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Microbial Load Analysis and Anti-Nutritional Assessment of Ready-to-eat Fruits and Nuts Vended in Selected Motor Parks in Ado-Ekiti Metropolis Ekiti-State Nigeria

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Abstract

Fruits and nuts are an important portion of the human diet and are often being contaminated with both pathogenic and non-pathogenic microorganisms. This study assessed the microbial load on the surface of some Fruits and Nuts sold by fruit vendors in selected motor parks in Ado Metropolis, Ekiti State. The studied fruits and nuts (F& N) include *Cola nitida*, *Garcinia kola*, *Malus domestica*, *Phoenix dactylifera*, *Daucus esculentus*, *Solanum ovigerum and Cyperus esculentus* were purchased from fruit vendors. The fruits and nuts in were rinsed in sterile water and 1ml of the water in which the fruits and nuts were washed was inoculated in sterile culture media using pour plate techniques. Different microbial species from the media in which the waters were inoculated were then identified using their morphological and biochemical characteristics. The microbes recovered belong to the phylum fungi, and bacteria and they include *Aspergillus niger, Aspergillus flavus, Pseudomonas* spp, *Proteus* spp, *Klebsiella* spp and Saccharomyces. The most frequently isolated contaminant was *Staphylococcus* spp, followed *by Aspergillius niger* and *Aspergillius. flavus*. Others are *Pseudomonas* spp, *Proteus* spp, *Klebsiella* spp, and *Saccharomyces*, the most contaminated sample was (*Cola nitida* and *Malus domestica*) had the least microbial contamination. The concentration of Anti-nutritional factors of fruits in this finding showed that Date plant has the highest concentration of Oxalate, Kola nut recorded the highest concentration in Alkaloids while bitter kola has the highest concentration of Cyanide. This study had shown that direct consumption of ready-to-eat fruit and nut vended at motor parks can be a potential risk factor for food-related illness and food poisoning in the area. Awareness of hygienic practices by vendors must be created

Keywords: Fruits and nuts, fruit vendors, microbial load, Antinutritional factor and microbial species

Citation

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1. Introduction

Ready-to-eat foods (RTEs) are foods that do not require further processing or preparations before consumption which include some vegetables, fruits and nuts. They are a special dietary source of nutrients, vitamins and fibre for humans and are thus vital for health and wellbeing (Eni *et al.,* 2010). Diets rich in fruit and vegetables are known to prevent nutritional related sicknesses such as scurvy, rickets, exophthalmia and vitamin C and A deficiencies among others (Eni *et al.,* 2010). Well balanced diets that are rich in fruits and vegetables, are especially valuable for their ability to prevent and reduce the risk of several diseases The anti-nutritional factors include: saponins, tannins, flavonoids, alkaloids, trypsin (protease) inhibitors, oxalates, phytates, hemagglutinins (lectins), cyanogenic glycosides, cardiac glycosides, coumarins and gossypol (Ekpa and Sani, 2017).

In contrast to having several beneficial effects, Fruits and nuts can act as important reservoirs for different microorganisms (Makinde e*t al.*, 2020). Some of these microorganisms maybe find their way into the consumers during consumption and hence, causing food-related infections in the consumers. Some studies have reported Food-borne illnesses which are due to the consumption of ready to eat fruits and nuts. In Nigeria, most fruit and nuts are commonly sold on the street, which are laden with microbes and as such fruit and nuts are readily infested by microbes being carried around in the air or surrounding dust. World Health Organization reported that ready to eat fruit and nut contributes a high percentage to food-borne illness (WHO, 2010). The potential health risk associated with fruit and nuts commonly sold on the street cannot be overemphasized. Oje *et al*. (2010) showed that street vended foods are a potential source of microbial infections owing to poor hygiene and improper handling (Oje *et al.*2018).

Fruits and nuts are naturally colonized by normal florals on their surfaces while many pathogenic strains such as *Salmonella* and *Escherichia coli* O157:H7 found in sprouted seeds and fruit juices (Health & Consumer Protection Directorate-General, 2002) other pathogenic species that have also been isolated from fruit and nuts include some *Bacillus* spp, Coagulase-negative *Staphylococci*, *Klebsiella pneumoniae*, *Listeria monocytogenes*, Pseudomonas spp., and *Staphylococcus aureus* (Annan-Prah *et al.*,2011; Fadahunsi & Makinde, 2018), *Aspergillus, Alternaria, Fusarium*, and *Penicillium* (Gdoura-Ben Amor *et al.*, 2018). Recent studies have shown an increase in the rate of recovery of pathogenic microorganisms from fruit and nuts (Oje *et al.*, 2018). The risen concern on the frequent report of food illness associated with fruits and nuts has led to continuous surveillance studies that can help in monitoring the microbial loads on fruits and nuts and hence, helping in preventing them from being a potential vehicle for transporting pathogenic microorganisms into human (Annan-Prah *et al.*,2011). Although there have been many studies on fruit and nuts most of these studies have limited themselves to the profiling of the microorganisms' resistance to antimicrobial agents (Osuntokun & Akele, 2018; Wedajo *et al.*, 2013). None of these studies has focused on understanding the total microbial load on fruit and nuts as well as their antinutritional factors. This study thus seeks to evaluate the microbial loads and antinutritional factors in F & N commonly sold in a selected motor park in Ado Ekiti, Ekiti State Nigeria.

2. Material and Methods

Collection of Sample: Kolanut (*Cola nitida*), bitter kola (*Garcinia kola*), apple (*Malus domestica*), date (*Phoenix dactylifera*), carrot (*Daucus esculentus*), white eggplant (*Solanum ovigerum*) and tiger nut (*Cyperus esculentus*) were bought from fruit hawkers in five major motor parks in Ado, Ekiti State, Southwestern part of Nigeria. The motor parks include Ijigbo motor park, Shasha motor park, Oja Oba motor park, Basiri motor park and Ilesha garage in Ado South Local Government,

The control samples were obtained from wholesaler in the case of apple, date and carrot, while others were bought from local farmers and then sterilize with alcohol. These were transported in sterile universal plastic bags to the postgraduate Laboratory at the Federal University of Technology Akure for further processing.

Media Used/ Preparation: Nutrient Agar and Potato Dextrose Agar were prepared according to the manufacturer's specification and were sterilized using autoclave at 121ºC for 15minutes.

Microbiological Evaluation: The fruit samples were rinsed in sterile water and the water in which the samples had been washed was serially diluted up to the factor of 10^{-6}. One ml of each rinse water was then carefully pipetted into a sterile petri-dish. 15ml of already sterilized Nutrient Agar (NA) and Potato Dextrose Agar (PDA) was poured into the plates respectively. 1ml of 10% Lactic acid was used in PDA as an inhibitor of bacteria growth. The set-up was properly rotated for uniform distribution and allowed to solidify before incubation at 37ºC for 24hours for NA and 72hours for PDA. Morphologically distinct colonies growing on each plate were repeatedly sub-cultured until pure cultures were obtained.

Morphological and Biochemical Characterization of the bacteria was done using; Grain Staining, Citrate Test, Catalase Test, Motility Test, Fermentation of Sugars according to the procedures of Characterization of the isolates were based on Physiological, morphological and biochemical characteristics according to Bergey's Manual of Determinative Bacteriology

Determination of Anti-Nutritional Components

***Tannin*:** About 200mg of finely grand sample was weighed into a 500ml sample bottle; 10ml of 10% aqueous acetone was added and properly covered. The bottle was put in a water bath and shaken for two hours at 30ºC. Each solution was then centrifuged, and supernatant stored in ice, 0.2ml of each solution was pipetted into test tubes and 0.8ml of distilled water was added. The standard tannic acid solution was prepared from a 5mg/ml stock and the solution was made up to 1ml with distilled water. Folin reagent of 0.5ml was added to both the sample and the standard, followed by 2.5ml of 20% Na_2CO_3. The solutions were then vortexed and incubated for ten minutes at room temperature after which the absorbance was read at 725nm against a reagent blank concentration of the sample from a standard tannic acid curve (Makkar & Godchild, 1996).

Oxalate: One gram of each sample was weighed into a 100ml conical flask. 0.75ml H_2SO_4 was added and the solution mixture was stirred intermittently with a magnetic stirrer for about 1hour and then filtered using Whatman No. 1 filter paper. The filtrate (extract) of 2.5ml was collected and titrated hot (80-90ºC) against 0.1ml $KMnO_4$ solution until a faint pink colour appeared and persisted for at least 30 seconds.

Phenol: The Phenol present in the F & N was determined using ferric acid chloride test, the filtered solution extract was treated with three drops of freshly prepared 1% Ferric acid Chloride and Potassium Ferrocyanide. The formation of bluish green colour indicated the presence of phenolic compounds.

Cyanide: About 10g of each sample was weighed into Kjeldahi flask with 200ml distilled water and allowed to stand for 4hours. The whole volume was transferred into a 500ml round bottom flask on the heating mantle with distillation apparatus connected; 100ml distillate was received into conical flask containing 10ml of 2.5% NaOH solution and was diluted to 250ml with distilled water. Aliquot of 100ml was set for titration 0.02M $AgNO_3$ after the addition of 8ml 6M NH_4OH and 2.5%, 2ml 5% KI was titrated in the micro burette, the end point is faint but turbid and may easily be observed especially against a black background.

3. Result

The result shows the microbial load on selected Ready-to-eat fruits in Ado-Ekiti Metropolis. The ability of some microorganisms to metabolize a variety of sugar as a carbon source was used as criteria for the characterization of the isolated organisms. Table 1 shows the outcomes of fermentation and other biochemical tests used in the identification of bacteria.

Bacterial Load on Eggplant: Staphylococcus was isolated from eggplant fruit from all motor parks studied. The bacterial load ranged between 1.27×10^1 and 1.06×10^2 colony forming units (Table 2). The highest Staphylococcal load was obtained in the control sample with a mean of 1.06×10^2. This was significantly different ($P<0.05$) from the microbial load obtained from all other motor parks. The least was obtained on eggplant fruits purchased at Shasha motor park, Ado Ekiti in Ekiti State with a mean of 1.27 x10^{1} (Table 2). This load was significantly high (P<0.05) comared to was what was obtained in Ijigbo and Shasha parks with mean values of 1.53 x10^{1} and 1.27 x10^{1} respectively. The colony forming units of Staphylococcus isolated from Ijigbo and Shasha parks are not significantly different (Table 2). Proteus was also isolated from eggplant fruits from Basiri motor park and the control with an average of 1.966 x10^{1} and 3.2 x10^{1} CFU respectively. Klesbsiella was from isolated eggplant from Shasha and Old garage with means of 1.07 x10^{1} and 2.7 x10^{1} respectively (Table 2). Lactobacillus was isolated from only eggplant collected from Basiri with a mean colony-forming unit of 21.7x10^{-1}.

Table 1. Probable Identification of bacterial isolates,

Isolates	**Shapes**	**Gram Reaction**	**Gas from glucose**	**Acid from glucose**	**Acid from maltose**	**Gas from maltose**	**Lactose gas**	**Lactose acid**	**Citrate test**	**Acid from sucrose**	**Galactose Acid**	**Galactose gas**	**Citrate test**	**Motility**	**Catalase**	**SUSPECTED ORGANISMS**
A	Rod	+	+	+	-	-	+	+	+	+	+	+	+	-	+	*Klebsiella* spp
B	Cocci	+	+	+	+	+	+	+	+	+	+	+	-	-	+	*Staphylococcus* spp
C	Rod	-	+	+	-	-	-	-	-	-	-	-	-	+	-	*Proteus* spp
D	Long rod	+	-	+	+	+	-	+	+	+	+	+	+	-	-	*Lactobacillus* spp
E	Chain cocci	+	+	-	+	+	-	+	+	+	-	-	+	-	+	*Streptococcus* spp
F	Short rod	+	-	-	+	+	-	-	+	+	+	+	-	+	+	*Bacillus* spp
G	Rod	-	+	+	-	-	-	-	-	-	-	-	+	+	+	*Pseudomonas* spp
H	Rod	-	+	+	-	-	-	-	+	+	+	+	-	+	-	*Serratia* spp

Key, + = positive, - = negative

Table 2: Bacteria load of the Fruit and Nut from a base on sampling location

Bacteria load on eggplant

Location	*Staphylococcus* spp	*Proteus* spp	*Klebsiella* spp	*Lactobacillus* spp
Control	106±10.26^{d}	32.00±0.00^{e}	0.00±0.00^{a}	0.00±0.00^{a}
Basiri	32.00±6.43^{ab}	19.66±3.28^{b}	0.00±0.00^{a}	21.66±4.33^{b}
Oja Oba	34.66±11.25^{ab}	30.00±3.2^{c}	0.00±0.00^{a}	0.00±0.00^{a}
Ijigbo	15.33±2.90^{a}	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}
Shasha	12.66±2.66^{a}	0.00±0.00^{a}	10.66±2.40^{b}	0.00±0.00^{a}
Ilesha Park	18.00±4.35^{a}	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}
Old garage	43.33±6.11^{b}	0.00±0.00^{a}	26.66±3.75^{c}	0.00±0.00^{a}

Bacteria load on apple obtained from motor parks and fruits vendor in Ado-Ekiti Metropolis

	Staphylococcus spp	Bacillusspp	*Klebsiella* spp	*Lactobaccillus* spp
Control	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}
Basiri	26.66±5.23^{b}	0.00±0.00^{a}	14.66±2.40^{b}	0.00±0.00^{a}
Oja Oba	48.00±7.09^{c}	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}
Ijigbo	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}
Shasha	30.00±1.15^{b}	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}
Ilesha Park	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}

Bacteria load on carrot obtained from motor parks and fruits vendor in Ado-Ekiti Metropolis

	Staphylococcus spp	*Serratia* spp	*Pseudomonas* spp	*Proteus* spp
Control	53.66±9.24^{e}	37.66±11.78^{c}	0.00±0.00^{a}	28.66±10.89^{b}
Basiri	17.66±5.36^{b}	0.00±0.00^{a}	0.00±0.00^{a}	6.66±2.72^{a}
Oja Oba	8.33±0.66^{ab}	4.00±0.57^{ab}	1.66±0.66^{a}	0.00±0.00^{a}
Ijigbo	NS	NS	NS	NS
Shasha	45.33±2.90^{de}	13.33±2.72^{b}	0.00±0.00^{a}	20.00±1.15^{b}
Ilesha Park	NS	NS	NS	NS

Bacterial load on Date Fruit

	Staphylococcus spp	*Bacillus* spp	*Klebsiella* spp	-
Control	20.00±2.00^{b}	0.00±0.00^{a}	0.00±0.00^{a}	-
Basiri	NS	NS	NS	-
Oja Oba	56.6±64.33^{e}	0.00±0.00^{a}	32.66±4.33^{b}	-
Ijigbo	NS	NS	NS	-
Shasha	45.33±2.90^{d}	0.66±0.33^{a}	0.00±0.00^{a}	-
Ilesha Park	NS	NS	NS	-

Bacteria load on bitter kola obtained from motor parks and fruits vendor in Ado-Ekiti Metropolis

	Staphylococcus spp	*Klebsiella* spp	*Pseudomonas* spp	*Baccillus* spp
Control	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}
Basiri	3.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}
Oja Oba	1.66±0.33^{a}	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}
Ijigbo	4.33±1.76^{a}	3.33±0.88^{b}	0.00±0.00^{a}	0.00±0.00^{a}
Shasha	4.00±1.54^{a}	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}
IleshaPark	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00^{a}

Bacteria load on tiger nut obtained from motor parks and fruits vendor in Ado-Ekiti Metropolis

	Staphylococcu spp	*Klebsiella* spp	*Pseudomonas* spp	*Baccillus* spp
Control	70.00±2.88^{de}	0.00±0.00^{a}	0.00±0.00^{a}	56.66±0.00^{c}
Basiri	NS	NS	NS	NS
Oja Oba	20.00±4.04^{bc}	18.66±3.17^{b}	0.00±0.00^{a}	0.00±0.00^{a}
Ijigbo	NS	NS	NS	NS
Shasha	10.00±2.88^{ab}	0.00±0.00^{a}	7.66±0.66^{ab}	0.00±0.00^{a}
Ilesha Park	NS	NS	NS	NS

Bacteria load on kola nut obtained from motor parks and fruits vendor in Ado-Ekiti Metropolis

	Staphylococcus spp	*Seratia* spp	*seudomonas* spp	*Klebsiella* spp
Control	$14.00{\pm}4.40^{ab}$	$0.00{\pm}0.00^{a}$	$0.00{\pm}0.00^{a}$	$0.00{\pm}0.00^{a}$
Basiri	$0.00{\pm}0.00^{a}$	$57.33{\pm}10.49^{d}$	$0.00{\pm}0.00^{a}$	$50.00{\pm}6.02^{c}$
Oja Oba	$8.33{\pm}3.33^{ab}$	$0.00{\pm}0.00^{a}$	$5.00{\pm}1.15^{a}$	$0.00{\pm}0.00^{a}$
Ijigbo	$14.00{\pm}1.00^{ab}$	$0.00{\pm}0.00^{a}$	$0.00{\pm}0.00^{a}$	$10.66{\pm}2.60^{b}$
Shasha	$15.33{\pm}2.90^{ab}$	$11.33{\pm}0.66^{ab}$	$12.00{\pm}3.05^{b}$	$0.00{\pm}0.00^{a}$
IleshaPark	$23.33{\pm}3.38^{bc}$	$16.66{\pm}1.66^{b}$	$0.00{\pm}0.00^{a}$	$0.00{\pm}0.00^{a}$

* Readings on the Table present mean values ± standard error of the mean, *NS –not Sampled*

Values with the same superscript letter(s) along the same column are not significantly different at P<0.05 using Duncan's new multiple range test.

Bacterial load on Apple: No bacteria isolate was observed on the control sample, Iigbo and Ilesha motor park in Ado-Ekiti Metropolis. While *Staphylococcus spp* with a population of 48 $x10^{1}$ colony-forming unit was isolated from Oja oba' s apple.

Bacterial load on carrot: Four bacterial species include Staphylococcus species, Serratia species. *Pseudomonas* species and *Proteus* species were isolated from carrot samples from Basiri, Oja Oba, Shasha in Ado-Ekiti. Carrot was not sampled at Ijigbo Motor Park and Ilesha Motor Park in Ado-Ekiti due to the unavailability of carrots at the time of sampling. Meanwhile, Staphylococcus species was repeatedly found in all the carrots sampled with the control having the highest mean value of 5.37 $x10^{1}$ (cfu) while the least mean value of $83.3x10^{-1}$(cfu) was recorded from Oja Oba's samples. Serratia species was isolated from all the sampled plates collected from Basiri, Ado-Ekiti. Proteus species was not detected in carrot samples from Oja Oba, Ado-Ekiti.

Bacterial load on date fruit: Date fruit was not sampled at Basiri, Ijigbo Motor Park and Ilesha Motor Park in Ado-Ekiti because it only sold around Oja-Oba. Staphylococcus was the only bacterial isolate found on the control plate with a mean value of 20.0 $x10^{-1}$ (cfu) (Table 2) Oja Oba in Ado-Ekiti recorded the highest *Staphylococcus* species with a man value of 5.66 $x10^{1}$ (cfu) and *Klebsiella* species with a mean value of $3.27x10^{1}$(cfu) (Table 2).

Bacterial load on bitter kola: The control sample plate and sample obtained from Ilesha motor park, Ekiti recorded no colony growth on the plate. Generally, in all the plate sampled, the number of bacterial colony was very low compared to other samples. The least *Staphylococcus* species with a mean CFU value of 16.6 $x10^{-1}$ was found at Oja Oba in Ado-Ekiti. The highest *Staphylococcus* mean of CFU value of $40x10^{-1}$ were obtained at Shasha park in Ekiti (Table 2).

Bacterial load on tiger nut; Basiri, Ijigbo and Ilesha Motor Park in Ado-Ekiti were not sampled due to its unavailability as at the time of visit. *Staphylococcus* spp. *Klebsiella* spp. *Pseudomonas* spp. and Bacillus spp were the isolated bacterial obtained. The most abundant was *Staphylococcus* spp with a mean value of 7 $x10^{1}$ on the control plate, the least mean value obtained was 10 $x10^{2}$ from Shasha park (Table 2). *Klebsiella* spp was only bacterial obtained at Oja Oba in Ekiti with a mean value of 18.66 while *Pseudomonas* was obtained on the plate from Shasha in Ekiti with colony mean count of 7.66.

Bacterial load on kola nut: *Staphylococcus* species, *Serratia* species, *Pseudomonas* species, *Klebsiella* species and Proteus species were bacteria isolated from kola nut sample plates. The control sample was significantly different (P<0.05) from other location samples. Mean colony forming unit value of Staphylococcus species was $1.40x10^{-1}$. it was the only organism observed on the control plate. Oja Oba, Ado-Ekiti had lowest Staphylococcus load of 83.3x10^{-1}cfu, *pseudomona* was 50 x10^{-1}cfu while *proteus* was 66.6 x10^{-1} cfu (Table 2). The highest *staphylococcal* count was observed from Ilesha motor park samples with mean value of 2.33 x10^{1}. *Proteus* species was the only colony observed on nuts obtained from Oja Oba plate in Ado-Ekiti at a mean of 66.6 x10^{-1} (Table 2).

Fungi load on eggplant: There was no fungi growth observed on the Shasha, Ado-Ekiti plate. However, the control sample plate recorded the highest means value of 3.33 colony of *Aspergillus flavus* and 5.33 x10^{2} colony of *Aspergillus niger*. Oja Oba, Ado-Ekiti recorded the least mean value of 6.6 x10^{-1} and 3.3x10^{1} colonies of *Saccharomycess* species and A. flavus respectively. The most abundant in occurrence was *A niger* while the least in abundant was *Saccharomycess* species (Table 3).

Fungi load on kolanut: The four fungi species isolated were *Saccharomyces* species, *A.niger, Rhizopus* spp and *penicillum* species. The highest Saccharomyces mean value of 2.1 x10^{1} was recorded at Basiri motor park, Ado- Ekiti. *Aspergillus niger* was recorded at Oja Oba, and Ilesha motor park in Ado-Ekiti and Oja Oba. *Penicillium* species was recorded at Ijigbo, Ilesha motor park, Ado-Ekiti (Table 3).

Fungi load on Bitter kola: One species of fungi was recorded on the control plate, Basiri and Oja Oba in Ekiti State, while Ijigbo, Ado-Ekiti had the four 4 species of fungi in occurrence. Two fungi species were observed at Shasha Ado-Ekiti. The control sample was significantly different (P <0.5) from other samples having the least *A. niger* mean value of 0.66 while Basiri, Ado-Ekiti had the highest *Penicillium* mean value of 1.57 x10^{1} (Table 3).

Fungi load on carrot: *A. niger* and *A. flavus* were the two isolated fungi on carrot. The highest A. niger mean value of 40 x10^{1}and *A. flavus* mean value of 16.6 x10 ^{1} were found on the control sample, while the least *A. flavus* values of 10 x10^{1} were found on the plate obtained from Basiri and Oja Oba, Ado-Ekiti. Ijigbo and Ilesha park (Table 3).

Table 3: Fungi load on eggplant obtained from motor parks and fruits vendor in Ado-Ekiti Metropolis

	Saccharomyces	***A.flavus***	***A. niger***
Control	$0.00±0.00^{a}$	$3.33±0.66^{b}$	$5.33±1.20^{c}$
Basiri	$1.66±0.33^{b}$	$0.00±0.00^{a}$	$0.00±0.00^{a}$
Oja Oba	$0.66±0.33^{ab}$	$0.33±0.33^{a}$	$0.00±0.00^{a}$
Ijigbo	$0.00±0.00^{a}$	$0.00±0.00^{a}$	$1.00±0.57^{ab}$
Shasha	$0.00±0.00^{a}$	$0.00±0.00^{a}$	$0.00±0.00^{a}$
Ilesha Park	$0.00±0.00^{a}$	$0.00±0.00^{a}$	$2.00±0.00^{b}$
Fungi load on kola nut obtained from motor parks and fruits vendor in Ado-Ekiti Metropolis			
	Saccharomyces	*A. niger*	*Rhizopus*
Control	$0.00±0.00^{a}$	$4.00±0.57^{bcd}$	$0.00±0.00^{a}$
Basiri	$21.66±1.66^{d}$	$0.00±0.00^{a}$	$0.00±0.00^{a}$
Oja Oba	$2.66±0.66^{ab}$	$5.67±2.02^{cde}$	$1.66±0.33^{b}$
Ijigbo	$0.00±0.00^{a}$	$1.67±0.38^{b}$	$1.66±0.33^{b}$
Shasha	$0.00±0.00^{a}$	$2.00±0.00^{abc}$	$0.00±0.00^{a}$
Ilesha Park	$7.33±1.76^{c}$	$9.00±2.64^{e}$	$0.00±0.00^{a}$
Fungi load on bitter kola obtained from motor parks and fruits vendor in Ado-Ekiti Metropolis			
	A. niger	*A. flavus*	*Penicillium spp.*
Control	$0.66±0.33^{ab}$	$0.00±0.00^{a}$	$0.00±0.00^{a}$
Basiri	$0.00±0.00^{a}$	$0.00±0.00^{a}$	$15.66±2.33^{c}$
Oja Oba	$2.66±0.33^{cde}$	$0.00±0.00^{a}$	$0.00±0.00^{a}$

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Ijigbo	3.66 ± 0.88^{de}	2.66 ± 0.33^{cd}	4.66 ± 0.66^{b}
Shasha	2.33 ± 0.33^{cd}	3.33 ± 0.33^{cd}	0.00 ± 0.00^{a}
Ilesha Park	2.33 ± 0.33^{cd}	0.00 ± 0.33^{a}	1.00 ± 0.57^{a}
Fungi load on carrot obtained from motor parks and fruits vendor in Ado-Ekiti Metropolis			
	A. niger	***A. flavus***	
Control	4.00 ± 1.15^{c}	1.66 ± 0.33^{c}	
Basiri	NS	NS	
Oja Oba	1.00 ± 0.07^{ab}	1.00 ± 0.00^{b}	
Ijigbo	NS	NS	
Shasha	2.00 ± 0.05^{b}	0.00 ± 0.00^{a}	
Ilesha Park	NS	NS	
Fungi load on Date fruit			
	A. niger	***A. flavus***	
Control	2.00 ± 0.00^{bcd}	1.33 ± 0.33^{bc}	
Basiri	NS	NS	
Oja Oba	1.00 ± 0.57^{ab}	0.00 ± 0.00^{a}	
Ijigbo	NS	NS	
Shasha	1.66 ± 0.33^{abc}	1.00 ± 0.57^{b}	
Ilesha Park	NS	NS	
Fungi load on Tiger nut			
	A. niger	*A. flavus*	
Control	7.00 ± 0.57^{d}	2.00 ± 0.00^{ab}	
Basiri	NS	NS	
Oja Oba	1.66 ± 0.33^{b}	1.00 ± 0.57^{a}	
Ijigbo	NS	NS	
Shasha	0.00 ± 0.00^{a}	0.33 ± 0.88^{a}	

*Table presents mean values ± standard error of the mean. *NS –Not Sampled* Values with the same letter along the same column are not significantly different at $P<0.05$ using Duncan's new multiple range test.

Fungi load on Date plant fruit: Two species of fungi were isolated which were *A. niger* and *A. flavus*. Date plant fruit was not available for sampling at Basiri motor park, Ijigbo and Ilesha motor park in Ado-Ekiti. A. niger colony court was also observed from a sample from Oja Oba in Ado-Ekiti (Table 3).

Fungi load on Tiger nut: The *A. niger* mean value of 70 $\times 10^{-1}$ was observed to be highest in the control sample of tiger nut and lowest in the sample from Oja Oba in Ado-Ekiti with a mean value of 1.66. Lowest CFU value in the sample plate from Shasha in Ekiti. However, the sample of tiger nut was not available at Basiri, Ijigbo park, Ilesha motor park, Ado-Ekiti (Table 3).

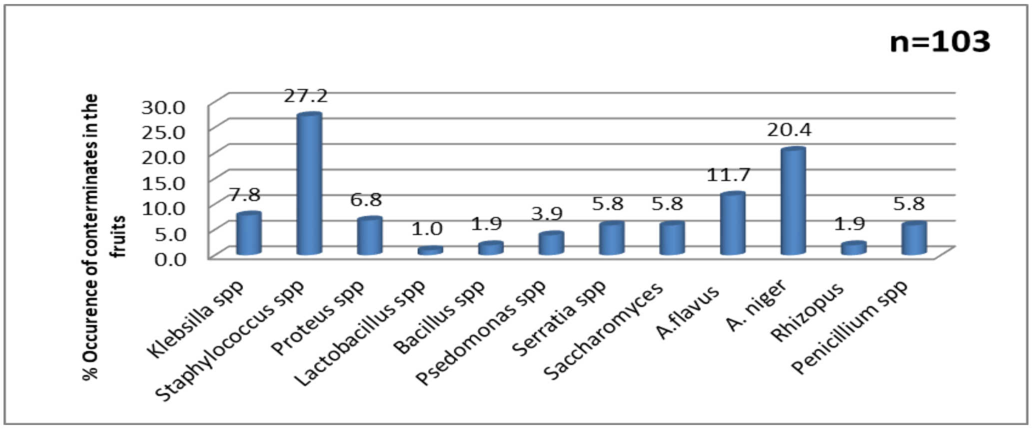

Fig 1: Occurrence of contaminants in the fruits/nuts samples.

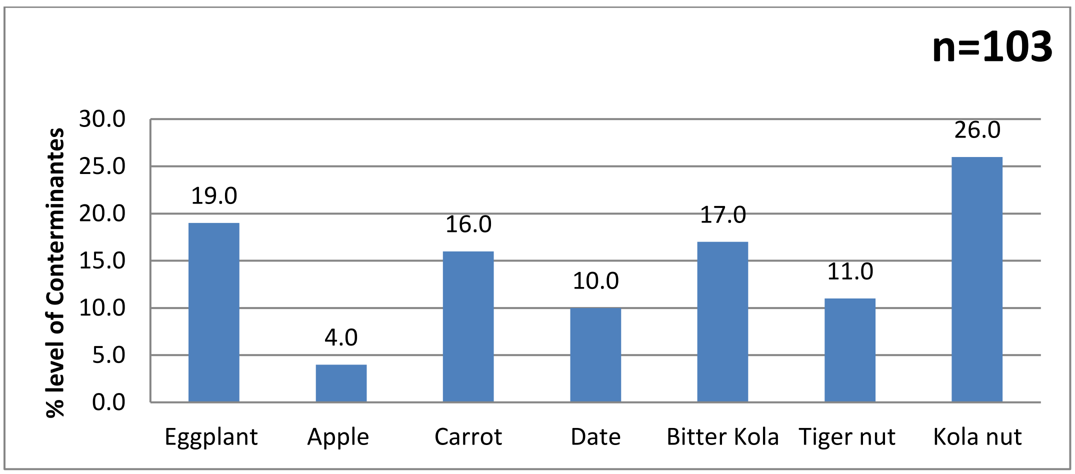

Fig 2: Contamination level of the fruit and nut collected from commercial motor packs in Ado Ekiti

Anti-Nutritional Screening: The anti-nutritional composition was analysed in bitter kola, apple, date, tiger nut, carrot, eggplant and kola nut. Bitter kola had the highest concentration of tannin at 0.258mg/g, while the lowest concentration of tannin was assayed in kola nut at 0.0024mg/g. Bitter Kola was observed to have the highest phenol concentration of 0.282mg/g, while the lowest phenol concentration of 0.017mg/g was recorded in kola nu (Table 4). The highest alkaloid concentration of 3.353% was recorded in kola nut and lowest concentration of 0.025% in garden egg. The oxalate concentration of 5.267mg/g was highest in date plant fruit while kola nut had the lowest concentration of oxalate. Bitter kola had the highest concentration of cyanide was assayed in egg plant at 0.33mg/g (Table 4).

Table 4: Anti-nutritional composition of fruits, nuts and root tuber sold around Ado-Ekiti metropolis.

Fruits	Tannin (mg/g)	Phenol (mg/g)	Alkaloid (mg/g)	Oxalate(mg/g)	Cyanide (mg/g)
Bitter kola	0.258 ± 0.017^{c}	0.282 ± 0.129^{b}	0.063 ± 0.003^{a}	0.880 ± 0.173^{b}	2.025 ± 0.000^{g}

Apple	0.124 ± 0.001^{c}	0.105 ± 10.000^{a}	0.090 ± 0.058^{a}	2.726 ± 0.026^{d}	1.013 ± 0.000^{c}
Date	0.101 ± 0.002^{b}	0.125 ± 0.001^{ab}	0.210 ± 0.058^{b}	5.267 ± 0.355^{e}	6.66 ± 0.66^{b}
Tiger Nut	0.229 ± 0.002^{d}	0.147 ± 0.002^{ab}	0.110 ± 0.006^{ab}	1.957 ± 0.882^{c}	1.688 ± 0.000^{e}
Carrot	0.102 ± 0.000^{b}	0.126 ± 0.015^{ab}	0.130 ± 0.851^{ab}	1.290 ± 0.288^{b}	0.675 ± 0.000^{b}
Garden Egg	0.125 ± 0.015^{c}	0.150 ± 0.001^{ab}	0.025 ± 0.003^{a}	1.200 ± 1.670^{b}	0.338 ± 0.000^{a}
Kola nut	0.024 ± 0.015^{a}	0.017 ± 0.000^{a}	3.353 ± 0.291^{c}	0.225 ± 0.026^{a}	1.856 ± 0.600^{f}

The table presents the means of three replicates ± standard error of the mean. Values with the same letter along the same column are not significantly different at $P<0.05$ using Duncan's new multiple range test.

4. Discussion

The study has shown the presence of some microorganisms in the studied fruits and nuts. These microorganisms include bacteria such as *Proteus spp, Klebsiella spp, Staphylococcus spp, Lactobacillus spp, Bacillus spp, Pseudomonas spp, Serratia spp* and fungi (*Saccharomyces, A. flavus and A. niger*). The findings in this study showed that *Staphylococcus spp*. (27.2%) was the most frequently isolated organism in all the fruits. The prevalence of *Staphylococcus* spp observed in this study agrees with Iyoha & Gorey (2015). The high prevalence could be due to the fact that these organisms are normal flora of the human skin and nasal cavity and might have entered the fruit during packaging (Iyoha and Agoreyo, 2015). Many of the fruit's vendors' sampled display their fruits and nuts on tables in the open square without covering and often touch them with the same hand that had been used to collect dirty money (Douye *et al.*, 2013). Poor unhygienic hands could be a factor contributing to the high microbial load. The dusty environments of the motor parks and busy roads, coupled with water of questionable quality which are often sprinkled on the fruits to keep them fresh may also be parts of contributing factors that can the survival and possible multiplication of contaminants on fruit surfaces (Oranusi *et al.*, 2012).

Another important sources through which the microbes might have got to the F&Ns are transportation and packaging (Hunter, 1993). The presence of *Staphylococcus* spp. in fruits is of public health significance because it is associated with Staphylococcal food poisoning, severe soft tissue infections, and toxic shock syndrome (TSS) (Weems *et al.*, 2001). Vendors of fruits and nuts usually make use of simple facilities like wheelbarrows, trays, tables, thus further increasing the risk of food contamination. The presence of *Staphylococcus* spp. in fruits may be a reflection of storage conditions for some of the samples and how long the produce was stored before they were obtained for sampling (Oranusi and Nubi,2016). Isolation of *Pseudomonas aeruginosa* in fruit samples is of public health concern. This is because the bacterial in concern is highly pathogenic and causes several diseases. The most serious infections include malignant external otitis, endophthalmitis, endocarditis, meningitis, pneumonia, and septicemia. According to Aguoru *et al* (2015), it causes between 10% and 20% of infections in most hospitals (Aguoru *et al.*, 2015).

The occurrence of *Bacillus*, *Penicillium* and *Aspergillus* spp. in this study could be associated with the presence of their spores in the surrounding air These microbes has been reported to be responsible for some food borne infections (Eni *et al.*, 2010; Aboloma, 2018). *Pseudomonas, Klebsiella,* and *Proteus* spp. are environmental contaminants; they have been isolated from plants, human skin, animal and dairy products. Their presence on the ready to eat fruits could be through unclean hands of the vendor and contact with contaminated water (Beuchant, 1995; Beuchant *et al.*, 1995). The presence of these microorganisms, no matter the level, raises considerable public health concern because of their pathogenicity. Most of the isolates found on the fruits and nuts have been reported to cause foodborne disease (Wanyenya *et al.*, 2004). Cross-contamination of food during preparation has been identified as an important factor associated with food-borne illness (Wanyenya *et al.*, 2004). This calls for concern as these organisms are frequently associated with poor sanitary practices and could be a pointer to the danger of possible food borne infection. The involvement of many of these pathogenic bacterial and fungi in the majorities of reported food related illnesses make an important to constantly evaluate their microbial load and prevalence in ready to eat fruit, and nuts Oluwadamilola *et al.* (2020). Many studies have shown fruit handlers as one of the sources of contaminations in fruits and nuts especially as regard to enterotoxin-producing *S. aureus* couple with their poor storage system (Madueke *et al.*, 2014; Zahra *et al.,* 2016).

According to Aneta & Mihaylova, (2019) antinutritional factors are primarily associated with compounds or substances of natural or synthetic origin, which interfere with the absorption of nutrients and act to reduce nutrient intake, digestion, and utilization and may produce other adverse effects. The level of antnutritional compound observed in this present study agree with previous findings. According to Ekpa & Sani (2017), Anti-nutritional factors are present in different food substances in varying amounts depending on the kind of food, mode of its propagation, chemicals used in growing the crop as well as those chemicals used in storage and preservation of the food substances (Umaru *et al.*, 2007). The nutritional importance of a given food depends on the nutrients and anti-nutritional constituents of the food (Aletor *et al.,* 2017).

Also, Date plant recorded the highest values of Oxalate, Kola nut recorded the highest values of Alkaloids while bitter kola recorded the highest values of Cyanide. The values of Oxalate recorded in this research is evidence that utilization of Date palm might not have any negative impact that is connected with abundance utilization of Oxalate, for example, complex development with divalent metals, which may have an impact on the natural action of the metal particles in the body. Oxalates have been reported to have a negative impact on the accessibility of some minerals which prompt assimilation of essential minerals into the body particularly calcium by forming insoluble salts (Wasagu *et al.*, 2013).

The concentration of the value of saponins from the analysis in this study is within the WHO permissible limit (48.50mg/100g) (WHO. (2003). High saponin level has been associated with gastroenteritis manifested by Diarrhea and dysentery (Ekpa & Sani, 2017). However, it was reported that saponin reduces body cholesterol by preventing its reabsorption and suppresses rumen protozoan by reacting with cholesterol in the protozoan cell membrane thereby causing it to lyse. Tannin in fruits imposes an astringent taste that affects palatability, reduce food intake and consequently body growth (Muhammad *et al.*, 2011). It also binds to both exogenous and endogenous proteins including enzymes of the digestive tract, thereby affecting the utilization of protein (Ali *et al.,* 2009).

5. Conclusion

The result shows a low level of most frequently isolated bacterial and fungal floral of fruits and nuts, the result from this investigation reviewed that the fruits and Nuts from these selected Motor parks in Ado Ekiti are safe for consumption although more hygienic practices are still encouraged among the fruit vendors. Are most frequently isolated floral of the fruits and nut. The anti-nutritional factors values observed in the fruits analyzed in this research suggests that this fruit may be safe for consumption. This indicates that the fruit can be used effectively since the anti-nutritional composition is low and there would be no interference with the nutrient like protein and minerals in the body.

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