



## Microbial profile and proximate composition of commonly used food thicker

Omorodion Nnenna\* and Nwala Chinyere

Department of Microbiology, University of Port Harcourt, P.M.B 5323, Port Harcourt, Rivers State, Nigriia  
nnennaomorodion@gmail.com

Available online at: [www.isca.in](http://www.isca.in), [www.isca.me](http://www.isca.me)

Received 16<sup>th</sup> April 2019, revised 11<sup>th</sup> September 2019, accepted 5<sup>th</sup> October 2019

### Abstract

The study evaluated the nutritional and microbial profile of five food condiments *Brachyegiaurycoma* (Achi), *Detariummicrocapum* (ofor), *Mucunasloani* (ukpo), *Cocoyam* (Ede) and starch used as food thickening agents bought from different markets. The total bacteria counts of the food thickening agent ranged from  $4.0 \times 10^6$  to  $2.12 \times 10^8$  cfu/g. However Achi had the highest bacteria count. The *Staphylococcus* counts of the food thickening agent ranged from  $1.5 \times 10^5$  to  $1.08 \times 10^6$  cfu/g. The *Salmonella shigella* counts ranged from  $3.0 \times 10^4$  to  $6.5 \times 10^5$  cfu/g, no visible growth was observed for Starch and Cocoyam but shigella was present in Ukpo, Fungal counts ranged from  $2.0 \times 10^6$  to  $5.4 \times 10^7$  cfu/g. These values when compared to the standards for food condiments which is  $10^3 < 10^5$  is unsatisfactory. Microorganism isolated were *Shigella* (5%), *Escherichia coli* (20%), *Staphylococcus spp* (29%), *Proteus spp* (10%), *Klebsiella spp* (18%) and *Bacillus spp* (18%). *Staphylococcus spp* had the highest frequency of occurrence due to its present on the skin, hands and mucous membrane. The presence of these organism indicates poor sanitary practices during the production process, however in order to improve the quality of these food thickening agents good sanitary practices must be adopted especially by the market sellers.

**Keywords:** Microbial, profile, proximate, composition, commonly, food thicker.

### Introduction

Thickening agents or thickeners can be defined as substances which when put into a mixture increase its viscosity without substantially changing other properties such as taste and aroma<sup>1</sup>. Their seed flour has a sticky texture when used in soups, which is a desirable attribute necessary for the eating of garri, pounded yam, etc.<sup>2</sup>. Food thickeners or thickening agents can be utilized in food to take up the dampness of the sustenance without changing its physicochemical composition. There are distinctive sorts of food thickeners that are being utilized and this relies on the kind of nourishment and reason, for instance, some are utilized to make the food delicious, and have enhance and nutritive esteem. food thickener are significant sources starch, protein and fats. Food thickeners has a high protein content when contrasted with that of some significant protein sources like fish and pork<sup>3</sup>. There various kinds of thickening specialists accessible in Nigeria, which are for the most part Achi (*Brachystegiaeurycoma*), Ofor (*Detariummicrocarpum*), and Ukpo (*Mucunaflagellipes*) and so on, and are right now utilized by numerous Nigerians. As of now, these are delivered on a cabin industry scale, which shifts starting with one ethnic gathering and area then onto the next. Legumes (family: Fabaceae) have been perceived to be the second most important plant hotspot for human and creature nourishment. Legumes have been featured as a powerful substitute to animal protein just as being practical. Practise of guaranteeing a safe products during harvesting, stockpiling, transport, preparing and bundling from the purchasing to selling point is negligible. The handling

techniques, vary starting with one area then onto the next, accordingly, the clean nature of the items likewise contrast. Practically all food can be contaminate by fungal organism that are equipped for producing at least one mycotoxins, especially Aflatoxins<sup>4</sup>. Aflatoxins produced by strains of *Aspergillus flavus* and *Aspergillus parasiticus* are hepatocarcinogenic, teratogenic and mutagenic, and have additionally been connected with growth stunning, underweight and change of insusceptible capacity in West African children<sup>5</sup>. The level of *Aspergillus* contamination and aflatoxin generation is known to be impacted by numerous biotic and abiotic natural components<sup>6</sup>. Fungal strain types, substrate, pH, temperature, relative humidity, moisture content of the substrate and air circulation have been found to impact the quality and amount of aflatoxin created, notwithstanding the collaboration among host and invading fungi. At present, no information is recorded on fungal contamination of prepared to-utilize food thickeners sold to shoppers in Nigeria. The customary strategies for preparing and poor treatment of these operators are the cause of contamination by aflatoxigenic type of *Aspergillus* species. Health hazard are related with eating mildew covered food thickeners by Nigerian individuals. It has not been conceivable to make compelling administration methodologies to stop contagious contamination and mycotoxin creation. Past work has shown that different microorganisms such as *Micrococcus*, *Bacillus*, *Klebsiella*, *Escherichia coli* and *Staphylococcus* are commonly linked with the spoilage of food thickening agents<sup>2</sup>. Food thickening agents are substances used by many homes in soup as it has a high gummy impairing ability. It is used because of its water holding

capacity without changing or altering the taste or flavour of the food. Over the years, pathogenic organisms have been isolated from ice cream samples from different market sellers. The presence of these microorganisms can be attributed to unclean water used during its processing, unsterilized equipments, unhealthy or poor hygienic environment and handling processes. These organisms have serious detrimental effect of the consumers of these thickening agents which could be Cholera, Dysentery, Typhoid, Stomach pain, Vomiting and Diarrhoea. However, it is necessary for the public to be informed about the microbiological qualities and health implications associated with consuming contaminated food thickening agents sold in markets around the University of Port Harcourt as a case study. This research study was carried out to detect the fungal and bacterial species lined or common with food thickeners in markets around University of Port Harcourt.

## Materials and methods

**Sample collection:** Commonly used food thickeners were collected from four (4) different open markets located around Uniport. From each market, five (5) samples (*Detariummicrocarpum* (Ofor), Cocoyam (colocasia and xanthosomaspp), *Mucunasloane* (Ukpo), *Brachystegiaeurycoma* (Achi), and Starch) were collected with sterile polyethylene bags and taken to the laboratory aseptically for analysis.

**Microbiological analysis of the samples:** 0.1ml of each tenfold dilution was transferred and spread on duplicate plate count agar using a fresh pipette for each dilution. The sample was spread as soon as possible on the surface of the plate with sterile glass spreader on plate count agar for total viable count, Mannitol salt agar for *Staphylococcus* count and Salmonella shigella agar for *Salmonella* and *Shigella* counts. The plates were incubated at 37°C for 24-48 hours. Potato dextrose agar for fungal count and the plates were incubated at 37°C for 3-5 days. Following incubation, plates exhibiting 30-300 colonies were counted. The average number of colonies in a particular dilution was multiplied by the dilution factor to obtain the total viable counts. The results of the total viable count were expressed as the number of organisms or colony forming units per gram.

**Isolation and purification of pure culture:** Distinct colonies that developed on the Plate count agar after incubation was sub cultured into freshly prepared Nutrient agar for bacteria isolates and Potato Dextrose agar for fungi isolates respectively. The plates were incubated at 37°C for 48 hours.

**Preparation of Stock Cultures:** Pure cultures were sub-cultured onto Nutrient agar slants and incubated at 37°C for 24-48 hrs; these served as stock cultures for further use in bijou bottles and were stored at 5-20°C.

**Calculation of colony forming unit per millilitre or gram:** At the end of incubation, the number of colonies was counted.

Average of duplicate plates were counted and recorded as the number colony forming units (cfu/g) of each thickening agent.

**Identification of Fungi: Microscopy:** Morphological characteristics such as shape, color, structure of mycelium and hyphae, arrangement and motility of spores were used in identifying isolates. A drop of lactophenol blue was used in staining the isolates and viewed under the microscope using  $\times 10$  and  $\times 40$  objectives.

## Results and discussion

Food thickening agents are linked to microbial contamination due to the way they are prepared and handled. They are often made in big batches by market sellers and may be kept in-use for inappropriately long periods or not under temperature control. This increases the likelihood of harmful bacteria proliferating to sufficient levels to cause food poisoning. In this study, the total bacteria count of the food thickener are as follows: Ofor ranged between  $9.7 \times 10^7$  cfu/g to  $1.05 \times 10^8$  cfu/g, Achi  $1.6 \times 10^8$  cfu/g to  $2.12 \times 10^8$  cfu/g, Starch  $5.2 \times 10^7$  cfu/g to  $1.08 \times 10^8$  cfu/g, Cocoyam  $4.0 \times 10^6$  cfu/g to  $1.6 \times 10^7$  cfu/g, Ukpo  $2.8 \times 10^7$  cfu/g to  $1.04 \times 10^8$  cfu/g, however, Achi had the highest count, which could be as a result of lack of sanitary practices during the preparation of this food thickening agent. The total *Staphylococcal* count of Ofor ranged between  $1.0 \times 10^6$  cfu/g to  $1.08 \times 10^6$  cfu/g, Achi  $1.5 \times 10^5$  cfu/g to  $2.5 \times 10^5$  cfu/g, Starch  $2.6 \times 10^5$  cfu/g to  $3.6 \times 10^5$  cfu/g, Cocoyam  $3.0 \times 10^5$  cfu/g to  $4.1 \times 10^5$  cfu/g, Ukpo  $1.5 \times 10^5$  cfu/g to  $2.3 \times 10^5$  cfu/g; and the *Salmonella Shigella* Count obtained in Ukpo ranged from  $3.0 \times 10^4$  cfu/g to  $5.0 \times 10^4$  cfu/g, Achi  $4.0 \times 10^4$  cfu/g, Ofor  $6.5 \times 10^5$  cfu/g, and no visible growth observed for Cocoyam and Starch, but *Shigella* was present more in Ukpo. Microorganisms isolated were *Shigella* (5%), *Escherichia coli* (20%), *Staphylococcus sp* (29%), *Proteus* (10%), *Klebsiella* (18%) and *Bacillus* species (18%). Analysis of microorganisms in the sample showed that *Staphylococcus sp.* had the highest frequency of occurrence. This is in contrast to the result obtained by Adenike *et al.*, who observed more growth of *Escherichia coli* in food condiments. The predominance of *Staphylococcus* could be as a result of its presence in the nose and is usually found, hands, skin, and clothing of handlers. According to Jones *et al.*<sup>7</sup>, the presence of *Staphylococcus* in food could be as a result of its existence in air, water, food equipments, and environmental surfaces not necessarily failure to thoroughly clean after each use or neglect to sanitize equipment before use. *Staphylococcus spp.* has been known to cause food poisoning due to consumption of improperly cooked food in which the organism has infected. According to Ananthanarayana *et al.*<sup>8</sup>, as little as  $1.0 \mu\text{g}$  of *Staphylococcal* toxin in food produces symptoms of illness. The occurrence of this organism in food thickeners implies that it can be a source of serious illness like cholera, dysentery, stomach pain, vomiting, diarrhoea etc and their presence could be as a result of unclean water used during processing, unsterilized and improper sterilized equipment, unhealthy production process, poor

hygienic practices and mishandling of food thickening agents before use. According to ICMSF<sup>9</sup> the microbial limit for total bacterial count ranges from  $<10^3$  as its satisfactory range,  $10^3$ - $<10^5$  as its Borderline and  $\geq 10^5$  as the unsatisfactory range, comparing the total bacterial count from this study which ranges from  $10^6$  to  $10^8$  with the above standard, it exceeded the permissible limit. According to Oranusi *et al.*, contamination by microorganism of a product is rely on the environment it passed through and the sanitary quality of the cultivation water, harvesting, transportation, storage, and processing of the produce. The isolation of *Escherichia coli* from thickening agents is in agreement with the findings of Adenike *et al.*<sup>10</sup> who worked on the microbial load of food condiments and isolated more of *Escherichia coli*. The presence of coliforms (*Klebsiella* and *Escherichia coli*) is indicative of contamination and hence the sanitary condition of the sample is questionable. Primary sources of these coliforms are water and soil<sup>11</sup>. *Escherichiacoli* are a commonly used faecal indicator organism. Its occurrence in food is linked to direct or indirect faecal contamination. Substantial number of *E. coli* in food clearly indicates a general lack of cleanliness in handling and improper storage<sup>12</sup>.

Fungal counts for Ofor ranged from  $4.0 \times 10^6$  cfu/g to  $1.04 \times 10^8$  cfu/g, Cocoyam  $3.0 \times 10^6$  cfu/g to  $1.1 \times 10^7$  cfu/g, Ukpo  $2.0 \times 10^6$  cfu/g to  $1.6 \times 10^7$  cfu/g, Starch  $9.0 \times 10^6$  cfu/g to  $1.4 \times 10^7$  cfu/g, Achi  $7.0 \times 10^6$  cfu/g to  $5.4 \times 10^7$  cfu/g. This result is low when compared with the results of Oranusi *et al.*<sup>13</sup> who reported that Achi had a fungal count of  $8.2 \times 10^9$  cfu/g and Ofor  $9.3 \times 10^8$  cfu/g. Some of the fungi isolated during this research were *Aspergillus* and *Penicillin*; which is similar to the findings obtained by Oranusi *et al.*<sup>13</sup>. According to Beatriz and Eliana<sup>4</sup>, almost all the food items can be contaminated by fungal

organisms and many of the food borne fungi capable of generating one or more mycotoxins, particularly aflatoxins. The high fungal counts of  $\geq 10^6$  recorded for the samples are above standard specification<sup>9</sup>, hence the products are therefore not recommended for consumption without further treatments. The result of the proximate analysis carried out are as follows: Starch had moisture content of 23.59%, carbohydrate 72.92%, Ash 0.10%, Lipid 0.90%, crude fibre 0.96%, Protein 1.53%, Achi had moisture 8.60%, carbohydrate 41.60%, Ash 2.70%, Lipid 17.30%, Protein 12.25%, Ukpo: moisture 10.40%, carbohydrate 51.04%, Ash 3.30%, lipid 9.30%, crude fibre 7.15%, protein 18.81%, Cocoyam: moisture 33.10%, carbohydrate 62.50%, ash 1.90%, lipid 0.40%, crude fibre 2.10%, and protein 1.75% respectively. However, the result obtained from this research work is similar to the findings obtained by Nwosu<sup>14</sup> and contrary to the findings of Donatus *et al.*<sup>15</sup> who had the value of lipids in Mucuna as  $8.4 \pm 0.20$  and carbohydrate in Mucuna as  $71.74 \pm 0.10$  higher than the result obtained in my research work. From the result above food thickening agents have been noticed to be a good source of protein especially Ukpo, Achi and Ofor. Although these thickening agents are good for consumption and have high nutritional contents, they could also be a source of food intoxication thereby causing harm to its consumers. The process of handling and grinding increase the risk of contamination, and also the powdered seeds are not dried after processing to reduce water activity, this available water increases survival and growth of fungal and bacteria contaminants and aflatoxins formation.

**Proximate Analysis:** The samples were analyzed for proximate compositions using the Official methods of AOAC<sup>16,17</sup>.

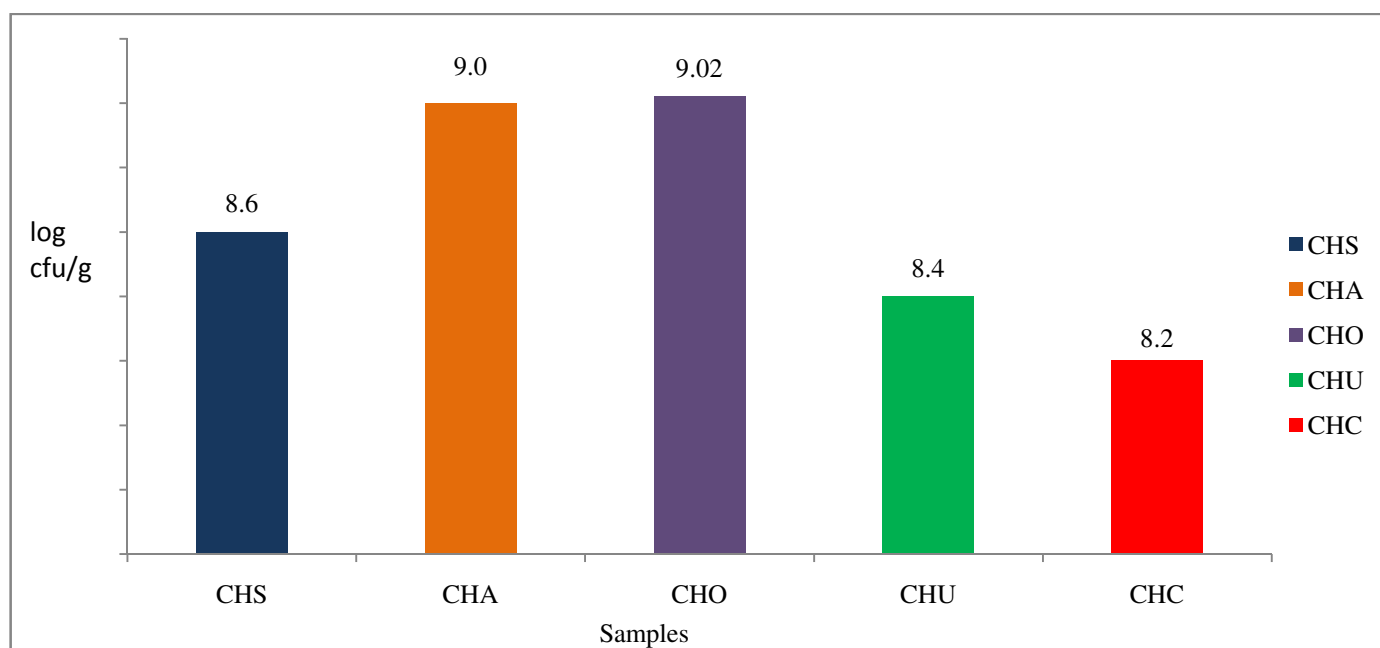
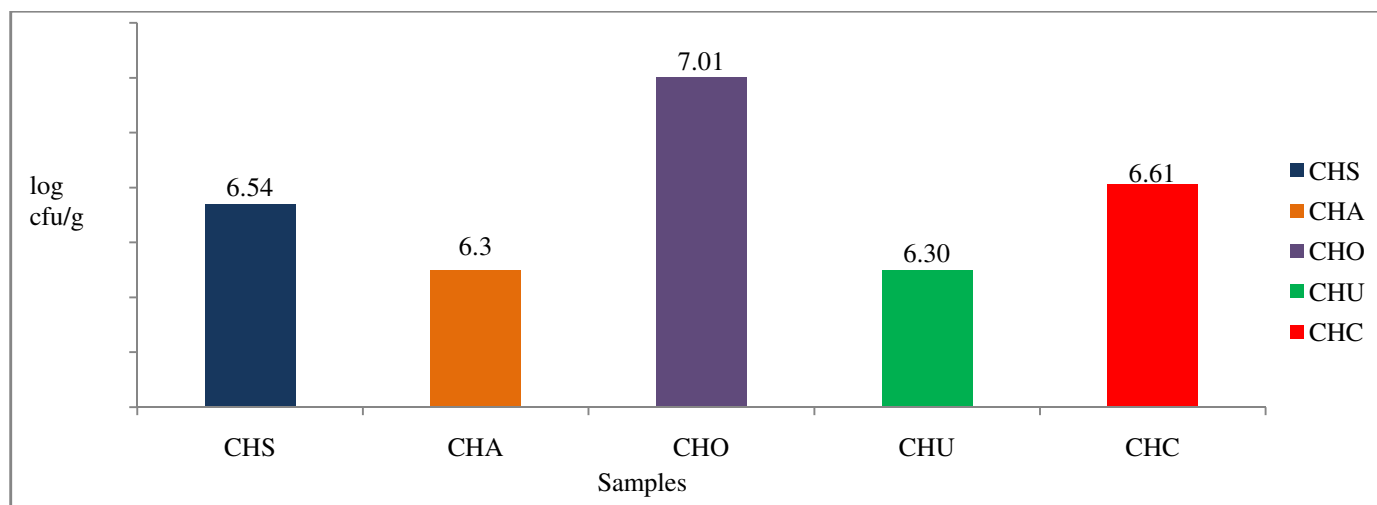
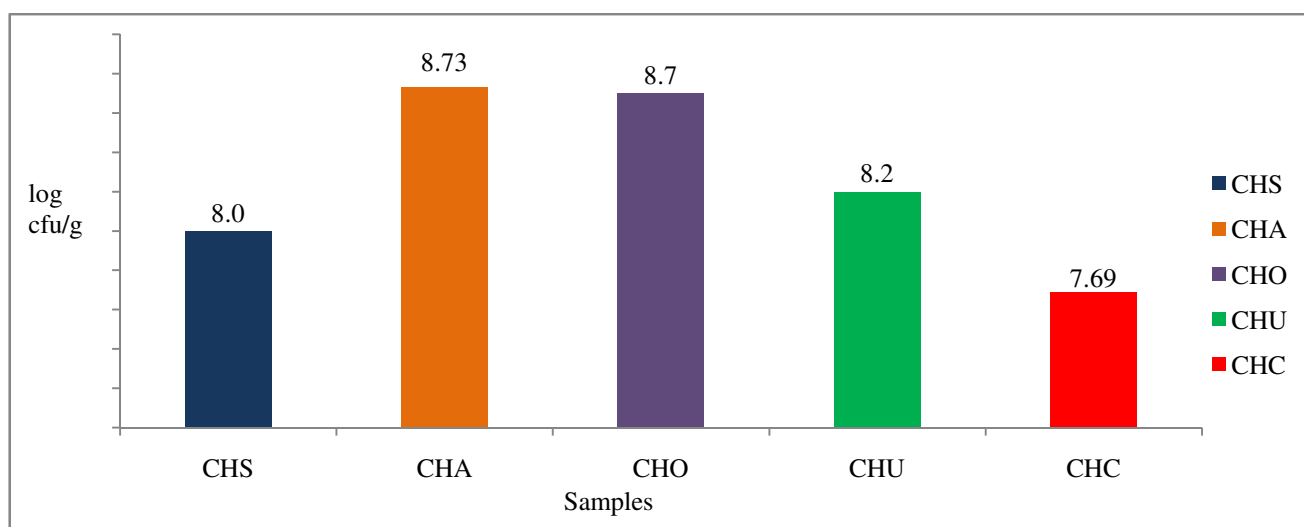


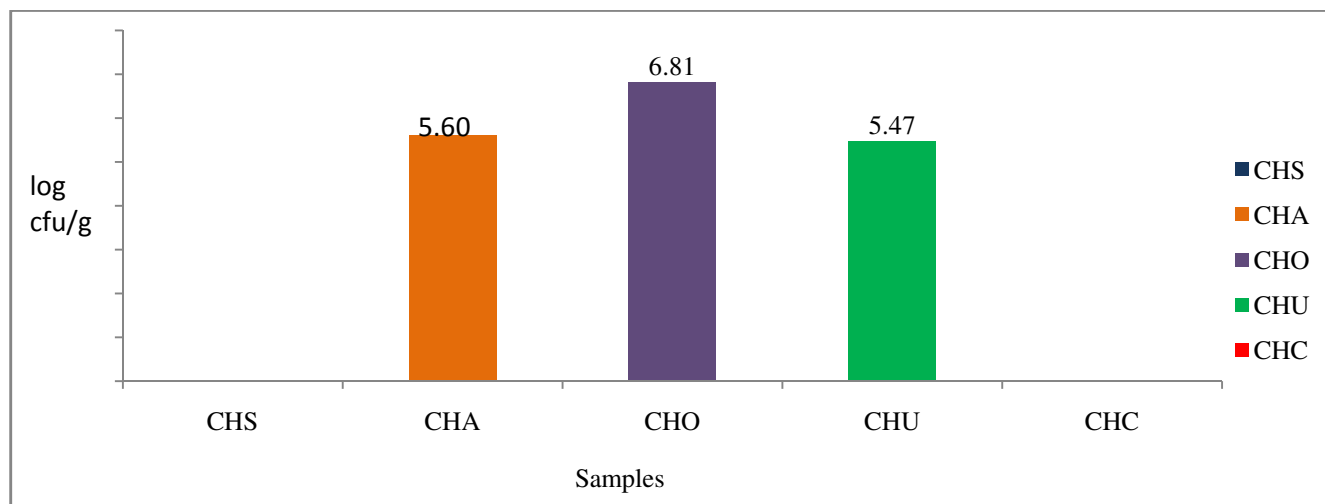
Figure-1: Total Viable Count from Choba Market Samples.



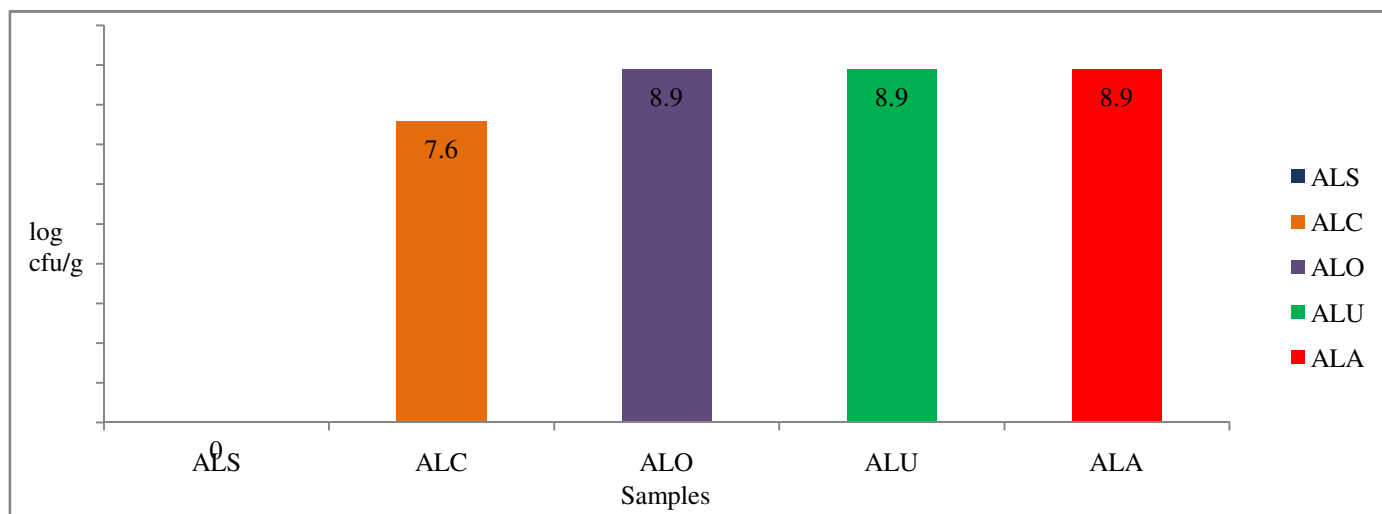
**Figure-2:** Staphylococcal count from Choba Market Samples (Key: CHS-Choba Starch, CHA-Choba Achi, CHO-ChobaOfor, CHU-ChobaUkpo, CHC-Choba Cocoyam).



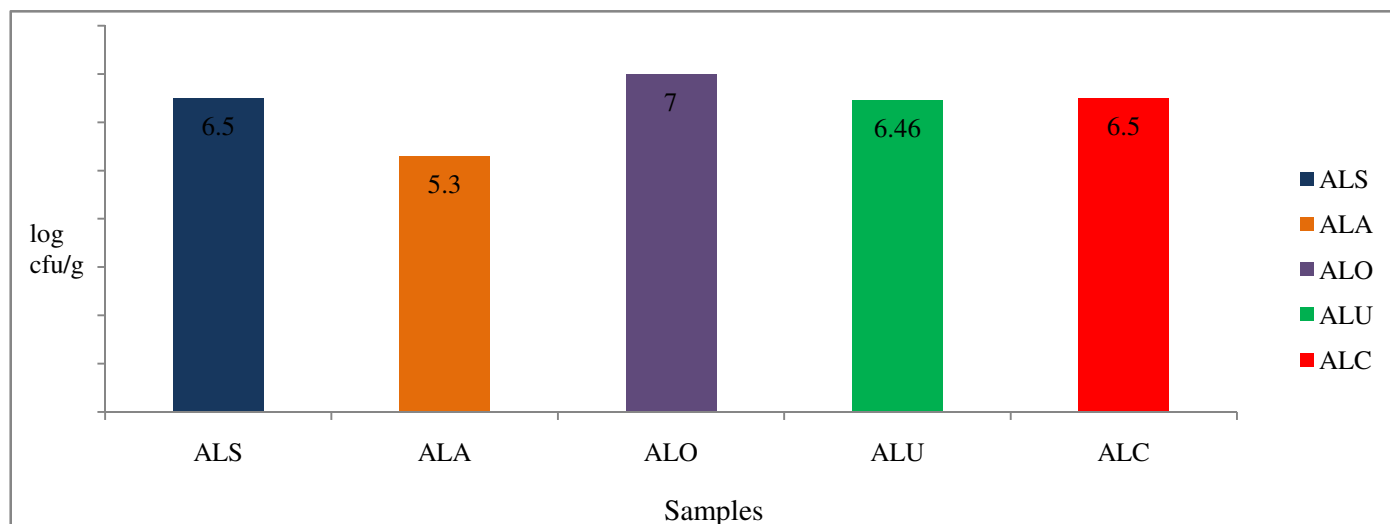
**Figure-3:** Fungal count from Choba Market Sample.



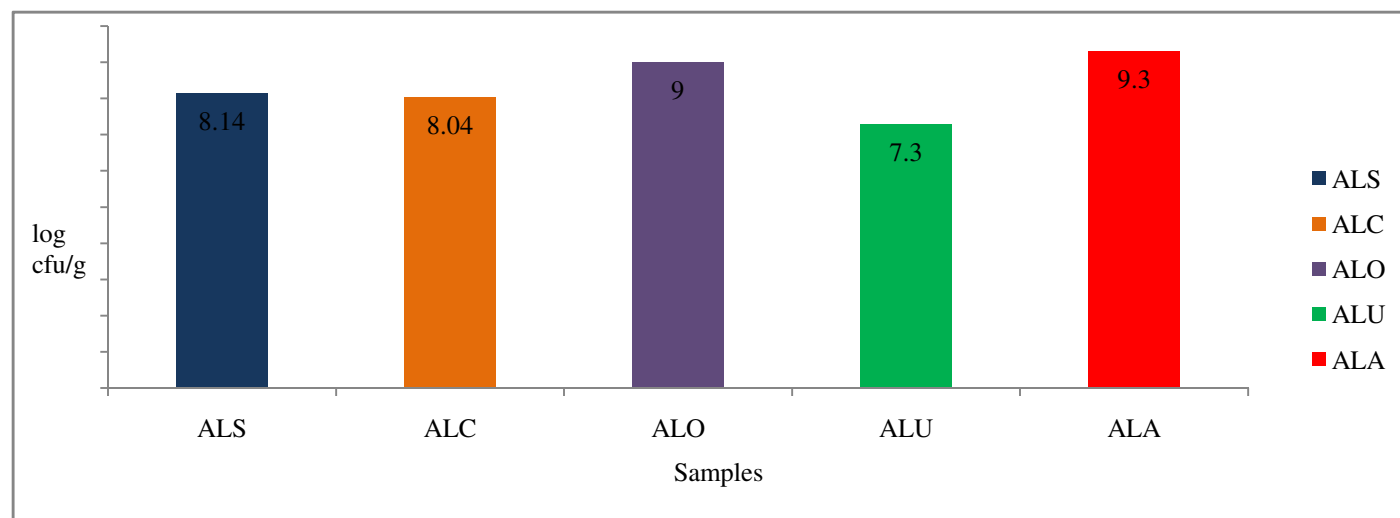
**Figure-4:** *Shigella* count from Choba Market Sample.



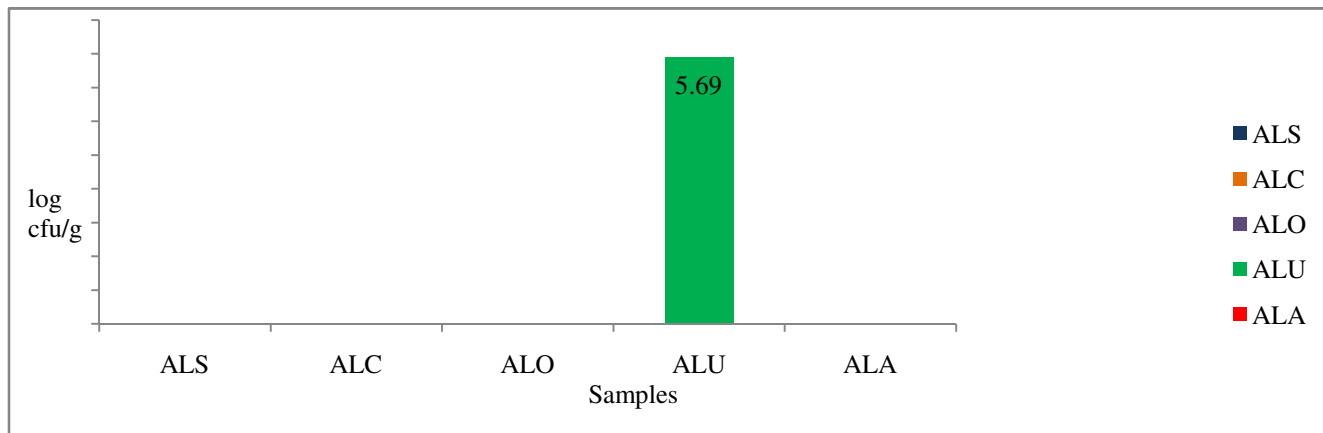
**Figure-5:** Total Viable Count from Alakahia Market Samples.



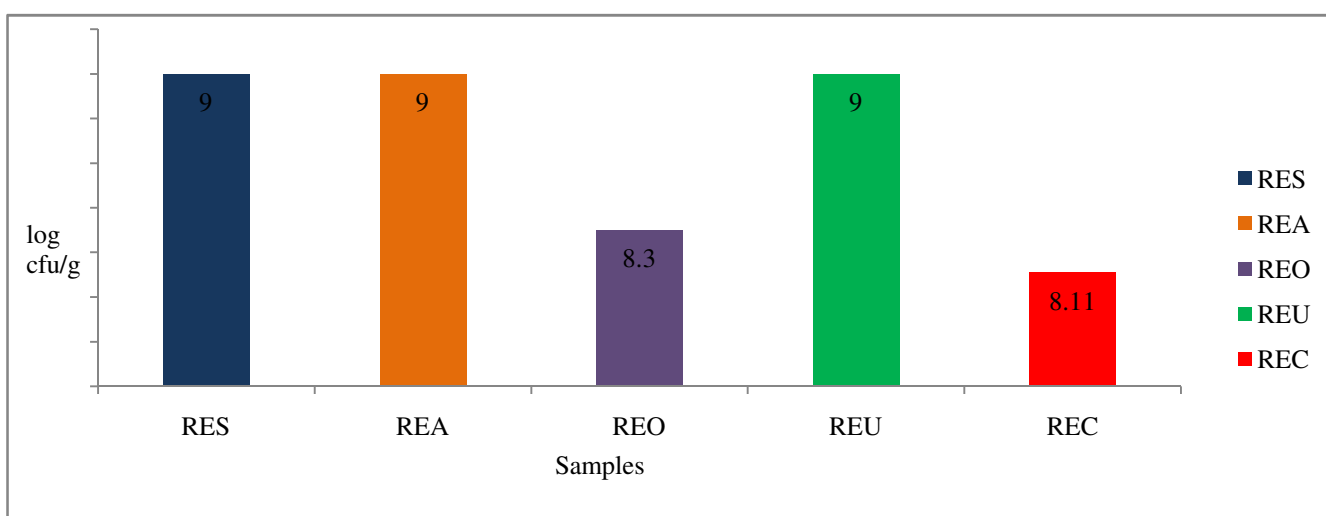
**Figure-6:** Staphylococcal count from Alakahia Market Samples.



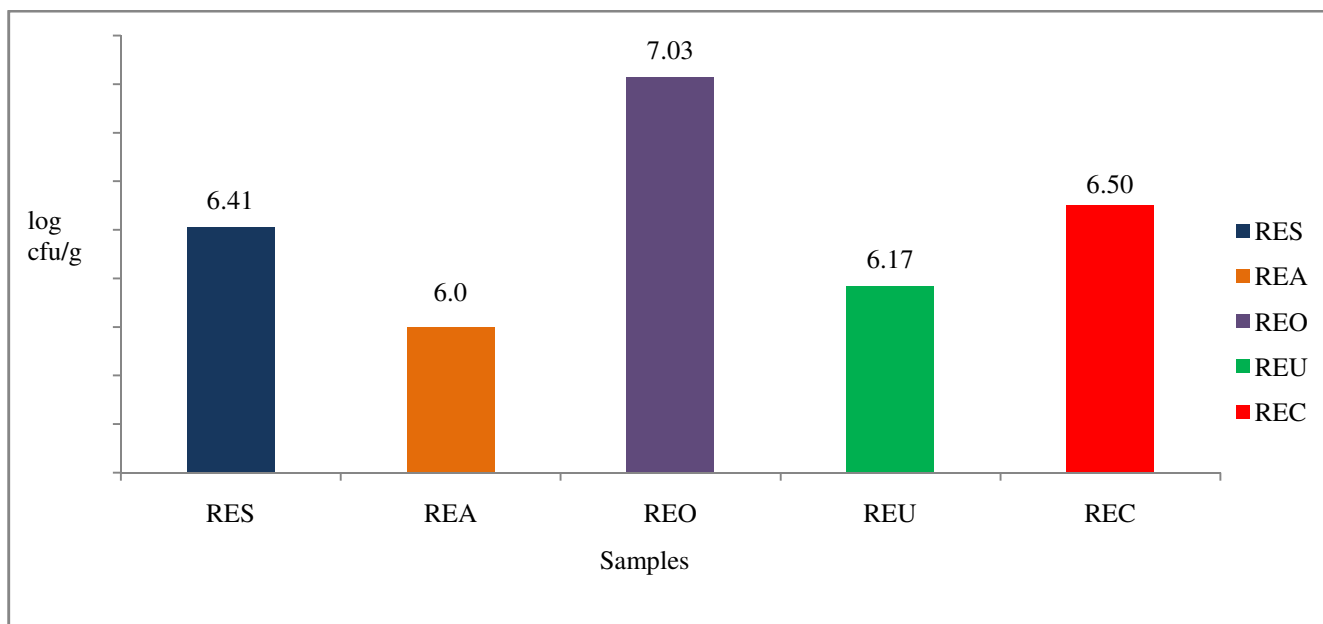
**Figure-7:** Fungal count from Alakahia Market Samples.



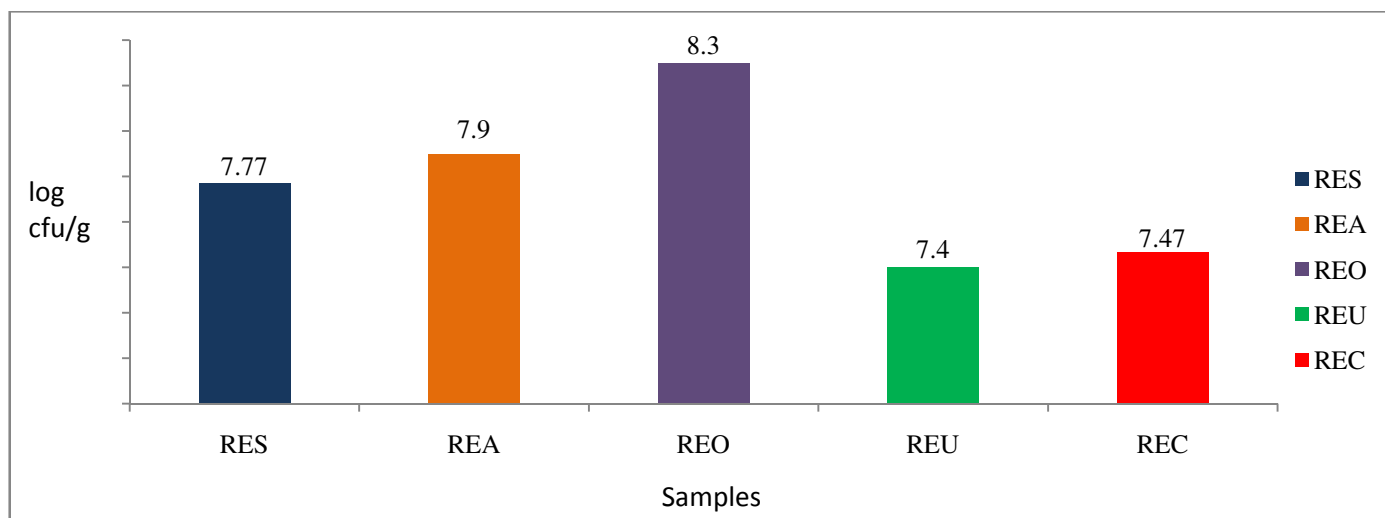
**Figure-8:** Shigella count from Alakahia Market Sample.



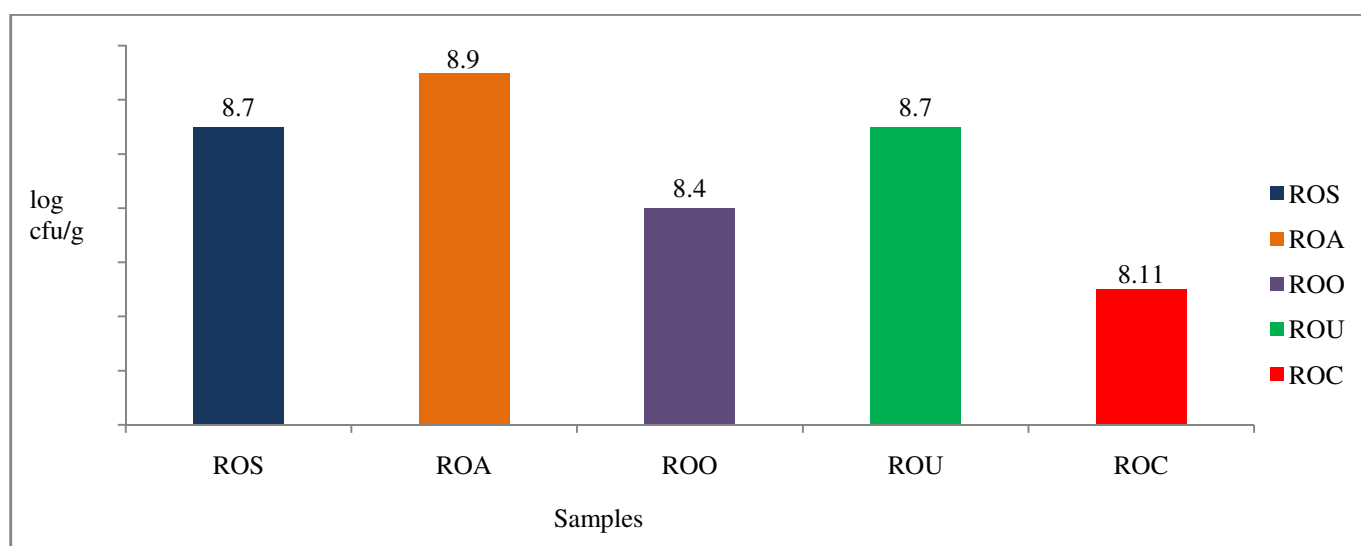
**Figure-9:** Total Viable Count from Rumuekini Market Samples.



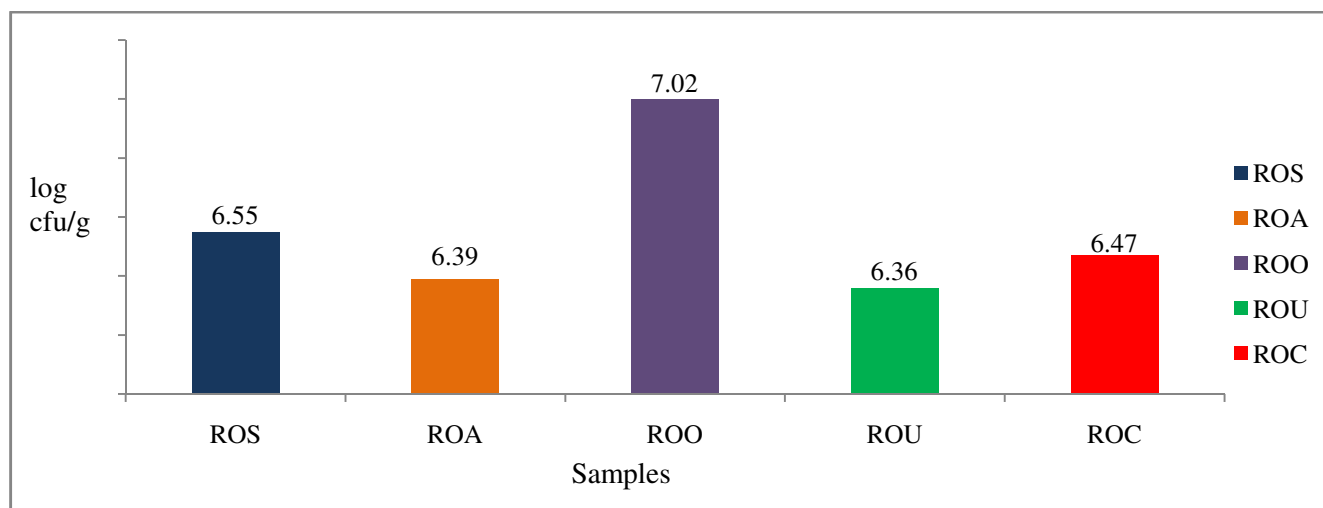
**Figure-10:** Staphylococcal count from Rumuekini Market Samples.



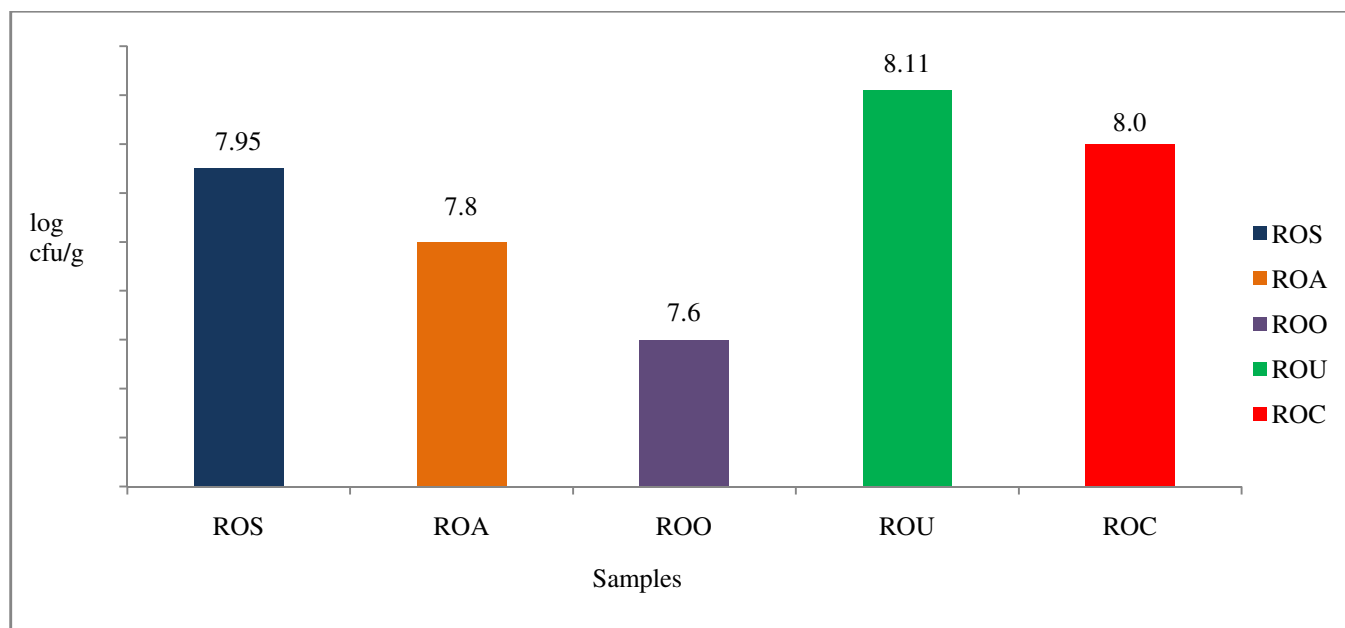
**Figure-11:** Fungal Count from Rumuekini Market Samples.



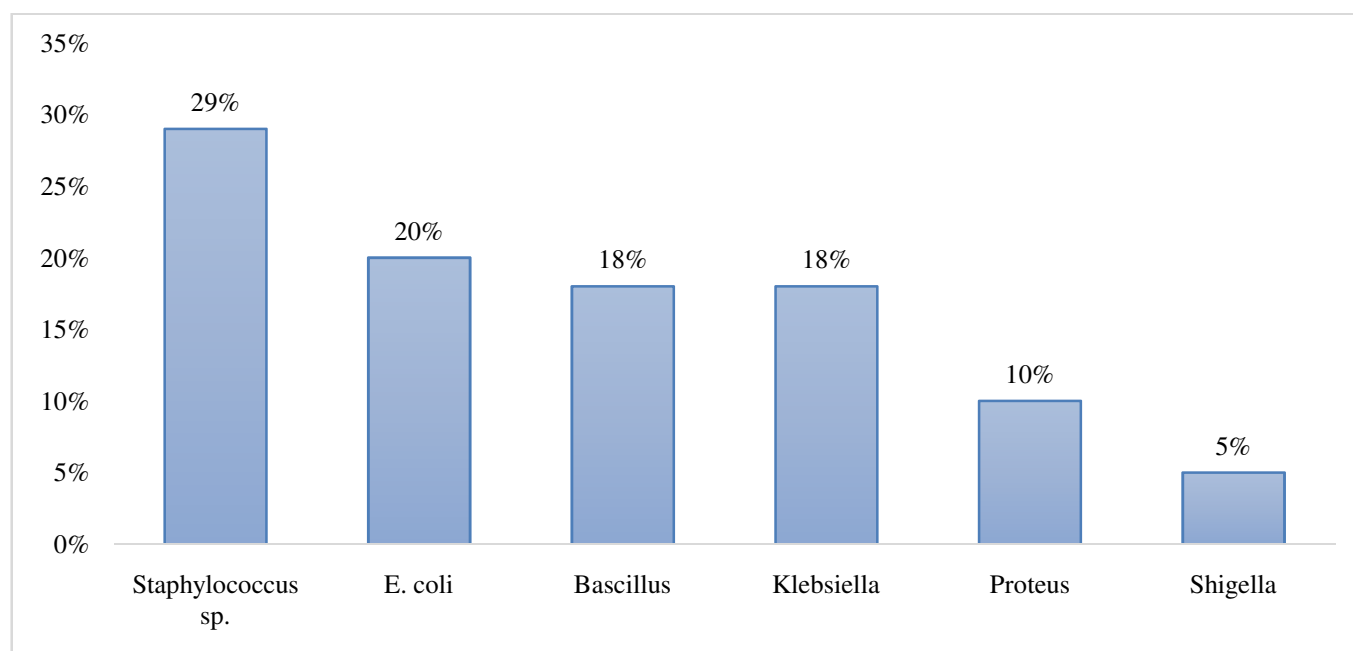
**Figure-12:** Total Viable Count from Rumuosi Market Samples.



**Figure-13:** Staphylococcal count from Rumuosi Market Samples.



**Figure-14:** Fungal count from Rumuosi Market Samples.



**Figure-15:** Percentage occurrence of the different organisms from the food thickening agen.

**Table-1:** Proximate composition (%) of the different food thickener.

Sample identity	Moistue	CHO	Ash	Lipid	fibre	Protei
Starch	23.59	72.92	0.10	0.90	0.96	1.53
Achi	8.60	41.60	2.70	17.30	13.99	15.73
Ofor	14.67	46.88	1.10	19.50	5.60	12.25
Ukpo	10.40	51.04	3.30	9.30	7.15	18.81
Cocoyam	33.10	62.50	1.90	0.40	2.10	1.75



## Conclusion

Food thickening agents are widely used by many individuals because of its water holding ability and nutritional benefits. However, it can be a source of food intoxication if not properly prepared or cooked. The isolation of *Staphylococcus*, *Escherichia coli* and *Shigella* spp. suggests that thickening agents could be a source of infection to human by the enteric organisms such as *Klebsiella* spp., *Shigella* spp., and *Proteus* spp. In conclusion adequate sanitary practices must be adopted during the processing of food thickening agents.

**Recommendation:** Thickening agents are mostly contaminated during its processing. However, thickening agents should be bought in their seed form and prepared at home to avoid contamination, awareness and sanitary practices should be taught to market traders and finally food should be cooked properly before consumption.

## References

1. Okwu G.I., Achar P.N. and Sharma S.K. (2010). Quantification of aflatoxin B1 in ready-to-use food thickeners in South-east geo-political zone in Nigeria. *African Journal of Microbiology Research*, 4(16), 1788-1793.
2. Uhuegbu F.O., Onwuchekwa C.C., Iweala E.E. and Kanu I. (2009). Effect of processing methods on nutritive and antinutritive properties of seeds of *Brachystegia eurycoma* and *Detarium microcarpum* from Nigeria. *Pakistan Journal of nutrition*, 8(4), 316-320.
3. Lund B.M. and Parker T.C. and Gould G.W. (2000). The microbiological safety and quality of food. Toxigenic fungi and mycotoxin Aspen Inc Publishers, 1490-1517.
4. Pinho B.H. and Furlong E.B. (2000). The occurrence of molds, yeasts and mycotoxins in pre-cooked pizza dough sold in Southern Rio Grande do Sul. *Brazilian Journal of Microbiology*, 31(2), 99-102.
5. Turner P.C., Moore S.E., Hall A.J., Prentice A.M. and Wild C.P. (2003). Modification of immune function through exposure to dietary aflatoxin in Gambian children. *Environmental health perspectives*, 111(2), 217-220.
6. Molina M. and Giannuzzi L. (2002). Modelling of aflatoxin production by *Aspergillus parasiticus* in a solid medium at different temperatures, pH and propionic acid concentrations. *Food Research International*, 35(6), 585-594.
7. Jones T.F., Kellum M.E., Porter S.S., Bell M. and Schaffner W. (2002). an outbreak of community acquired food borne illness caused by methicillin-resistant *Staphylococcus aureus*. *Emerging infectious disease*, 8(1), 82-84.
8. Anantharayan (2011). Diagnostic value of mannitol for sugar fermentation in *Staphylococcus aureus*. *Textbook of Microbiology*, 31-33.
9. ICMSF (2009). International Committee on Microbiological Specifications for Foods' Microorganisms in food 1, Their Significance and Methods of Enumeration. 2nd Ed., University of Toronto Press, Toronto, Buffalo and London.
10. Ogunshie A.A. and Olasugba K.O. (2008). Microbial loads and incidence of food-borne indicator bacteria in most popular indigenous fermented food condiments from middle-belt and southwestern Nigeria. *African Journal of Microbiology Research*, 2(12), 332-339.
11. Jay J.M. (2000). Modern Food Microbiology. Van Nostrand Reinhold, New York, 3<sup>rd</sup> edition 642.
12. Codex (2011). Code of Hygienic Practice for collecting, processing and marketing of Natural mineral waters. (CAR/RCP33-1985 REVISED 2011) 110-112
13. Oranusi S.U., Braide W., Nwodo C.F. and Nwosu U.P. (2013). Assay for aflatoxins in some local food condiments. *International Journal of Biology, Pharmacy and Allied Sciences (IJBPAS)*, 2(3), 529-537.
14. Nwosu J. (2011). The Effect of Storage Condition on the Rheological/Functional Properties of Soup Thickener *Mucuna sloanei* (Ukpo). *Researcher*, 3(6), 27-32.
15. Donatus E.O. and Ezinna Okoro (2007). Phytochemical composition of *Brachystegia eurycoma* and *Mucuna flagellipes* seed. *Medicinal and Aromatic plant Science and Biotechnology*, 1(1), 103-106.
16. AOAC (1980). Official methods of Analysis. 13th edition Association of Official Analytical Chemists, Washington, D.C.
17. Igwenyi I.O. and Azoro B.N. (2014). Proximate and phytochemical compositions of four indigenous seeds used as soup thickeners in Ebonyi state Nigeria. *J. Environ. Sci. Toxicol. Food Technol*, 8(6), 35-40.