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Reducing Potentials of *Pennisetum Glaucum* and *Sorghum bicolor*

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ABSTRACT

Oxidative stress, resulting from an imbalance between free radicals and antioxidants, is a key factor in the development and progression of various diseases. Antioxidants, which neutralize free radicals, play a crucial role in maintaining cellular homeostasis. Understanding the antioxidant properties of food components can help mitigate oxidative stress and related diseases, highlighting the importance of dietary antioxidants in promoting overall health and well-being. This study investigates the antioxidant properties of *Pennisetum glaucum* (millet) and *Sorghum bicolor* (guinea corn) using the ferric ion reducing power assay (FRAP). The aqueous 1M acetic acid extracts of the samples were tested at varying concentrations (15 µl/ml, 30 µl/ml, and 45 µl/ml) and compared to standard antioxidants, butylated hydroxyl toluene (BHT) and α-tocopherol. The results showed that both millet and guinea corn possess antioxidant potentials, with guinea corn exhibiting higher reducing power at lower concentrations. Millet demonstrated a concentration-dependent increase in reducing power, with values of 0.268, 0.413, and 0.488 at 15 µl/ml, 30 µl/ml, and 45 µl/ml, respectively. Guinea corn showed reducing power values of 0.452, 0.473, and 0.485 at the respective concentrations. The study's findings suggest that both *Pennisetum glaucum* and *Sorghum bicolor* have antioxidant capacities, which can help mitigate oxidative stress and related diseases. The results support the recommendation of incorporating these cereals into daily diets to harness their antioxidant benefits.

KEYWORDS: Auto-oxidation, Antioxidants, Reductants, Reducing-power

INTRODUCTION

Antioxidants significantly delay or prevent oxidation of oxidizable substrates when present at lower concentrations than the substrate (Halliwell, 2007). Antioxidants can be synthesized *in vivo* (e.g., reduced glutathione (GSH), superoxide dismutase (SOD), etc.) or taken as dietary antioxidants (Halliwell, 2007). Plants have long been a source of exogenous (i.e., dietary) antioxidants. It is believed that two-thirds of the world's plant species have medicinal importance, and almost all of these have excellent antioxidant potential (Krishnaiah, 2011). Plants have an innate ability to biosynthesize a wide range of non-enzymatic antioxidants capable of attenuating ROS- induced oxidative damage (Kasote *et al.*, 2015).

Plants synthesize low molecular weight antioxidants such as glutathione and ascorbate within the chloroplast stroma and cytosol using NADPH as the ultimate electron donor (Carocho and Ferreira ,2013). These low molecular weight antioxidants function as redox buffers that interact with numerous cellular components and influence plant growth and development by modulating processes from mitosis and cell

elongation to senescence and death (Oladiji *et al.*, 2007; Foyer, 2005).

S. bicolor is an annual plant having its different parts widely used in traditional medicine. Other names for sorghum include large millet, Indian millet and *jowar*. This is a grass plant believed to originate in Africa, where it remains a major crop. It is grown and sold for a number of purposes except for human consumption. *S. bicolor* is a less well-known grain on the global food market. It ranks in the distant fifth place among most famous cereals - after barley, rice, wheat and corn. Although less popular, sorghum is an important culture that has long played a vital role in certain diets.

Sorghum is known to be rich in phenolic compounds, many of which act as antioxidants. It has also been shown to be useful for reducing certain forms of inflammation due to its antioxidant properties. Several of the phenolic compounds in sorghum are associated with anticancer effects. Sorghum is a popular food for animals as well as emerging biofuel. Sorghum can be cooked and eaten in many ways, but it is also often processed into ingredients for other dishes. A

quarter cup of whole grain sorghum contains approximately the following nutritional values:

Calories: 163 kcal, Protein: 5 g, Fat: 2 g, Carbohydrates: 36 g, Fiber: 3 g.

Ethno-botanical reports showed that a preparation from *S. bicolor* seed possessed demulcent, diuretic, emollient, remedy for cancer, epilepsy, flux, and stomach ache (Okoye et al., 2014). Sorghum is used in traditional medicine in developing countries, including primary care of anemia, cancer, and a variety of infectious diseases, including viral diseases.

Pearl millet, commonly known as bulrush millet (*Pennisetum glaucum* (L.) R. Br.), also classified as *P. typhoides*, *P. americanum*, or *P. spicatum*, is a cultivated, small-grain, tropical cereal grass. Vernacular names include: “bajra” (India), “gero” (Nigeria, Hausa language), “hegni” (Niger, Djerma language), “sanyo” (Mali), “dukhon” (Sudan, Arabic), and “mahangu” (Namibia). Pearl millet is quantitatively the most important millet, with world annual production ~14 million tons (Mt). It is cultivated mainly in the semiarid tropics, almost exclusively by subsistence and small-scale commercial farmers.

MATERIALS AND METHODS

Extraction of samples

Extracts of ground air-dried samples were prepared following method of Swain and Hills (1959), with some modifications. Four grams of ground sample was mixed with 25 ml of 1 M acetic acid and homogenized using an orbital Rocker and Shaker. The homogenate was centrifuged at 3000 rpm for 20 minutes using centrifuge. The supernatant was gently recovered with a pastuer pipette, and assayed with ferric ion reducing antioxidant power method. *P. glaucum* and *S. bicolor* were used as samples.

The Ferric ions (Fe^{3+}) reducing antioxidant power method (FRAP)

The reducing power of *P. glaucum* and *S. bicolor* and standards (BHT and α -tocopherol) were determined by the method of Oyaizu (1986) as modified by Gulcin 2006a. 15, 30 and 45 μl extracts and standards were treated differently.

For example, to 15 μl of *P. glaucum* extract added with 1 ml of distilled water were mixed with 2.5 ml of 0.2 M sodium phosphate buffer pH 6.6 and 2.5 ml of 1% potassium ferricyanide [$\text{K}_3\text{Fe}(\text{CN})_6$]. The mixture was

incubated at 50 °C for 20 minutes. Aliquots 2.5 ml of 10 % trichloroacetic acid were added to the mixture. The upper layer of solution (2.5 ml) was mixed with distilled water (2.5 ml) and FeCl₃ (0.5 ml, 0.1%), and the absorbance was measured at 700 nm in a spectrophotometer. Absorbance of different concentration of the extracts (15, 30 and 45 µl/ml) were compared. Increase in absorbance of the reaction mixture indicates an increase of reduction capability (Buyukokuroglu *et al.*, 2001; Gulcin *et al.*, 2005a). The solutions without sample extracts or standards were used as blank samples. Reducing antioxidant power determination was performed in triplicate.

RESULTS AND DISCUSSIONS

Results

Total ferric power determined according to the ferric ions (Fe³⁺) - ferrous ions (Fe²⁺) transformation. Results from fig. 1 showed the reducing potentials of *P. glaucum* and standards (BHT and α-tocopherol) using ferric reducing antioxidant power (FRAP) assay. *P. glaucum* showed an increasing reducing potential as concentration increases.

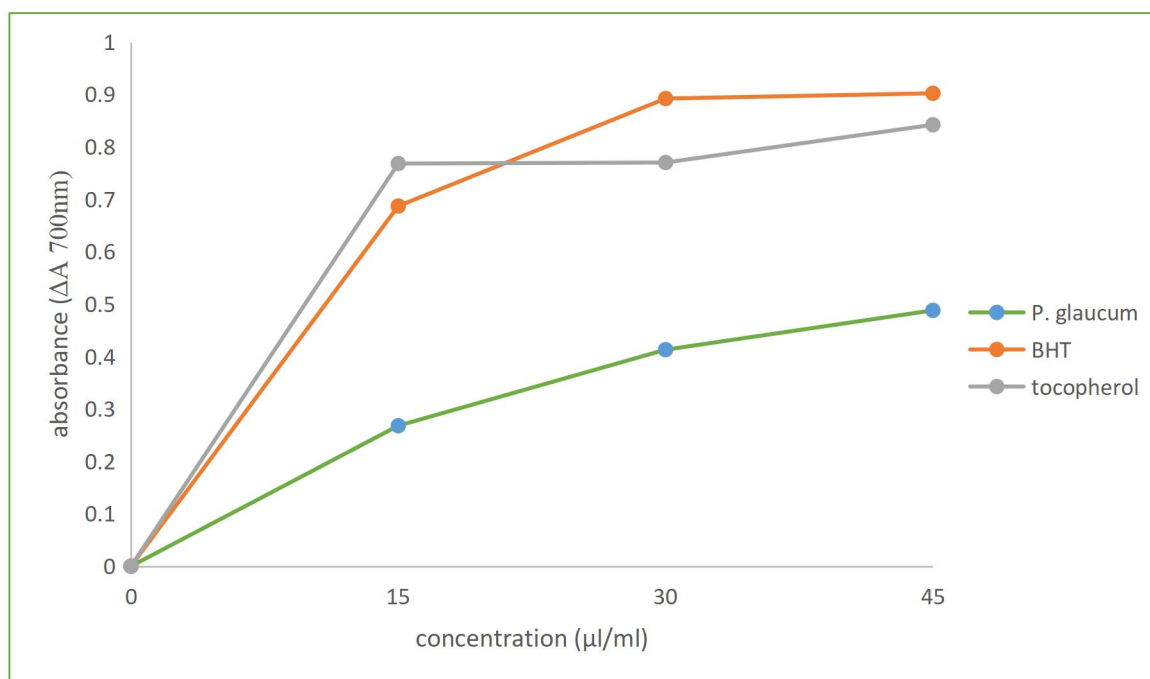


Fig. 1. Reducing potential of *P. glaucum* as compared with standard (BHT and α - tocopherol) using Fe^{3+} - Fe^{2+} conversion.

S. bicolor showed a much lower reducing potentials when compared with BHT and α - tocopherol (standards) as shown in figure 2. The reducing power was estimated based on the absorbance reading at 700 nm with a spectrophotometer. Reducing power will increase accordingly to the increase in absorbance.

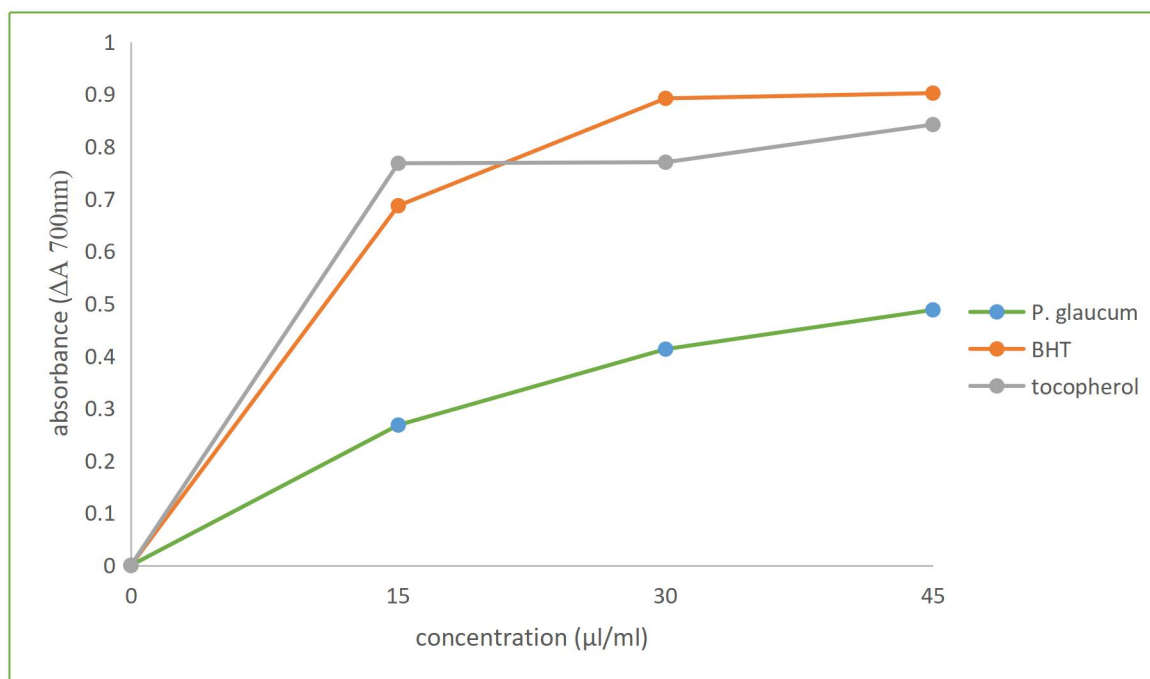


FIG 2: Reducing potential of *S. bicolor* as compared with standard antioxidant (BHT and α -tocopherol) using Fe^{3+} - Fe^{2+} conversion

A combined result for the samples and standards (fig 3) showed that at the concentration of 15 $\mu\text{l/ml}$, *P. glaucum* had the lowest reducing potential but highest among the samples at the concentration of 45 $\mu\text{l/ml}$. the standards (BHT and α -tocopherol) showed very higher reducing ability when compared to the samples (*P. glaucum* and *S. bicolor*). The reducing potential values are expressed as mean \pm standard deviation of three replicate determinations.

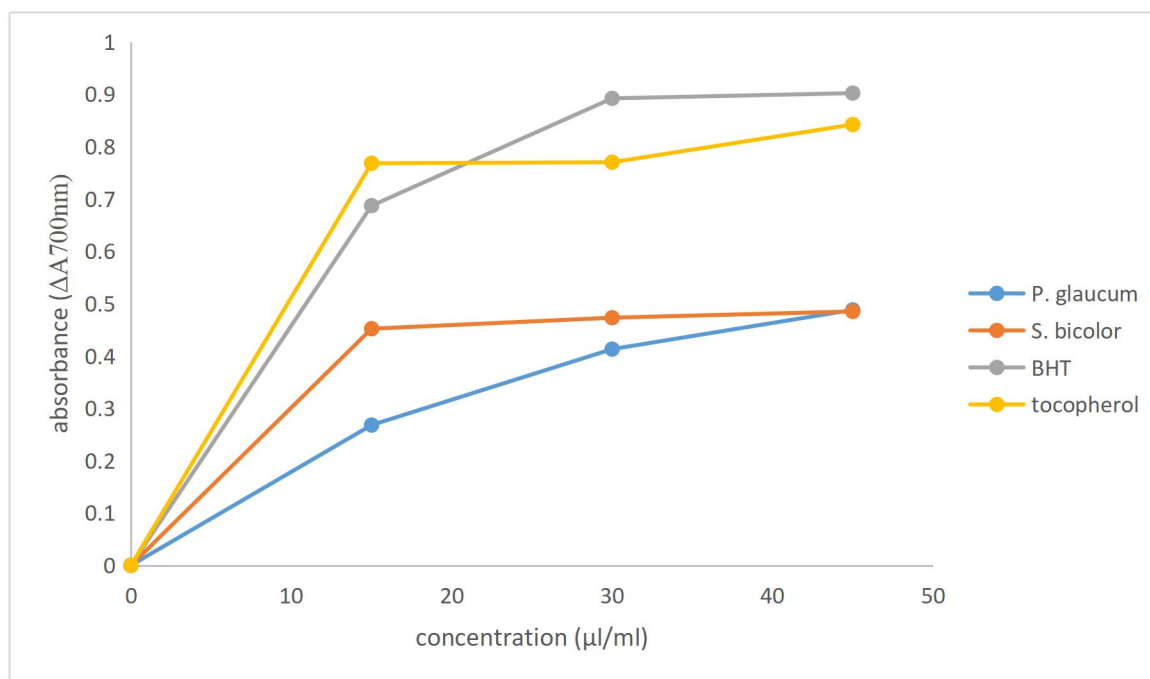


Fig 3: Reducing potentials of *P. glaucum*, *S. bicolor*, Butylated hydroxytoluene (BHT) and α -tocopherol

Discussion

Antioxidants can be explained as reductants, and inactivation of oxidants by reductants can be described as redox reactions in which one reaction species is reduced at the expense of the oxidation of the other. The presence of reductants such as antioxidant substances in the antioxidant samples causes the reduction of the Fe^{3+} /ferricyanide complex to the ferrous form. Therefore, Fe^{2+} can be monitored by measuring the formation of Perl's Prussian blue at 700 nm (Chung *et al.*, 2002). In the Ferric ions (Fe^{3+}) reducing antioxidant power (FRAP) method, the yellow colour of the test solution changes to various shades of green and blue depending on the reducing power of the samples. The reducing capacity of a compound may serve as a significant indicator of its potential antioxidant activity.

As shown in fig 1, *P. glaucum* had a steady increasing reducing power using potassium ferricyanide method when compared to *S. bicolor* but much lower reducing potentials when compare with the standards (BHT and α -tocopherol). For the measurement of the reductive ability of *P. glaucum* and *S. bicolor*, the Fe^{3+} - Fe^{2+} transformation was

investigated in the presence of the samples using the method of Oyaizu (1986). Reducing power will increase accordingly to the increase in absorbance. As more Fe^{3+} are reduced to the ferrous form or when more electrons are donated by antioxidant components. Reductants also react with certain precursors of peroxide, thus preventing the formation of peroxide.

At different concentrations (15-45 $\mu\text{l/ml}$), *P. glaucum* demonstrated an increasing reducing ability ($r=0.997$) as concentration increases and these differences are statistically significant ($p<0.05$). It was observed that *S. bicolor* had higher reducing potential than *P. glaucum* but it did not significantly increase at increasing concentration. The reducing power of *P. glaucum*, *S. bicolor* and standard compounds (BHT and α -tocopherol) exhibited the following order: BHT > α -tocopherol > *S. bicolor* > *P. glaucum*. Guinea corn showed an increasing reducing power as concentration increases.

The results on reducing power demonstrate the electron donor properties of samples thereby neutralizing free radicals by forming stabilized products. The outcome of the reducing reaction is to terminate the

radical chain reactions that may otherwise be very damaging.

Conclusion

Antioxidants play crucial roles in the cellular metabolism of aerobes in that they reduce oxidant species formed during normal oxidative processes. Thus, it is of use to note common cereals that have better reducing potentials (antioxidant capacities) for recommendations for use in daily consumption. *P. glaucum* showed a promising reducing potential than *S. bicolor* as its absorbance which correlates with reducing potential increases significantly with increase in concentration.

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