IPS Journal of Nutrition and Food Science
IPS J Nutr Food Sci, 3(2): 131-134 (2024)
DOI: https://doi.org/10.54117/ijnfs.v3i2.42

Identification of Foodborne Pathogens on Four Species of Tomatoes (*Lycopersicon esculentum*)

Christiana Ngozi Opara[1] and Kennedy Ahamefula Okoronkwo[2]

[1]Department of Microbiology, Faculty of Science, Federal University, Otueke, Bayelsa State, Nigeria.
[2]Department of Food Science and Technology, Faculty of Science and Computing, University of Agriculture and Environmental Sciences, Umuagwo, Imo Sate, Nigeria.

**Corresponding author:* xtyo2000@gmail.com

Abstract

Tomato (*Lycopersicon esculentum)* contains large amounts of water which makes it more susceptible to foodborne pathogens especially (bacteria and fungi). These pathogens produce *mycotoxins* that are detrimental to human health. This study was therefore carried out to identify foodborne pathogens, of tomatoes sold in Swali market, Bayelsa state, Nigeria. The isolates from the tomatoes after culturing with Potato Dextrose Agar using pour plate method are: *Escherichia coli, Shigella, Salmonella, Enterobacter, Proteus, Corynabacter, Staphylococcus, Pseudomonas, Fusobacterium, Lactobacillus, Vibrio, Streptomyces, Bacillus cereus and Clostridium*. While the macroscopic and microscopic examinations were used to identify the morphologies of the fungi isolated from the tomatoes. The following were isolated; *Vericolor, flavus, Emericella rugulosis, Rhizopus spp, Aspergillus phoreolina, Micropholina phaseolina, Rhizopus solarin, Penecillum oxalina* and *mould*. The percentage occurrence of the isolates from all the markets locations were; *flavus* 33.33%, *vugulosin* 33.33%, *Microphoslina* 33%, *rugulosis* 67% and *Rhizopus* spp 13.33%. The decay diameter of *flavus i*s 18mm, the decay diameter of *Microphoslina* is 15mm, while the decay diameter of *Rhizopus* spp is 11mm. Proper handling and adequate storage facilities must therefore be employed to prolong the shelf life of tomatoes.

Keywords: *Tomatoes, Foodborne pathogens, Mycotoxins, Swali market, Bayelsa state*

Article History
Received: 26 Jul 2023
Accepted: 12 Aug 2023
Published: 08 Mar 2024

Scan QR code to view♦

How to cite this paper: Opara, C. N., & Okoronkwo, K. A. (2024). Identification of Foodborne Pathogens on Four Species of Tomatoes (Lycopersicon esculentum). *IPS Journal of Nutrition and Food Science*, *3*(2), 131–134. https://doi.org/10.54117/ijnfs.v3i2.42.

1. Introduction

Tomatoes are exceptional dietary source of nutrients and fiber for human beings, hence vital for good health and fitness (Abarca *et al*., 2015). The importance of fruit in human nutrition cannot be overestimated as it provides essential growth factors such as vitamins and minerals necessary for proper body metabolism (Abd-Allah, 2013). Humans and many animals have become dependent on tomatoes and vegetables as a source of food (Liu, 2010). However, tomatoes and vegetables are easily spoilt and usually have active metabolism during harvest, transportation, sales and storage stage. The high concentration of various sugars, minerals, vitamins, amino acids, and low pH also enhances the successful growth and survival of various parasitic and saprophytic forms of fungi and bacteria (Denton, and Swarup, 2014).

Tomato contamination refers to several changes which make the food to be toxic and less palatable to consumers, and these could be associated with alterations in appearance, texture, taste or smell (Agrios, 2005; Sylwia and Tokarczyk, 2022). Tomatoes (*Lycopersicon esculentum*) which is of juicy flesh endocarp belonging to the fruit class, berry are naturally very rich in vitamins, minerals, dietary fiber and protein are however classified as either fruits or vegetable, they do not only serve as fruits/vegetables for food but as medicine, nutrient supplement, flavoring ingredient, detoxificant and human system cleanser. It is a perishable food that is widely cultivated and consumed worldwide (Villareal, 2008; Alissa, 2023). It is the third most cultivated and world widely grown vegetable crop and rich in nutrients, vitamins, dietary fibers, and phytochemicals. It is known to be a very profitable crop that provides high returns for small scale farmers in most developing countries (Adeoye *et al*., 2009; Popescu *et al*., 2022). Due to its nutritive value, taste, affordability, and accessibility, there has been an increase in demand by consumers. However, isolation and identification of microorganisms that are associated with the contamination of tomatoes have gained some research focus as the household consumption is on high increase worldwide. The microbial deterioration activities on tomato fruits propagated by microorganisms such as *Bacillus, Staphylococcus spp, Escherichia coli, Salmonella spp., Enterobacter spp., Citrobacter spp., Acinetobacter spp., Klebsiella spp., Aeromonas spp., Listeria spp., Micrococcus spp., Aspergillus niger* and *Penicillium notatum* causes reduction in market values and nutritional qualities, and at times rendered the fruits

non-fit for consumption. This is due to contaminations with exotoxins and mycotoxins (naturally occurring toxic chemical usually of aromatic structure) that produces aflatoxins in human, following inhalation or ingestion and so resulting to food poisoning. In developing countries at the open markets, tomatoes are often displayed in baskets and on benches for the prospective customers, thereby exposing them to opportunistic microbial infections (Mariga *et al*., 2014). The proliferation of bacteria more especially in fresh and damaged tomatoes could be considered to be more harmful when such contaminated tomatoes are consumed in improperly cooked food (Flood, 2016).

Some studies have been carried out on bacteria associated with tomato and tomato products in some countries. A study carried out by (Bugel, 2013) in the United State has revealed that *Clostridium spp., Staphylococcus spp.,* and *Bacillus spp*. were predominant bacteria isolated from both canned and raw tomatoes. In India, a study carried out on tomato puree revealed the presence of *Klebsiella spp., Proteus mirabilis, Vibrio spp., and Pseudomonas sp*. (Garg *et al*., 2013). In Nigeria, Wilson *et al*., 2007 isolated *Bacillus subtils, Klebsiella aerogenes, Pseudomonas aeruginosa, Salmonella typhi, Proteus mirabilis, and Staphylococcus aureus* from spoilt tomatoes in Benin City. A similar study also revealed high levels of *Staphylococcus* spp. (22.5%), *Bacillus* spp. (20%), and *Escherichia coli* (15%) in Lagos State, Nigeria (Snowdon, 2016). The succulent nature of the tomatoes requires peculiar storage conditions to prevent microbial infection as microbiological diseases are a major limiting factor for tomato production and availability and could be contagious, hence, may spread from plant to plant, often very rapidly when environmental conditions are favorable (Duarte, 2011) as foods of plant origin such as fruits and vegetables have heterogeneous characteristics with regard to their compositions, environment and short shelf-life and tomatoes (*Lycopersicon esculentum*) has some of this heterogeneous characteristics that consequently makes it easier for microbial contamination by these pathogenic microorganisms either during their growing in fields, orchards, vineyards, or greenhouses, or during harvesting, postharvest handling, distribution, sales and storage (Wang *et al*, 2008).

Therefore, the safety of this food is thus called to question, as regulatory bodies in Nigeria do not enforce sanitary conditions in handling and preservation of food. Also, without much formal education on hygiene and sanitation, the food vendors acquire their handling techniques traditionally, considering the above conditions in which this edible fruit is handled and sold. It is expected that the safety of this food is not guaranteed, thus not meeting the health standard. Since there is a rapid increase of the already large number of people involved in the consumption of tomatoes (*Lycopersicon esculentum*) in the study area, there is every need to have first-hand information on the safety of these fruits by investigating and identifying the type and volume of potential foodborne pathogenic microbes present and capable of causing rot spoilage in tomatoes (*Lycopersicon esculentum*) and foodborne disease in human.

However, this study seeks to experimentally investigate, isolate and identify potentially pathogenic foodborne microorganisms associated with four species of raw tomatoes (*Lycopersicon esculentum*) samples. Also, this research seeks to ascertain this assertion in tomatoes sold in the major market of Swali market in Bayelsa State Nigeria.

2. Materials and Methods

Description of Study Area

Swali market is located in Yenagoa, Bayelsa State in the South-South geographical zone of Nigeria. It is geographically located within the latitude coordinates of 4º 15' N and latitude 5º 23' South and longitude of 5º 22' West 6º 45' East. The sample study area which is the community market of Swali is an ever busy market uniquely sits on the bank of Yenagoa River in the Bayelsa State capital.

Sample Collection

Four species of tomatoes sample (*Lycopersicon esculentum)* which include Plum (Roma or paste tomato), Grape (super sweet baby tomato), Beefsteak (green tomato), and Cherry (black tomato), were randomly collected in triplicates from Swali market to minimize experimental errors in analysis. Market location were labeled as site location A, site location B, site location C and site location D where individual tomato specie were collected. A total number of twelve (12) tomatoes samples were collected, (3) three for each species into a sterile polythene bag. The samples were labeled according to the specie collected and the site collection location.

Microbial (Bacterial) Enumeration

After 24 hours of incubation, the discrete bacteria colonies on each of the plates were counted, using a digital colony counter and the media plates having microbial colonies within the range of 25-250 were calculated for their colony forming unit (CFU) per gram (g). Nutrient agar was used for total bacteria count and *Staphylococcus* count, Salmonella Shigella agar was used for total Salmonella/Shigella counts, MacConkey agar for total gram negative bacterial count, potato dextrose agar was used for fungi identification and counts. Frequency of individual microbial species was calculated in percentage as follows; Microbial frequency (%) = number of colony of the species appeared ×100/ Total number of all colony isolated from each sample.

Sample Preparation

The tomatoes samples were washed and rinsed with clean water to remove surface dirty. The tomato samples were also sterilized with 70% ethanol to remove surface microorganisms and to prevent cross contamination of the sample with external bacteria pathogen. Preparation of stock for serial dilution were done and with some modifications by homogenizing the edible portion of the samples. The tomato were homogenized by adding 9ml of sterile peptone water in a heat sterilized glass cup until a homogenous slurry mixture was obtained, this was then filtered and used to determine the pH values of the samples. One gram (1g) of the slurry was weighed and then added to sterile glass tube containing 9ml peptone water.

Media Preparation

The media used for the study were; Nutrient Agar, MacConkey Agar (MCA), Potatoes Dextrose Agar (PDA), Salmonella Shigella Agar (SSA). The agar media were prepared mainly based on the manufacturer's instructions, heated in water bath till the agar powder melted, and the medium was sterilized in

an autoclave and was kept in the incubator over the night for sterility test and kept in refrigerator for further use.

Statistical Data Analysis

The mean and standard error of the colony counts were calculated using Microsoft (Excel, 2016) for each sample specie and results were presented in tables. Data generated were subjected to standard statistical test using statistical package for social science (SPSS) version 17.

3. Results and Discussion

The total viable count was done using MacConkey agar (Oxoid, England) by streak plate method. Among the various samples of tomatoes analyzed, beef steak [green tomatoes] samples sourced from Swali market location B had the highest total viable count of 2.9 x 108 cfu/g (Table 1). This was higher than the total viable count of 1.9 x 106 cfu/g for tomatoes samples reported by Umeh and Oyedun (2015). Haighton et al. (2012) suggested that a limit of 10cfu/g should be standard with market raw tomatoes. This finding implies that since tomatoes used for cooking usually harvested from the farm, hence can become contaminated by pathogenic organisms in the farm. Among the four different tomatoes samples analyzed, Cherry samples had the lowest bacterial load. The highest total viable count for Grape [super tomatoes] samples was sourced from swali market location C with a load of 1.6 x 102 cfu/g while those sourced from same Swali market location A had the load of 1.6 x 103 cfu/g. This result was comparable to the bacterial load of 1.9 x 106 cfu/g reported by Abd-Allah et al. (2013). Unlike tomato A and tomato D are rarely contaminated. Contamination with these pathogens could be due to poor hygiene practices by handlers. This was higher than a load of 6.9 x 106 cfu/g reported for tomatoes samples by Mendgen et al. (1996). The high bacterial load in tomato B can be attributed to the large surface area of the tomatoes suitable for water contact, making them susceptible to bacterial contamination.

Table 1: Total viable count for tomatoes

No of sampling	Sampling site	Type of Sampling	Total Viable Count (cfu/g)
1	Swali Market (location A)	Plum (Roma tomatoes)	1.6×10^3 SD 5.20×10^7
2	Swali Market (location B)	Beefsteak (green tomatoes)	2.9×10^8 SD 5.70×10^3
3	Swali Market (location C)	Grape (super tomatoes)	1.6×10^2 SD 4.50×10^8
4	Swali Market (location D)	Cherry (black tomatoes	1.50×10^1 SD 3.90×10^1

Key: SD = Standard deviation

The biochemical test (Table 2) indicates the presence of *E-coli, Shigella, Lactobacillus, Enterobacte, Proteuss, Corynabacter, Staphylococcus, Pseudomonas, Fusobacterium, Clostriduim, Bacillus cereus, Streptomyces, Vibrio. S. aureus* observed in the tomato samples is of serious public health importance because of its ability to cause a wide range of infections especially food- borne intoxication (Talvas *et al*., 2010). Contamination with *S. aureus* has been linked to carriage in nasal passages of food handlers or by infected workers. The presence of *S. aureus* and some Gram negative rods have been reported to contaminate some tomatoes such as Plum [Roma tomatoes), Beefsteak, Grape and Cherry (Baker, 2016). The presence of *E. coli* in the analyzed samples is indicative of faecal contamination. *E. coli* are part of the normal flora of the human intestines. Some strains of *E. coli* have been linked to diarrhoea, gastro-enteritis and urinary tract infections (Hongyin *et al*., 2011). *Proteus* is second only to *E. coli* as a urinary tract pathogen. It is well known in the environment and can be cultured from soil, water and vegetables when consumed raw as in tomatoes.

Table 2: Results of biochemical tests of the pathogenic isolates

S/n	Motility	Oxidase	Catalase	Indole	Citrate	Grams staining	V.P	Organisms
1	+	-	+	+	-	-	-	*E.coli*
2	-	-	+	-	-	-	-	*Shigella*
3	+		+		-		-	*Salmonella*
4	+	-	+	+	-	-	+	*Enterobacte*
5	+	-	+	-	+	-	-	*Proteuss*
6	+	-	+	-	+		-	*Corynabacter*
7		-	+	+	-	+	+	*Staphylococcus*
8	+	+	+	-	+	-	-	*Pseudomonas*
9	+	-	+	+	-	--	--	*Corynaebacter*
10	--	--	-	+		-	--	*Fusobacterium*
11	--	--	+	--	--	+	--	*Lactobacillus*
12	+	+	+	+	+	--	--	*Vibrio*
13		+	+		+		--	*Streptomyces*
14	+	--	+	--	+	+	+	*Bacillus cereus*
15	+		--	+		+		*Clostriduim*

Key: + = Positive; - = Negative

The occurrence of fungi in the sample (Table 3) is due to the pathogen in the environment, the heavy presence of *Microphomina phaseolina, Aspergillus terreous, Emericella regulasis* in grape (super) tomatoes can be attributed to the concentration of the pathogen in the sample from the environment. Beef steak (green) tomatoes and cherry (black) tomatoes has less concentration of the fungal count, this is due to their small surface area in the environment.

Table 3: Number of occurrence for Fungal Counts

Samples		*Organisms*
Plum (Roma tomatoes)	1^A	*Microphomina phaseolina*
	1^B	*Microphomina phaseolina, Aspergillus flevus*
	1^C	*Microphomina phaseolina, Aspergillus*
Beefsteak (green tomatoes)	2^A	*Aspergillus vsicolor*
	2^B	*Microphina plasenta*
	2^C	*Emericella vugulosis*
Grape (super tomatoes)	3^A	*Microphomina phaseolina, Rhizopus solarin*
	3^B	*Microphomina phaseolina, Aspergillus terreous, Emericella regulasis*
	3^C	*Microphomina phaseolina, Rhizopus solarin*
Cherry (Black tomatoes)	4^A	*Penicillium oxolicia*
	4^B	*Emericella regulasis*
	4^C	*Aspergillus flevus mould*.

4. Conclusion

The present study showed that many bacterial and fungal pathogens are associated with tomatoes. In this investigation, *Aspergillus veguralsis*, *Aspergillus flavus*, *Microphsolila* spp, *Rhizopus spp and mould* were found.

Declarations

Competing Interest

The authors declare no competing of interest.

References

Abarca, M., Bragulat, M., Castellá and G., Cabañes, F. (2015). "Ochratoxin A production by strains of *Aspergillus niger var. niger". Applied Environmental Microbiology*. 60(7):2650-2.

Abd-Allah, E. F., Ezzat, S. M. and Tohamy, M. R. (2019). *Bacillus subtilis* as an alternative biologically based strategy for controlling fusarium wilt disease in tomato: A histological study. *Phytoparasitica*. 3**5:** 474-478.

Adeoye, I. B., Odeleye, O. M. O., Babalola, S. O. and Afolayan, S. SoO. (2009). Economic Analysis of tomato losses in Ibadan metropolis, Oyo State, Nigeria. *African Journal of Basic and Applied Science*. **1:**87-92. 144.

Agrios G.N. (2005). Plant pathology, Academic press, New York, USA.

Alissa Dillon (2023) Navigating California's 2024 Tomatoe crop with caution and optimism.

Baker, R. J. and Pusey, C. D (2016). The changing profile of acute tubulointerstitial nephritis. *Nephrol Dial Transplant*.19**:** 8-10.

Bartz J. A., Sargent, S. A. and Mahovic, M. (2016). Guide to Identifying and Controlling Postharvest Tomato Diseases in Florida. Horticultural Sciences Department, Florida Cooperative Extension Service. University of Florida/IFAS, Gainesville.

Brandwagt, and Bas, F. (2019). "A longevity assurance gene homolog of tomato mediates resistance to *Alternaria alternata f. sp. lycopersici* toxins and fumonisin B1". Proceedings of the National Academy of Sciences. 94**(9):**4961-4966.

Bugel, S. (2013). Vitamin K and bone health. Proc. Nutri Soc.CABI. (2011) Distribution Maps of Plant Diseases CABI, Wallingford, UK.

Denton, O. A. (2014). Tomato cultivation and its potential in Nigeria. *Acta Hort*, **123:** 257-263.

Duarte, S. C., Pena, A. and Lino, C. M. Human ochratoxin A biomarkers--From exposure to effect. *Critical Rev. in Toxicology*. (2011); **41:** 187-212.

Flood Julie. A review of Fusarium wilt of oil palm caused by *Fusarium oxysporum f.* sp. *elaeidis*. *Phytopathology* (2016); 96(6): 660-662.

Hongyin, Zhang., Renping, Li. and Weimin, Liu, (2011). Effect of chitin and its derivative chitosan on postharvest decay of fruits. *International Journal of Molecular Science*. 12**:** 917-934.

Kader, A. A. (2016). Effects of postharvest handling procedures on tomato quality. Acta Horticulture. 190: 209-221.

Kutama, A. S., Aliyu, B. S. and Mohammed, I. (2017). Fungal pathogens associated with tomato wicker baskets. *SWJ*. **2:** 38-39.

Liu, Y., Wu, F (2010). Global burden of aflatoxin-induced hepatocellular carcinoma: a risk assessment. *Env. Health Perspection* **118:** 818-824.

Mariga, M., Neri, F. and Bertolini, P. (2014). Novel approaches to prevent and control postharvest diseases of fruits. *Stewart postharvest*. **3:** 1-7.

Mendgen, K., Hahn, M., Holger Deising (1996). Morphogenesis and mechanisms of penetration by plant pathogenic fungi. *Annual review of phytopathology*.34 (1)**:** 367-386.

Olanrewaju, D. and Swamp, V. (2016). Tomato cultivation and its potential in Nigeria. National Horticulture Res Institute Bulletin. Ibadan, Nigeria. 20pp.

Olayemi, F. F., Adegbola, J. A., Bamishaiye, E. I. and Daura, A. M. (2012). Assessment of postharvest challenges of small scale farm holders of tomatoes, Belland Hot pepper in some LGA of Kano State Bayero. *Journal of pure and Applied Science*. **3:** 39-42.

Popescu, M., Iancu, P., Plesu, V., Maria-Cristina Todasca, Isopencu, G and Costin Sorin Bildea (2022) Valuable Natural Antioxidant Products Recovered from Tomatoes by Green Extraction. *Molecules*. 27(13): 4191

Snowdon, A. L. (2016). Color Atlas of postharvest diseases and disorders of fruit and vegetables. Vol. 2. Vegetable.

Sylwia P. and Tokarczyk, G (2022) Lycopene in the prevention of Cardiovascular Diseases. *Int. Journal of Molecular Sciences*. 24(4):1957

Talvas, J., Caris-veyrat, L., Guy, M., Rambeau, B., Lyan, R. and Minet-Quinard, J. A.(2010) Differential effects of lycopene consumed in tomato paste and lycopene in the form of a purified extract on target genes of cancer prostatic cells. *American Journal of Clinical Nutrition*. 91(6): 1714

Umeh, V. C. and Oyedun, O. (2015). Effects of spacing, staking and insecticide applications on the abundance of tomato fruit worms Spodoptera and Helicoverpa species and on infection by *Alternaria* sp. Proceeding 10th Annual Conference of the Horticultural Society of Nigeria, Kwara state ADP, Nigeria.

Villareal,. R. I. (2008). Tomato in the tropics. Wesview Press Boulder, Colorado U.S.A

Wang, Yifei., Bao, Yihong., Shen, Danhong., Feng, Wu., Yu, Ting., Zhang, Jia and Zheng, Xiao Dong, (2008). "Biocontrol of *Alternaria alternata* on cherry tomato fruit by use of marine yeast *Rhodosporidium paludigenum* Fell & Tallman". *International Journal of Food Microbio*. 123(3)**:** 234-239.

Wilson.C. L and Wisniewski, M. E. (2007). Biological control of postharvest diseases of fruits and vegetables: an emerging technology. *Annual Review on phytopathology*. **27:** 425-441.