Direct Research Journal of Agriculture and Food Science
Vol. 12(1), Pp. 69-75, February 2024
ISSN 2354-4147
DOI: https://doi.org/10.26765/DRJAFS15212970
Article Number DRJAFS15212970

https://journals.directresearchpublisher.org/index.php/drjafs

Original Research

Preservative Potential of Aqueous Garlic Extract on the Biochemical Characteristics and Organoleptic Qualities of Functional Soymilk

Malomo, A. A.*, Fabiyi, O. I., Adejumo, S. D., and Adewolu, H. A.

Department of Food Science and Technology, Obafemi Awolowo University, Ile-Ife, Nigeria.
Corresponding Author E-mail: adepojuadekunbi@gmail.com

Received 2 April 2023; Accepted 25 February 2024; Published 29 February 2024

ABSTRACT: Soymilk, a plant-based milk, is a cheap alternative to dairy milk. It has a short shelf life and is highly perishable. This study examined the effect of aqueous garlic extract on the shelf life of soymilk to produce a functional food with a longer shelf life. The microbial count, pH, and titratable acidity of the refrigerated soymilk samples were determined weekly for 21 days as well as the organoleptic properties. Microorganisms isolated from the soymilk samples during storage were also identified using standard methods. *Bacillus spp.*, *Staphylococcus spp.*, *Streptococcus spp.*, and *Aspergillus niger* were identified during storage and the total viable count obtained for the freshly prepared soymilk was lower compared to the acceptable standard of bacterial limit (4.30 log CFU/ml) for Soy foods. The preserved soymilk's initial and final fungal count ranged from 4.46 - 4.79 and 8.30 - 8.70 log CFU/ml respectively. The score for taste decreased with an increase in the concentration of garlic extract. However, the overall acceptability of the soymilk samples was not significantly different ($p > 0.05$) with the addition of garlic extract. Incorporating 3 and 4% aqueous garlic extract into soymilk, as a preservative, increased the product's shelf life beyond 7 days. Thus, garlic extract could be explored as a potential preservative in extending the shelf life of soymilk.

Keywords: Functional food, soymilk, garlic, shelf life, *bacillus*
Citation: Malomo, A. A., Fabiyi, O. I., Adejumo, S. D. and Adewolu, H. A. (2024). Preservative Potential of Aqueous Garlic Extract on the Biochemical Characteristics and Organoleptic Qualities of Functional Soymilk. Direct Res. J. Agric. Food Sci. Vol. 12(1), Pp. 69-75. https://doi.org/10.26765/DRJAFS15212970

INTRODUCTION

Soybean (*Glycine max* Merrill) is a cheap leguminous plant that is rich in essential amino acids, micronutrients and unsaturated fatty acids. Protein is an essential macronutrient needed daily and it is very important in the diet for proper growth and development of the body (Kabiru *et al*., 2016). Likewise, soybean is rich in polysaccharides, soluble fibres, saponins, and isoflavones. Products such as soymilk, tofu, sufu (furu), soybean-based cereal, root and tuber products are derived from soybeans to increase access to healthy and affordable diets (Farinde *et al*., 2016; Malomo *et al*., 2018). Soymilk is a soybean product that originated from the Orients and is now consumed all over the world as an alternative source of protein. It is a creamy filtrate produced from soaked, dehulled, milled, and sieved soybeans. It is highly acceptable because of its nutritional composition and resemblance to cow milk (Tunde-Akintunde and Souley, 2009; Borode, 2017). According to Mazumder and Begum (2016), soymilk has about 4 - 7 mg total isoflavones, a rich source of B-vitamin complex such as niacin (0.15 mg/100 g), folacin (1.5 µg/100 g), Vitamin E (0.01 mg/100 g) and a minimal amount of vitamin A (3.0 µg/100 g). It is a rich source of minerals like calcium (4.7 mg/100 g), iron 1.0 mg/100 g), zinc (0.23 mg/ 100 g), magnesium (19.0 mg/ 100 g), phosphorus (49.0 mg/ 100g), sodium (12.0 mg/ 100 g) and potassium (141.0 mg/ 100 g). The use of synthetic preservatives in food processing is declining due to the awareness about

its adverse effect on the health of consumers thereby increasing the demand for bio-preservatives (Mahmood *et al*., 2019; Malomo, and Abiose, 2019). Herbs and spices contain phenolic compounds that serve as powerful antioxidants against free radicals. They have been known for years as the treatment for stomach ache, convulsions, cough, loss of appetite and rheumatism. Spices have been used since time past in many cultures as seasonings, preservatives and the knowledge of their medicinal properties has been recorded and is still in use till date (Ene-Obong, 2018).

Garlic (*Allium sativum*) is a functional spice known for its nutritional and health benefits. It has anti-inflammatory, and anti-oxidative properties and it has been reported to lower blood pressure, and cholesterol levels and prevent clotting of blood (Ayar and Gurlin 2014; Alolgaa *et al*., 2021). It is a popular additive in food processing both at domestic and industrial levels because of its desirable flavour, appetizing properties and increase in the flow of gastric juice. It also contains a bioactive substance known as allicin which is an antioxidant and antimicrobial substance that reduces/prevents the growth and activities of spoilage microorganisms thereby extending the shelf life of food (Sohany *et al*., 2022). It has been confirmed that garlic and garlic products keep the taste and flavour of foods by preventing bacterial and fungal growth. In addition, garlic extract acts as an antioxidant. The application of naturally occurring antioxidants such as garlic and garlic products in food preservation and their benefits on human health has gained wide attention (Gündoğdu *et al*., 2009).

Addition of a significant level of biologically active substances to foods has increased the physiological advantages enjoyed by the consumer and broadens the metabolism homeostasis (Cha *et al*., 2020). Over the years dairy products have been exploited in the development of functional foods whereas non-dairy products are under-studied. Functional soymilk has been produced through enrichment with B vitamins (Zhu *et al*., 2020), fermentation using lactic acid (Kumari *et al*., 2022) supplementation with microorganisms such as *Bifidobacterium longum* (Zhao and Shah, 2014). Yet, there is little or no information about the application of spices such as garlic as a preservative and bio-enrichment molecule in soymilk. Hence, this study examined the effect of aqueous garlic extract on the shelf life of soymilk and its influence on some of the microbial, physicochemical and sensory qualities.

MATERIALS AND METHODS

Preparation of aqueous garlic extract and soymilk

Garlic and Soybean were bought from a local market in Ile-Ife, Osun state Nigeria. The aqueous extract of garlic (*Allium sativum*) was produced using the method of Gbodi *et al*. (2002). Soymilk was produced using the method of Iwe (2003) (Modified).

Sample design and storage

The soymilk was divided into four parts. Garlic extracts (2%, 3% and 4%) were added to the three samples while the fourth sample was untreated (control sample). The samples were designated as SG0- Soymilk without preservative (Control); SG2- Soymilk preserved with 2% garlic extract; SG3- Soymilk preserved with 3% garlic extract; SG4- Soymilk preserved with 4% garlic extract. The treated and untreated samples were kept at refrigerated temperature (4 ± 2 ˚C) for 21 days.

Analyses

Microbiological, physicochemical analyses and sensory evaluation were carried out using standard methods. The total viable count and fungal count were determined using the pour plate method (Harrigan, 1998). pH values of preserved soymilk samples were determined with an electronic digital pH meter after calibration of the equipment and titration method was used for the determination of titratable acidity (AOAC, 2000). Fifteen (15) semi-trained panelists were used for sensory evaluation of the soymilk samples using a 9-point Hedonic scale (Larmond, 1977).

Data analysis

Data obtained were analysed statistically using SPSS (Analysis of Variance -ANOVA). The means for the scores were separated with Duncan Multiple Range Test (DMRT). The significant difference in the means was determined at $p<0.05$.

RESULTS AND DISCUSSION

Biochemical characteristics and identity of microorganisms isolated from garlic extract-preserved soymilk

All the bacteria isolates associated with soymilk preserved with garlic extract were Gram-positive. These belong to the genera *Bacilli, Staphylococci* and *Streptococci*. The species of the bacteria were identified as *Bacillus licheniformis*, *B*. spp., *B. subtilis*, *Staphylococcus epidermidis* and *Streptococcus* spp. All the organisms isolated from the treated and untreated soymilk samples were lactose-fermenters. No coliform was isolated from the soymilk samples. This is an indication of the observation of good manufacturing practices during the production of the milk.

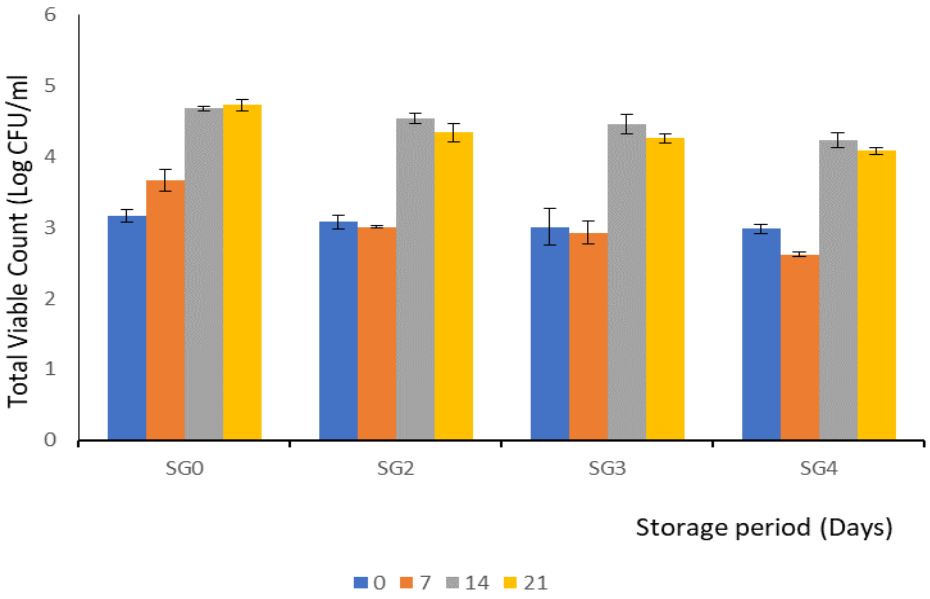

Figure 1: Total viable counts of aqueous garlic extract-treated soymilk during cold storage.

Arekemase and Babashola (2019) reported the dominance of the genera *Bacillus* and *Staphylococcus* in ginger-preserved soymilk. The fungi isolated from the soymilk was identified as *Aspergillus* sp. Several studies (Kabiru *et al*., 2012; Arekemase and Babashola, 2019) have reported the presence of *Aspergillus* spp. in preserved soymilk.

Total viable count of preserved soymilk during storage at refrigeration temperature

The initial total viable count obtained for the freshly prepared soymilk (untreated and treated) samples ranged between 2.62 – 4.08 log CFU/ml (Figure 1). The soymilk sample treated with 4% aqueous garlic extract (sample SG4) had the lowest initial total viable count while the untreated soymilk had the highest initial total viable count. The total viable count obtained for the fresh soymilk was below the acceptable limit of 4.30 log CFU/ml bacterial count recommended by the Soy Foods Association of America (SFAA). After 21 days of storage, the untreated soymilk sample, samples SG2 and SG3 had exceeded the acceptable bacterial limit. Sample SG4 had a total viable count lesser than the acceptable bacterial limit for Soy Foods after 21 days of storage at refrigerated temperature. The addition of 4% aqueous garlic extract to soymilk was effective in limiting bacteria multiplication. Thereby, increasing the shelf life of the soymilk product by days compared with the untreated sample. Inhibition of bacterial growth in clove-preserved soymilk was reported by Kabiru *et al*. (2012) as observed in this study.

pH and titratable acidity of soymilk during storage at refrigeration temperature

The initial pH of garlic extract treated soymilk ranged from 6.77 – 7.21. The soymilk sample without garlic extract had the highest pH value (7.21) while the sample with 4% garlic extract had the lowest pH. At the end of 21 days of refrigerated storage, the pH value had decreased to a range of 6.25 – 6.49 as shown in Figure 2. The control sample had the highest pH value (6.49). Gündoğdu *et al*., (2009) also reported a decrease in pH and an increase in TTA with the addition of garlic. Likewise, Arekemase and Babashola (2019) reported a drop in the pH of soymilk preserved with ginger and clove extracts during storage.

The titratable acidity of garlic-treated soymilk ranged from 0.28 - 0.54% before storage and 0.34 - 0.52% after storage for 21 days at refrigerated temperature. The soymilk sample treated with 4% garlic extract had the highest TTA values while the untreated soymilk sample had the lowest TTA values before and after storage. The TTA increased with a decrease in the pH of the stored soymilk samples (Figure 3). Production of lactic acid during storage could increase the titratable acidity level of the soymilk samples. Akakpo *et al*. (2019) also reported similar findings in the preservation of soymilk with essential oil from citrus.

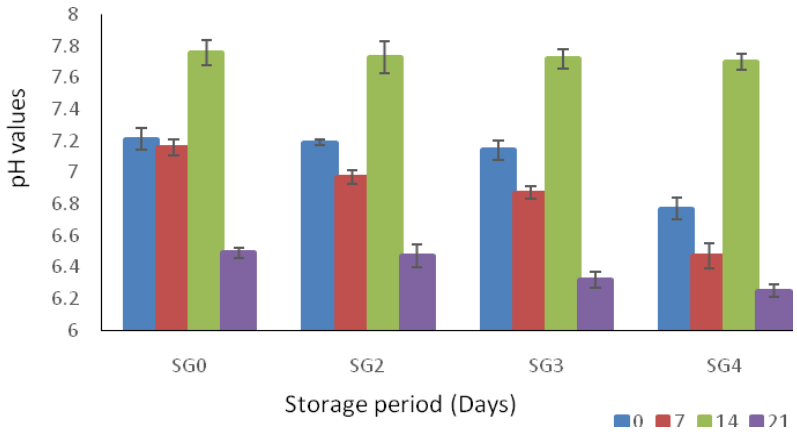

Figure 2: pH of aqueous garlic extract-treated- soymilk.

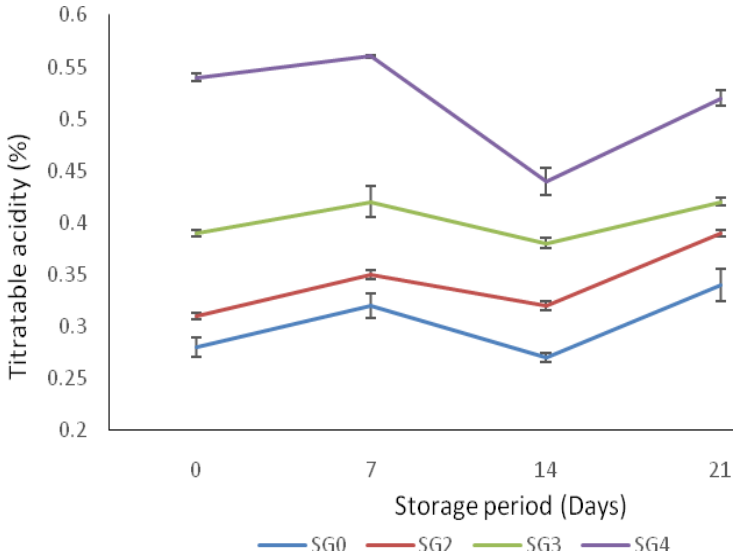

Figure 3: Titratable acidity of aqueous garlic extract-treated soymilk during refrigerated storage.

Sensory qualities of soymilk samples preserved with aqueous garlic extract

The colour of soymilk samples with or without garlic was not significantly different ($p < 0.05$) but the score of the colour of soymilk samples increased as the concentration of garlic extract increased in the samples. Soymilk with 4% garlic extract (Sample SG4) had the highest score for colour (Table 1). The appearances of all the soymilk samples were not different statistically. The anti-oxidative property of garlic could be responsible for the stability of colour and improvement in the appearance of soymilk. It has been reported that garlic is rich in phenolic compounds that serve as antioxidants (Daka, 2011). There was an increase in the mean score for taste of soymilk with the increase in garlic concentration. A slight decrease in the score for taste of sample SG4 when the concentration increased to 4% was observed. Sample SG4 had the highest score while sample SG0 had the lowest score for taste. The score for taste also decreased with an increase in the concentration of garlic extract. Soymilk sample (SG0) which had no garlic had the

Table 1: Sensory Scores of Soymilk Samples treated with Garlic Extract.

Samples	Colour	Appearance	Taste	Aroma	Oiliness	Viscosity	Mouth coating	After taste	Overall acceptability
SG0	7.07±0.17^{a}	6.93±0.19^{a}	7.27±0.04^{a}	6.40±0.15^{a}	5.53±0.61^{a}	6.07±0.18^{a}	5.93±0.32^{a}	6.47±0.02^{a}	6.60±0.12^{a}
SG2	6.80±0.52^{a}	6.80±0.22^{a}	5.93±1.61^{ab}	5.33±1.02^{a}	5.33±0.90^{a}	5.93±0.62^{a}	5.73±0.70^{a}	5.07±0.51^{b}	5.87±0.62^{a}
SG3	6.67±0.73^{a}	6.60±0.38^{a}	5.20±1.43^{b}	5.53±0.96^{a}	5.20±1.10^{a}	5.60±0.84^{a}	5.40±0.95^{a}	5.47±1.00^{ab}	6.20±0.34^{a}
SG4	7.13±0.09^{a}	6.67±0.32^{a}	5.53±1.30^{b}	5.87±0.73^{a}	5.47±0.55^{a}	5.53±0.97^{a}	5.27±1.01^{a}	5.40±0.95^{ab}	6.27±0.27^{a}

All values with different alphabets at the superscript in the same row are significantly different at (P< 0.05)
Key: SG0- Soymilk without preservative (Control); SG2- -Soymilk preserved with 2% garlic extract; SG3-Soymilk preserved with 3% garlic extract; SG4-Soymilk preserved with 4% garlic extract

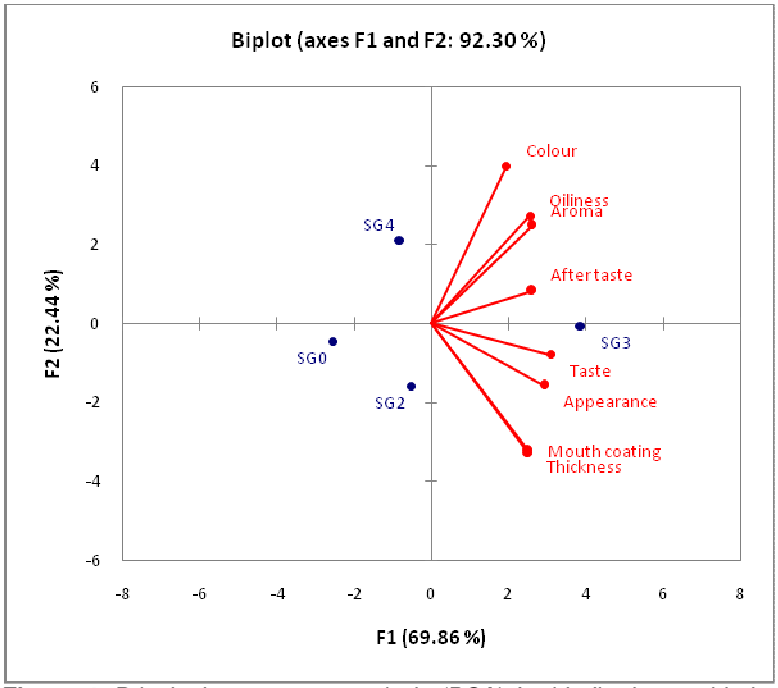

Figure 4: Principal component analysis (PCA) for bi-pilot in considering two factors.

highest score for taste (7.27). The addition of garlic to soymilk had a negative effect on the taste at 4% (5.53) (SG4). The aroma of the soymilk samples was not different statistically either in the presence or absence of garlic extract), although sample SG0 had the highest score (6.40) for aroma. Some authors (Gündoğdu *et al*., 2009; Nielsen and Rios, 2000) have reported that garlic improves and preserves the taste and flavour of food.

Statistically, there was no significant difference in the viscosity of the soymilk after the addition of the garlic extract (2 - 4%). The sample SG4 had a lower score (5.53) than sample SG0 (6.07) for viscosity. This could be due to the absence of garlic extract in sample SG0. The oil on the perception of astringency during the ingestion of soymilk was not significantly different with the addition of garlic extract to the samples as judged by the panelists. The ability of a consumer to detect the oil on perception for soymilk has been established to increase maximally with continuous sips according to Courregelongue *et al*. (1999).

The addition of garlic extract to soymilk (2 - 4%) had no significant difference in the mouth coating of the samples. The mean sensory scores for mouth coating of the soymilk samples decreased with an increase in concentration (2- 4%) of garlic extract. The sample SG4 had a lower score (5.27) for mouth coating than sample SG0 (5.93). The concentration of garlic used for this study had no influence on the mouth coating of the soymilk sample. This could probably be because garlic could mask the astringency of soymilk at concentrations of 2 - 4%.

Sample SG4 had the lowest score while sample SG0 had the highest score for aftertaste. Sample SG4 probably had a lower score for aftertaste because of the high concentration of garlic extract. The high concentration of garlic (4%) could impart a pungent smell and astringent

taste of garlic in soymilk. It has been reported that garlic contributes strong odour, appetizer properties and bitter taste, which contributes flavours of food consumed (Courregelongue *et al*., 1999). There was no significant difference in the overall acceptability of the samples. The addition of 4% garlic extract to soymilk was the most preferred followed by sample SG3 which contains 3% garlic extract. Nevertheless, there was no significant difference in the overall acceptability of the soymilk samples preserved with garlic extract and the untreated sample.

The soymilk preserved with aqueous garlic extract compared favourably with the control that had no garlic. None of the samples scored less than 5.00 on the 9-point Hedonic scale which means the addition of garlic (2 – 4%) as a bio-preservative and functional ingredient to soymilk had a positive effect on consumer acceptability.

Principal component analysis of the sensory attributes of garlic-treated- soymilk

The PCA separated the soymilk samples into three sections. Figure 4 shows that the first component accounted for 68.86%, while the second component accounted for 22.44%. It was discovered that sample SG3 had the highest connection with all of the sensory qualities tested. It also meant that SG0 had the least relationship with the sensory characteristics. The sample SG4 demonstrated a poorer correlation than the samples SG3 and SG2. This could be attributed to a rise in garlic extract concentration, which revealed the pungent smell and astringent taste of soymilk. When antioxidants such as ascorbic acid and phenolic compounds are given to food at increasing concentrations, they have been shown to act as pro-oxidants (Malomo *et al*., 2022).

Conclusion

The addition of aqueous garlic extract at 3 and 4% concentration extended the shelf life and stability of soymilk at a refrigerated temperature beyond 7 days. Garlic extract has a promising application as a biopreservative for soymilk. Also, the sensory qualities of soymilk were not negatively influenced by the addition of garlic extract; hence it could be included as a functional food for consumers.

Acknowledgment

Profound gratitude goes to all the technical staff at the Department of Food Science and Technology, Obafemi Awolowo University, Ile-Ife, Nigeria for their contributions towards the success of this research.

Funding sources

This research was not funded by any institution.

REFERENCES

Akakpo, A., Somda, M., Kabore, D., Mihin, H., Taale, E., Ouattara, C., and Ouattara, A. (2019). Biopreservation and sensory quality of soymilk (*Glycine max*) by using essential oil from *Cymbopogon citratus* (DC) Stapf in Burkina Faso. Nutrition and food, 131.

Alolgaa, R. N., Osae, R., Essilfiec, G., Saaliad, F. K., Akaba, S. and Chikarib, F. (2021). Sonication, osmosonication and vacuum-assisted osmosonication pretreatment of Ghanaian garlic slices: Effect on physicochemical properties and quality characteristics. Food chemistry, Vol. 343 (2021), 1- 11.

AOAC (200). Assn. of Official Analytical Chemists. 2000. Coffee and tea. In: Official methods of analysis. 17th ed. Gaithersburg, Md.: AOAC.

Arekemase, M. O. and Babashola, D. R. (2019). Assessment of the effectiveness of ginger (*Zingiber officinale*), clove (*Syzygium aromaticum*) and sodium benzoate on the shelf life of soymilk. Notulae scientia biologicae, Vol. 11(4), 2019.

Ayar, A., and Gurlin, E. (2014). Production and sensory, textural, physicochemical properties of flavored spreadable yogurt. Life science journal, Vol. 11(4), 58-65.

Borode, O. F. (2017). The effect of water and ethanol extracts of ginger and garlic on the nutritional quality and physico-chemical properties of stored soymilk. International journal of food science and biotechnology, Vol. 2(2), 43-50. doi: 10.11648/j.ijfsb.20170202.12.

Cha, K. H., Yang, J. S., Kim, K. A., Yoon, K. Y., Song, D. G., Erdene-Ochir, E., Kang, K., Pan, C. H. and Ko, G. (2020). Improvement in host metabolic homeostasis and alteration in gut microbiota in mice on the high-fat diet: a comparison of calcium supplements. Food research international, Vol. 36, 109495.

Courregelongue, S., Schlich, P., and Noble, A. C. (1999). Using repeated ingestion to determine the effect of sweetness, viscosity and oiliness on temporal perception of soymilk astringency. Food quality and preferences, Vol. 10(4-5), 273-279.

Daka, D. (2011). Antibacteria effect of garlic (*Allium sativum*) on *Staphylococcus aureus*: An *in vitro* study. African journal of biotechnology, Vol. 10(4), 666-669.

Ene-Obong, H., Onuoha, N. O., Aburimea, L. and Mbah, O. (2018). Chemical composition and antioxidant activities of some indigenous spices consumed in Nigeria. Food chemistry, Vol. 238 (2018), 58–64.

Farinde, E. O. Ejigbo, E. A. and Sulaiman P. O. (2016). Production of yoghurt analogue from soymilk and different fermented cereal based water filtrates (omi ogi). Food science and quality management, Vol. 56 (2016), 72 -78.

Gbodi, T.A; Akanya, H.O.; Makun, H.A; Kabiru, Y.A. and Yakubu, Y. (2002). Preservation of soymilk using extract of ginger and garlic. Nigeria journal of technological research, Vol. 1, 67-74.

Gündoğdu, E., Çakmakçi, S. and Dağdemir, E. (2009). The effect of garlic (*Allium sativum* L.) on some quality properties and shelf- life of set and stirred yoghurt. Turkish journal of veterinary animal science, Vol. 33(1), 27-35.

Harrigan, W. F. (1998). Laboratory methods in food microbiology. Gulf professional publishing.

Iwe, M. O (2003), The science and technology of soybean. Journal of science and food agriculture, Vol. 4(4), 79-145.

Kabiru, A. Y., Makun, H. A., Saidu, A., Muhammad, H. L., Nuntah, L. C., and Amoo, S. A. (2012). Soymilk preservation using extracts of cloves (*Syzygium aromaticum* Myrtaceae) and guinea-pepper (*Xylopia aethiopica* Annonaceae).http://repository.futminna.edu.ng:8080/jspui/handle/123456789/14817.

Kabiru, Y. A., Makun, H. A., Saidu, A. N., Muhammad, L. H.,

Nuntah, L. C and Amoo, S. A. (2016). Soymilk Preservation Using Extracts of Cloves (*Syzygium aromaticum* Myrtaceae) And Guinea-Pepper (*Xylopia aethiopica* Annonaceae). Journal of pharmacy and biological sciences, Vol. 3(5), 44-50.

Kumari, M., Kokkiligadda, A., Dasriya,V. and Naithani, H. (2022). Functional relevance and health benefits of soymilk fermented by lactic acid bacteria. Journal of applied microbiology, Vol. 133(1), 104-119.

Larmond, E. (1977). Laboratory methods for sensory evaluation of food. Research Branch, Canada Dept. of Agriculture.

Mahmood, A., Tuan Zainazor, T.C. and Anuar, N.R (2019). Effect of garlic (*Allium sativum* L.) on the physicochemical, microbiological and sensory properties of chili sauce. Food research, Vol. 3(5), 416 – 421.

Mahmood, H., Ali, Q., Hafeez, M., and Malik, A. (2021). Antioxidant activity of *Syzygium aromatium* and *Cinnamomum verum* seed extracts. Biological and clinical sciences research journal, Vol. 2021(1). https://doi.org/10.54112/bcsrj.v2021i1.63

Malomo, A. A. and Abiose, S. H. (2019). Protein quality and functional properties of *masa* produced from maize, *acha* and soybean. Food research, Vol. 3(5), 556 - 563. https://doi.org/10.26656/fr.2017.3(5).223.

Malomo, A. A., Abiose, S. H. and Adeniran, H. A. (2018). Microbiological changes during the production of Maize-Acha *Masa* Fortified with Soybean. Annals. Food science and technology, Vol. 19(2), 349 - 357.

Malomo, A. A.,Odubanjo, A. F., Olawoye, O., Olaniyi, O. I., and Lawal, M. A. (2022). Effect of turmeric on the quality of canned African catfish in tomato sauce during storage at 25°C and 45°C. Food science and applied biotechnology, Vol. 5(1), 12-21.https://doi.org/10.30721/fsab2022.v5.i1.162.

Mazumder, M. A. R. and Begum, A. A. (2016). Soy milk as source of nutrient for malnourished population of developing country: A review. International journal of advanced scientific and technical research, Vol. 5(6), 192-203.

Nielsen, P. V., and Rios, R. (2000). Inhibition of fungal growth on bread by volatile components from spices and herbs, and the possible application in active packaging, with special emphasis on mustard essential oil. International journal of food microbiology, Vol. 60(2-3), 219-229.

Sohany, M., Halim,M.A., Akhter, M.J., Yasmin, S. and Noor, F.(2022). Effect of garlic paste on the physicochemical attributes of Cheese. Food and nutrition sciences, Vol. 13, 6-16.

Tunde-Akintunde T. Y. and Souley A. (2009). Effect of processing methods on quality of soymilk. Pakistan journal of nutrition, Vol. 8 (8), 1156-1158.

Zhao, D. and Shah, N. P. (2014). Antiradical and tea polyphenol stabilizing ability of functional fermented soymilk-tea beverage. Food chemistry, Vol. 158, 262-269.

Zhu, Y. Y., Thakur, K., Feng, J. Y., Cai, J. S., Zhang,J. G., Hu, F. and Wei, Z. J. (2020). B vitamin enriched fermented soymilk: A novel strategy for soy-based functional foods development. Trends in food science and technology, Vol. 105, 43-55.