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To cite this article: Obinna C Nwinyi and Phoebe O Umane 2019 IOP Conf. Ser.: Earth Environ. Sci. 331 012057

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Review On Probiotics Potentials, Nutritional Composition Of Bambara Nut (Vigna Subterranea (L.) - An Underutilized LegumE.

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Abstract. Bambara nut is one of the underutilized legumes. Conversely this underutilized legume could be utilized to solve the ravaging food security issues in sub-Saharan Africa owing to its balanced nutritive composition. Bambara nut limited use, is because of the antnutritional factors, hard to cook phenomenon and difficulty in dehulling. Because of high global demand on food security, there is an urgent need to review possible methods that could reduce the limiting factors to the widespread use of this crop and enunciate other benefits derivable from this legume. In this review, we appraised the nutritional composition of Bambara nut from different geographic locations, commonly associated bacteria species that predominate mostly during Bambara fermentation and their potential uses as probiotics. Expedient areas for further research were highlighted in this paper

1. Introduction

Bambara groundnuts (Vigna subterranea) varieties are of two types; V. subterranea var. spontanea (the wild type) and V. subterranea var. subterranean (the domesticated type). Bambara groundnut is an under-utilized legume mostly grown by the female gender in tropical and sub-tropical areas where it serves as a means of livelihood[1,2]. This legume is also cultivated in some Asian countries such as India, Malaysia, Philippines and Thailand [3]. It belongs to Fabaceae family under the genus Vigna. The crop takes its name from the Bambara tribe in Mali and in the Sahelian provinces of West Africa [4-6]. It has been rated as the third most significant pea in West Africa, in terms of its versatility [7-8]. The husk may be crumpled and it is 1.5cm in length enclosing 1 to 2 seeds [9]. However, the pea itself is to a great extent oval or round. The pods are yellowish-white when unripe, and yellowishbrown or purple when ripe [10]. The seeds come in different sizes and colours such as dark-brown, black, white, red, creamy or mixture of the colors. It may be spotted with or without hilum coloration [11]. The nuts appear smooth, round and tough.

Bambara groundnut has various vernacular names based on different tribes/areas. These include but not limited to: indhlubu, Nzama, Njama (Malawi), jugo beans, Ndlowu, Nduhu nwa tzidzimba (South Africa), Ntoyo cibemba, Juga bean (Republic of Zambia), Njogo bean(Central Africa). In Nigeria the vernacular names include: Gurjiya or Kwaruru, Okpa and Epa- Roro. In Zimbabwe, it bears Nyimo beans while is called Aboboi akyii in Ghana. [12-14].

2. Nutritional composition

Bambara nut has nutritional benefits which could improve the level of malnutrition and boost food security. The Bambara nut seed is comprised of (%) 63 carbohydrate, 19 protein and 6.5 fat [14, 16-18]. Other works on the nutritional benefits of Bambara nuts obtained from various geographical location revealed similar trends. [19] reported Bambara nut from the South East Nigeria to comprise



International Conference on Energy and Sustainable Environment

IOP Publishing

IOP Conf. Series: Earth and Environmental Science **331** (2019) 012057 doi:10.1088/1755-1315/331/1/012057

of (%) 2.86 moisture, 32 protein, 7.4 fat, 5.78 ash, 2.68 crude fibre and 53 carbohydrates . In Northern Nigeria, [20] revealed that Bambara nut from Kano contained approximately (%) moisture content 13, ash 3.52, protein 19, fat 7; fibre 6 and carbohydrate 63. In the North central (Benue State), [21] reported of Bambara nut seeds containing approximately (%) 6 moisture content, 4 ash content; 6 % crude fibre, 20% protein content, 6% lipid composition and 56% carbohydrate. Also Commercial Bambara nut collected from South- West Nigeria by [22] revealed to possess moisture content of (%) 7 Protein (%) 18 Fat (%) 6, Ash (%) 4, Crude fiber (%) 4 and Carbohydrate 60%. Bambara seeds are highly proteinous, with malted seeds consisting of ten dissimilar proteins and dry seeds consisting of twelve different protein [4].

Boiled bambara nuts has antidiarrheic properties, it's mostly applied by the Lio tribe of Kenya. Bambara leaves serves in healing infected wounds. Its sap is used on the eye for epilepsy treatment [23] . Amongst others, the roots when eaten has an aphrodisiac effect, Igbos in Nigeria make use of its plant for venereal ailments. In Senegal, cataract is managed using a mixture of minced bambara nut and water. The raw seed suppress nausea when eaten by pregnant women, and also suppresses vomiting [14,16]. In Botswana, black land races treat impotence with Bambara plant [24]. Bambara nut possesses other nutrients, these include: Vitamin A, thiamine, riboflavin, niacin and carotene. The nut also contains trace amounts of vitamin C [25-26]. The seeds also contain micronutrients such as zinc, calcium, iron and potassium [27], but have trace quantities of phosphorus and magnesium. Seed with red coloration have about two-fold measure of iron as cream seeds; thus, they are fundamental for the remedy of iron deficiency [26]. Germinating Bambara nuts hold nutrients and also take out antinutrients efficiently at the same time [28] Studies have shown high levels of flavonoids, saponins and alkaloids present in the seed could assist in the general well - being of human [29]. In their study, they showed the beneficial effects of consuming Bambara nut which was inferred to due to flavonoids, saponins and alkaloids that occur at high levels in the seed. Anthocyanins, phenols and carotenoids could occur at low concentrations; of which their presence could be favorable since they act probably in synergy with other secondary metabolites.

2.1. Health benefits

• Reduction in dangers of stomach cancer

• Fatty acids present in nuts provide antioxidant properties capable of suppressing development of carcinogenic constituents in the stomach. Thus, the nuts could prevent stomach cancer [30].

• Antimicrobial action

• Bambara milk can serve as a probiotic. Probiotics minimize proliferation and activity of pathogenic organisms [26, 30].

- Rich radix of lysine
- Lysine Production

Bambara nut produce lysine naturally which humans are unable to produce [30]. As stated in [31] Bambara nut contains high levels of Lysine content. Lysine required for body enhancement and critical roles in making carnitine that functions in creation of energy and brings about decreased cholesterol level in blood via breaking down fatty acids. Furthermore, lysine aids in bone development by infusing calcium and assist in collagen formation that is essential in creation of connective tissue (e.g., tendon, skin. and cartilage) and bones[30]. Fights Kwashiorkor

Bambara groundnut contains high levels of protein that aid in the prevention of protein deficiency typically seen in undernourished children often known as Kwashiorkor [30]. Wholly cereal-based meals can be supplemented with Bambara as a protein source. Thus, malnutrition due protein deficiencies could be prevented.

2.2. Limiting factors to use of Bambara nut

Anti-nutritional factors: There is a connection between colours of the seeds and levels of antinutrients found in them. For example, cream Bambara nuts have low tannin content while brown and red nuts have high tannin content [13-14]. Other anti-nutrients present include: trypsin inhibitor, tannic acid, tannin, phytin oxalate, haemagglutinin and phytic acid. These lead to the decrease in nutritive significance of this legume [8, 28]. **Figure 1** shows anti-nutrients that could be present in different forms of bambara nut during its preparation for other uses. Hard to cook phenomenon (HTC): HTC factor is a principal reason behind the underutilization of this legume. The alterations in the cotyledons and seed coat result in the HTC factor. These factors could advance under storage conditions such as high temperature (30-40 °C) and high relative humidity (RH) (> 75%) in tropical and subtropical areas [32-36]. Boiling time could be increased to about 3-4 hours due to this HTC factor and this is a major problem for those residing in rural areas that use fire wood for its preparation. Due to long duration of cooking, it demands more use firewood/increase in costs of the woods that are rarely available in most rural areas [37]. Aside from the upsurge in the cost of wood, deforestation of trees will be on the increase [38].

Bambara nuts ought to be preserved at low temperature and humidity to avoid the HTC factor. Some latest methods to deal with this HTC phenomenon include chemical methods (cooking aids), physical (grinding, broiling and canning) and biological methods (germination and fermentation) [26,39].

2.3. Cultural disposition

Among the Mzimba culture, Bambara nut consumption is tied to some beliefs. It is believed that if the right persons does not handle it, it could result to evil. The Malawi culture views Bambara nut as difficult to ingest, and with terrible scent and in some cases taste-less. Their convictions influence them to stay away from the Bambara nut. In some instance, the bullet shape of Bambara nut portends danger that one could be shot via strayed bullets. In some cultures they believe bambara nut plays a role in witchcraft [40].

2.4. Ignorance of appropriate agronomic practices and prospects as a cash crop

Bambara nut are generally developed by rustic ranchers and the greater part of them have no training on the great benefits of this crop (Ibrahim and Ogunwusi, 2016). Consequently they need to be trained on recent agronomic methods to improve production of this legume. Most farmers do not consider the legume to be a cash crop because typically, it is grown around the farmers' households (William et al., 2016).

2.5. Availability and access to markets

According to [8]; Bambara nut could be limited in supply due to several reasons. These include but not limited to:

Inaccessibility of nuts lasting through the year because of little conservation and preparations employed to store the nuts when harvest time is over

Little scale agriculturists have a tendency to cultivate some other crops that they believe would be in demand by customers than Bambara nut

Effect of Hoarding: The bambara nut can withstand environmental dryness when there is scarcity of water supply. Different crops cannot survive such conditions thus farmers will rather save the legume for personal use than distribute it for sales.

Export rejection and poor marketing networks for local and especially international markets limit the widespread use of bambara nut. Government support and sponsorship is also at low level as governments do not seem to value Bambara nut as a cash crop. The government can assist in creating awareness about the benefits of bambara nut as a cash crop rather than the lack of awareness of the

benefits the crop is having. Urbanisation is deemed an additional factor as it appears to confine the crop to mostly rural, subsistence-agriculture life [8].

2.6. Bambara nut a good means of achieving Food Security

Farming Bambara crop is not capital intensive; hence it could provide new vistas of opportunities for agriculturists and farmers. It is a remarkable wholesome quality crop. Since is mostly cultivated by female genders, it provides a functional way to economically empower women, thus improve the living conditions of communities. The seeds are drought resistant, pest- and disease-resistant. Bambara nut is perfect for intercropping and crop rotation; they do not require huge land space but do enhance nitrogen fixation and consequently soil fertility [27].

2.7. Fermented Bambara nut

[41] stated that fermentation modifies leguminous seeds physically, helps lower anti-nutrients, improves nutrients, digestibility of seeds and also adjusts flavor. Fermentation has been able to cause a decrease in under-utilization of several legumes [42]. A traditional condiment such as those produced from African locust bean(Parkia biglobosa) daddawa/Iru can be produced from Bambara groundnuts [41-48] proposed that bambara flour could made from using Rhizopus oligosporus and Rhizopus nigricans as starter cultures during bambara nut fermentation. The product of fermentation could act as good supplement with enhanced nutritional composition. The researchers inferred that the product could provide avert obesity, cardiovascular diseases, heart attack and other related ailments that are predominant in developing nations. [49] reported that when bambara nut is fermented into Bambara-Ogi, It can adequately serves as food for infants since the pattern of amino acids created by this blend compares favourably to those recommended for infants.

In all fermentation procedures, there is need to properly comprehend the microbial ecology of legume fermentations. This usually requires one to know fermentation substrates i.e. seeds of the different plants and the end products [43]. For example, Bacillus subtilis, B. licheniformis (Antai and Ibrahim, 1986), Staphylococcus saprophyticus, and Leuconostoc spp. are predominant organisms during African locust bean fermentation into Dawadawa/Iru [50-51]. Dadawa/Soyiru contains Bacillus subtilis (Omafuvbe et al., 2002) and Bambara-daddawa contains Staphylococcus spp., Streptococcus, Enterococcus, Bacillus subtilis, Bacillus pumilus and B. lichenformis [46]. Traditionally, fermented legumes have differences in quality and stability of product due to lack of starter cultures (Achi, 2005). Conversely with Hazard Analysis Critical Control Points (HACCP) generated for improvement of Microbial Safety Quality during the fermentation processes, the variations can be averted [53].

2.8. Common organisms associated with Fermentation of the Bambara nut

Information on succession studies of the fermentation protocols for Bambara groundnut solid state fermentation or liquid state fermentation has been scarcely reported. However few studies on Bambara nut fermentation revealed that the predominant organisms are mostly Bacillus species. The notable bacteria species include: Bacillus subtilis, Bacillus licheniformis, Bacillus pumilus, Staphylococcus spp. and Streptococcus Enterococcus [47].

2.9, Common bacteria species associated with fermented bambara nut with probiotic potential

Globally, statistics indicate that about 1.8 million people die annually as a result of food borne diseases caused by contamination. Gastrointestinal disorders are major health challenge when microbes, chemical, or physical agents are ingested.

Bambara groundnut an underutilized crop could have probiotic potential with high nutritional composition [4, 47]. According to Cutting, (2011); for over 50 years Bacillus species have been known to provide probiotics potentials. Some Bacillus species with probiotic function from naturally fermented bambara groundnut have been identified and shown to prevent gastrointestinal disorders See Table 1, [54-55] See Table 1.0). These bacteria species could assume a critical part in the gut as a

result of their high metabolic actions and ability to survive in the gastro-intestinal tract due to presence of certain properties. For example, the spore structure has an outermost layer called the exosporium which is made up of proteins, lipids and carbohydrates that is responsible for hydrophobic properties. The higher hydrophobicity and the lower energy barrier of spores results in significantly higher adhesion forces, compared to vegetative cells [56-59]. Various workers [60-62] reported of the probiotic potential of some organisms they worked with, although no detailed scientific proof to support these claims. Recently, scientific evidence of probiotic bacteria ability to prevent and treat variety of diseases have emerged[63]. Generally, probiotics have been known for their safety, but in rare cases bacteria-host contact and reactions may arise [64-65].

To ensure of the safety of potential probiotic organism the following tests could be carried out:

- Choice of antibiotic resistance schemes
- Evaluation of definite metabolic activities (e.g., D-lactate production, bile salt deconjugation)
- Assessment of reactions amid human analysis
- Epidemiological reconnaissance of antagonistic episodes in customers
- Toxin generation
- Hemolytic potential could be assessment

Table 1.0 Common bacteria species associated with fermented bambara nut with probiotic potential

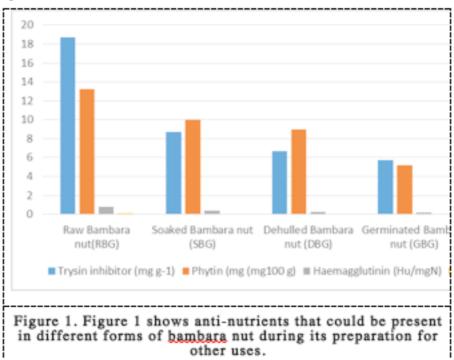
	Predominant microorganisms	Product source	source
Naturally found during	Bacillus subtilis,	Iru-like condiment	Fadahunsi and
fermentation process	Bacillus licheniformis	from Bambara nut,	Olubunmi, 2010
*	Bacillus pumulus	Bambara daddawa	
	Staphylococcus spp.		
	Streptococcus,		
	Enterococcus		
Artificially induced	Lactobacillus	Bambara Yoghurt	Murevantemas and
(starter culture)	bulgaricus,	C	Jideani 2013
	streptococcus		
	thermophiles L.		
	plantarum		Pahare et al., 2017
	-		Olanipekun et al., 2012
	Rhizopus (R.	Bambara flour	*
	oligosporus, R oryzae,		
	R.nigricans		

3. Bacillus spp as a Probable Probiotic

Bacillus species have been found to have probiotic abilities and can survive the low pH of the gastric environment, making them preferable to non-spore formers as a probiotic (Cutting, 2011).

The ability of spores to withstand heat makes spores more advantageous than vegetative cells. For example, Lactobacillus spp. and Bacillus strains have improved digestion, immune modulation and stimulates growth in many animals [66-67]. [68] reported Bacillus subtilis makes use of 4-5% its genome for antibiotic production. To function admirably as a probiotic, the organism must possess basic requirements for survival in the gastrointestinal tract, notwithstanding great adherence and bio remedial characteristics. B.subtilis has been reported to acclimatize to prevailing conditions in the

human gastrointestinal tract. They can cope with antimicrobials, create biofilms, sporulate anaerobically and could be viewed as gut commensals [69-70]. Bacillus spores can tolerate serious gastric acids, bile salts and other antagonistic conditions of the gastrointestinal tract [71-72]. Furthermore, Bacillus species have been reported to support human well-being by aversion and cure of Heliocbacter pylori contamination, looseness of the bowels, support of gut homeostasis and prevent gingivitis [73-75].



Source: (Omoikhoje et al., 2006)

3.1. Possible mechanisms of actions of Bacillus species:

Direct antagonism

The creation of antimicrobials by probiotics is thought to be one of the main mechanisms thwarting the development of pathogenic organisms in the gastrointestinal tract (Hong et al., 2005). Bacillus spp. convey different antimicrobial substances, for example, coagulin, bacteriocins, subtilin and antiinfection agents [45]. B. subtilis antimicrobial products are dynamic against an extensive variety of irresistible microorganisms, and unwanted side effects or the event of pathogen opposition are decreased because of the similarity of probiotic microbes to that in the human antimicrobial resistance framework. Bacillus subtilis are strain specific and can produce lytic catalysts. [62-71] revealed that B. subtilis had a noteworthy deterrent result on the growth of Candida spp. proposing it as a substitution treatment for oral candidiasis. In poultry farming, controlled trials have shown that oral utilization of Bacillus subtilis spores decreases diseases, for example, diseases caused by Salmonella enterica, Salmonella enteritidis, Clostridium perfringens and Escherichia coli (La Ragione et al., 2001; La Ragione and Woodward, 2003).

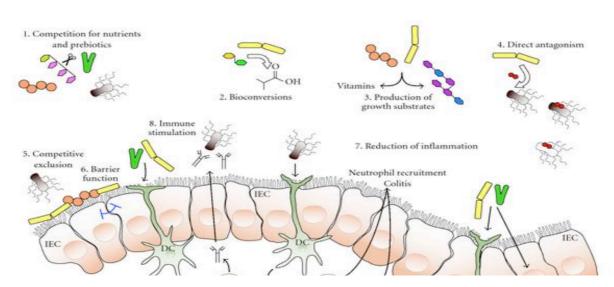


Figure 2: Mechanisms of Action for Bacillus Probiotics Source: Custom Probiotics, 2018

3.2. Immunomodulatory Effect

Immune stimulation can be activated during treatment of oral infection using B. subtilis spores because the spores cause increased expression of activation markers on the lymphocytes. Spores also have the ability to cause humoral immunological responses (Sorokulova, 2013, Suva et al., 2016). Various studies report enhanced immune responses in the gastrointestinal tract of mice brought about by growing spores, furthermore, this immune incentive could be the hidden driver of the probiotic impact of spores (Cutting, 2011, Sorokulova, 2013).

3.3. Competitive Exclusion of Pathogens

Non-pathogenic probiotic organisms when ingested into the gastrointestinal tract, creates favourable conditions that support the growth of other beneficial microbes but hinder the growth of pathogenic organisms. Probiotic Bacillus species hinder the growth of pathogenic organisms by taking most of the nutrients and space, leaving little or none left to sustain growth of the pathogen. They also outcompete pathogenic microbes with adherence and help in hastening the development of natural flora in the intestine (Hong et al., 2005; Suva et al., 2016; Bernardeau et al., 2017). It should be known that Bacillus can also make use of extra-cellular enzymes and vitamin products to initiate a beneficial effect on growth and make up of commensal and useful species in the gut (Elshaghabee et al., 2017). Some other mechanisms include detoxification, inhibition of intestinal inflammation (Hong et al., 2005; Bernardeau et al., 2017) and antidiarrheal effect (Suva et al., 2016)

3.4. How safe is Bacillus?

Bacillus species cause disease or deterioration of food. B. anthracis and B. cereus are both pathogenic strains of the genus Bacillus (Cutting, 2011, Elshaghabee et al., 2017). Certain Bacillus strains, such as B. subtilis, B. indicus, and B. licheniformis have been considered non-toxic and safe for consumption if in food, according to European standards (Sorokulova et al., 2008; Hong, 2008; Sorokulova, 2013; and Olmos and Paniagua-Michel, 2014).

4. Conclusion

Bambara nut is a legume categorized as underutilized however it holds considerable potential in solving the issue of food security in Nigeria and other sub-Saharan African countries. What is most important is to explore the benefits of this legume, assess its probiotic potential in model animals. In addition, elucidate on the proper phylogenetic studies on predominant bacteria species associated with the fermented Bambara nut. This could be a veritable way of establishing potential bacteria species that could serve as probiotic organisms. We anticipate that when these investigations are carefully established, widespread use of this underutilized crop will be adopted in most homes, more so, new food products developed from this crop.

Acknowledgements

The authors would like to acknowledge Covenant University, Ota, Nigeria for defraying the cost of this publication.

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