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EFFECT OF NEEM LEAVES AND GARLIC BULBS EXTRACTS ON BACTERIA CAUSING POST-HARVEST SPOILAGE OF TOMATOES AT SOKOTO

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ABSTRACT
This study attempts to overcome the challenges of postharvest losses of tomato fruits by examining some botanical extracts as alternative means to the use of synthetic chemicals. Step by step procedures were followed during conducting this work. Two locations were chosen, Sokoto and Shuni, and the samples of spoiled tomato fruits were collected randomly, materials that were used for the analysis were sterilized. bacterial pathogens were isolated at Microbiology Laboratory respectively. The pathogens were also identified after isolation. The identified pathogens were, Saccharomyces cerevisiae, Enterobacter aurogene, Citrobacter freudii Providencia spp. Preparation of crude extracts of neem leaves and garlic bulb and determination of the efficacy of these extracts on identified pathogens were conducted. Each identified pathogens were treated with three different concentrations of both neem and garlic extracts, at 100mg/ml, 200mg/ml, and 300mg/ml and streptomycin was used as control. The zones of inhibition were observed after 24hours in each and recorded in each sample.

KEYWORDS: *Neem Leaves and Garlic Bulbs Extracts, Bacteria, Post-Harvest Spoilage, Tomatoes*

INTRODUCTION

Tomato (*Solanum lycopersicum* L.) belongs to the family *solanaceae* and it is an annual sub-tropical fruit vegetable crop. The crop originated from South America and was introduced to Europe in the 16th Century and later to East Africa by colonial settlers in early 1900 (Wamache, 2005). Nigeria is the 14th largest producer of tomatoes in the world. On the continent, the country is ranked second after Egypt with about 1.8 million metric tons (MT), which she produces annually. With over 48 million tomato farmers across the country, Nigeria accounts for 65 per cent of tomatoes produced in West Africa (Eno-Abasi *et al.*, 2018). Ironically the country is the largest importer of tomato paste in the world, importing an average of 150,000mt of concentrate per annum, which value at $170m. (Eno-Abasi *et at.*, 2014) Tomato plays a vital role in meeting the nutritional food requirements, generation of income, foreign exchange earnings and creation of employment (Sigei *et al.,* 2014). The crop is grown for fresh domestic but there is increasing demand for processed tomato products (Mungai *et al.*, 2000). The crop is grown either on open field or under greenhouse technology. Open field production account for 95 % while greenhouse technology

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accounts for 5% of the total tomato production (Seminis, 2007). Tomato crop does well in warm climate with an altitude range of 0 – 2100m above sea level. It requires rainfall ranging between 760 mm to 1300 mm and deep fertile loam soil that is well drained, with high content of organic matter and a pH ranging between 5-7 (Rice *et al.,* 1994). Fruits are used in salads or cooked as a vegetable, processed into tomato paste, sauce and puree. The nutritional value of tomato makes it a widely accepted vegetable by consumers. Fruits are rich in calcium, phosphorus, magnesium, copper, niacin, iron, folate, Vitamin A, B6, Vitamin E, Vitamin B2, Vitamin C, iron and carbohydrates (Wamache, 2005). Furthermore, the fruit has medicinal value as a gentle stimulant for kidneys, and washing off toxins that contaminate the body systems. It improves the status of dietary anti-oxidants (lycopene, ascorbic acid and phenols) in diet (George *et al*., 2004). Tomato juice is known to be effective for intestinal and liver disorders (Wamache, 2005).

Tomato production is constrained by factors such as poor pre-harvest practices, adoption of poor production techniques, rough handling and moisture condensation causing pathogen infestation (Kader, 1992). Packaging in bulk without sorting and grading of produce, damage during transport and storage due to mechanical injuries are other factors contributing to post-harvest losses (Kader, 1992). Inadequate storage, distance and time consuming market distribution, poor access to the market, post-harvest spoilage micro-organisms and cultivars disposition to diseases causes high post-harvest losses of tomatoes (Kader, 1992).

According to FAO (2002), records of post-harvest losses do not exist and if available they do not cover enough period of time and the figures are only estimates made by observers. It has been estimated that 20-50% of tomato fruits harvested for human consumption are lost through microbial spoilage while other losses result from damage by dynamic stresses during transit, and through rough handling during loading and unloading (Kader,1992; Okezie, 1998). Thirupathi *et al.* (2006) estimated the magnitude of post-harvest losses in fresh fruits to be 25-80 %. Post-harvest decay remains a major challenge in tomato production. The magnitude of post-harvest losses varies from one country to another, one season to another and even one day to another (Mujib *et al.,* 2007). There are numerous micro-organisms that cause post-harvest decay of tomatoes. Among these, fungi and bacteria are the most destructive.

Most of the tomato fruits are also damaged after harvesting because of inadequate handling and preservation methods (Wills *et al.,* 1981). Fruits, due to their low pH, high moisture content and nutrient composition are very susceptible to attack by pathogenic fungi, which in addition to causing rots, may also make them unfit for consumption by producing mycotoxins (Stinson *et al*., 1981; Moss, 2002). Mycotoxins are potential health hazards to man and animals and in most cases they are unnoticed. Control of fruit rot also remains a major challenge in tomato production.

Tomatoes are an attractive cash crop for small scale farmers and provide potential source of employment to many rural and urban Nigerians. The tomato fruits have been marketed freshly picked from the field and is the bestselling fresh market vegetable crop (AVRDC, 2006). Despite the human need of tomato, damage as a result of post-harvest spoilage micro-organisms has been of serious concern. Microbial decay is one of the main factors that determine losses and compromises the quality of the produce. The extent of the losses especially through microbial decay has not been quantified in most areas and where this has been quantified the results are short lived. Therefore, the study aims at evaluating ways of managing the post-harvest losses of tomatoes using crude plant extracts.

Several kinds of synthetic fungicides have been successfully used to control the post-harvest decay of fruits and vegetables (Adaskaveg *et al.,* 2000; Kanetis *et al.,* 2007). However, there are three major concerns: Firstly, the increasing consumer concern over pesticide residues on foods which are toxic and carcinogenic. Secondly, predominance of fungicide resistant strains of fungi due to excessive use of fungicides and thirdly, environmental pollution.

Therefore, there is need for new effective means of post-harvest disease control that possess less risk to human health and the environment.

Natural plant products and their analogues have been found as important sources of agricultural bio-pesticides which serve as anti-microbial properties of the plant extracts (Cardelina, 1995; Okigbo, 2009). It has been reported that plants are sources of natural pesticides that lead in new pesticide development. Arokiyaraj *et al.* (2008), Shanmugavalli *et al.* (2009), Swarnalatha and Reddy (2009). Anti-fungal and anti-bacterial compounds of neem plant leaf and garlic bulb crude extracts on rot pathogens of post-harvest tomato fruits are the main target of this study.

Objective of the Study

The objective of the study was to evaluate the effect of the selected crude plant extracts on major micro-organisms causing post-harvest spoilage of tomatoes in Sokoto, and specifically the study was aimed at:

- Identification and collection of four spoiled tomato fruits sample from two different locations around Sokoto metropolis

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- Isolation and identification of pathogenic bacteria causing post-harvest spoilage of tomato
- Preparation of neem leaves and garlic cloves extract
- Determining the effect of crude plant extract on growth of bacteria colonies.

MATERIALS AND METHODS

Experimental Location

The research was conducted at Usmanu Danfodiyo University Sokoto, at Microbiology Laboratory for effects of neem leaves and garlic bulbs extracts on the bacterial isolates. Sokoto is located in the Sudan savannah zone in the extreme northwestern part of Nigeria. It lies between latitudes 12^0N, 13^0N and 58^0N and longitudes 4^0E, 8^0E and 5^0E (Mamman *et al.,* 2000). Sokoto has low humidity and high solar radiation, it records annual rainfall between 300mm-800mm and mean temperature of 34.5^{o}C. The dry season temperatures do exceed 45^0C during the day time which is the highest recorded in Nigeria, (Adegboyega, *et al.*, 2016).

Sample Collection

In this analysis, four (4) spoiled tomato fruit samples were collected from different places. Two samples from Sokoto vegetable market (Kasuwan Daji) in Sokoto metropolis, and the remaining two (2) others from Dange Shuni Local Government. Infected tomato fruit samples were identified by physical examination and then collected randomly in a sterilized polythene bags. The samples were then brought to Usmanu Danfodiyo University at Microbiology Laboratory for bacterial analysis.

Sterilization of Materials

Different Laboratory materials that were used for this analysis were first washed with detergent, rinsed with clean water and air dried. The Petri dishes were sterilized in a hot air oven at a temperature of 160^0C for 1hour. For test tubes, 9ml of distilled water were measured using sterilized syringe and poured into the test-tubes. The test tubes were then inserted into an autoclave heater and heated at a temperature of 121^0C for 15minutes.

Media Preparation

Sabouraud Dextrose Agar (SDA) and Nutrient Agar (NA) were the standard media that were used to isolate bacterial pathogens from the tomato samples respectively. Preparations of both Media were conducted according to the manufacturer's instruction.

For bacterial isolation, 2.3g of the NA media was measured using sensitive weighing scale and poured into a conical flask, 100ml of distilled water was also measured using measuring cylinder. It was then heated using hot plate with frequent agitation till it boils to completely dissolve the powder. The media was inserted into an autoclaving for sterilization at 121^0c for 15minutes it was allow to cool and poured into Petri dishes.

Isolation of Pathogenic Bacteria from Rotting Tomato Fruits

For bacterial isolation, about 0.1ml of the aliquot from the serial dilution of the sample was inoculated using sterile syringe. A bend glass rod was then sterilized to spread the aliquot on nutrient agar surfaces and the same procedure was repeated for the other samples. The inoculated plates were incubated inverted at 37^0 C for 24 hours.

Colony Count

Bacterial count was carried out on each of the plate using colony counter to determine the number of bacterial growth. This was obtained by counting the whole plate and using the number obtained to multiply by the dilution factor.

Identification

Gram Staining

This was carried out as described by (Chesbrough, 2000). A drop of water was placed on a clean slide, and a speck of bacterial growth was taken from the culture. The speck was emulsified on the slide to make a thin smear. It was then flooded with crystal violet for 60seconds washed with water, and lugols iodine was added for 30seconds, and washed with water and it was decolorized with alcohol. The smear was finally flooded with safranin to counter stain for 60seconds. It was then washed with water and allowed to air dried. Oil immersion was added and it was viewed at x100 objectives. Gram positive stains purple while gram negative stain red.

Biochemical Tests

Catalase test

Catalase test was carried out as described by (Fawole and Oso, 2001). Catalase is an enzyme which catalyzes the decomposition of hydrogen peroxide to water and oxygen. It will be formed by most aerobic bacteria. A speck of bacterial growth will be transferred with a wire loop to a drop of hydrogen peroxide on the slide. The presence of catalase is indicated by bubbles of gas (Singleton, 1997).

Indole test

Indole test was carried out as described by (Singleton, 1997). Indole test detect the ability of an organism to detect Indole from amino acid tryptophan. A speck of each isolate was inoculated into sterile peptone water enriched with 1%

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tryptophan in a test tube which was inoculated at 37°C for 48hours. After inoculation, 0.5kovac's reagent was added and shaked gently. In a positive test indole was dissolved in the reagent which then becomes pink.

Methyl red test

Methyl red test was carried out as described by (Chesbrough, 2000). Methyl red test detect the ability of an organism growing in a phosphate buffer medium to produce sufficient acid to reduce pH of the medium (Singleton, 1997). A speck of the isolate was inoculated into the medium and incubated at 37^{0}C for 48hours. Few drop of methyl red was added to the culture. Methyl red positive was indicated by red color formation.

Citrate Test

This was carried out as described by (Fawole and Oso, 2001). Citrate detects the ability of an organism to use citrate as sole source of carbon (Singleton, 1997). A speck of each isolate was inoculated into citrate medium in test tube and incubated at 37^{0}c for 48hours. A positive test was indicated by blue color while a green color denote negative.

Urease Test

This was carried out as described by (Chesbrough, 2000). Urease is an enzyme which hydrolyzes urea to carbon dioxide and ammonia (Singleton, 1997). A speck of each isolate was inoculated into urea agar slant and incubated at 37°c for 24hours. Red color was indicated urease positive while yellow color denote negative.

Hydrogen Sulphide Production

This was carried out as described by (Chesbrough, 2000). This test detects the ability of bacterial species to produce hydrogen sulphide (Singleton, 1997). A speck of each isolate will be inoculated by streaking and stabbing into triple sugar ion (TSI) agar and was incubated at 37^{0}C for 24hours. Evolution of blackening on the medium indicates a positive test.

Motility Test

This was carried out as described by (Singleton, 1997). Motility can sometimes be inferred from the way organism growing on solid media. A speck of each isolate was stab on triple sugar phosphate agar which was incubated at 37^{0}C for 24hours. Motility was observed by spreading of the organism outward from the stab area.

After identifying different organisms, both fungi and bacteria on samples collected, the Petri dishes were labeled, wrapped with masking tape and kept in a refrigerator before preparing the media for anti-fungal and anti-bacterial tests.

Preparation of Plant Crude Extracts

Crude plant extracts were obtained from neem leaves and garlic cloves. The extraction process was followed the procedure described by Handa *et al.* (2008). Neem leaves were collected from Usmanu Danfodiyo University Plantation and were brought to Agricultural Physical Laboratory for drying. The leaves were washed under tap water, rinsed in three changes of sterile distilled water and dried using sterile blotting paper. They were then placed in the oven and dried at a temperature of 40^{0}C for three days. Garlic bulbs were obtained from University mini market and brought to the same laboratory. Garlic cloves were peeled washed in sterile distilled water and dried using sterile blotting papers. The cloves were cut into smaller pieces and placed in the oven to dry at a temperature of 40^{0}C for seven days. The neem leaves were grounded to powder using sterile mortar and pestle so as to rapture leaf tissues and cell structures to release the active cell contents. The extracts were placed in sterile specimen bottles. The garlic was also grounded into powder by use of a sterile motor and pestle and place in the sterile specimen bottles. This was done to maximize the surface area which in turn enables the mass transfer of active ingredients from the plant material to the solvent.

Fifty (100gms) of each of the powder was put into separate sterile conical flasks and 300ml of distilled water was added to each of the plant powder ensuring that the powder is completely immersed into the solvent, then it was shaken vigorously and allowed to stand on the bench at room temperature but with continued shaking at different intervals for two days. A sterile funnel was placed into a 500mls conical flask and then a Whitman's (No.2) filter paper was folded and placed into the funnel. The extract was poured gradually into the filter paper and allowed to trickle into the conical flask. The filtrate was then poured into stainless plate and covered with a foil paper. The stainless plates were labeled and taken to biochemistry laboratory, Usmanu Danfodiyo University and dried in an oven at 40°C for four days until a powder like substance remains at the bottom of the stainless plates.

Effect of Crude Plant Extracts on Growth of Bacterial Colonies.

Screening of Extracts for Anti-Bacterial Activities

The agar diffusion method was used. Sterilized nutrient agar plates were prepared and different concentrations of the extraction were also prepared and dissolved in distilled water. 1ml of each

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of the 3 identified organisms were added to 6 plates containing 19mls of the nutrient agar and labeled. A sterile cork borer (12mm) was used to make four ditches in each plate. The base of each plate was filled with sterile Mueller Hinton agar to seal the bottom and left for some time to allow it to gel. 0.2mls of the extracts were dispensed into each ditch. The plates were left to allow diffusion of the extracts before being placed in the incubator at 37^0C for 24hrs. The zone of inhibition produced after incubation were measured and recorded.

Control Used for the Screenings

Positive control was made using the antibiotic streptomycin in order to check if the organisms are not resistance to treatment with broad spectrum antibiotic.

Data Analysis

The biocidal activities of the plant extracts on susceptibility of the tomato samples to the pathogens was analyzed using completely randomized design (CRD) and least significant difference (LSD) was used for mean separation.

RESULTS AND DISCUSSIONS

Isolation and Identification of Pathogens Associated with Post-Harvest Losses of Tomatoes Bacteria

Bacteria Count

Table 1 Colony count of bacteria in the tomato samples collected from different locations.

S/N	Samples	Plate I count	Plate II count	Mean	Standard CFU/ml
1	Shuni	916	484	700	$7.0x10^5$
2	Sokoto	235	696	466	$4.66x10^5$

From the Table 1 above, the result showed that samples collected from Shuni local government contained higher bacterial population than those of Sokoto, plate I collected from Shuni contained the highest bacterial population (916) followed by plate II collected from Sokoto with (696), the third and fourth were plate II from Shuni and plate I from Sokoto with population of 484 and 235 respectively.

Biochemical Test

After conducting different biochemical tests, the tests indicated the presence of two pathogenic bacteria in each sample. In two samples collected from Sokoto vegetable market, *Klebsiella aerogene* previously known as *Enterobacter aerogene* and *Citrobacter freundii* were identified. While *Providencia spp* and *Enterobacter aerogene* were identified in the samples collected from Shuni local government. *Enterobacter aerogene* was present in both samples.

Table 2 Identified bacterial isolates from tomato samples

S/N	Sample code	GS	L	G	S	Ci	M	I	U	Ca	Mr	Confermed spp
1	TMT SKT_S	-	+	+	+	+	+	-	-	+	-	*Klebsiella aerogene*
2	TMT SKT_L	-	+	+	+	+	+	-	-	+	+	*Citrobacter freudii*
3	TMT SHN_S	-	-	+	+	+	+	+	-	+	+	*Providencia spp*
4	TMT SKT_L	-	+	+	+	+	+	-	-	+	+	*Citrobacter freudii*

Key:
TMT SKT_S:Tomato sample collected from Sokoto which smaller bacterial pathogens were observed
TMT SKT_L: Tomato samples collected from Sokoto which larger bacterial pathogens were observed.
TMT SHN_S: Tomato samples collected from Dange Shuni local government in which smaller bacterial pathogens were observed
TMT SHN_L: Tomato samples collected from Dange Shuni local government in which larger bacterial pathogens were observed
G S: Gram staining, L: Lactose, G: Glucose, S: Sucrose, Ci: Citrate, M: Motility,
I: Indole, U: Urease, Ca: Catalase, Mr: Methyl red.

Similar previous researches have also confirmed the appearance of these identified organisms on tomatoes. Brigitte *et al.* (2016) reported

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that the dominant species associated with tomatoes were those belonging to the genera Enterobacter, Klebsiella, and Citrobacter.

Klebsiella aerogenes

Klebsiella aerogenes previously known as *Enterobacter aerogene* (Tindall *et al.,* 2017), is a Gram-negative, oxiadase negative, catalase positive, citrate positive, indole negative, rod-shaped bacterium. The bacterium is approximately 1-3microns in length, and is capable of motility via peritrichous flagella. These characteristics were observed in the present study and are similar to what was observed by (Tindall *et al.,* 2017).

Citrobacter freudii

Citrobacter freudii is a gram-negative, catalase positive, citrate positive, indole negative,methyl red positive, it is a species of facultative anaerobic bacteria of the family *Entrobacteraceae*. The bacteria have a long rod shape with a typically length of 1-5microns in length. Most *C. freudii* cells generally have several flagella for locomotion but some are non-motile. (Wang *et al.,* 2000).

Providencia spp

Providencia spp Is a gram-negative, catalase positive, indole positive, citrate positive, urease negative, it is a motile bacterium which belongs to the family *morganellacea* (Stuart *et al.,* 1943).

Table 3 Anti-bacterial activities of aqueous extract of neem and garlic on bacterial pathogens isolated from the tomato samples

S/N	Name of plant used	Part of plant used	Extractant	Diameter of inhibition mm/bacterial spp			
				Conc. mg/ml	*E. aerogene*	*C. freudii*	*Providencia spp*
1.	Garlic	Bulb	H_2O	40	00	00	00
				80	14	16	15
				120	17	19	19
2.	Neem	Leaves	H_2O	40	00	00	00
				80	00	14	00
				120	15	16	14
3.	Streptomycin			5	23	26	25

Size of cork borer = 12mm

The anti-bacterial activities of neem and garlic extracts on *E. aerogenes*, from the Table 3 above, shows that garlic is more active than neem because it produces 14mm and 17mm zones of inhibition at 80mg/ml and 120mm/ml concentration of the extract. While neem produces effect only at 120mg/ml concentration with 15mm zone of inhibition.

Similar result was observed on *Providencia spp* because neem does not show any effect at 40mg/ml and 80mg/ml concentrations; it only shows its effect at 120mg/ml concentration with 14mm zone of inhibition which was less than that of garlic at 80mg/ml concentration that produced 15mm zone of inhibition and 19mm at 120mg/ml concentration.

The result also shows that garlic extract is more effective on *C. freundii* than neem, by showing 15mm and 19mm zones of inhibition at 80mg/ml and 120mg/ml respectively, it is only on this pathogen that effect of neem extract at 80mg/ml was observed, the result from the table shows that neem extract inhibited bacterial growth with 14mm and 16mm zones of inhibition at 80mg/ml and 120mg/ml respectively.

The overall activities of both neem and garlic on all isolated bacterial species have shown less effect at all concentrations used compared to streptomycin which was used as the control.

Streptomycin is an antibiotic used to treat a number of bacterial infections. These include tuberculosis, *Mycobacterium avium* complex, endocarditis, brucellosis, *Burkholderia* infection, plague, tularemia, rat bite fever (ASPHSP, 2016). However, streptomycin contains several side effects which include feeling like world is spinning, vomiting, numbness of the face, fever and rash. (ASHSP, 2016). When used during pregnancy it may result in permanent deafness in the developing baby, (WHO, 2008). The above mentioned side effects and many others found in several synthetic chemicals used in the treatment of pathogenic fungi and bacteria were among the main reason why scientist started to think of other alternatives like producing resistance varieties and treatment with natural pesticides.

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Table 4 Effect of Neem and Garlic extracts on isolated bacteria species from tomato fruit samples.

Treatments	*Citrobacter freudii*	*Enterobacter aurogene*	*Providencia spp*
Neem	10.00	5.00	4.67
Garlic	11.67	10.33	11.33
Control	25.98	22.98	24.98
SE±	7.75	7.24	7.43
Significance	NS	NS	NS

NS=Not Significant

From the table 4 above, the result shown that the inhibition of growth of bacterial pathogens by both neem leaves and garlic bulb extract is not significant compared to the control. However, since zone of inhibition increases with increase in concentration rate, further research needed to be done using higher concentration than what were used in this research and that might produce a significance effect when compared to the control.

SUMMARY, CONCLUSION AND RECOMMENDATIONS

Summary

This research was conducted to analyze the efficacy of neem leaves and garlic bulbs extracts on identified bacterial pathogens isolated from tomato fruit samples collected from Sokoto vegetable market and Shuni local government Sokoto state. Nutrient agar was used for bacterial isolation at Microbiology Laboratory as well to analyze the efficacy of the efficacy of neem and garlic extract extracts on bacteria.

all at Usmanu Danfodiyo University Sokoto.

Three bacterial species were identified after the isolation and both were treated with the extracts that were prepared at different concentrations of 100mg/ml, 200mg/ml and 300mg/ml.

The three bacterial species identified after the isolation were also treated with neem and garlic extracts, both neem and garlic extracts have shown a positive result on identified bacterial species, the bacterial species that were identified includes; *Providensia* spp, *C. freudii* and *E. aerogenes.* However, both extracts have shown less effect compared to streptomycin which was used as the control, although it contained several side effects.

Conclusion

At the end of this study, it was observed that there are bacteria that cause post-harvest losses of tomatoes in both Shuni and Sokoto. The identified bacteria were *Enterobacter aerogene, Citrobacter freudii and Providencia spp.*

Neem leaves and garlic bulb extracts were found to have potential anti-microbial compound that inhibit the growth of pathogens isolated on tomato fruits at various concentrations. Result of this study can be an important step in developing plant biopesticides for management of fruit rots.

Recommendations

This study recommends that;

i. Farmers should disinfect the tomato fruits after harvesting to reduce chance of infection by pathogens. This could be done by use of sodium hypochlorite.
ii. Further research need to be carried using higher concentrations of extracts than the quantity used in this research, which might be enough to substitute the used of synthetic chemicals.
iii. Government and other research organizations need to sponsor researches on this aspect since these medicinal plants are abundant and affordable, this can help greatly in reducing several diseases such as cancer that might occur as a result of ingesting chemical residues.

REFERENCES

1. *Adaskaveg, J. E., Forste, H. and Sommer, N. F. (2000). Principle of Post-harvest pathology management of decays of edible horticultural crops. In: post- harvest technology of horticultural crops, 331: 163-195.*
2. *Adeboyega, S. A., Olajuyigbe, A. E., Balogun, I. and Olatoye, O. (2016). Ethiopian journal of Environmental studies and Management. g(1): 56-69. 2. 16.*
3. *Arokiyaraj, S., Martin, S., Perinbam, K., Arockianathan, P. M. and Beatrice, V. (2008). Free radical scavenging activity and HPTLC finger print of Pterocarpus santalinus L. –an in vitro study. Indian Journal of Scientific Technology, 1 (7): 1-7.*
4. *AVRDC. (2006). Tomato and pepper production in the tropics. Nigerian Food Journal, 10:19-27.*
5. *Cardellina, J. H. (1988). Natural product in the search for new agro-chemicals. In H. G. Gutter. Biologically Active New York Science Journal, 2011:4*
   Cheesbrough, M. (2000). District Laboratory Practice in Tropical Countries. Cambridge University Press, UK. Pp: 250-253.
6. *Eno-Abasi, S., Gbenga, A., Joke M, and Abdulganiu, A. (2018). Still a long way to self-*

SJIF Impact Factor 2021: 8.013| ISI I.F.Value:1.241| Journal DOI: 10.36713/epra2016 ISSN: 2455-7838(Online)
EPRA International Journal of Research and Development (IJRD)
Volume: 6 | Issue: 4 | April 2021 - Peer Reviewed Journal

sufficient in tomato production. Sunday magazine, 50: 7-8

7. *FAO, (2002). www.fao.org/decrep/t0073e/too73e00.*
8. *Fawole, B, and Oso, K. (2001). Principle of microbiology for student of food and technology, 7th edition (University press, Nigeria) Pp 25-30.*
9. *George, B., Kaura, C., Khurdiya, D. S., Kapoor, H. C. (2004). Anti-oxidants in tomato (Lycopersicum esculentum) as a function of genotype. Indian Food Chemistry, 84: 45-51.*
10. *Handa, S. S., Khanuja, S. P. S., Longo, G. and Rakesh, D. D. (2008). Extraction technologiesfor medicinal and aromatic plants. International centre for science and high technology, 1:21-25.*
11. *Kader, A. A. (1992). Post-harvest biology and technology: In: A.A. Kader Post-harvest technologyof horticultural crops. University of California, Agriculture and Natural Resources, 3311:15-20.*
12. *Kanetis, L., Förster, H., and Adaskaveg, J. E. (2007). Comparative efficacy of the new post-harvest fungicides azoxystrobin, fludioxonil, and pyrimethanil for managing citrus green mold. Plant Diseases, 91:1502-1511.*
13. *Mamman, A.B, Oyebanji, J.O and Peters, S.W. (2000). Nigeria A people united A future assured (survey of states) vol.2 Gabumo publishing co. Ltd calabar Nigeria*
14. *Moss, M. O. (2002). Mycotoxin Review Journal on Aspergillus penicillium. Mycologist, 16:116-119.*
15. *Mujib, U. R., Naushad, K. and Inayatullah, J. (2007). Post-harvest losses of tomato crop. Sarhad Journal of Agriculture, 23(4): 1279.*
16. *Mungai, J., Ouko, J. and Heiden, M. (2000). Processing of fruits and vegetables in Kenya. Printed by Agricultural Information Centre. Nairobi. Kenya*
    Okezie, B. O. (1998). World food security: the role of post-harvest technology. Food Technology, 52: 64-69.
17. *Okigbo, R. N. (2009). Variation in phytochemical properties of selected fungicidal aqueous extract of some plant leaves in Kogi State, Nigeria. American European Journal of Sustainable Agriculture, 3 (3): 407-409.*
18. *Rice, R. W., Rice, L. W. and Tindall, H. D. (1993). Fruit and vegetable production in warm climates. Macmillan Press, London, 222-230.*
19. *Samson, J. A. (1984). Tropical fruits. Tropical Agricultural Series, 20:64-118.*
20. *Seminis-Kenya. (2007). Retrieved from: http://www.freshplaza.com/news*
21. *Shanmugavalli, N., Umashankar, V. and Raheem, A. (2009). Anti-microbial activity of Vanilla planifolia. Indian Journal of Science and Technology, 2 (3), 37-40.*
22. *Sigei, K. G., Ngeno, K. H., Kibe, M. A., Mwangi, M. M. and Mutai, C. M. (2014). Challenges and strategies to improve tomato competitiveness along the tomato value chain in Kenya. International Journal of Business and Management, 9(9): 230-245*
23. *Singleton, P. (1997): Bacteria in Biology, Biotechnology and Medicine, (4th edition). John Wiley and sons Ltd, Chichester, England. Pp 140-203, 208-367.*
24. *Stinson, E. E., Osman, S. F., Heisler, E. G., Sicihano, J. and Bill, D. D. (1981). Mycotoxin production in whole tomatoes, apples, oranges and lemons. Journal of Agriculture and Food Chemistry, 29:790-792.*
25. *Stuart, C. A., Wheeler, K. M., Rustigian, R. and Zimmerman, A. (1943). "Biochemical and Antigenic Relationships of the Paracolon Bacteria". Journal of Bacteriology. 45 (2): 101–19*
26. *Swarnalatha, L. and Reddy, P. N. (2009). Anti-microbial, anti-diarrhoeal and analysis of phytochemical constituents of Sphaeranthus amaranthoides.Indian Journal of Science and Technology, 2 (3): 45-48.*
27. *Thirupathi, V., Sasikala, S. and John Kennedy, Z. (2006). Preservation of fruits and vegetables by wax coating. Journal of science and food agriculture, 55:1-10.*
28. *Tindall, B. J., Sutton, G. and Garrity, G. M. (2017). "Enterobacter aerogenes". International Journal of Systematic and Evolutionary Microbiology. 67 (2): 502–504.*
29. *Wamache, A. (2005). Vegetable seeds handbook. Regina seeds Seminis. Printed by Bizone ltd. Nairobi Kenya, 23-25.*
30. *Wang, J. T., Chang, S. C., Chen, Y. C. and Luh, K. T. (2000). "Comparison of antimicrobial susceptibility of Citrobacter freundii isolates in two different time periods". Journal of Microbiology, Immunology, and Infection. 33 (4): 258–262.*
31. *Wills, R. H., Lee, T. H., Graham, D., Mcglassom, W. B. and Hall, E. G. (1981). An Introduction to the physiology and handling of fruits and vegetables. London, 432-438.*