

The Assessment of Bacterial and Protozoan Contamination of Local Soup Condiment (*Daddawan Batso*) Sold in Sakaba Local Government Area, Kebbi State, North-Western Nigeria

Daniel Dan-Inna Attah^{1*}, Yusuf Muhammad Sanyinna², Regina Doro Jabaka³

^{1,2}Department of Animal and Environmental Biology, Faculty of Life Sciences, Kebbi State University of Science and Technology, Aliero, Nigeria

³Department of Microbiology, Faculty of Life Sciences, Kebbi State University of Science and Technology, Aliero, Nigeria

*Corresponding Author: danielattah10@gmail.com, Tel.: +234-80383-50460

Available online at: www.isroset.org

Received: 10/Oct/2021, Accepted: 20/Nov/2021, Online: 31/Dec/2021

Abstract—*Daddawan Batso* is a condiment produced by a traditional uncontrolled alkaline fermentation of the seeds of *Hibiscus sabdariffa*. They are abundantly produced in Nigeria especially in the North-West. The study was carried out to assess the bacterial and protozoan contamination of local local soup condiment (*Daddawan Batso*) sold in Sakaba Local Government Area, Kebbi State, North-Western, Nigeria. Samples were purchased and collected at random from the markets of Laraba, Makuku, Janbirni, Dankolo, Doka and Dirin-Daji towns. Clean hand gloves were used aseptically and sterilized polythene bags were labelled correctly according to the markets. The samples were then transported to the Laboratory for analysis. About 0.1 ml of the samples was inoculated on the nutrient media and incubated at 37°C for 24 hours. Gram staining technique was also employed and the samples observed under microscope using ×100 objective lenses. For the identification of protozoa, 1 g of the sample was soaked in 9 mls of distilled water for 7 - 10 minutes, centrifuged and observed using ×10 and ×40 objective lenses. The bacteria present were counted in colony forming unit per gram per sample. A total of six bacterial genera were isolated (*Proteus* sp., *Escherichia* sp., *Klebsiella* sp., *Micrococcus* sp., *Staphylococcus* sp. and *Bacillus* sp.). Further characterization revealed the organisms to be *Proteus mirabilis*, *Escherichia coli*, *Klebsiella aerogenes*, *Micrococcus roseus*, *Staphylococcus epidermidis*, *Bacillus licheniformis* and *Bacillus subtilis* respectively. Again, the morphological features of the isolates revealed that some are gram +ve (*Micrococcus roseus*, *Staphylococcus epidermidis* and *Bacillus* spp.) while others are gram -ve (*Proteus mirabilis*, *Escherichia coli* and *Klebsiella aerogenes*). A total of eight protozoa was isolated and all were *Entamoeba* spp. The overall prevalence of the protozoa was 44.4%. *Bacillus* spp. and *Entamoeba* spp. were observed to be the key spoilage organisms of the condiments. Sellers should ensure that they do not expose the fermented foods during display for sale.

Keywords—Assessment, Microbes, Contamination, Local Soup Condiment, Sakaba Local Government Area.

I. INTRODUCTION

Daddawan Batso is a condiment produced by a traditional uncontrolled alkaline fermentation of the seeds of *Hibiscus sabdariffa* (Roselle). It adds aroma and flavour to dishes such as baobab soup (*Miyar Kuka*), fish pepper soup and chicken pepper soup [1].

Soup condiments are edible food items which are added to dishes, used as thickeners for soup and also as food supplements such as sauce that is added to food to impact specific flavours [2]. In some cultures, they are used to compliment some dishes. The terms originally describe the condiments as preserved foods, but their meaning has changed over time [3]. They are abundantly produced in Nigeria especially in North-Western part of the country [4].

Food condiments or spices are strong smelling, sharp tasting substances usually used to improve or adjust the

flavour of food. They are usually of vegetable origin. Common examples include mustard, nutmeg, ginger, garlic, coriander, locust bean etc. [5].

Food condiments made from vegetable protein may be a good source of certain vitamin B, but are deficient in ascorbate and some fat-soluble vitamins, which are lost during fermentation. It is evident that fermented condiments are good source of nutrients and could be used to produce complementary food supplements and contribute to enhance food quality [6].

Condiments add flavour, make the food tasty and palatable. They also serve as appetizers in food [7]. Condiments possess essential stimulating qualities, rendering them peculiarly fitted for inducing reflex action, the secretion of alimentary juice. As the function of condiment, such as pepper, mustard, spices, potherbs, that besides their stimulating properties, they give flavour to

food and then indifferently food is made palatable, and digestion accelerated. Letheby enumerated it as the aid of digestion – proper selection of food, according to the taste of the individual [3].

Condiments were known in ancient Rome, ancient India and ancient China, where there was myth that, before food preservation techniques became widespread, pungent spices and condiments were sold as wet paste, used to give the very strong traditional taste, making the food palatable [8].

There are different types of condiments which include pickle foods, which date back to the ancient world in Europe as well as Asia. Common pickle foods used as condiments today include ginger used by Japanese, chutney in Asia and cucumbers. Almost all the vegetables have been pickled and used as condiments in some forms. They are served whole, in slices or diced in relish [8].

There are spicy and hot condiments which are black pepper, chilli pepper, mustard garlic and onions [9]. Black pepper is a product from Asia which is commonly used from shakers throughout the western world in a dried state and is usually ground and coarse or fine state before consumption. Like salt, pepper is used in dishes and also as based ingredient in some pepper sauce. Chilli pepper is a product of the Americas employed in dried form on diverse foods [10].

Fresh chopped chilli pepper is also the based ingredient in a number of other based condiments, including salsa. In Pre-Columbian times, a number of sauces based on chilli pepper were employed along with tomatoes and grounded pumpkin seeds. After the Spanish conquest in 1521, fusion foods developed including salsas, which combined salt with ground tomatoes and chilli peppers from the new world and vinegar from the old world [4].

II. RELATED WORK

It is generally known that illnesses due to the consumption of contaminated foods are the most common health problems in the contemporary world today, and a significant cause of reduced economic productivity [4]. Recent studies examining the morbidity and mortality of food-borne diseases have confirmed the significant public health burden posed by these diseases [4].

Microbial food safety is an increasing public health concern worldwide [15]. In developed countries, it is estimated that one-quarter to one-third of the population are made sick each year because of food-borne diseases. In the developing countries, including Nigeria, the burden is much more severe; and therefore, there is need to regularly survey the food preparation, handling and storage techniques to ensure the safety of the food on one hand, and the health of the consumers on the other hand [4].

Most previous studies were mainly on investigating the nutrient composition of fermented condiments [2], [6], [16], [17], [18], [19].

However, Sanyinna *et al.* conducted a recent study on “Evaluation of Bacterial and Protozoan Contamination of Local Soup Condiment (Daddawa) Produced by North-Western Community, Nigeria” [4].

III. METHODOLOGY

Study Area/Location

The study was conducted in Sakaba Local Government Area (LGA) of Kebbi State, North-Western Nigeria. Sakaba Local Government Area is located in the Zuru Emirate Council; one of the four (4) Emirates in the State. Sakaba has an area of 1,260 km² and a population of 91,728 people based on 2006 national population census [11]. The area council of Sakaba covers communities of Dokan-Kambari, Dirin-Daji, Dirin-Gari, Dokan-Hausawa, Doka, Doka-Bere, Maza-Maza, Gelwasa, Janbirni and Jandutse [11].

The main occupation of the people in the study area especially in the rural areas is agriculture; farming and rearing of animals, which become their source of income.

Sample Collection

The local soup condiments (*Daddawan Batso*) were purchased and collected at random from the markets of different places in Sakaba Local Government Area. Six (6) major villages in the LGA were randomly selected which include: Laraba, Makuku, Janbirni, Dankolo, Doka and Dirin-Daji.

The condiments were purchased from different sellers at different selling spots in the various markets selected during the market days. Clean hand gloves were used to place the samples of the condiment aseptically into sterilized polythene bags and were labelled correctly according to the markets. The samples were then transported to Microbiology Laboratory, Kebbi State University of Science and Technology, Aliero for bacterial and protozoan isolation and identification [12].

Sample Processing

Bacterial Isolation

The local soup condiment was cut into pieces then placed in a conical flask containing distilled water. About six test tubes were arranged accordingly on a test tube rack, each containing 9 ml of distilled water. 1 ml from the sample was transferred into the first test tube (10¹) and shaken. Again, 1 ml was transferred into the second test tube (10²). This was repeated up to the last test tube (10⁶). The fifth test tube (10⁵) was selected for inoculation.

Inoculation of Bacteria

About 0.1 ml of the samples was inoculated on the prepared, solidified and sterile nutrient media using streak plate method. The inoculated petri-dishes were incubated at 37°C for 24 hours. The growth was observed after 24 hours incubation. The colony was counted using colony counting machine.

Smear Preparation

A loop full of colony was placed on a clean grease-free microscope slide. A drop of distilled water was added and mixed until a homogenous mixture was obtained; it was heat fixed. The smear was allowed to air dry and then placed on a staining rack.

Gram Staining Technique

The smear was flooded with crystal violet for 60 seconds and washed with water. It was then flooded with Lugol's iodine for 60 seconds and washed with water. The smear was decolorized with acid-alcohol drop wise and washed with water. It was counter stained with Safranin for 60 seconds and washed with water. The smear was allowed to air dry and a drop of immersion oil was placed on it. The smear was examined under the microscope using $\times 100$ objective lenses [13].

Isolation and Identification of Protozoa

One gram (1 g) of local soup condiment was soaked in 9 ml of distilled water for 7 - 10 minutes. The suspension was transferred into another clean test tube removing large debris of the condiment. It was then centrifuged at 2500 rpm for 5 minutes. The supernatant was discarded and sediments of each sample were placed on a clean grease-free glass slide and examined microscopically. 0.9% saline smear for parasite cysts and trophozoites using $\times 10$ and $\times 40$ objective lenses was prepared. Samples were stained with Lugol's iodine and then examined under the microscope [14].

Statistical Analysis

Data were analysed using Statistical Package for the Social Sciences (SPSS) version 20.0 and results were presented as mean \pm standard error of mean of triplicate results. Frequency and percentage were used to calculate the occurrence and total number of prevalence rates of the identified protozoa.

IV. RESULTS AND DISCUSSION

A total of six (6) bacterial genera were isolated from various samples collected from the six (6) different markets in the study area namely: *Proteus* sp., *Escherichia* sp., *Klebsiella* sp., *Micrococcus* sp., *Staphylococcus* sp. and *Bacillus* sp. (Table 1).

Table 1: Isolation of Bacteria Associated with Contamination of Local Soup Condiment in the Study Area

S/N	Bacteria (Genus)
1.	<i>Proteus</i> sp.
2.	<i>Escherichia</i> sp.
3.	<i>Klebsiella</i> sp.
4.	<i>Micrococcus</i> sp.
5.	<i>Staphylococcus</i> sp.
6.	<i>Bacillus</i> sp.

Key: S/N = Serial number; sp. = species (singular)

The bacteria associated with contamination of local soup condiment in the study area were counted in colony forming unit per gram per sample as illustrated below

(Table 2). Dirin-Daji: Sample (1) 4.0×10^3 , Sample (2) 3.8×10^4 , Sample (3) 3.2×10^5 ; Doka: Sample (1) 4.4×10^4 , Sample (2) 4.0×10^5 , Sample (3) 3.6×10^6 ; Laraba: Sample (1) 3.2×10^4 , Sample (2) 4.8×10^5 , Sample (3) 2.0×10^6 ; Makuku: Sample (1) 4.4×10^3 , Sample (2) 3.3×10^4 , Sample (3) 2.8×10^5 ; Janbirni: Sample (1) 4.0×10^3 , Sample (2) 3.6×10^4 , Sample (3) 2.4×10^5 and Dankolo: Sample (1) 3.6×10^4 , Sample (2) 3.8×10^5 , Sample (3) 2.4×10^6 respectively.

Table 2: Bacterial Count of the Bacteria Associated with Contamination of Local Soup Condiment in the Study Area

Town (cfu/g)	Sample	Bacterial Count
Dirin-Daji	Sample 1	4.0×10^3
	Sample 2	3.8×10^4
	Sample 3	3.2×10^5
Doka	Sample 1	4.4×10^4
	Sample 2	4.0×10^5
	Sample 3	3.6×10^6
Laraba	Sample 1	3.2×10^4
	Sample 2	4.8×10^5
	Sample 3	2.0×10^6
Makuku	Sample 1	4.4×10^3
	Sample 2	3.3×10^4
	Sample 3	2.8×10^5
Janbirni	Sample 1	4.0×10^3
	Sample 2	3.6×10^4
	Sample 3	2.4×10^5
Dankolo	Sample 1	3.6×10^4
	Sample 2	3.8×10^5
	Sample 3	2.4×10^6

Key: cfu = colony forming unit; g = gram

Further characterization of the bacterial isolates revealed the organisms to be *Proteus mirabilis*, *Escherichia coli*, *Klebsiella aerogenes*, *Micrococcus roseus*, *Staphylococcus epidermidis*, *Bacillus licheniformis* and *Bacillus subtilis* respectively (Table 3).

Again, the morphological features of the isolates revealed that some are gram +ve such as *Micrococcus roseus*, *Staphylococcus epidermidis* and *Bacillus* spp. while others are gram -ve such as *Proteus mirabilis*, *Escherichia coli* and *Klebsiella aerogenes* as illustrated in Table 3 below.

Table 3: Morphological Features and Characterization of Bacterial Isolates from Local Soup Condiment in the Study Area

Gram Stain/Shape Identification	Characterization			Probable Identification
	Mortality	Spore	Lactose Fermentation	
Gram -ve/rods	+	-	-	<i>Proteus mirabilis</i>
Gram -ve/short rods	+	ND	+	<i>Escherichia coli</i>
Gram -ve/rods <i>aerogenes</i>	-	-	+	<i>Klebsiella</i>
Gram +ve/cocci, single <i>roseus</i>	+	central spore	-	<i>Micrococcus</i>
Gram +ve/cocci, clusters <i>epidermidis</i>	-	-	-	<i>Staphylococcus</i>
Gram +ve/long rods <i>licheniformis</i>	+	central spore	-	<i>Bacillus</i>
Gram +ve/long rods <i>subtilis</i>	+	spore	-	<i>Bacillus</i>

Key: -ve = negative; +ve = positive; ND = not detected; + = present; - = absent

Table 4: Biochemical Reaction Patterns in Primary Tests for the Identification of Isolated Bacteria from Local Soup Condiment in the Study Area

Probable Organisms	Biochemical Reactions						
	Glu	Cit	Indo	Urea	Oxi	Coagu	Cata
<i>Proteus</i> sp.	-	+	+	+	-	-	+
<i>Escherichia</i> sp.	Gas	-	+	-	-	-	+
<i>Klebsiella</i> sp.	Gas	+	-	-	+	-	+
<i>Micrococcus</i> sp.	Acid	-	-	-	-	-	+
<i>Staphylococcus</i> sp.	Acid & Gas	ND	-	ND	-	-	+
<i>Bacillus licheniformis</i>	Acid & Gas	ND	+	ND	+	-	+
<i>Bacillus subtilis</i>	Acid & Gas	-	-	ND	+	-	+

Key: Glu = Glucose; Cit = Citrate; Indo = Indole; Urea = Urease; Oxi = Oxidase; Coagu = Coagulase; Cata = Catalase; sp. = species (singular); ND = not detected; + = present; - = absent

Table 5: Identification of Protozoa Associated with Contamination of Local Soup Condiment in the Study Area

Town	Sample	Protozoa	Frequency	Percentage
Dirin-Daji	Sample 1	-	0	0.0%
	Sample 2	-	0	0.0%
	Sample 3	<i>Entamoeba</i> spp.	1	12.5%
Doka	Sample 1	<i>Entamoeba</i> spp.	1	12.5%
	Sample 2	-	0	0.0%
	Sample 3	<i>Entamoeba</i> spp.	1	12.5%
Laraba	Sample 1	-	0	0.0%
	Sample 2	-	0	0.0%
	Sample 3	<i>Entamoeba</i> spp.	1	12.5%
Makuku	Sample 1	<i>Entamoeba</i> spp.	1	12.5%
	Sample 2	-	0	0.0%
	Sample 3	<i>Entamoeba</i> spp.	1	12.5%
Janbirni	Sample 1	-	0	0.0%
	Sample 2	<i>Entamoeba</i> spp.	1	12.5%
	Sample 3	-	0	0.0%
Dankolo	Sample 1	<i>Entamoeba</i> spp.	1	12.5%
	Sample 2	-	0	0.0%
	Sample 3	-	0	0.0%
Total			8	100%

Key: spp. = species (plural); % = percent

Table 6: Prevalence of Protozoa Associated with Contamination of Local Soup Condiment in the Study Area

Town	No. of Samples	No. Positive	No. Negative	% Positive	% Negative
Dirin-Daji	3	1	2	5.6%	11.1%
Doka	3	2	1	11.1%	5.6%
Laraba	3	1	2	5.6%	11.1%
Makuku	3	2	1	11.1%	5.6%
Janbirni	3	1	2	5.6%	11.1%
Dankolo	3	1	2	5.6%	11.1%
Total	18	8	10	44.4%	55.6%

Key: No. = number; % = percent

Discussion

It is generally known that illnesses due to the consumption of contaminated foods are the most common health problems in the contemporary world today, and a significant cause of reduced economic productivity [4]. Recent studies examining the morbidity and mortality of food-borne diseases have confirmed the significant public health burden posed by these diseases [4].

Microbial food safety is an increasing public health concern worldwide [15]. In developed countries, it is

estimated that one-quarter to one-third of the population are made sick each year because of food-borne diseases. In the developing countries, including Nigeria, the burden is much more severe; and therefore, there is need to regularly survey the food preparation, handling and storage techniques to ensure the safety of the food on one hand, and the health of the consumers on the other hand [4].

In the present study, a total of six (6) bacterial genera were isolated from various samples collected from the six (6) different markets in the study area namely: *Proteus* sp., *Escherichia* sp., *Klebsiella* sp., *Micrococcus* sp., *Staphylococcus* sp. and *Bacillus* sp. (Table 1). However, this study contradicts the findings of Sanyinna *et al.* who in a similar study on Evaluation of Bacterial and Protozoan Contamination of Local Soup Condiment (Daddawa) Produced by North-Western Community, Nigeria isolated four bacterial species only namely: *Staphylococcus aureus*, *Shigella dysenteriae*, *Escherichia coli* and *Salmonella typhi* [4]. Only *Escherichia* sp. and *Staphylococcus* sp. were re-isolated in this study in comparison to the previous study by Sanyinna *et al.* [4]. In addition, *Proteus* sp., *Klebsiella* sp., *Micrococcus* sp. and *Bacillus* sp. were also isolated in this study.

The bacteria associated with contamination of local soup condiment in the study area were counted in colony forming unit per gram per sample as illustrated above (Table 2). Dirin-Daji: Sample (1) 4.0×10^3 , Sample (2) 3.8×10^4 , Sample (3) 3.2×10^5 ; Doka: Sample (1) 4.4×10^4 , Sample (2) 4.0×10^5 , Sample (3) 3.6×10^6 ; Laraba: Sample (1) 3.2×10^4 , Sample (2) 4.8×10^5 , Sample (3) 2.0×10^6 ; Makuku: Sample (1) 4.4×10^3 , Sample (2) 3.3×10^4 , Sample (3) 2.8×10^5 ; Janbirni: Sample (1) 4.0×10^3 , Sample (2) 3.6×10^4 , Sample (3) 2.4×10^5 and Dankolo: Sample (1) 3.6×10^4 , Sample (2) 3.8×10^5 , Sample (3) 2.4×10^6 respectively. However, this is inconsistent with the result of Sanyinna *et al.* who obtained 1.50×10^2 , 1.30×10^3 and 9.60×10^4 for sample A; 1.30×10^2 , 8.50×10^3 and 4.00×10^4 for sample B; 9.00×10^2 , 7.60×10^3 and 6.00×10^4 for sample C; 6.40×10^2 , 3.00×10^3 and 2.50×10^4 for sample D, respectively [4].

Further characterization of the bacterial isolates revealed the organisms to be *Proteus mirabilis*, *Escherichia coli*, *Klebsiella aerogenes*, *Micrococcus roseus*, *Staphylococcus epidermidis*, *Bacillus licheniformis* and *Bacillus subtilis* respectively (Table 3). Again, the morphological features of the isolates revealed that some are gram +ve such as *Micrococcus roseus*, *Staphylococcus epidermidis* and *Bacillus* spp. while others are gram -ve such as *Proteus mirabilis*, *Escherichia coli* and *Klebsiella aerogenes* (Table 3). This result is supported by previous study of Sanyinna *et al.* who confirmed *Staphylococcus* sp. and *Escherichia* sp. as gram +ve and gram -ve bacteria respectively [4].

The biochemical reaction patterns in primary tests for the identification of isolated bacteria from local soup condiment in the study area (Table 4) revealed that

Glucose is absent, Citrate present, Indole present, Urease present, Oxidase absent, Coagulase absent, Catalase present and the probable organism being *Proteus* sp. For *Escherichia* sp., Glucose is Gas, Citrate absent, Indole present, Urease absent, Oxidase absent, Coagulase absent and Catalase present. For *Klebsiella* sp., Glucose is Gas, Citrate present, Indole absent, Urease present, Oxidase absent, Coagulase absent and Catalase present. For *Micrococcus* sp., Glucose is Acid, Citrate absent, Indole absent, Urease absent, Oxidase absent, Coagulase absent and Catalase present. For *Staphylococcus* sp., Glucose is Acid & Gas, Citrate not detected, Indole absent, Urease not detected, Oxidase absent, Coagulase absent and Catalase present. For *Bacillus licheniformis*, Glucose is Acid & Gas, Citrate not detected, Indole present, Urease not detected, Oxidase present, Coagulase absent and Catalase present. This study agrees with the previous study of Sanyinna *et al.* for *staphylococcus* sp. and *Escherichia* sp. where *Staphylococcus aureus* had gram stain of +Cocci, positive catalase, positive coagulase, positive urease and negative oxidase and *Escherichia coli* had gram stain of –Rod, positive catalase, negative coagulase, negative urease and negative oxidase respectively [4]. Although, *Shigella dysenteriae* and *Salmonella typhi* were not isolated in this study, previous study indicated that they had gram stain of –Rod, positive catalase, negative coagulase, negative urease and negative oxidase and –Rod, positive catalase, negative coagulase, negative urease and negative oxidase respectively [4].

Most previous studies were mainly on investigating the nutrient composition of fermented condiments [2], [6], [16], [17], [18], [19]. However, the relatively high recovery of the gram positive bacteria and Mannitol-fermenting or Mannitol Salt Agar (MSA) are of clinical importance. Many gram-negative bacteria such as *Escherichia coli*, *Salmonella serovars*; *Campylobacter* spp., *Enterobacter* spp. *Klebsiella* spp. etc. have been implicated in food borne diseases [20]. Similar report of Ogunshe *et al.* also isolated such gram-negative bacteria of clinical importance from a fermented food condiment [21]. *Staphylococci* have been known worldwide to cause food borne intoxication and poisoning [22]. There is perhaps no documented information available on the involvement of *S. aureus* in food borne disease out-breaks of fermented food condiment origin due to virtually non-existing system in Nigeria. *E. coli* isolated in this study has become a significant public health concern with a worldwide distribution [20]. Other gram-negative bacteria isolated in this study have also been implicated in acute bacterial diarrhoea and food poisonings. *S. dysenteriae* and *S. typhi* although not isolated in this study, had also been previously implicated as opportunistic pathogens and have become of increasing importance [22]. The fermented condiment preparation is still a traditional family art done in households and the fermentation that do not require conscious introduction of microbial flora in fermenting environment, thereby leading to the relative significant recovery of these indicator bacteria from the fermented condiment as indicated by the study [23]. The high

recovery rates of *S. aureus* and the total coliform in the fermented condiments; the wholesomeness in the fermented condiments, which may result in an increased risk of transmission of the disease to the humans who consume them.

The identification of protozoa associated with contamination of local soup condiment in the study area (Table 5) showed a total of eight (8) protozoa isolated from Dirin-Daji, Doka, Laraba, Makuku, Janbirni and Dankolo. For all the six (6) towns in the LGA, only *Entamoeba* spp. were isolated. This result however, is consistent with the result obtained in previous study (Sanyinna *et al.*, 2021). Although they isolated *Entamoeba histolytica* (50.0%) alongside *Giardia lamblia* (25.0%), but, *E. histolytica* was the most prevalent in their study area. This also revealed that *E. histolytica* or generally *Entamoeba* spp. are the most serious parasitic protozoa responsible for the contamination of local soup condiments in the study area. However, there is dearth of published data on protozoan contamination of local soup condiments in Nigeria. Estimates of food-borne disease deaths are subject to uncertainty because the number of deaths caused by unidentified pathogenic agents in the food supply is unknown. However, in the influential study of food-borne diseases in the United States by Mead *et al.*, it was estimated that unknown food-borne agents caused 3400 deaths per year or 65% of the estimated 5200 annual deaths from food-borne illnesses [20]. No matter how alarming these estimates from a developed country like the United States may be, more alarming will be the estimate from developing countries like Nigeria [4].

The overall prevalence of the protozoa associated with contamination of local soup condiment in the study area (Table 6) was 44.4%. This result corroborates the findings of Sanyinna *et al.* who obtained an overall prevalence rate of 50.0% for *Entamoeba histolytica* in Zuru LGA [4].

Common sources of food-borne diseases are bacterial and protozoan contamination of food by food handlers. However, the safety aspects of fermented condiments are not adequately documented and appreciated in developing countries like Nigeria [24].

The results obtained in this study agree with the study conducted by Liman *et al.*, who demonstrated the impact of environmental conditions on the quality of some processed condiments where contaminants like bacteria and protozoa can be gotten either from the water used in washing the seeds, the handlers or from utensils, flies and a lot [19]. Therefore, there is need to prepare and process the condiments under better hygienic processes.

V. CONCLUSION AND FUTURE SCOPE

From the results of the experiments, *Bacillus* spp. and *Entamoeba* spp. were observed to be the key spoilage organisms of local soup condiment. Spoilage was noted to be due to continuous activities of the organisms after

desired fermentation was reached. Other organisms isolated from the local soup condiment are likely contaminants exposed to the food after boiling, prior to fermentation through the air, water or poor aseptic measures during handling. To ensure a simple method of maintaining local soup condiment quality with a longer shelf life, good hygienic practices, sterile water and clean environment should be maintained. Additionally, experiments should be targeted towards simple, uncomplicated methods for the removal, killing or inactivation of the pathogens or their enzymes present in the finished products to maintain local soup condiment quality for a longer period of time. Sellers of the condiments should ensure that they do not expose the fermented foods during display for sale because this may predispose them to contamination. During sales and production, sellers and local processors must always wear gloves to discourage and limit contamination. Fermentation should be considered safe for consumption because food pathogens such as *Salmonella typhi*, *Shigella dysenteriae*, *Entamoeba coli*, *Campylobacter jejuni* and *Clostridium* were not isolated from them, although their safety can be ensured. Fermentation of local soup condiments should be mostly carried out during the dry seasons so as to increase fermentation and also reduce the difficulty of drying them during rainy seasons. Information concerning the use of seasonings in some areas/homes should be made available through public awareness campaign.

ACKNOWLEDGEMENTS

We sincerely acknowledge the efforts of Sakaba Local Government Communities particularly the people of Laraba, Makuku, Janbirni, Dankolo, Doka and Dirin-Daji for providing the local soup condiment samples. Equally, the laboratory technicians of Microbiology Laboratory, Kebbi State University of Science and Technology, Aliero, Nigeria are highly appreciated for their contribution in making this research a success.

REFERENCES

- [1]. A. A. Yagoub and A. A. Abdalla, "Effect of domestic processing methods on chemical composition, *in-vitro* digestibility of protein and starch and functional properties of Bambara Groundnut (*Voandzeia subterranea*) seed", *Research Journal of Agriculture and Biological Sciences*, Vol. 3, No. 1, pp. 24-34, 2007.
- [2]. M. Collins, A. Bere, A. Traore, "The chemical composition of Bikalga, a traditional fermented Roselle (*Hibiscus sabdariffa* L.) seeds condiment. Part II. Evaluation of minerals, total polyphenols and phytic acid content, predicting the iron bioavailability", *Electronic J. Food Plants Chem.*, Vol. 2006, No. 1, pp. 7-11, 2014.
- [3]. A. D. Smith, "The Oxford Companion of American Food and Drinks", *Oxford University Press*, Oxford, UK, 2007.
- [4]. Y. M. Sanyinna, D. D. Attah, M. K. Kele and R. D. Jabaka, "Evaluation of Bacterial and Protozoan Contamination of Local Soup Condiment (Daddawa) Produced by North-Western Community, Nigeria", *International Journal of Scientific Research in Biological Sciences*, Vol. 8, No. 5, pp. 25-31, 2021.
- [5]. W. A. Redmond, "Spices", Microsoft Student 2008 [DVD]. *Microsoft Corporation*, 2007.
- [6]. O. A. Akoma, N. Y. Nma, S. A. Musa and A. B. Salihu, "Nutritional and phytochemical composition of *Vitellaria paradoxa* (shea fruit pulp)", *International Journal of Biochemistry Research and Review*, Vol. 22, No. 1, pp. 1-7, 2018.
- [7]. K. T. Farrell, "Species Condiments and Seasoning", 2nd edition, *USA publishers*, USA, pp. 291, *Fecal Specimen*, Vol. 4, No. 1, pp. 37-56, 1990.
- [8]. A. Zeldes and A. Leah, "Chicago Restaurants' and Entertainment Guide", 2009.
- [9]. A. D. Smith and F. Andrew, "The Oxford Comparison to American Food and Drink", *Oxford University Press*, Oxford, UK, 2007.
- [10]. S. Dakwa, E. Sakiyi-Dawson, C. Diako, N. T. Annon and W. K. Amoa-Awua, "Effect of boiling and roasting the fermentation of beans locust beans into Daddawa", *International Journal of Food Microbiology*, Vol. 104, No. 1, pp. 69-82, 2005.
- [11]. NPC, "National Population Commission", National Population and Housing Census, 2006.
- [12]. I. A. Takon, S. P. Antai and E. U. Eyong, "Comparative studies on the content of active ingredients of contaminated and non-contaminated Aspirin tablets sold in patent medicine stores in Calabar", *Journal of Medical Pharmaceutical and Allied Sciences*, Vol. 5, No. 8, pp. 142-150, 2016.
- [13]. M. Cheesbrough, "District Laboratory Practice in Tropical Countries", 2nd edition, *Cambridge University Press*, Cambridge, UK, pp. 47-54, 2006.
- [14]. M. Cheesbrough, "District Laboratory Manual for Tropical Countries", Vol. 2, 2002.
- [15]. C. Zhao, G. Beilei, J. Villena, R. Sudler, E. Yeh, S. Zhao, D. G. White, D. Wagner, A. Meng, "Prevalence of *Campylobacter* spp., *Escherichia coli*, and *Salmonella serovars* in retail chicken, turkey, pork and beef from the greater Washington D.C. area", *Appl. Environ. Microbiol.*, Vol. 66, No. 0, pp. 431 – 5436, 2001.
- [16]. S. A. Odunfa, "Carbohydrate changes in fermenting locust bean during Iru preparation", *Plant Foods Hum. Nutrition*, Vol. 32, No. 0, pp. 1-10, 1983.
- [17]. S. A. Odunfa, "African fermented foods. In: Wood, B.J.B. (ed.) Microbiology of fermented foods", *Applied Science Publishers*, Amsterdam, Elsevier, Vol. 11, pp. 155-191, 1985.
- [18]. B. O. Omafuvbe, O. S. Falade, B. A. Osuntogun and S. R. A. Adewusi, "Chemical and biochemical changes in African locust bean (*Parkia biglobosa*) and melon (*Citrullus vulgaris*) seeds during fermentation to condiments", *Pakistan Journal of Nutrition*, Vol. 3, No. 3, pp. 140-145, 2004.
- [19]. A. A. Liman, P. Egwin, M. A. Vunchi and C. Ayansi, "Lipase activity in fermented oil seeds of African locust bean (*Parkia biglobosa*), castor seeds (*Ricinu communis*) and African oil bean (*Pentaclethra macrophylla*)", *Nigerian Journal of Basic and Applied Science*, Vol. 18, No. 1, pp. 136-140, 2010.
- [20]. P. S. Mead, L. Slutsker, V. Dietz, L. F. McCaige, J. S. Bresee, P. Shapiro, P. M. Griffin and R. V. Tauxe, "Food-related illness and death in the United States", *Emerging Infectious Diseases*, Vol. 5, No. 5, pp. 607-625, 1999.
- [21]. A. A. O. Ogunshe, A. E. Ayodele, O. Okonko, "Microbial studies on *Aisa*- a potential indigenous laboratory fermented food condiment from *Albizia saman* (Jacq.)", *F. Mull Pak. J Nutr.*, Vol. 5, No. 1, pp. 51-58, 2005.
- [22]. L. M. Prescott, J. P. Harley and D. A. Klein, "Detection of disease-causing microorganisms", In: *Microbiology*, 4th edition, McGraw-Hill, New York, NY, pp. 916-917, 1999.
- [23]. L. J. Ogbadu and R. N. Okagbue, "Fermentation of African locust bean (*Parkia biglobosa*) seeds: involvement of different species of *Bacillus*", *Food Microbiology*, Vol. 5, No. 0, pp. 195-199, 1988.
- [24]. A. A. O. Ogunshe, M. A. Omotosho and J. A. Adeyeye, "In vitro antimicrobial characteristics of Bacteriocin-producing *Lactobacillus* strains from Nigerian indigenous fermented foods", *African Journal of Biotechnology*, Vol. 6, No. 4, 445-453, 2007.

AUTHORS' PROFILE

Dr. Daniel Dan-Inna Attah is currently an Associate Professor of Parasitology and Head of Zoology Unit of the Department of Animal and Environmental Biology, Faculty of Life Sciences, Kebbi State University of Science and Technology, Aliero, Nigeria. Formerly, he was the Postgraduate Coordinator of the same department. He has supervised many Undergraduates, Masters and Ph.D students. He had a lot of publications in several reputable journals both locally and internationally.



Mr. Yusuf Muhammad Sanyinna pursued B. Sc. Ed. (Hons.) Biology degree from Usmanu Danfodiyo University, Sokoto, Nigeria and currently undergoing M. Sc. Zoology with specialization in Parasitology from Department of Animal and Environmental Biology, Faculty of Life Sciences, Kebbi State University of Science and Technology, Aliero, Nigeria. He is currently working as the Principal, Secondary Conventional Section as well as Biology and Animal Husbandry Teacher with Imam Malik Academy Nigeria (IMAN), Sokoto State, Nigeria. He is a member of Teachers' Registration Council of Nigeria (TRCN), Parasitology and Public Health Society of Nigeria (PPSN) and Zoological Society of Nigeria (ZSN). His main research works focus on Medical and Veterinary Parasitology, Bacteriology, Protozoology, Helminthology, Molecular Epidemiology, Entomology, Animal Physiology and Microbiology. He had taught in Government Secondary School, Sanyinna; Government Girls Day Secondary School, Sanyinna; Shehu Shagari College of Education Staff Secondary School, Sokoto; Sani Dingyadi Unity Secondary School, Sokoto; Bichi Academy, Kano; Nady Academy Schools, Sokoto and Brilliant Footsteps Int'l Academy, Sokoto. He has 5 years of teaching experience and 2 years of research experience. He has a lot of experiences in diverse fields of endeavour and leadership qualities.



Mrs. Regina Doro Jabaka is currently an academician in the Department of Microbiology, Faculty of Life Sciences, Kebbi State University of Science and Technology, Aliero, Nigeria. She is also currently undergoing her Ph.D in Federal University of Technology Minna, Niger State, Nigeria. She had a lot of experience in the Microbiology Laboratory. She equally had a lot of publications in reputable journals.

