Fermentation Technology Enhances the Nutritional Quality of Underutilized Bambara Groundnut – A Review

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Abstract

Bambara groundnut (BG) offers great potential for sustainable food and nutritional security in the face of a growing population and changing climate. However, its utilization remains restricted by the hard-to-cook phenomenon of seed and the presence of anti-nutritional factors (ANFs). Fermentation has been reported to enhance the nutritional value and reduce ANFs in leguminous seeds. The induced fermentation technology exhibits greater control over the process parameters and ensures the fermented product's quality, reproducibility, and safe consumption. Hence, this review highlights the importance of fermentation technology in overcoming the challenges of processing hard-to-cook BG while retaining its nutrients and flavor. This paper also reviews the food safety measure and risks of consuming fermented BG. Future research on the fermented BG's nutrient digestibility should be of concern to unlock its full potential as human food or animal feed.

Keywords: Bambara groundnut; underutilized legume; fermentation; food processing; nutritional value; anti-nutritional factors

1. Introduction

Bambara groundnut (*Vigna subterranea* (L.) Verdc; BG) is an indigenous African legume. It is a promising underutilized crop due to its unique nutritional quality (Tan et al. 2020) and intrinsic stress tolerance attributes (Mayes et al. 2019). Its seed is often regarded as a "complete food" as it contains a range of important macro- and micronutrients (Gulzar and Minnaar 2017; Halimi et al. 2019; Nwinyi and Umane 2019). As a nitrogen-fixing legume, it helps to replenish soil fertility and improve crop productivity (Hasan et al. 2018; Hillocks, Bennett, and Mponda 2012). It is also of potential value in climate change adaptation strategies, owing to its resistance to drought and heat stresses (Halimi et al. 2019; Mayes et al. 2019). Despite its

nutritional and agronomic benefits, BG remains underutilized because it is hard-to-cook and hard-to-mill (Hillocks, Bennett, and Mponda 2012; Mubaiwa et al. 2017; Mubaiwa et al. 2018; Nti 2009). Besides, BG seeds contain anti-nutritional factors (ANFs) (Hillocks, Bennett, and Mponda 2012; Mubaiwa et al. 2018; Popoola et al. 2019; Unigwe et al. 2018), which negatively impact the nutritional and sensorial quality of the finished product and may influence consumer acceptance.

Through years of research, different conventional legume processing methods, such as thermal, physical and biological treatments, have been utilized to overcome the abovementioned challenges (Ndidi et al. 2014; Mubaiwa et al. 2017; Mubaiwa et al. 2018; Okafor, Ani, and Okafor 2014; Xiang et al. 2019). However, the thermal and physical treatments involve complex procedures, modern equipment, as well as high operational and labor costs. In addition, these treatments may also cause some losses in water-soluble and heat-labile nutrients, such as ascorbic acid (vitamin C), B vitamins, phosphorus, calcium, iron, zinc and β-carotene (Raptopoulou et al. 2017; Drulyte and Orlien 2019; Yong, Amin, and Dongpo 2019). All these negatives have limited their applications, thus directing research to focus on biological treatments such as fermentation.

Fermentation is a natural decomposition process in which complex organic substrates are broken down into simpler compounds by the action of enzymes produced by naturally occurring microorganisms or those added as starter culture (Joshi 2016; Mehta, Kamal-Eldin, and Iwanski 2012; Xiang et al. 2019). Fermentation is intentionally performed to improve food properties, such as texture, flavor, shelf-life, and nutritional content (Sharma et al. 2020). This method has been reported to enhance the nutritional value of leguminous seeds by reducing the ANFs that hinder nutrient bioavailability and bioaccessibility (Olanipekun, Otunola, and

Oyelade 2015; Mubaiwa et al. 2017; Ogodo et al. 2018a; Mayes et al. 2019). It can also modify the legume's physical and chemical structures, making legumes more edible (Ogodo et al. 2018a, 2018b; Olanipekun, Otunola, and Oyelade 2015; Xiang et al. 2019). In addition, fermentation can also enhance the organoleptic properties of food (Chude, Chidiebere, and Okpalauwaekwe 2018; Jibril and Egwim 2019; Ola and Adewole 2019) and confer a preservative effect by inhibiting the growth of spoilage microorganisms and thereby extending product shelf-life (Anal 2019; Singh 2018).

With all the benefits listed above, fermentation appears to have considerable potential as an accessible and affordable method of improving the nutritional value of BG. Therefore, we reviewed the published literature on the fermentation of BG, mainly focusing on the impacts of fermentation on the nutritional, anti-nutritional, and sensory properties, to ascertain whether fermentation is a viable method to process this legume. We also assessed the risks and safety concerns of consuming fermented BG products.

It is hoped that this review can (i) help overcome the challenges of processing the hard-to-cook BG using green fermentation technology while retaining the nutrients and enhancing the flavor of the fermented products; and (ii) serve as baseline data for the commercial production of fermented BG products to promote the broader use of this underutilized legume and to contribute to solutions to global food security challenges.

2. Fermentation of Bambara Groundnut

Fermentation of BG can be carried out using two methods, (i) the natural fermentation method using starter cultures naturally present in the processing environment or indigenously found on the legume, and (ii) the induced fermentation method using well-defined starter cultures to drive the process. Table 1 shows different types of natural and induced fermented BG products

reported over the years. Traditionally, fermented BG condiments, such as *Dawadawa*, *Iru* and *Ogirri*, are produced using natural fermentation methods (Adamu Shahidah et al. 2018; Adebiyi, Kayitesi, and Njobeh 2020; Adebiyi, Njobeh, and Kayitesi 2019; Ademiluyi and Oboh 2011; Aigbodion 2017; Barimalaa and Anoghalu 1997; Barimalaa et al. 1994; Fadahunsi and Olubunmi 2010; Oboh, Ademiluyi, and Akindahunsi 2009; Omogbai and Aghahowa 2017). However, as fermentation technology evolves, induced fermentation has been widely applied for the production of *Dawadawa*, a famous food flavoring condiment in Africa (Akanni, De Kock, et al. 2018; Akanni, Naudé, et al. 2018), and BG milk has been fermented by lactic acid bacteria (LAB) to produce probiotic beverages (Hardy and Jideani 2020). The induced fermentation method offers greater control throughout the process and improves product quality and reproducibility. For example, BG fermented milk has higher sensory acceptability than milk from other legumes, and its nutritional profile is substantial in sustaining probiotics' growth (Murevanhema and Jideani 2013).

Tables 2 and 3 summarize the conditions used for natural and induced fermentation of BG, respectively. The studies on natural fermentation employed temperatures ranging from 25 - 45°C and a fermentation duration of 1.5 - 5 days (Table 2). The microorganisms and the exact amounts involved in natural fermentation process are usually unknown. Only 10 out of 19 of the reported studies performed microbial isolation and identification to determine what microorganisms were present (Adamu Shahidah et al. 2018; Adeoye, Obayemi, and Akinola 2018; Aigbodion 2017; Barimalaa and Anoghalu 1997; Barimalaa et al. 1994; Fadahunsi and Olubunmi 2010; Mbata, Ikenebomeh, and Alaneme 2009; Ogodo et al. 2018a, 2018b; Omogbai and Aghahowa 2017). The starter cultures of natural fermentation are discussed later in Section 3.

The natural fermentation process is often unpredictable as there is no standard protocol. This is due to variations in pre-fermentation processing methods, the fermentation conditions, and the involvement of unknown microbial diversity, which could pose risks of contamination and failure, leading to inconsistent product quality and safety (Bidemi 2017; Capozzi et al. 2017; Ray and Joshi 2014). For these reasons, research has shifted toward the induced fermentation method.

Studies on induced fermentation of BG have been performed using temperatures ranging from 25 - 45°C, similar to that used for natural fermentation (Tables 2 and 3). However, the duration used for induced fermentation can be as short as 5 hours or up to 14 days, depending on the starter culture used and the type of fermented BG product being produced (Table 3). All studies involved the application of specific starter cultures, consisting of either single or multiple strains of culture organism, which can be the same or different species (see Section 3 for more details), depending upon the types of food substrates, desired metabolic activities, and preferred characteristics (e.g., chemical, physical, functional properties) of the finished products. The inoculum size of the starter culture ranged from 0.05 - 15% (Adebanke et al. 2017; Amadi, Barimalaa, and Omosigho 1999; Amadi et al. 1999; Ani, Amove, and Igbabul 2018; Chude, Amadi, and Iro 2018; Chude, Okoyeuzu, and Amadi 2018; Enyidi and Etim 2020; Fadahunsi 2009; Ogodo et al. 2018a, 2018b; Ola and Opaleye 2019; Olanipekun, Otunola, and Oyelade 2014, 2015; Olanipekun et al. 2012). The induced fermentation method exhibits greater control over the process parameters (e.g., duration, temperature) than natural fermentation. In addition, the induced fermentation involves a sterile fermentation environment, a known inoculum size, as well as known identity and characteristics of starter microorganisms, which could ensure the quality, reproducibility, and safety of the product (Capozzi et al. 2017; Sharma et al. 2020).

3. Starter Cultures of the Bambara Groundnut Fermentation

3.1 Bacterial Starter Cultures

The *Bacillus* species are the dominant starter cultures present in the natural fermentation, whereas LAB are prevalent in the induced fermentation of BG (Table 1).

The notable *Bacillus* species involved are *B. subtilis*, *B. licheniformis*, *B. pumilus*, *B. cereus* and *B. amyloliquefaciens*, which have been found in the fermented BG-based condiments, such as *Dawadawa*, *Iru* and *Ogirri*. *Bacillus* species are endospore-forming bacteria. Their spores remain dormant and resistant to harsh environmental conditions (e.g., extreme pH, heat, cold and desiccation) (Turnbull 1996). Hence, they are often not destroyed even after thermal processing and tend to regain metabolic functions and vegetative growth under favorable fermentation conditions (Adesulu-Dahunsi, Dahunsi, and Banwo 2019; Olasupo, Okorie, and Oguntoyinbo 2016).

Bacillus species utilize simple sugars or organic acids present in the fermentation substrates as carbon and energy sources. However, ammonium is generally the preferred nitrogen source of *Bacillus* species for use in protein and nucleic acids synthesis (He et al. 2022). During fermentation, simple sugars, such as glucose, will be metabolized through a series of enzymatic reactions, including glycolysis, pentose phosphate pathway and Krebs (citric acid) cycle. Meanwhile, organic acids need to be converted into the intermediates of the Krebs cycle before gluconeogenesis and the pentose phosphate pathway (Stülke and Hillen 2000).

Bacillus species are known to carry out an alkaline fermentation and produce a wide range of enzymes (e.g., protease, amylase, lipase, cellulase, fibrinolytic enzymes), organic acids (e.g., α-ketogenic acid, acetic acid, propionic acid, succinic acids), amino acids (e.g., L-glutamate,

L-lysine), vitamins (e.g., vitamin D, E, B complex) and bacteriocins (D'Este, Alvarado-Morales, and Angelidaki 2018; Nagarajan, Rajasekaran, and Venkatachalam 2022; Yan et al. 2013). During fermentation, complex starch and cellulose in the legume are broken down into sugars via amylase and cellulase (Gopikrishna et al. 2021). Proteases help degrade the legume proteins into peptides and amino acids while releasing ammonia as a by-product (Anal 2019). Accumulation of ammonia elevates the pH and thereby prevents the colonization of spoilage microorganisms (Akanni et al. 2018, 2018).

The *Lactobacillus* (e.g., *L. plantarum*, *L. acidophilus*, *L. fermentum*, *L. reuteri*, *L. bulgaricus*, *L. rhamnosus*, *L. nantensis*, *L. brevis*, *L. delbruieckii* and *L. casei*), *Lactococcus* (e.g., *Lc. lactis*), *Leuconostoc* (e.g., *L. mesenteroides*), and *Pediococcus* (e.g., *P. acidilactici*) are the common LAB genera involved in the production of fermented BG-based complementary flour, yoghurt, semolina (*dackere*), probiotic beverage, and animal feed meal. LAB are non-spore forming and acid-tolerant bacteria that have the generally recognized as safe (GRAS) status. LAB are found predominantly in fermented carbohydrate-rich foods (e.g., legumes, cereals, starchy vegetables) owing to their fast-growing and strong acidifying properties (Khandelwal et al. 2016; Mozzi, Raya, and Vignolo 2015).

LAB utilizes glucose or other simple sugars to obtain cellular energy, resulting in the production of organic acids, primarily lactic acid, which initiates rapid acidification in the fermentation substrates (Florou-Paneri, Christaki, and Bonos 2013; Liptáková, Matejčeková, and Valik 2017). Besides, they also produce acetic acid, propionic acid, 3-hydroxypropionic acid, formic acid and succinic acid as part of the flavor compounds (Wang et al. 2021). The acidic conditions will inhibit the growth of spoilage microorganisms (Vinderola et al. 2019). In addition, LAB are known to synthesize a variety of metabolites during fermentation, such

as enzymes (e.g., protease, amylase, amylopullulanase, aldolase, lactase, lipase), vitamins (e.g., folic acid, riboflavin, pyridoxal, cobalamine, vitamin C, vitamin K), bacteriocins (e.g., nisin, Helveticin-M, Helveticin-J, Sakacin P, Gasserin), exopolysaccharides and gamma-aminobutyric acid (Hayek and Ibrahim 2013; Nagarajan, Rajasekaran, and Venkatachalam 2022; Wang et al. 2021).

Several less common bacterial species, including *Staphylococcus saprophyticus*, *Phyllobacterium leguminum* and *Lysinibacillus fusiformis*, have also been detected in the naturally fermented BG *Dawadawa* and *Ogirri* (Adamu Shahidah et al. 2018; Omogbai and Aghahowa 2017). However, their involvement in the fermentation of BG still need to be expanded and clarified.

3.2 Fungal Starter Cultures

The most common fungal starter cultures associated with the fermentation of BG are *Rhizopus*, *Aspergillus* and *Saccharomyces* species, with other cultures, including *Penicillium*, *Trichoderma* and *Candida* species, being used less frequently (Table 1).

The *Rhizopus* (e.g., *R. oligosporus*, *R. stolonifer*, *R. arrhizus*, *R. oryzae* and *R. nigricans*), *Aspergillus* (e.g., *A. niger* and *A. flavus*), *Penicillium* and *Trichoderma* are the fungal species which have been detected in both natural and induced fermented BG-based condiments, flour, *tempeh*, feed meal and feedstuff ingredients. These fungi possess some unique characteristics that make them well suited for fermentation, including the ability to (i) reproduce asexually by producing large amounts of stress-resistant spores (Dijksterhuis 2019; Warnock 2012), (ii) colonize and penetrate food substrates easily due to their hyphae (Puvanakrishnan,

Sivasubramanian, and Hemalatha 2019), and (iii) survive at very low water activities (Hui et al. 2004).

Fungi utilize sugars (e.g., glucose, xylose, or sucrose) and starch as carbon and energy sources while consuming amino acids and urea for nitrogen. They synthesize various enzymes, such as protease (acidic, neutral, and alkaline), α- and β-amylase, glucoamylase, glutamidase, pectinase, hemicellulase, and cellulase, to breakdown complex macromolecules in substrates (Yang et al. 2021). For example, *Rhizopus* has been reported to produce vitamins (e.g., niacin, riboflavin, cobalamin, pyridoxine, thiamine, biotin, pantothenic acid, ergosterol, tocopherol, and vitamin K) during *tempeh* fermentation (Feng 2006; Sharma et al. 2020). Organic acids, such as citric acid, fumaric acid, lactic acid and gluconic acid, are also metabolites secreted by *Rhizopus* and *Aspergillus* species (Nagarajan, Rajasekaran, and Venkatachalam 2022).

Saccharomyces cerevisiae and *Candida utilis* are the two fungal species commonly used in the fermentation of BG. They have been found in fermented BG-based condiments (Aigbodion 2017), flour (Ogodo et al. 2018a, 2018b) and feed meals (Enyidi and Etim 2020). *S. cerevisiae* is a unicellular eukaryotic fungus reproduces asexually by budding (McGinnis and Tyring 1996). They can replicate quickly to achieve high cell biomass within food substrates and possess high metabolic rates, often dominating the natural fermentation process. With adequate sugars, *S. cerevisiae* can grow in the absence or presence of oxygen, carrying out either anaerobic or aerobic fermentations. Hence, they can initiate a fermentation process under oxygen conditions or participate in the later stage of fermentation (alcoholic fermentation) after the oxygen has been depleted (Faria-Oliveira et al. 2015; Josephsen and Jespersen 2004).

S. cerevisiae can grow on various compounds as energy sources and carbon-containing precursors for anabolic metabolism. However, it prefers glucose and fructose over mono-, di-, and trisaccharides, such as sucrose, raffinose, and trehalose (Broach 2012). Yeasts mainly utilize ammonium, amino acids, di- and tripeptides for their nitrogen needs (Crépin et al. 2012; Roca-Mesa et al. 2020). During growth and fermentation, *S. cerevisiae* produces different types of metabolites, including enzymes (e.g., amylase, invertase, maltase, protease, and lipase) that catalyze the release of simple sugars, amino acids and fatty acids, vitamins (e.g., ergosterol and riboflavin) and organic acids (e.g., α-ketogenic acid and lactic acid) (Gu and Li 2016; Nagarajan, Rajasekaran, and Venkatachalam 2022).

4. Effect of Fermentation on the Nutritional Quality of Bambara Groundnut

Table 4 summarises the effect of fermentation on the proximate nutrient contents of BG products.

4.1 Protein

Numerous studies reported that fermentation could increase the protein content of BG, with an increase of 0.5 - 7.4% observed in different fermented BG products (Table 4). Although the minute increments are negligible, the more significant increment of 7.4% could be attributed to the active proliferation of fermenting microorganisms since they utilize carbohydrates in the legume substrate as an energy source for cell metabolism (Adamu Shahidah et al. 2018; Mahamat et al. 2016; Ogodo et al. 2018b; Olanipekun et al. 2012). In addition, these microorganisms can synthesize a wide range of protein-based metabolites during growth (e.g., hydrolyzed peptides, amino acids, enzymes), further adding to the total crude protein content (Ogodo et al. 2018b).

To the authors' knowledge, only two studies have investigated whether fermentation improves the *in vitro* protein digestibility of BG. Ogodo et al. (2018b) found that natural and LAB-induced fermentation increased the protein digestibility of BG by 11.5 - 18.0%, while the *Rhizopus*-fermented BG exhibited 19.4 - 43.1% increases (Olanipekun, Otunola, and Oyelade 2015).

The increase in protein digestibility in fermented BG products could be ascribed to the production of enzymes by fermenting microorganisms (Ogodo et al. 2018b; Olanipekun, Otunola, and Oyelade 2015). Additionally, reducing the pH during LAB fermentation helps enhance the activity of phytases and proteases, thus promoting the release and/or degradation of proteins into readily digestible amino acids or smaller peptides (Batt and Tortorello 2014; Nkhata et al. 2018). Indeed, the activation of endogenous ANF-degrading enzymes (e.g., phytase) releases the proteins that are bound (e.g., to phytate), thereby improving protein digestibility (Adebiyi, Njobeh, and Kayitesi 2019). Similarly, removing protease inhibitors, such as Bowman-Birk inhibitors and Kunitz-type inhibitors, likely to form stable insoluble complexes with digestive enzymes, also enhances protein digestibility (Avilés-Gaxiola, Chuck-Hernández, and Saldívar 2018).

4.2 Amino Acid

There is little research on the fermentation-induced changes in the amino acid composition of BG (Adebiyi, Njobeh, and Kayitesi 2019; Ijarotimi and Esho 2009). Adebiyi, Njobeh, and Kayitesi (2019) observed an improvement in the content of some essential (isoleucine, leucine, methionine, phenylalanine, threonine, and valine) and non-essential amino acids (arginine, serine, aspartic acid, glutamic acid, glycine, alanine, and proline) in the 84 h-fermented BG (*Dawadawa*). Ijarotimi and Esho (2009) reported that the naturally fermented BG flour

exhibited higher levels of aspartic acid (10.12 g/100g), isoleucine (4.05 g/100g) and glycine (4.03 g/100g) compared to the raw, germinated and roasted samples. Such changes in amino acid content could be due to the transamination during fermentation and/or free amino acid production by the fermenting microorganisms (Adebiyi, Njobeh, and Kayitesi 2019). In contrast, Adebiyi, Njobeh, and Kayitesi (2019) noted a reduction in some amino acids (histidine, lysine, tyrosine, and HO-proline) in the 84 h-fermented BG (*Dawadawa*), suggesting that the microorganisms may utilize certain amino acids as nitrogen sources for their growth and anabolic requirements (Adebiyi, Njobeh, and Kayitesi 2019; Liu et al. 2020).

Both studies reported glutamic and aspartic acids as the most abundant non-essential amino acids in fermented BG products, contributing to the pleasant umami or savory taste development in foods (Zhu et al. 2020). The increase in glutamic acid resulting from fermentation is desirable, as this can translate into higher levels of gamma-aminobutyric acid (GABA), a bioactive compound with beneficial physiological functions (Adebiyi, Njobeh, and Kayitesi 2019). In addition, the essential sulfur-containing amino acid methionine, which is present at low levels in raw BG, has also been found to increase after fermentation, further improving the protein profiles of the fermented product.

4.3 Carbohydrate

Most studies reported a decrease in carbohydrates, ranging from 0.3 - 34.4% in BG up to the end of fermentation (Adamu Shahidah et al. 2018; Aigbodion 2017; Amadi, Barimalaa, and Omosigho 1999; Amadi et al. 1999; Barimalaa et al. 1994; Esiegwu and Obih 2019; Ogodo et al. 2018b; Olanipekun et al. 2012; Pahane et al. 2017) (Table 4).

The significant 34.4% reduction in carbohydrate content in the fermented BG products is likely due to amylase and sucrase production by fermenting microorganisms, which promote the hydrolysis of complex carbohydrates into fermentable sugars for microbial utilization (Adamu Shahidah et al. 2018; Fadahunsi and Olubunmi 2010; Ola and Adewole 2019; Osman 2011). For example, the *Bacillus* species isolated from naturally fermented BG was shown to produce carbohydrate-degrading enzymes, such as amylase, galactase, galactosidase, glucosidase and fructofuranosidase (Aigbodion 2017; Kakou et al. 2017).

In contrast, Ijarotimi and Esho (2009) reported higher carbohydrate levels (68.72%) in the 96 h-fermented BG flour compared with raw (68.52%), germinated (67.53%) and roasted (65.82%) samples. Mahamat et al. (2016) and Ola and Adewole (2019) observed elevated levels of total reducing sugar in the fermented BG-sweet potato *dackere* and –wheat flour blends, respectively. Olanipekun et al. (2012) showed a steady carbohydrate rise up to 48 h of fermentation, followed by a slight reduction at the end of the process. These fluctuations in carbohydrate content could be due to the different types of microorganisms involved, as they have different growth and sugar utilization patterns. On the other hand, the increases in carbohydrate content might be due to the production of carbohydrate-based metabolites, such as exopolysaccharides, by fermenting microorganisms. For example, yeasts and LAB are reported to produce exopolysaccharides during the fermentation of quinoa sourdough (Franco et al. 2020), soybean and fava bean flour (Xu et al. 2017).

Leguminous starches are poorly digested compared to cereals and tubers (Tomasik and Horton 2012). Inadequate starch digestion in raw BG often limits its use in human and animal diets, with very little published research on the starch digestibility of fermented BG. For instance,

Ogodo et al. (2018b) reported a positive effect of fermentation on the *in vitro* starch digestibility of BG flour. However, more research is required to further validate these findings.

4.4 Lipid

Several published reports showed that fermentation enhances the lipid content of BG, ranging from 0.1 - 13.7% (Adamu Shahidah et al. 2018; Aigbodion 2017; Amadi, Barimalaa, and Omosigho 1999; Amadi et al. 1999; Barimalaa et al. 1994; Ijarotimi and Esho 2009) (Table 4). The significant enhancement of 13.7% of the lipid content may be due to the accumulation of lipids or lipid-related compounds during the growth and metabolism of the fermenting microorganisms (Amadi, Barimalaa, and Omosigho 1999). In contrast, three studies have reported a minor decrease of 0.1 - 1.4% in the lipid content of BG after fermentation (Esiegwu and Obih 2019; Ogodo et al. 2018b; Olanipekun et al. 2012) (Table 4). Although such a minute reduction might be insignificant, the reduction in lipid content could be due to the lipolytic activity of endogenous or microbial enzymes, which promote the hydrolysis of fats into fatty acids and glycerol for microbial utilization (Fadahunsi and Olubunmi 2010; Ogodo et al. 2018b; Olanipekun et al. 2012).

To date, information about the fatty acid composition of fermented BG is limited. Olanipekun et al. (2012) studied the changes in fatty acid profiles of BG flour within 72 h of fermentation. The unsaturated fatty acid content showed an initial decline at 48 h of fermentation, followed by a significant increase from 41.9 to 61.5% (with a concomitant decrease in saturated fatty acids) during the later stages of fermentation. Such changes in the fatty acid composition could be due to the disparate microbial communities during the fermentation. Different fermenting microorganisms secrete lipolytic enzymes with varying degrees of activity, while their fatty acid synthesis ability is also strain-specific (Olanipekun et al. 2012; Vieira et al. 2015). For

example, Olanipekun et al. (2012) observed high levels of saturated fatty acids (e.g., stearic acid) and certain unsaturated fatty acids (e.g., oleic, linoleic, linolenic, eicosenoic, eicosatrienoic, eicosapentaenoic, and nervonic acids) in the fermented BG flour.

4.5 Ash

Ash content is commonly used to measure a sample's total amount of minerals. Several studies have shown that fermentation enhances the ash content of BG by 0.3 to 1.1% (Esiegwu and Obih 2019; Ogodo et al. 2018b; Olanipekun et al. 2012; Pahane et al. 2017) (Table 3). This small increment in ash content of the fermented BG products could be due to the degradation of phytates, tannins and oxalates via the enzymes secreted by fermenting microorganisms (Ertop and Bektaş 2018). For example, phytase and alpha-amylase enzymes break down any phytate and starch present in the food matrix, releasing minerals bound or trapped by them and preventing the formation of insoluble mineral chelates. Indeed, fermentation provides an optimum pH for phytase to completely hydrolyze phytate (Bishai et al. 2015; Gupta, Gangoliya, and Singh 2015), while the fiber-degrading activity of fermenting microorganisms also contributes to cell wall disruption and further loss of the food matrix (Nkhata et al. 2018).

In contrast, three studies reported a decrease of 0.7 - 8.2% in the ash content of BG after fermentation (Adamu Shahidah et al. 2018; Aigbodion 2017; Ijarotimi and Esho 2009) (Table 4). The significant 8.2% reduction in ash content might be due to the addition of water during fermentation, resulting in the leaching of soluble minerals into the water (Nwanna et al. 2005). Furthermore, pre-fermentation processings, such as dehulling or dehusking, also result in considerable ash losses, as the removed seed coat contains a relatively high amount of minerals (Felix and Francis 2019; Iheke et al. 2017). In addition, the fermenting microorganisms may

have utilized some mineral ions as energy sources or electron sinks, causing lower ash content in the fermented products (Adebiyi, Njobeh, and Kayitesi 2019).

5. Effect of Fermentation on the Anti-nutritional Factors Present in Bambara Groundnut

ANFs are biological compounds found mainly in plant-based foods that can interfere the nutrient bioavailability and bioaccessibility (Popova and Mihaylova 2019; Singh and Basu 2012). Like most legumes, BG contains various ANFs, including trypsin inhibitor, phytate, tannin and oxalate (Mubaiwa et al. 2018; Popoola et al. 2019; Unigwe et al. 2018). Table 5 summarizes the effect of fermentation on the levels of ANFs in BG products.

5.1 Trypsin Inhibitor

Trypsin inhibitor (TI) is a serine protease inhibitor that competes with proteins to bind to the enzyme trypsin and inhibit its activity. It also partially interferes with chymotrypsin activity and hinders protein digestion and absorption (Hill 2003; Popova and Mihaylova 2019). Most studies reported that fermentation could reduce the TI content, or trypsin inhibitory activity (TIA), of BG by 37.3 - 87.1% (Barimalaa and Anoghalu 1997; Barimalaa et al. 1994; Fadahunsi 2009; Ijarotimi and Esho 2009; Ogodo et al. 2018a; Ola and Opaleye 2019; Olanipekun, Otunola, and Oyelade 2015) (Table 5).

Olanipekun, Otunola, and Oyelade (2015) observed a 70.7 - 84.6% reduction in TI content of the BG flour fermented for 72 h using different *Rhizopus* starter cultures. In addition, Ola and Opaleye (2019) noted a decrease in BG TI content, from 3.25 mg/100 g in the unfermented sample to 0.42 mg/100 g in the 72 h fermented sample. Similarly, Ijarotimi and Esho (2009) showed a lower TIA level (4.2 mg/100 g) in the 96 h naturally fermented BG flour compared to the raw (6.7 mg/100 g), germinated (5.6 mg/100 g) and roasted (4.8 mg/100 g) samples. The

TIA of BG flour in the study of Ogodo et al. (2018a) also decreased by 80.3 - 86.6% after natural and LAB-induced fermentation.

The decrease in TI or TIA is likely due to the fermenting microorganisms enzymatically breaking down the TI into an inactive form (Adeyemo and Onilude 2013; Boroojeni et al. 2018; Ijarotimi and Esho 2009). Indeed, LAB have unique proteolytic systems involving (i) cell-wall bound proteinases, (ii) peptide transporters, and (iii) various intracellular peptidases (Liu et al. 2010; Kieliszek et al. 2021), which can degrade the protease inhibitors and reduce their inhibitory activity (Avilés-Gaxiola, Chuck-Hernández, and Saldívar 2018; Ji et al. 2020).

In contrast, Fadahunsi (2009) reported a significant increase in TIA from 0.71 mg/100 g protein in the boiled-soaked BG to 1.33 mg/100 g protein after 24 h of fermentation induced by *R. oligosporus*. They suggested that this increase was due to the release of bound TI and that active TI was liberated from a heat-resistant, inactive, bound form by the *R. oligosporus* proteases during the fermentation process (Fadahunsi 2009; Wang, Vespa, and Hesseltine 1972).

5.2 Phytate

Phytate (phytic acid) is inositol-6-phosphate (IP6) and is regarded as the main storage form of phosphate and inositol in plants, including legumes (Gemede and Ratta 2014). As a polyanionic molecule, phytate strongly chelates the positively charged cations (e.g. minerals) and proteins, forming stable insoluble complexes impairing nutrient bioavailability and absorption (Golam Masum Akond et al. 2011; Popova and Mihaylova 2019). It also tends to hinder the activity of digestive enzymes, such as pepsin, chymotrypsin, trypsin, and amylase (Adebiyi, Njobeh, and Kayitesi 2019).

Several studies have shown that fermentation reduces the phytate content of BG by 18.1 - 95.9% (Adebiyi, Njobeh, and Kayitesi 2019; Ijarotimi and Esho 2009; Ogodo et al. 2018a; Ola and Opaleye 2019; Olanipekun, Otunola, and Oyelade 2015; Pahane et al. 2017) (Table 4). A reduction in phytate content could be due to the activation of phytase and phosphophytase enzymes, which are either naturally occurring in the BG or synthesized by the fermenting microorganisms and subsequently hydrolyze phytate into lower esters (e.g., IP5, IP4), thereby releasing phosphorus as well as any phytate-bound nutrients (Adebiyi, Njobeh, and Kayitesi 2019; Ola and Opaleye 2019; Olanipekun, Otunola, and Oyelade 2015). In addition, the lactic acid produced during LAB fermentation leads to a rapid decrease in pH, favoring phytase stability and activity (Castro-Alba et al. 2019; Ogodo et al. 2018a). The inactivation or removal of phytate from legumes helps improve their nutritional value by increasing the bioavailability of minerals and proteins (Ojo 2021).

5.3 Tannins

Tannins, including tannic acids, are water-soluble polyphenolic compounds found mainly in the bark of trees and the seed coat of legumes. Tannin content varies across different seed varieties and seed coat colors, with a darker color usually containing higher tannin levels (Elias, De Fernandez, and Bressani 1979; Gemede and Ratta 2014). Tannins bind with enzyme and non-enzyme proteins, forming tannin-protein complexes that cause digestive enzyme inactivation and impairment of protein digestibility (Barr and Sutton 2019; Popova and Mihaylova 2019). Tannins also precipitate organic compounds, including alkaloids and amino acids. Their chelating ability with metal ions (e.g., iron, copper, and zinc) contributes to reduced mineral bioavailability and bioaccessibility (Adamczyk et al. 2017; Naumann et al. 2017). In addition, tannins may contribute to the hard-to-cook properties of legume seeds. They

may migrate from the seed coat to cotyledon and cross-link with macromolecules from the cell wall or middle lamella during storage at high temperature and humidity, thus leading to a hardening effect (Coelho et al. 2007; Ojo 2022).

Several studies suggested that fermentation can effectively decrease the tannin content of BG from 25.6 to 98.3% (Adebiyi, Njobeh, and Kayitesi 2019; Ijarotimi and Esho 2009; Ogodo et al. 2018a; Ola and Opaleye 2019; Olanipekun, Otunola, and Oyelade 2015) (Table 5). The reduction in tannin content is likely due to the action of tannase or polyphenol oxidase (PPO) enzymes that are either present in the legume or are secreted by the fermenting microorganisms. Tannase degrades the hydrolyzable tannins into glucose and gallic acid, while PPO polymerizes and inactivates tannins (Mäkelä et al. 2015; Ogodo et al. 2018a; Ola and Opaleye 2019). The removal of tannins from fermentation enhances the nutritional value while overcoming the hard-to-cook problem in legumes.

5.4 Oxalate

Oxalic acid is an organic compound commonly found in plants that forms both soluble (e.g., potassium, ammonium, and sodium) and insoluble (e.g., calcium, magnesium, and iron) salts with minerals, namely oxalates (Popova and Mihaylova 2019). The insoluble chelate complexes' formation makes these essential minerals unavailable or inaccessible for absorption. Therefore, excessive oxalate consumption can lead to kidney stones, nutritional deficiencies, and severe inflammation of the gut lining (Gemede and Ratta 2014).

Only a few studies have investigated the effect of fermentation on the oxalate content of BG. Most findings showed a reduction of 15.8 - 74.0% (Adebiyi, Njobeh, and Kayitesi 2019; Ijarotimi and Esho 2009; Ola and Opaleye 2019; Olanipekun, Otunola, and Oyelade 2015)

(Table 5). However, the exact mechanism for the decrease in oxalate content remains unclear. Wadamori, Vanhanen, and Savage (2014) suggested that oxalate reduction in kimchi during natural fermentation may be due to oxalotrophic bacteria, which tend to utilize oxalate as an energy or carbon source. Certain fermenting microorganisms (e.g., fungi and LAB) have also been found to have the oxalate-degrading ability, most probably through microbial enzymatic actions (Adegbehingbe 2015; Batra, Lomash, and Ganguli 2018).

6. Effect of Fermentation on the Sensory Quality of Bambara Groundnut

Sensory attributes also affect consumers' acceptance and preference when selecting food products (Mihafu, Issa, and Kamiyango 2020). Hence, sensory evaluation should be included in research to provide valid and reliable information for new fermented BG product development and existing product improvement.

To date, there are several studies evaluated the sensory attributes of fermented BG based-products, such as yoghurt (Adebanke et al. 2017; Ani, Amove, and Igbabul 2018; Falade et al. 2014), complementary flour (Chude, Amadi, and Iro 2018; Ijarotimi and Keshinro 2013), biscuit (Ola and Adewole 2019; Otunola, Ade-Omowaye, and Akanji 2007), croissant (Arise, Taiwo, and Malomo 2020), tempeh (Amadi et al. 1999), dackere (Mahamat et al. 2016), and probiotic beverage (Murevanhema 2012).

The BG plain yoghurt produced in the study of Falade et al. (2014) was comparable to the commercial (farm fresh) plain yoghurt, with the mean scores of aroma (5.13), color (5.43), taste (5.33) and consistency (4.83). Chude, Amadi, and Iro (2018) reported that the sensory attributes (overall acceptability of 4.5 - 5.9) in the diets formulated with different proportions of fermented BG flour and corn cruel were significantly lower than commercial (Nutrend) and

traditional (Ogi) weaning food samples. Ola and Adewole (2019) indicated that the biscuit produced from blends of wheat flour and fermented BG flour at a ratio of 70:30 was acceptable, while further addition of fermented BG flour resulted in decreased overall acceptability scores. Besides, the panelists generally accepted all croissant samples with fermented BG inclusion (Arise, Taiwo, and Malomo 2020). The incorporation of fermented BG flour makes the croissant more appealing and attractive. According to Mahamat et al. (2016), *dackere* made from 50% sweet potatoes and 50% ungerminated BG flour blends, followed by fermentation, showed acceptable color, flavor and texture attributes.

The number of studies on sensory analysis is much lesser than those investigating the impact of fermentation on the nutritional and anti-nutritional composition of BG. This indicates an existing research gap that needs to be addressed further to understand the sensory profiles of fermented BG-based products and how they are affected by different fermentation methods or conditions. This will promote the broader applications of fermented BG in the food industry.

The ratings for appearance, aroma, taste, texture, and overall acceptability of BG after fermentation varied across studies. Most studies reported sensory scores ranging from 4.05 to 7.45, showing a moderate-slightly high acceptance of fermented BG as a potential future food source. The disparity between the sensory scores could be due to the distinctive sensory properties of the end product and the panelists' adifferent viewpoints. The panelists will usually be more familiar with and prefer the commercial (marketed) fermented foods to the new fermented BG-based products.

7. Potential Risks of Bambara Groundnut Fermentation

Undoubtedly, the fermentation of BG can improve the nutritional, physicochemical and functional qualities. However, these benefits should always be balanced against the risks of consuming fermented products. The use of heterogeneous microbiota, either those present naturally or added as starter cultures, is associated with some risks since some microorganisms may synthesize undesirable toxic by-products, such as biogenic amines, ethyl carbamate and mycotoxins (Capozzi et al. 2017; Wilburn and Ryan 2017). The species that exhibit this ability and to what extent secretion is induced are highly variable and often strain-specific.

Biogenic amine (BA) is a nitrogenous compound formed through microbial decarboxylation of free amino acids during fermentation, with the most common BA being histamine and tyramine. BA in small amounts are usually detoxified by intestinal amine oxidases but consuming foods containing high concentrations of BA may trigger toxicological health consequences (Restuccia 2017; Sarkadi 2017). Among other decarboxylase-positive microorganisms, LAB are the main producers of BA in many fermented dairy products (ripened cheese), meat products (sausages) and vegetables (sauerkraut or table olives) (Barbieri et al. 2019; Sarkadi 2017). So far, limited information is available on fermented legumes and their BA levels, which are believed to be within a safe range (e.g., in fermented soybean and bean curd) (Yang et al. 2014). Nevertheless, evaluating the BA levels is deemed essential for ensuring the safety of fermented products.

Ethyl carbamate (EC) forms naturally in fermented foods during fermentation or storage (Jha 2016). It was reclassified as a probable human carcinogen (Group 2A) from possible human carcinogen (Group 2B) by the International Agency for Research on Cancer (IARC) in 2007 (IARC 2010). Several animal studies reported that EC could trigger an increased incidence of

568 tumors in various tissue sites, including liver, lung, and blood vessels (Beland et al. 2005;
569 Mirvish 1969). EC has been detected in many fermented foods (e.g., cheese, bread, and soy
570 sauce) and alcoholic beverages (e.g., wine, beer, spirit, and whisky) (Gowd et al. 2018; Li et
571 al. 2017; Weber and Sharypov 2009). Humans can be exposed to EC through these fermented
572 products, and long-term exposure can threaten health (Gowd et al. 2018). While there is no
573 evidence suggesting that EC levels in fermented BG products are likely to be higher than those
574 already in the human food chain, it may be prudent to confirm this in the future.
575
576 Aflatoxins belong to the group of toxic secondary metabolites called mycotoxins. These
577 compounds are produced by fungi, primarily *Aspergillus* species, such as *A. flavus* and *A.*
578 *parasiticus*. Aflatoxins are potent carcinogenic compounds, and their contamination of food
579 sources poses a real threat to human and animal health (Sivamaruthi, Kesika, and Chaiyasut
580 2019; Wilburn and Ryan 2017). Aflatoxins have high thermal stability and are difficult to
581 destroy using normal processing methods (Kumar et al. 2017). Interestingly, several studies
582 showed that LAB could effectively inhibit fungal growth and reduce mycotoxin production
583 (Ghanbari et al. 2018; Taheur et al. 2019; Wilburn and Ryan 2017). For example, *L. plantarum*
584 isolated from fermented cereal gruels exhibited maximum inhibition on the vegetative growth
585 and sporulation of aflatoxin-producing *Aspergillus*, mainly due to the secretion of phenyl-acids
586 that have strong antifungal properties (Onilude et al. 2005).
587
588 Although the reported studies on mycotoxin content in fermented BG are scarce, Adebiyi,
589 Kayitesi, and Njobeh (2020) showed that fermentation effectively reduces mycotoxins
590 (aflatoxins B_1 and G_1; fumonisins B_1 and B_2) that were initially present at high concentrations
591 in the raw BG. However, Olagunju et al. (2018) found that fermentation failed to eliminate
592 fungal contaminants and aflatoxins in BG flour, whereas roasting at 140°C for 20 minutes

successfully degraded the fungal toxins. These contradictory findings indicate the need for more research to investigate the effect of fermentation on the levels of aflatoxins and mycotoxins before such processing is widely adopted.

Concern about the possibility of opportunistic microorganisms or pathogens in fermented products is continuously being focused on and discussed. These unknown pathogenic microorganisms may carry along antibiotic-resistance genes and/or secrete toxic compounds, which can seriously threaten public health at excessive levels. Naturally fermented products exhibit great uncertainty in safety limits compared to those produced using the induced fermentation method. The risks of contamination by pathogens in induced fermentation will be far lower as it involves a sterile/controlled fermentation environment, optimized fermentation parameters and known starter cultures (usually with GRAS status) (Capozzi et al. 2017; Sharma et al. 2020).

Fermenting microorganisms like LAB and *Rhizopus* synthesize lactic acids, which can lower the pH of the substrate and act as a hurdle to the growth of acid-sensitive pathogens (e.g., *Campylobacter jejuni, Vibrio cholera,* and *Salmonella* spp.). In contrast, *Bacillus* releases ammonia as a by-product during fermentation and creates an alkaline environment that prevents the colonization of acidophilic spoilage microorganisms. In addition, some exhibit the ability to produce bacteriocins and antimicrobial peptides (Nagarajan, Rajasekaran, and Venkatachalam 2022). All these mechanisms serve as biopreservative means to ensure the safety of fermented products. Besides, the *in vitro* and *in vivo* (e.g., animal acute, subchronic and chronic toxicity testing) safety/quality control measurements are of high necessity before commercializing or marketing the fermented products. Furthermore, cooking, frying, and boiling before consuming the fermented foods (e.g., *tempeh* and miso) also help remove those

heat-labile pathogens and their toxic by-products, thus minimizing the safety concerns of the fermented products.

8. Conclusion

Fermentation has been shown to enhance the nutritional quality of BG, potentially increasing protein and other nutrient content and digestibility, as well as reducing the undesired ANFs due to the metabolic and enzymatic activities of the indigenous fermenting microorganisms or the inoculated well-defined starter cultures. This review suggests that fermentation could represent an affordable and efficient way of maximizing the nutrient content of this underutilized protein-rich legume, which could thrive in harsh climatic conditions. The induced fermentation method using known starter cultures and optimized process parameters are recommended to ensure end-product safety and quality rather than the natural fermentation method. While further research is required to confirm the safe use of fermented BG products, the *in vivo* nutrient digestibility using animal models should be assessed to fully understand the exact amount of bioavailable and bioaccessible nutrients in fermented diets.

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Table 1. Types of fermented Bambara groundnut products and their starter cultures involved in fermentation.

Fermented Bambara groundnut product	Natural starter cultures	Induced starter cultures	References
Condiment (e.g., *Dawadawa*, *Iru*, *Ogirri*)	*Bacillus cereus*, *Bacillus cereus* BTD4-1, *Bacillus cereus* GBPS9, *Bacillus subtilis*, *Bacillus licheniformis*, *Bacillus pumilus*, *Lysinibacillus fusiformis* LMG 18474, *Phyllobacterium leguminum* ORS1419, *Staphylococcus saprophyticus*, *Streptococcus* spp., *Aspergillus niger*, *Penicillium* spp., *Rhizopus* spp., *Saccharomyces cerevisiae*	*Bacillus amyloliquefaciens* subsp. *plantarum* SFBA2, *Bacillus cereus* PALB7, *Bacillus licheniformis*, *Bacillus licheniformis* OALB2, *Bacillus subtilis*, *Bacillus subtilis* subsp *subtilis* SFBA3	Adamu Shahidah et al. 2018; Adebiyi, Kayitesi, and Njobeh 2020; Adebiyi, Njobeh, and Kayitesi 2019; Ademiluyi and Oboh 2011; Aigbodion 2017; Akanni, De Kock, et al. 2018; Akanni, Naudé, et al. 2018; Amadi, Barimalaa, and Omosigho 1999; Barimalaa and Anoghalu 1997; Barimalaa et al. 1994; Fadahunsi and Olubunmi 2010; Oboh, Ademiluyi, and Akindahunsi 2009; Omogbai and Aghahowa 2017
Infant complementary food	ND	*Lactobacillus fermentum*, *Lactobacillus plantarum*	Chude, Amadi, and Iro 2018; Ijarotimi and Keshinro 2013
Flour	*Lactobacillus* spp., *Lactobacillus bulgaricus*, *Lactobacillus fermentum*, *Lactobacillus plantarum*, *Lactobacillus reuteri*, *Lactococcus lactis*, *Leuconostoc* spp., *Leuconostoc mesenteroides*, *Rhizopus* spp., *Candida utilis*, yeasts	*Lactobacillus brevis* ATCC 14869; *Lactobacillus fermentum* CIP 102980, *Lactobacillus nantensis* LP33, *Lactobacillus plantarum* WCFS1, *Lactobacillus reuteri* DSM 20016, *Lactobacillus rhamnosus* GG, ATCC 53/03, *Pediococcus acidilactici* DSM 20284, *Rhizopus nigricans*, *Rhizopus oligosporus*, *Rhizopus oligosporus* NRRL 2710, *Rhizopus oryzae*	Arise, Taiwo, and Malomo 2020; Fadahunsi 2009; Ijarotimi 2008; Ijarotimi and Esho 2009; Mbata, Ikenebomeh, and Alaneme 2009; Ogodo et al. 2018a, 2018b; Ola and Adewole 2019; Ola and Opaleye 2019; Olanipekun, Otunola, and Oyelade 2014, 2015; Olanipekun et al. 2012; Otunola, Ade-Omowaye, and Akanji 2007
Meal (e.g., experimental rat feed meal, broiler feed meal, fish meal)	ND	*Lactobacillus fermentum*, *Lactobacillus plantarum*, *Saccharomyces cerevisiae*	Chude, Okoyeuzu, and Amadi 2018; Enyidi and Etim 2020; Esiegwu and Obih 2019; Fualefac et al. 2014

Feed ingredient	ND	*Aspergillus flavus*, *Aspergillus niger*, *Penicillium* sp., *Trichoderma* sp., *Yeast*	Nwanna et al. 2005; Oloyede, Minari, and Muhammad 2010; Oloyede et al. 2007
Tempeh	ND	*Rhizopus arrhizus* NRRL 1527, *Rhizopus oligosporus* NRRL 2710, *Rhizopus stolonifer* NRLL 1477	Amadi et al. 1999
Yoghurt	ND	*Lactobacillus bulgaricus*, *Lactobacillus delbruieckii* subspp. *bulgaricus*, *Lactobacillus plantarum* A6, *Streptococcus thermophilus*, *Streptococcus salivarus* subspp. *thermophilus*	Adebanke et al. 2017; Ani, Amove, and Igbabul 2018; Falade et al. 2014; Pahane et al. 2017
Semolina (e.g., *Dackere*)	ND	*Lactobacillus plantarum* A6	Mahamat et al. 2016
Bran	*Lactobacillus acidophilus*, *Lactobacillus* mesenteroides, Lactobacillus *plantarum*	ND	Adeoye, Obayemi, and Akinola 2018
Probiotic beverage	ND	*Lactobacillus acidophilus*, *Lactobacillus bulgaricus*, *Lactobacillus casei*, *Lactobacillus plantarum*	Hardy and Jideani 2020; Murevanhema 2012; Murevanhema and Jideani 2020

Table 2. Summary of conditions for studies using natural fermentation of Bambara groundnut products.

Fermented Bambara groundnut product	Fermentation duration (h)	Fermentation temperature (°C)	Isolated fermenting microorganisms present	References
Condiment (e.g., *Dawadawa*, *Iru*, *Ogirri*)	48 - 120	25 - 45	*Bacillus cereus*, *Bacillus cereus* BTD4-1, *Bacillus cereus* GBPS9, *Bacillus subtilis*, *Bacillus licheniformis*, *Bacillus pumilus*, *Lysinibacillus fusiformis* LMG 18474, *Phyllobacterium leguminum* ORS1419, *Staphylococcus saprophyticus*, *Streptococcus* spp., *Aspergillus niger*, *Penicillium* spp., *Rhizopus* spp., *Saccharomyces cerevisiae*	Adamu Shahidah et al. 2018; Adebiyi, Kayitesi, and Njobeh 2020; Adebiyi, Njobeh, and Kayitesi 2019; Ademiluyi and Oboh 2011; Aigbodion 2017; Barimalaa and Anoghalu 1997; Barimalaa et al. 1994; Fadahunsi and Olubunmi 2010; Oboh, Ademiluyi, and Akindahunsi 2009; Omogbai and Aghahowa 2017
Infant complementary food	72	ND	ND	Ijarotimi and Keshinro 2013
Flour	48 - 96	28 - 32	*Lactobacillus* spp., *Lactobacillus bulgaricus*, *Lactobacillus fermentum*, *Lactobacillus plantarum*, *Lactobacillus reuteri*, *Lactococcus lactis*, *Leuconostoc* spp., *Leuconostoc mesenteroides*, *Rhizopus* spp., *Candida utilis*, yeasts	Arise, Taiwo, and Malomo 2020; Ijarotimi 2008; Ijarotimi and Esho 2009; Mbata, Ikenebomeh, and Alaneme 2009; Ogodo et al. 2018a, 2018b
Broiler feed meal	36 - 72	ND	ND	Esiegwu and Obih 2019; Fualefac et al. 2014
Bran	96	28 - 32	*Lactobacillus acidophilus*, *Lactobacillus* mesenteroides, Lactobacillus *plantarum*	Adeoye, Obayemi, and Akinola 2018

Note: ND-Not determined

Table 3. Summary of conditions for studies using induced fermentation of Bambara groundnut products.

Fermented Bambara groundnut product	Fermentation duration (h)	Fermentation temperature (°C)	Composition of starter culture	References
Condiment (e.g., *Dawadawa*)	48 - 120	30 - 45	*Bacillus amyloliquefaciens* subsp. *plantarum* SFBA2, *Bacillus cereus* PALB7, *Bacillus licheniformis*, *Bacillus licheniformis* OALB2, *Bacillus subtilis*, *Bacillus subtilis* subsp *subtilis* SFBA3	Adamu Shahidah et al. 2018; Akanni, De Kock, et al. 2018; Akanni, Naudé, et al. 2018; Amadi, Barimalaa, and Omosigho 1999
Infant complementary food	72	25	*Lactobacillus fermentum*, *Lactobacillus plantarum*	Chude, Amadi, and Iro 2018
Flour	24 - 72	30 - 32	*Lactobacillus brevis* ATCC 14869; *Lactobacillus fermentum* CIP 102980, *Lactobacillus nantensis* LP33, *Lactobacillus plantarum* WCFS1, *Lactobacillus reuteri* DSM 20016, *Lactobacillus rhamnosus* GG, ATCC 53/03, *Pediococcus acidilactici* DSM 20284, *Rhizopus nigricans*, *Rhizopus oligosporus*, *Rhizopus oligosporus* NRRL 2710, *Rhizopus oryzae*	Fadahunsi 2009; Ogodo et al. 2018a, 2018b; Ola and Adewole 2019; Ola and Opaleye 2019; Olanipekun, Otunola, and Oyelade 2014, 2015; Olanipekun et al. 2012; Otunola, Ade-Omowaye, and Akanji 2007
Meal (e.g., experimental rat feed meal, fish meal)	72 - 96	25 - 32	*Lactobacillus fermentum*, *Lactobacillus plantarum*, *Saccharomyces cerevisiae*	Chude, Okoyeuzu, and Amadi 2018; Enyidi and Etim 2020
Feed ingredient	72 - 336	25	*Aspergillus flavus*, *Aspergillus niger*, *Penicillium* sp., *Trichoderma* sp., *Yeast.*	Nwanna et al. 2005; Oloyede, Minari, and Muhammad 2010; Oloyede et al. 2007
Tempeh	18 - 30	30	*Rhizopus arrhizus* NRRL 1527, *Rhizopus oligosporus* NRRL 2710, *Rhizopus stolonifer* NRLL 1477	Amadi et al. 1999
Yoghurt	5 - 10	38 - 45	*Lactobacillus bulgaricus*, *Lactobacillus delbruieckii* subspp. *bulgaricus*, *Lactobacillus plantarum* A6, *Streptococcus thermophilus*, *Streptococcus salivarus* subspp. *thermophilus*	Adebanke et al. 2017; Ani, Amove, and Igbabul 2018; Falade et al. 2014; Pahane et al. 2017
Semolina (e.g., *Dackere*)	60	37	*Lactobacillus plantarum* A6	Mahamat et al. 2016

Probiotic beverage	24	35	*Lactobacillus acidophilus*, *Lactobacillus bulgaricus*, *Lactobacillus casei*, *Lactobacillus plantarum*	Hardy and Jideani 2020; Murevanhema 2012; Murevanhema and Jideani 2020

Table 4. Effects of fermentation on the proximate nutrient contents of Bambara groundnut products.

Fermented Bambara groundnut product	Changes in proximate nutrient contents (%)				References
	Protein	Carbohydrate	Lipid	Ash	
Condiment (e.g., *Dawadawa*)	3.3 - 7.4% increment	2.3 - 34.4% reduction	0.1 - 13.7% increment	0.7 - 8.2% reduction	Adamu Shahidah et al. 2018; Adebiyi, Njobeh, and Kayitesi 2019; Aigbodion 2017; Amadi, Barimalaa, and Omosigho 1999; Barimalaa et al. 1994
Flour	3.9 - 6.2% increment	0.3 - 5.7% reduction	1.1 - 1.4% reduction	0.8 - 1.1% increment	Ogodo et al. 2018b; Olanipekun et al. 2012
	0.5% increment	0.2% increment	0.1% increment	0.8% reduction	Ijarotimi and Esho 2009
Fish meal	1.6% increment	2.5% reduction	0.1% reduction	0.3% increment	Esiegwu and Obih 2019
Tempeh	6.6 - 7.4% increment	13.0 - 13.2% reduction	5.0% increment	ND	Amadi et al. 1999
Yoghurt	0.8% increment	1.1% reduction	ND	0.9% increment	Pahane et al. 2017

Note: ND-Not determined

Table 5. Effects of fermentation on the levels of anti-nutritional factors in Bambara groundnut products.

Fermented Bambara groundnut product	Changes in levels of anti-nutritional factors				References
	Trypsin inhibitor	Phytate	Tannin	Oxalate	
Condiment (e.g., *Dawadawa*)	38.3 - 40.9% reduction	18.1 - 25.6% reduction	34.2 - 76.4% reduction	26.6 - 59.1% reduction	Adebiyi, Njobeh, and Kayitesi 2019; Barimalaa et al. 1994; Barimalaa and Anoghalu 1997
Flour	37.3 - 87.1% reduction	39.4 - 95.9% reduction	25.6 - 98.3% reduction	15.8 - 74.0% reduction	Ijarotimi and Esho 2009; Ogodo et al. 2018a; Ola and Opaleye 2019; Olanipekun, Otunola, and Oyelade 2015
	87.3% increment	ND	ND	ND	Fadahunsi 2009
Yoghurt	ND	89.7% reduction	ND	ND	Pahane et al. 2017

Note: ND-Not determined