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Antioxidant Potentials of *Adansonia digitata* (Baobab) Fruit Pulp: A Mini-Review

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Authors' contributions

This work was carried out in collaboration between both authors. Author ESO designed the study, wrote the protocol, and wrote the first draft of the manuscript. Author SAM managed the literature searches. Both authors read and approved the final manuscript.

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ABSTRACT

Background: Antioxidant is any substance that has the ability to delay, prevent or remove oxidative damage to a target molecule. The review is aimed at itemizing the components of baobab fruit pulp with antioxidant capabilities.

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

Results: Boabab fruit pulp is a very good source of nutrient with a lot of antioxidant components such as vitamin C, Beta carotene, flavonoids, phenols, zinc etc. These antioxidant components help to reduce reactive oxygen species including free radicals such as superoxide anion, hydroxyl radical, as well as non-radical molecules like hydrogen peroxide, singlet oxygen. Baobab fruit pulp carries 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 α -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.

Conclusion: Baobab fruit pulp is a super fruit with a lot of antioxidant capabilities due to the present of several components like vitamin C, Beta carotene, flavonoids, phenols, zinc etc. These components work in different ways to eliminate reactive oxygen species and by extension prevent oxidative stress.

Keywords: Baobab fruit pulp; antioxidant; reactive oxygen species; oxidative stress.

1. INTRODUCTION

Baobab fruit pulp is a source of food, and medicine, contributing to the livelihood of many populations in Africa [1.2], Several traditional uses have been documented collectively in Nigeria and other African countries [3]. The fruit pulp has been used traditionally as immunostimulant, anti-inflammatory as well as in the treatment of dysentery and diarrhea. Similarly, it is been compared and considered as a substitute for some western drugs [4,5]. In the past decade, several researchers, food and pharmaceutical companies have shown a lot of interest due to its various benefits (medicinal, nutritional and cosmetic). Interestingly, European Commission approved the import of baobab fruit pulp as a novel food [3] and it was authorised in 2009 by the Food and Drug Administration as a food ingredient in the United States of America [6]. The baobab fruit is called a super fruit because of its exotic nature and rich nutrient profile. Its fruit pulp has very high content of vitamin C, potassium, calcium and antioxidants [7].

Naturally, aerobic metabolism leads to the production of reactive oxygen species (ROS). These reactive oxygen species play important roles in the maintenance and regulation of cell survival, cell death, cell signaling, differentiation and inflammation-related factor production [8,9]. Reactive oxygen species include free radicals such as superoxide anion (O₂-), hydroxyl radical (OH'), as well as nonradical molecules like hydrogen peroxide (H_2O_2) , singlet oxygen $(^1O_2)$, etc [10]. Some environmental factors such as tobacco smoke, heavy metal exposure, UV radiation, xenobiotics, ionizing radiation together with superficial receptors activation in the cell, may cause more production of free radicals in the affected organisms [11,12]. Free radicals are highly attracted to DNA, proteins and lipids [13]. 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; they can also interact in various ways with other free radicals as well as bind to another molecule [14].

Oxidative stress occurs when the generation of free radicals exceeds the system's capacity to neutralize and eliminate them which can be as a result of either overproduction ROS or decreased antioxidant defense systems [15]. To contain this oxidative stress caused by aerobic metabolism, antioxidant defense systems have been developed by organisms. This consists of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and glutathione reductase together with a number of low molecular-weight antioxidants such flavonoids. ascorbate, α-tocopherol glutathione, cysteine, thioredoxin, vitamins, etc. [16]. In some cases ROS escape destruction and form highly reactive hydroxyl radicals due to the fact that the defense system is overpowered by various pathological or environmental factors [17]. An increase in ROS can lead to oxidative damage to DNA and other macromolecules (proteins, nucleic acids and lipids). It also plays a role in the pathogenesis of several diseases such as cardiovascular diseases, diabetes mellitus, cancer, neurodegenerative diseases as well as human aging [18].

Antioxidant is "any substance that delays, prevents or removes oxidative damage to a target molecule" [19]. In living cells, antioxidants can be categories into two groups: enzymatic antioxidants and non-enzymatic antioxidants. The enzymatic antioxidants are produced in vivo and are further subdivided into primary enzymatic defenses and secondary enzymatic defenses [20]. The primary enzymatic defense helps to prevent the formation of free radicals as well as neutralizing the already formed free

radicals. The primary enzymatic defense includes SOD, CAT and GPx. SOD helps to convert superoxide anions into hydrogen peroxide; CAT, breaks down hydrogen peroxide into water and oxygen molecule. GPx donates electrons to peroxides, thus maintaining it in a non-reactive form because peroxides are important component of Fenton reaction. Fanton reaction led to the production of ROS in the cell [21].

The secondary enzymatic defense does not neutralize free radicals directly, but support the primary enzymatic defense antioxidants. This includes glutathione reductase and glucose-6dehydrogenase. Glutathione phosphate reductase reduces glutathione peroxidase (primary enzymatic defense antioxidant) from its oxidized form to its reduced form. This recycling process is important for glutathione peroxidase to continue in neutralizing more free radicals. Glucose-6-phosphate regenerates Nicotinamide Adenine Dinucleotide Phosphate (NADPH), thereby creating a reducing environment for the primary enzymatic defense to act properly [22]. These two enzymes have supporting roles to the other endogenous antioxidants, but do not neutralize free radicals directly.

Non-enzymatic antioxidants can be classified into antioxidants and natural synthetic antioxidants. Natural antioxidants are nutrientderived 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 [23]. 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 live. Most processed foods contain synthetic antioxidants, which are reported to be safe, but recent studies indicate otherwise [24]. Some studies have indicated that most synthesized antioxidants generate substances that can lead to cancer or other diseases in the long run [25]. The aim of this review was to itemize the components of baobab fruit pulp with antioxidant capabilities.

2. 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 fruit pulp,

antioxidant properties of baobab fruit pulp, antioxidant, reactive oxygen species and oxidative stress. The articles were selected and reviewed base on the following criteria:

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

3. RESULTS AND DISCUSSION

Baobab fruit is termed super fruit because of its exotic nature and rich nutrient profile; several constituents have been identified from the fruit pulp including terpenoids, flavonoids, sterols, vitamins, amino acids, carbohydrates and lipids [26]. In the early fifties, the presence of organic acids such as ascorbic acid, citric, malic, succinic and tartaric in the fruit pulp has been identified. The pulp is about 14 to 28% of the total fruit weight with low water content which is less than 15% [27]. The fruit pulp has been shown to contains high amounts of carbohydrate (≈70%). crude fibre (≈11.2%), ash (≈5.7%) and protein (≈2.2%), and a considerable low quantity of fat (≈0.4%) [27]. Studies have shown other constituents like amino acids such as alanine, arginine, glycine, lysine, methionine, proline, serine, valine [28]; vitamins (B1, B2, B3, A, C) [2,29]; minerals (Cu, Fe, K,Mg, Mn, Na, P, Zn) [30] as well as beta carotene in the fruit pulp [31]. Ten aromatic compounds including isopropyl myristate and nonanal were identified in the fruit pulp using GC-MS [32]. Some of the constituents mentioned above are known to exhibit strong anti-oxidant activity [33].

3.1 Vitamin C (Ascorbic Acid)

Measurement of vitamin C content of baobab fruit pulp has been carried out jointly between the Novartis Foundation for Sustainable Development and the Malian Agronomic Research Institute. They discovered that vitamin C content of the bulk fruit pulp depends on the variety and geographical location: and that it varied from 1623mg/kg in one tree to 4991mg/kg in another [2]. According to De Caluwe et al. [34] the vitamin C content in the fruit pulp is almost ten times that of an orange, but Namratha and

Sahithi [35] observed that the vitamin C content is 6 times more than that of the oranges. In a particular study, it was observed that the vitamin C content of baobab fruit pulp (280-300 mg/100 g) is very high, and is seven to ten times more than oranges (51 mg/100 g) [36,37]. A different study demonstrated and recommended a daily consumption of 40 g of baobab fruit pulp for pregnant women (19-30 years) as this provides them with 100% of the recommended daily intake of vitamin C [38]. Vitamin C content of the baobab fruit was higher when compared to several other fruits including kiwi, orange, apple and strawberry [39]. A study by Eromosele et al., [40], recorded 337mg ascorbic acid/100g of the baobab fruit pulp in Nigeria. Vitamin C is seen to be one of the contributors to the acidity of the baobab fruit pulp [41].

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 [42]. Similarly, vitamin C is considered as one of the most vital dietary antioxidants that help to reduce freeradical and oxidative stress mediated damages [42]. Vitamin C is assimilated from the intestine by active transport: it functions as an important antioxidant by donating two of its electrons, thereby preventing the oxidation of other compounds [43]. Ascorbic acid contains two important components with antioxidant capabilities: L-ascorbic acid and 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 [44].

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 [45]. To be specific, vitamin C has shown the ability to interact with Fe³⁺ and reduce it to Fe²⁺ (or Cu³⁺ to Cu²⁺), which easily reacts with hydrogen peroxide or oxygen to form hydroxyl radicals and superoxide leading to some negative impacts on biomolecules [46]. Importantly, the pro-oxidant tendency of vitamin C is believed to be dependent on the dose as

well as availability of metal ions which act as catalyst during the reaction [45]. In addition, the antioxidant or pro-oxidant behavior of vitamin C has been shown to also depend on the concentration of vitamin E in the body or the system [47]. These two vitamins complement each other as they work together in the regeneration of vitamin E from the tocopheroxyl radical to an intermediate form thereby reinstating its antioxidant capability. Interestingly, this regeneration process is controlled by Vitamin C [48].

The high vitamin C and antioxidant content of the fruit pulp is believed to play a role in the extension of shelf-life for foods and beverages, as well as cosmetics. The food/beverage industry is therefore looking at the option of introducing baobab fruit pulp into foods in order to act as a preserving ingredient by preventing oxidation of lipids in the food, and by so doing prolong the shelf-life of the product [49].

3.2 Beta Carotene

Vitamin A or retinol is a carotenoid produced from the breakdown of β-carotene in the liver. It is known to have beneficial impact on the skin. eves and internal organs. What confers the antioxidant ability on vitamin A is its ability to combine with peroxyl radicals before they propagate peroxidation to lipids [50,51]. A study by Aluko et al., showed a concentration of beta carotene in baobab fruit pulp to be 3.03mg/100g and 3.19mg/100g from two different locations [52]. Carotenoids belong to the class of phytochemical compounds that has the capacity to scavenge free radicals such as superoxide anion (O₂-), hydroxyl radical (OH*), or lipid peroxyl radical (LOO') in plasma [53]. It is an interesting antioxidant component that is derived from plant: it functions as singlet oxygen quenchers, free radical scavengers, enzymes inhibitors, peroxide decomposers and synergies [54]. It has been observed that singlet oxygen quenching is the main antioxidant property of carotenoids. This happens when excited carotenoids that dissipate the newly acquired energy through a series of rotational and vibrational interactions with the solvent, thereby returning to the unexcited state and allowing them to quench more radical species. This can occur while the carotenoids have conjugated double bonds within. It is important to note that peroxyl radicals are the only free radicals that completely destroy these carotenoids. Carotenoids are seen to be relatively unreactive but may also decay and form non-radical compounds that may terminate free radical attacks by binding to these radicals [55]. Beta carotenes as well as other 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 synergistically with vitamin E [56]. Vitamin E is a major lipid-soluble antioxidant. It is the most effective chain-breaking antioxidant within the cell membrane where it protects membrane fatty acids from lipid peroxidation [57].

3.3 Flavonoids

Flavonoids are grouped into six major subgroups, catechins or flavanols. anthocyanidins, isoflavones, flavones, flavonols and flavanones. All these sub-groups of compounds share the same diphenylpropane (C6C3C6) skeleton [58]. Catechin is a flavonol (flavonoid) found in many plants such as grapes, cocoa and tea. It has also been isolated from the boabab fruit using column chromatography [59]. Catechin is known to exhibit strong anti-oxidant activity [60] and can also promote survival in diabetic mice [61]. Study has shown the flavonoid level of aqueous methanol extract of the baobab fruit pulp to be 42.7 mg QE (quercetin equivalents)/100g which is higher than the aqueous acetone extract of 31.7 mg QE /100g of the fruit [62]. The antioxidant potential of the flavonoids is as a result of the presence of phenolic hydroxyl groups attached to their ring structures. Flavonoids act as reducing agents, singlet oxygen quenchers, superoxide radical scavengers, hydrogen donators as well as metal chelators [58]. They also help in activating antioxidant enzymes, reduce α-tocopherol radicals, reduce nitrosative stress, prevent oxidases, and increase levels of uric acid and low molecular weight molecules which help in antioxidation in the long run [63].

It has been observed that the ability to directly scavenge the reactive oxygen species is the best-described antioxidant property of flavonoids. They have the ability to chelate free radicals immediately by single-electron transfer or by donating a hydrogen atom [63]. Also, chelation of transition metal elements is seen to be another possible mechanism of action of flavonoids [64]. Flavonoids with this chelating property enable them to chelate, or bind to metal ions present in the human body making them inaccessible for oxidation reaction. Some flavonoids have the ability to bind or chelate trace metal ions such as

Fe2+ and Cu+ that play a vital role in metabolism of oxygen leading to free radical formation [64]. Apart from scavenging free radicals directly and chelating of transition metal elements, flavonoids can also inhibit free radical generating enzymes such as protein kinase C, cyclooxygenase, microsomal monooxygenase, mitochondrial succinoxidase, xanthine oxidase, lipoxygenase, and NADPH oxidase: thereby functioning as an intracellular antioxidant [62]. Similarly, internal antioxidant enzymes induction is believed to be another possible pathway through which flavonoids function as an antioxidant. It is important to note that the most important defensive enzymes against intracellular toxicants and xenobiotic are phase II metabolizing enzymes (e.g. UDP-glucuronosyltransferases, glutathione S-transferases and methyltransferases. sulfotransferases. Nacetyltransferases,) which flavonoids tends to induce [63].

3.4 PhenoIs

Phenolic acids are made up of hydroxycinnamic and hydroxybenzoic acids. They are very common in plant material and in some cases present as glycosides and esters. Phenolic acid is well known for their good anti-oxidant activity [65]. It has been shown from a particular study that the total phenolic levels in the aqueous methanol of baobab fruit pulp was 4057.5mg GAE (gallic acid equivalents)/100 g of fruit which was significantly higher than the aqueous acetone extracts with 3518.3mg GAE /100 g of fruit [62]. (Lamien-Meda et al. 2008). In a different investigation, Nhukarume et al. [66] showed that phenolic content of baobab fruit pulp was ≈52 mg/100 ml GAE which was not statistically different from that of Citrus sinensis Pers: and he suggested the use of baobab fruit pulp as an anti-oxidant botanical dietary supplement. The antioxidant activities of phenolic acid include metal chelating and free radical scavenging with special impact on hydroxyl and peroxyl radicals, superoxide anions peroxynitrites [67].

3.5 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 [68]. Study has shown the present of certain minerals such as Zn, Cu, Fe, K, Mg, Mn, Na, P in the baobab fruit pulp. The concentration of zinc in the baobab

fruit pulp is seen to be 10.4 µg/g dry weights [69]. In a separate study, the concentration of zinc was found to be 1.8 mg/100g of the fruit pulp [70]. Kinuthia et al. [71] also measured the concentration of zinc and found it to be 71.6 µg/g of the fruit pulp. Interestingly, zinc is one of the minerals that take part in various metabolic reactions together with antioxidation. It plays quite an important role in the prevention of formation of free radical, but does not directly attack free radicals. 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 [68]. Zinc is also present in superoxide dismutase. Superoxide dismutase is an important antioxidant enzyme that converts the singlet oxygen radical into hydrogen peroxide. Zinc is seen to induce the production of some scavengers 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 [68].

3.6 General Antioxidant Capability

The most commonly used method for measuring antioxidant activity in any substance involves generating reactive oxygen species and using that substance to cause the disappearance of these radicals. The scavenging activity of antioxidants is measured against a reference compound, such as Trolox, a water-soluble equivalent of vitamin E. In a particular study, Vertuani et al. [72] investigated the antioxidant activity of baobab fruit pulp (PCL) method of photochemiluminescence aqueous/methanol extracts of baobab products [73,74]. This method allows for the measurement to be done on the antioxidant capacities of both water and lipid-soluble components. In watersoluble fractions, antioxidants such as vitamin C and flavonoids were measured; while in lipidsoluble fractions, antioxidants carotenoids and tocopherols were detected [73,75]. In this study, Vertuani et al. [72] observed that the water-soluble antioxidant capacity of the baobab fruit pulp was 6.96 mmol/g of Trolox, while the lipid-soluble antioxidant component of baobab fruit pulp was 4.15 mmol/g of Trolox. The higher value of watersoluble component suggests that vitamin C content is the major contributor to the activity.

Another method used to assess antioxidant capacity of a substance is ORAC assay (Oxygen Radical Absorbance Capacity). This method measures substance (antioxidant) capacity to inhibit peroxyl radical that induced oxidations and thus reflects classical radical chain breaking antioxidant activity by Hydrogen atom transfer [76]. Values in this method are also reported as Trolox equivalents. It was observed that seasonal variation in fruit products, different methods of extraction and treatment of a sample can lead to differences in the outcome value [77] In a particular study using ORAC method, the value of baobab fruit pulp was seen to be 200 ORAC units (µ mol TE/g). In a separate study using the same method, the antioxidant capacities of the fruit pulp from the six species varied between 109 and 159 µmol TE/g. Cissé et al. [78], Compared the value obtained from baobab fruit pulp with cranberries pomegranate, he discovered that the antioxidant capacity of baobab fruit pulp is twice that of cranberries and pomegranate. It was submitted that there is additional potential antioxidant benefit from the consumption of baobab fruitcontaining products [79].

4. CONCLUSION

Baobab fruit pulp is a super fruit with a lot of antioxidant capabilities due to the present of several components like vitamin C, Beta carotene, flavonoids, phenols, zinc etc. These components work in different ways to eliminate reactive oxygen species and by extension prevent oxidative stress. Therefore, optimal consumption of the fruit pulp is recommended for prevention of reactive oxygen species that cause oxidative stress.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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