell; cell signaling, cell differentiation, cell survival, cell death and inflammation-related factor production.<sup>11-12</sup> Reactive oxygen species include free radicals such as hydroxyl radical (OH<sup>•</sup>), superoxide anion (O<sub>2</sub><sup>•-</sup>), as well as non-radical molecules like singlet oxygen (<sup>1</sup>O<sub>2</sub>), hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>), etc.<sup>13</sup> Excess production of free radicals in the system is caused by some environmental factors such as heavy metal exposure, tobacco smoke, UV radiation, xenobiotics and ionizing radiation together with superficial receptors activation in the cell of the affected organisms.<sup>14,15</sup> A free radical is an atom, molecule, or compound with unpaired electron making it highly unstable because of its atomic or molecular structure. This instability causes free radicals to be very reactive, and by so doing tries to pair up with other electrons, atoms or even molecules to form a stable compound. For a more stable state to be attained free radicals can “steal” a hydrogen atom from another molecule; it can also interact in various ways with other free radicals as well as bind to another molecule.<sup>16</sup> Free radicals are highly attracted to DNA, proteins and lipids.<sup>17</sup> When the generation of free radicals exceeds the system’s capacity to neutralize and eliminate them, which can be as a result of either decreased antioxidant defense systems or over-

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production of ROS, end result is oxidative stress.<sup>18</sup> Normally organisms have developed antioxidant defense system to contain this oxidative stress caused by aerobic metabolism. The antioxidant defense system consists of enzymes such as catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase (GPx) and glutathione reductase as well as a number of low molecular-weight antioxidants such as flavanoids, vitamins, ascorbate,  $\alpha$ -tocopherol, glutathione, cysteine, thioredoxin, etc.<sup>19</sup> In some cases, due to some pathological and negative environmental factors, the defense system can be overpowered, causing ROS to escape destruction and forming highly reactive hydroxyl radicals.<sup>20</sup> An increase in ROS can lead to oxidative damage to DNA and other macromolecules (proteins, nucleic acids and lipids). It is also implicated in the pathogenesis of several diseases such as neurodegenerative diseases, diabetes mellitus, cancer, cardiovascular diseases as well as aging.<sup>21</sup>

Antioxidant is “any substance that delays, prevents or removes oxidative damage to a target molecule”.<sup>22</sup> Antioxidants can be divided into two groups: enzymatic antioxidants and non-enzymatic antioxidants. The enzymatic antioxidants are produced in the body of the organism and are further subdivided into primary and secondary enzymatic defenses.<sup>23</sup> The primary enzymatic defense includes catalase (CAT), glutathione peroxidase (GPX) and superoxide dismutase (SOD). In general, primary enzymatic defense helps to prevent the formation of free radicals as well as neutralizing the already formed free radicals. Catalase, breaks down hydrogen peroxide into water and oxygen molecule. Glutathione peroxidase forms selenols by donating electrons to reduce peroxides by so doing eliminates peroxides which is an important component of Fenton reaction. Superoxide dismutase helps to convert superoxide anions into hydrogen peroxide.<sup>24</sup> The secondary enzymatic defense includes glucose-6-phosphate dehydrogenase and glutathione reductase. They do not neutralize free radicals directly, but support the primary enzymatic defense antioxidants. Glucose-6-phosphate regenerates Nicotinamide Adenine Dinucleotide Phosphate (NADPH), thereby creating a reducing environment for the primary enzymatic defense to act properly. Glutathione reductase reduces glutathione peroxidase from its oxidized form to its reduced form; by so doing help in recycling glutathione peroxidase to continue in neutralizing more free radicals.<sup>25</sup>

Non-enzymatic antioxidants can be classified into two: natural antioxidants and synthetic antioxidants. Natural antioxidants are nutrient-derived antioxidants like ascorbic acid (vitamin C), tocopherols and tocotrienols (vitamin E), carotenoids, and other low molecular weight compounds that are found in plants materials that have the ability to protect the cell and organ from reactive oxygen species.<sup>26</sup> Synthetic antioxidants are pure substance like butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA) that are added to food to prevent food oxidation and prolong shelf life. Most processed foods contain synthetic antioxidants, which are reported to be safe, but recent studies indicate otherwise.<sup>27</sup> Some studies have indicated that most synthesis antioxidants generate substances that can lead to cancer or other diseases in the long run.<sup>28</sup> The aim of this review was to itemize the components of baobab leaf with antioxidant capabilities.

## Materials and Methods

Database were search for articles from the Directory of Open Access Journals, PubMed, Google Scholar, Science Direct using key words such as composition of baobab leaf, antioxidant properties of baobab leaf, reactive oxygen species, oxidative stress and

antioxidant. The articles were selected and reviewed base on the following criteria:

- i. Articles on baobab leaves from different sources.
- ii. Articles that described the composition of baobab leaves
- iii. Articles that described the antioxidant properties of baobab leaves
- iv. Articles on reactive oxygen species, oxidative stress and antioxidant,
- v. Articles that met the selection criteria were selected for the review.

## Results and Discussion

Baobab leaves are a pool of essential and protective compounds and minerals. Studies has shown that the leaves are rich in phytochemicals such as flavonoids, glycosides, saponins, steroids vitamins, amino acids, carbohydrates and lipids.<sup>29</sup> They are also important source of minerals such as zinc, iron, manganese and copper. In addition, they are rich in fiber, nitrogen, ash and crude protein.<sup>30</sup> The phytochemicals found in this plant are very crucial in the antioxidant activities, protection against infections and maintenance of electrolyte level.<sup>5</sup> Glycosides are stored in the gut flora where they may have a beneficial effect in animals perhaps in cholesterol reduction.<sup>9</sup> Similarly, steroids have also been reported to be involved in cholesterol reduction and may as well possess a hypocholesterolemic capability in mammals.<sup>9</sup> In addition, carotene, vitamin C, and flavonoids have high antioxidant properties while saponins are thought to have antimicrobial, anti-inflammatory, anti-oxidant and immune-stimulating properties. The presence of carotene, vitamin C, flavonoids, and saponin suggest that the samples may be beneficial in the reduction of reactive oxygen species, cancer risk, and heart diseases.<sup>10</sup>

## Vitamin A (Retinol)

Retinol or vitamin A is a carotenoid produced from the breakdown of carotene in the liver. It is known to have beneficial impact on the skin, eyes and internal organs. What confers the antioxidant ability on vitamin A is its ability to combine with peroxy radicals before they propagate peroxidation to lipids.<sup>31,32</sup> Extraction of vitamin A precursor from baobab leaves showed that Alpha carotene was 1.01mg/100g, beta-carotene was 5.92mg/100g, and cryptoxanthin was 0.81mg/100g of leaves.<sup>33</sup> In a separate study, vitamin A content of the Baobab sun dried leaves contain 9.9 $\mu$ g/g of  $\alpha$  carotene, 87.0 $\mu$ g/g of  $\beta$  carotene and 15.3 $\mu$ g/g retinol equivalent; while the shade dried leaves contain 19.4 $\mu$ g/g  $\alpha$  carotene, 157 $\mu$ g/g of  $\beta$  carotene and 27.2 $\mu$ g/g retinol equivalent.<sup>34</sup> Carotenoids has the ability to scavenge free radicals such as hydroxyl radical (OH $\cdot$ ), superoxide anion (O $_2^{\bullet-}$ ), or lipid peroxy radical LOO in plasma.<sup>35</sup> It is one of the potent antioxidant components that is derived from plant: it functions as singlet oxygen quenchers, free radical scavengers, enzymes inhibitors, peroxide decomposers and synergies.<sup>36</sup> It is important to note that singlet oxygen quenching is the main antioxidant property of carotenoids. This happens as a result of excited carotenoids that dissipate the newly acquired energy through a series of biochemical interactions with the solvent, thereby returning to the unexcited state and allowing them to quench more radical species. It is important to note that these carotenoids can be completely destroyed by free radicals called peroxy radicals. Carotenoids are relatively unreactive but may also breakdown and form non-radical compounds that

may terminate free radical attacks as they bind to these radicals.<sup>37</sup> Carotenoids are also believed to provide antioxidant protection to some tissues especially those that are rich in lipids. Research suggests that beta carotene may work in collaboration with vitamin E.<sup>38</sup> Vitamin E is a major lipid-soluble antioxidant. As the most effective chain-breaking antioxidant within the cell membrane, it helps to prevent lipid peroxidation in the fatty acids component of the cell membrane.<sup>39</sup>

## Vitamin C

Vitamin C is a very important cofactor in enzymatic reactions; an excellent electron donor which makes it a very strong water-soluble antioxidant in humans. Unfortunately, humans can only get Vitamin C from external sources as they cannot synthesize it by themselves due to the lack of gulonolactone oxidase.<sup>40</sup> A study conducted by Baobab Fruit company in Senegal observed that the concentration of vitamin C in baobab leaves ranged from 38 to 53mg/100g.<sup>41</sup> Similarly, vitamin C is considered as one of the most vital dietary antioxidants that help to reduce free-radical and oxidative stress mediated damages.<sup>40</sup> Vitamin C functions as an important antioxidant by donating two of its electrons, thereby preventing the oxidation of other compounds.<sup>42</sup> Ascorbic acid contains two important components with antioxidant capabilities: L-ascorbic acid and L-dehydroascorbic acid which are both assimilated in the intestinal tract and can be interchanged enzymatically *in vivo*. Ascorbic acid is very effective in scavenging and neutralizing free radicals like the hydrogen peroxide, hydroxyl radical, superoxide radical anion, singlet oxygen and reactive nitrogen oxide.<sup>43</sup> The antioxidant potential of Vitamin C has been shown in both *in vitro* and *in vivo* study conditions due to its interactions with reactive oxygen species. Interestingly, vitamin C can also function as a pro-oxidant in the presence of catalytic metal ions.<sup>44</sup> To be specific, vitamin C has shown the ability to interact with Fe<sup>3+</sup> and reduce it to Fe<sup>2+</sup> (or Cu<sup>3+</sup> to Cu<sup>2+</sup>), which easily reacts with hydrogen peroxide or oxygen to form hydroxyl radicals and superoxide leading to some negative impacts on biomolecules.<sup>45,46</sup>

## Flavonoids

Flavonoids can be categorized into six major subgroups, flavones, flavonols and flavanones catechins or flavanols, anthocyanidins, isoflavones. All these sub-groups of compounds share the same diphenylpropane (C<sub>6</sub>C<sub>3</sub>C<sub>6</sub>) skeleton.<sup>47</sup> Study on baobab leaves powder (stored, sun-dried & shade-dried) has shown flavonoid level of 0.15%, 0.12% and 0.22% respectively.<sup>30</sup> The presence of phenolic hydroxyl groups attached to their ring structures is what gives the antioxidant ability to flavonoids. These flavonoids function as reducing agents, metal chelators, superoxide radical scavengers, hydrogen donors as well as singlet oxygen quenchers.<sup>47</sup> They also play a role in activating antioxidant enzymes, reduce nitrosative stress, prevent oxidases, reduce  $\alpha$ -tocopherol radicals, and increase levels of uric acid and low molecular weight molecules which help in reducing oxidation in the process.<sup>48</sup>

## Zinc

Zinc is one of the most important minerals that are only found in trace quantity in humans, and forms small proportion of dietary

antioxidants. It plays a vital role in antioxidation.<sup>49</sup> In baobab leaves, zinc vary between 0.7mg/100g dw and 4.0mg/100g dw.<sup>6,50</sup> However, in a separate study, a higher value of 22.4mg/100g dw was reported.<sup>51</sup> Importantly, zinc is one of the minerals that are involved in various metabolic reactions including antioxidation. It does not directly attack free radicals, but plays quite an important role in the prevention of formation of free radical. It has been observed that zinc helps to prevent the production of the singlet oxygen radical from oxygen by inhibiting NADPH oxidases which catalyzes the reaction using NADPH as an electron donor.<sup>49</sup> Zinc is also an important component of superoxide dismutase. Superoxide dismutase is an important antioxidant enzyme that converts superoxide radical into hydrogen peroxide. Zinc is seen to induce the production of some scavengers like metallothionein that scavenges hydroxyl radical. Zinc can as well reduce the production of hydroxyl radicals by competing with copper for binding to the cell wall. Copper is believed to play a role in the production of hydroxyl radicals on the cell wall thereby inducing oxidation.<sup>49</sup>

## General antioxidant capability

The process of generating reactive oxygen species and using the test substance to cause the disappearance of these radicals is most commonly used method for measuring antioxidant activity in any substance. The scavenging activity of antioxidants is measured against a reference compound, such as Trolox which is a water-soluble equivalent of vitamin E. In a particular study, the integral antioxidant capacity which represents the sum of the antioxidant capacity of hydrophilic and lipophilic antioxidants, calculated as mmol equivalents in activity of Trolox of the baobab leaves was examined. The water-soluble antioxidant capacity of the baobab leaves was 6.39 mmol/g of Trolox, while the lipid-soluble antioxidant component of baobab leaves was 2.35 mmol/g of Trolox.<sup>52</sup> In water-soluble fractions, antioxidants such as vitamin C and flavonoids were measured; while in lipid-soluble fractions, antioxidants such as carotenoids and tocopherols were detected.<sup>53,54</sup>

## Conclusions

Baobab leaves are usually used in dry form in the preparation of a soup known as *miyan kuka* in Northern Nigeria. The leaves are rich in phytochemicals such as vitamin C, carotenes glycosides, saponins, steroids and flavonoids. Also, they are important source of minerals such as zinc, copper, iron and manganese. Some of these components work in different ways to regulate reactive oxygen species and by extension prevent oxidative stress. Therefore, consumption of *miyan kuka* can help to prevent or reduce reactive oxygen species thereby preventing oxidative stress in the long run.

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