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RESEARCH ARTICLE

ISOLATION AND CHARACTERIZATION OF FUNGI FROM THE FRUIT OF ORANGE AND TOMATO IN JIMMA TOWN MARKET SELLERS, SOUTH WEST ETHIOPIA.

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Abstract

The study was carried out from February, 2013 to May, 2013in Jimma to assess the common spoilage fungi from infected fruits of tomato *(Lycopersiconesculentum)* and orange *(Camellia sinensis)* in Jimma town (Agip, Kochi and Bishishe) market sellers. Mycological investigation on spoilage fungi from 40 (20 from each) apparently infected*C. sinensis*and*L.esculentum*was carried out. Potato Dextrose Agar was used for fungi isolation and purification following standard procedures. Pure cultures were identified morphologically to the genus level. A total of 56 spoilage moulds were isolated from the two different types of deteriorating fruit samples, from which 52 (92.85 %) isolates were identified and the remaining4(7.15) wereunidentified. Thefungal species *Aspergillus*spp., (13.46) *Penicillium*spp.,(19.2) *Rhizopus*spp.,(1.92) *Mucor*spp., (5.76*)* *Fusarium*spp., *(3.84)*, *Byssochlamys*spp.(3.84) and *Cladosporium*spp.(1.92) were found to be associated with deterioration of orange. The fungal isolates associated with the spoilage of tomatowere *Aspergillus*spp., (7.69) *Penicillium*spp., (9.61) *Rhizopus*spp., (7.69) *Mucor*spp., (9.61) Fusarium, (9.61) and *Monilia*spp. (5.76).Generally, of all isolated moulds *Penicillium*spp. was the dominant isolate 15(28.81%), followed by*Aspergillus*spp.11(21.15). *Byssochlamys*spp. And *Cladosporium*spp.were the least encountered 2 (3.84%) and 1(1.92%), respectively. The results of this study indicate that fruits sold atAgip, Kochi and Bishishe were massively infected with spoilage fungi due to lack of selling parameters of fruits found in Jimma town market sellers. So, appropriate measurement must be taken by responsible bodies to reduce the fungal load and to enhance the quality of fruits sold in Jimma town.

Introduction:-

Fruits and vegetables are very important and have high dietary and nutritional quality. Fruits play a vital role in human nutrition by supplying the essential growth factors such as vitamins and essential minerals to the regular diet, which is necessary for the good and normal health consumption of fruit and vegetable products has dramatically increased by more than 30% during the past few decades (Barth et al., 2009). Fresh fruit and vegetable consumption increased by 25.8 and 32.6% respectively and far exceeded the increases observed for processed fruit and vegetable products. Fruits and vegetables are a major source of macronutrients such as a fiber and micronutrients such as

minerals and vitamin C, thiamin, riboflavin, B-6 niacin, folate, A and E (Rickman et al., 2007). The fruits of tomato are popular throughout the world and are used in all kind of stews, soups and also eaten raw in salads. Ripe tomato fruits have high nutritive values, being a good source of vitamin A, B, C and minerals (Elsayed and Edrees, 2014). According to Wada and Ou (2002) fruits and vegetables are well known for their antioxidants compounds that protect against oxidative damage caused by free radicals, and they have been shown to be effective in helping to prevent retinal disease such as muscular degeneration. And also tomato plant contain antioxidant phytochemicals such as the carotene and lycopene (Wogu and Ofuase, 2014; Sravanthi and Gangadhar, 2015).

Traditional varieties of fruits like avocado, banana, orange and papaya are affected by a wide array of microorganisms. Spoilage refers to any change in the condition of food in which the food becomes less palatable, or even toxic: these changes may be accompanied by alteration in taste, smell, appearance or texture (Akinmusire, 2011). One of the limiting factors that influence health and economic values of the fruit is the relatively short lifespan. Fruits deteriorate rapidly after harvest and in some cases do not reach consumers at optimal quality after transport and marketing. The main causes of fruit deterioration are dehydration, weight loss, color change and microbial spoilage. However, deterioration rate is affected by different factors like temperature, relative Humidity and storage atmosphere condition (Sherrattet al., 2006).

Fungal fruits infection may occur during the growing season, harvesting, transport and post-harvest storage and marketing transport and post-harvest storage and marketing conditions, or after purchased by the consumer. The important post-harvest disease causing fungi include *penicillium, Aspergillus, Alteranaria species, Botrytis cinerea, Molinilinia lax and Rhizopus stolonifer* (Ogawa et al., 1995). Fruits particularly tomatoes and orange are eaten raw and tomatoes are also eaten cooked. Spoilage fungi that typically produce more diverse and greater amounts of extracellular depolymerases successfully attack both fruits (Barth et al., 2009). And also an abundance of extracellular pectinases and hemicelluloses produced by the fruits are important factors for fungal spoilage (Miedes and Lorences, 2004).

Although available literatures revealed that the importance of fruit is increasing daily, the incidence of microbial attack on this fruit demands attention. Janisiewicz and Korsten (2002) also remarked the exploration of plant extracts have shown to be great potential and may be used as alternative to synthetic fungicides. The purpose of the current investigation was to isolate and characterize some fruit spoilage fungi.

Materials and Methods:-

Description of study area:-

This research was conducted in Jimma town, which is the largest city in Southwestern Ethiopia. Jimma town is located 353km southwest of the capital city of Ethiopia, Addis Ababa. And the city islocated between 1,500 - 2,400 m above sea level and the total population was about 120, 969 in 2007. It lies in a climatic zone locally known as Woyna degas which is ideal for agriculture as well as human settlement. The town is characterized by warm climate with a mean annual maximum temperature of 30^0C and minimum of 14^0C. The laboratory based study was conducted at microbiological Laboratory of Department of Biology, Jimma University during the period from February, 2013 to May, 2013.

Fruit source:-

Fruits of orange and tomato which were found with symptoms of fungal infection were purchased from Agip, Kochi and Bishishe market sellers in Jimma town.

Sample size determination:-

Purposive sampling technique was used to determine the sample size and assess the common fungal pathogens from infected orange and tomato, from Jimma town market sellers. A total of 40 samples comprising 20 samples from each fruit were collected randomly from Agip, Kochi and Bishishe markets. Ten of the samples (5 orange and 5 tomato) were collected from Agip market, the other 10 samples consisting 5samples from each fruit were collected from Kochi market and the rest 20 samples comprising 10 samples from each fruit were collected from Bishishe market this is because Bishishe Market in Jimma town serves as the main depot where the major fruit dealers and suppliers sell to petty traders who in turn distribute to other local markets (Agip and Kochi) and cafeterias throughout the town. The fruits were sourced from areas like Mizan, Tepi, Addis Ababa and neighboring woredas with in the Jimma zone itself. All of the 40 fruit samples were purchased from different venders, so 40 fruit venders were visited to collect all the samples.

Collection of infected fruit:-
Fruit types of orange and tomato with the common post-harvest disease symptoms were collected using sterile polyethylene bag from Agip, Kochi and BishishemarketsinJimma town. A total of 40 samples comprising 20 samples from each fruit were collected randomly from the venders and it was transported aseptically to the laboratory of microbiology, department of Biology, Jimma University for further analysis.

Isolation of fungi:-
Potato dextrose agar medium was prepared using distilled water and it was sterilized within an autoclave. The petridishs were also sterilized to kill any microbial contamination. In order to suppress bacterial growth, chloramphenicol of 35-100mg $100ml^{-1}$ of PDA was poured on the already sterilized petridishs and it was left open for a few minute inside a laminar flow to be solidify, then the petridishs were closed to prevent other contamination. Fruits of orange and tomato were surface sterilized by exposing them in 90% ethyl alcohol for 1 min and 3 min to 1% sodium hypochlorite and then, it was rinsed three times in sterile distilled water. Segments (3-5cm), which cut with a sterilized scalpel was placed on previously prepared potato dextrose agar (PDA in case of both orange and tomato) in petridishs and they were incubated at 21^0C for 5 to 7 days. After incubation, the colonies were observed. To get a pure culture, each of the emerging mycelium was transferred to fresh solid medium. The pure culture of each colony on different plates were obtained and they were prepared for characterization

Characterization of the isolates:-
The pure isolated fungi were identified using cultural and morphological features according to the most documented keys in fungal identification (Domsch*et al.*, 1993; klich, 2002; Samson and Varga, 2007). The identification of fungal isolate was based on colony characteristics and conidial features of the isolates within genus level. The isolates were also identified by comparing their characteristics with those of known taxa, as described by Jolt *et al.* (1994) and Oyeleke and Manga (2008). To apply this, slide cultures were prepared for each of the isolates. The identified fungi were smoothly caught (2-3cm) with sterilized scalpel and they were placed on slide.

The technique of James and Natalie (2001) was applied for identification of the unknown isolated fungi using cotton blue in lacto phenol stain. The identification was achieved by placing a drop of the stain on clean slide with the air of mounting needle, where a small portion of mycelium from the fungal culture was removed and place on a drop of lacto phenol. The mycelium spread very well on the slide with the aid of the needle. A cover slip was gently applied with little pressure to eliminate air bubbles. The slides were mounted and observed under compound light microscope with magnification power of x10 and 40 objective lenses, respectively.

Results:-

Isolation of spoilage moulds from fruits:-
In this study, a total of 52 fruit spoilage moulds were isolated from 40 samples of deteriorating Orange (*Citrus sinensis*) and Tomato (*Lycopersicomesculentum*) fruits purchased from Kochi, Agip and Bishishe markets of Jimma town (Table 1). Of the total isolates, 26(50%) of the spoilage fruits were isolated from Orange (*Citrus sinensis*) and the remaining 26(50%) of the spoilage fruits were isolated from Tomato (*Lycopersicomesculentum*).

Table 1:- Total number of spoilage moulds isolated from deteriorating fruits sold at Kochi, Agip and Bishishe markets, Jimma town.

Fruits	No of samples	No of isolates	Isolates Proportion (%)
C. sinensis	20	26	50%
L. esculentum	20	26	50%
Total	40	52	100%

Characterization of the isolates
Physically observation of the diseased fruits revealed brownish, necrotic patches on the skin of the orange and tomato fruits. Table 2 shows that the colonial and morphological characteristic of the isolated spoilage moulds of fruits.

Table 2:- Colonial and morphological characteristics of moulds associated with the spoilage of fruits.

Isolates	Colonial characteristics	Microscopic morphology	Suggested Genus
JUO 01-02 JUT 03-07	Cottony, pink, purple, brown colonies.	Extensive septet mycelium and conidiophores simple or branched with ovoid to elongated conidia of variable size.Septate fusiform, slightly curved and pointed at both ends.	*Fusarium*spp.
JUO 08-17 JUT 18-22	Greenish or blue green colonies.	Conidia in long chains on repeatedly branched conidiophores resembling a brush like head (penicllus). Conidiophores smooth, relatively short. Penicillia mycelia arranged very irregular and asymmetrical with branches of various lengths.	*Penicillium* spp.
JUO 23-29 JUT 30-33	Colonies with loose white to yellow mycelium rapidly becoming dark brown to black on the development of conidia. Colonies light green-yellow.	Black, brownish black, purple brown Conidiophores and yellow to green conidia with dark sclerotia. Microscopically conidiophores arising from a foot-cell, catenate (basipetal) conidia on phialides (1or 2 series) on vesicle.	*Aspergillus*spp.
JUO 34 JUT 35-38	White to dark grey colonies, fast growing and filling the petri dish with dense cottony mycelium, producing mass of sporangia.	Nonseptate mycelium with root like rhizoids; black columellate, sporangiophores, in clusters and dark sporangia containing dark to pale spores.	*Rhizopus* spp.
JUO 39-41 JUT 42-46	Nonseptate hyphae, cottony colony, Smooth, non-striated Sporangiophores and Produce no rhizoids	Nonseptate or sparsely Septate, broad hyphae, sporangiophores, Sporangia and spores are visualized. Apophysis, rhizoid and stolon are absent.	Mucor spp.
JUO 00 JUT 47-49	Pink, gray, or tan conidia	Asterisk-like shape of the colony. Type "S" colonies grew slowly and stopped after days entirely.	*Monilia*spp.
JUO 50-51 JUT 00	Cottony colony Absence of ascocarps, asci in open clusters	Phialides with cylindrical bases that taper abruptly into long cylindrical necks and produce catenate conidia.	Byssochlamys spp.
JUO 52 JUT 00	Thick, velvet colony, green, olive green, darkblue, black or brown colony and some lemon shaped conidia variously branched	Hyphae are Septate and brown in color.Conidiophores are brown and often Septate. Conidiophores are erect, straight or flexuous, unbranched or branched only in the apical region	*Cladosporium*spp.

Legend: O= Orange; T= Tomato; n= Code no of isolates

Occurrence of Fungi Isolates Associated with the Spoilage of Fruits:-

Of the total 56 spoilage moulds isolated from the two different types of deteriorating fruits (*C. sinensis* and *L. esculentum*), 52(92.85 %) isolates were characterized and identified as *Aspergillus*, *Fusarium*, *Penicillium* and *Rhizopus,Mucor,MoniliaByssochlamys and Cladosporium*spp. The remaining 4(7.15) were uncharacterized since

they were totally contaminated with bacterial growth. Generally, of all isolated moulds*Penicillium* sp. was the dominant isolate 15(28.81%), followed by*Aspergillus* sp. 11(21.15). *Byssochlamys*and *Cladosporium*spp. were the least encountered 2(3.84%) and 1(1.92%), respectively (Table 3). In addition, most of the spoilage fungi were wide spread among all examined fruits. However, no *Monilia*was isolated from orange and no *Byssochlamys*and *Cladosporium*spp. were isolated from tomato (Table 3).

From this outcome, it can be concluded that, *Penicillium*was the most spoilage fungi of orange and tomato than other fungal species. Next to *Penicillium, Aspergillus*was the dominant one for the spoilage of orange. And *Monilia, Byssochlamys*and *Cladosporium*were found in small percentage compared to others.

Table 3:- Prevalence of spoilage moulds isolated from deteriorating fruits sold at Kochi, Agip and Bishishe, Jimma town, 2011/12.

	Orange (No of samples 20)	**Tomato (No of samples 20)**	
	No of isolates	**No of isolates**	**Total**
Fusarium sp.	2(3.84)	5(9.61)	7((13.45)
Penicillium sp.	10(19.2)	5(9.61)	15(28.81)
Aspergillus sp.	7(13.46)	4(7.69)	11(21.15)
Rhizopus sp.	1(1.92)	4(7.69)	5(9.61)
Mucorsp	3(5.76)	5(9.61)	8(15.37)
Monilia sp.	-	3(5.76)	3(5.76)
Byssochlamys sp.	2(3.84)	-	2(3.84)
Cladosporium sp.	1(1.92)	-	1(1.92)
al	26(50)	26(50)	52(100)

Note: Values in parenthesis are percentages of the isolated moulds.

Discussion:-

The findings of this study showed that *Aspergillus, Fusarium, Penicillium, Rhizopus,Mucor, Monilia, Byssochlamys and Cladosporium* spp. were isolated from fruits sold in main markets of Jimma town (Kochi, Agip, and Bishishe). All the post-harvest spoilage moulds except *Monilia* spp. were found to be associated with spoilage or deterioration of orange (Citrus sinensis). Similarly, Al-Hindi et al. (2011) have reported that *A.niger, P. digitatum* and *R. stolonifer, Byssochlamyssp, Cladosporium* spp. and *Mucor* were implicated in spoilage of *Citrus sinensis*. Tournas and Katsoudas (2005) also reported that *Fusarium* spp. were the most common fungi in citrus fruits. Bukar et al., (2009) also revealed that, the most predominant fungus isolated from the orange (*Citrus sinensis*) were, *Aspergillus* sp; others include *Mucor* sp, *Penicillium* sp, *Rhizopus* sp *Fusarium* sp, and *Alternaria* spp. A study conducted by Oviasogie et al., (2015) also confirmed that the fungal pathogens associated with the spoilage of orange (*Citrus sinensis*) were *Aspergillus* spp. *Penicillium* spp. *Mucor* spp. *Rhizopus* spp. *Candida tropicalis Saccharomyces cerevisiae* and *Alternaria* spp. in which *Aspergillus* spp. was the predominant fungal pathogen.

In the case of tomato fruits *(L. esculentum*), *Aspergillus, Penicillium, Rhizopus, Fusarium, Mucor* and *Monilias* spp. were implicated in its spoilage. In line with this, Mitra (1997) has discovered that the species of fungi associated with the spoilage of *Citrus sinensis, Asiminatriloba* and *L. esculentum* fruits include species of *Aspergillus, Fusarium,* Yeast, *Penicillium,* and *Rhizopus. A. flavusandA. Fumigatus* caused *L. esculentum* spoilage were also investigated by Al-Hindi et al. (2011). Seven fungal pathogens were associated and responsible for fruit rotting of *L. esculentum* caused by *Fusariumequiseti, A. flavusandA. niger.* Akinmusire (2011) also reported that fungi affecting *L.esculentum* includes *FusariumOxysporium, Fusariummoniliform,AspergillusnigerandRhizopusstolonifer*. Ugwu et al. (2014) also discovered that the responsible fungal pathogens for the spoilage of tomato were *Candida tropicalis, Penicillium notatum, Aspergillus niger, Fusarium oxysporum, Absidia corynbifera, Rhizopus stonolifer*

In the current study *Penicillium* spp. was the most spoilage fungi of orange and tomato than other fungal species and this result is in agreement with the study conducted by Mbajiuka and Enya (2014) in which abundant presence of *Penicillium nalgiovense, Penicillium notatum* and *Penicillim expansium* were found among other fungi species involved in deterioration of tomatoes fruit.

The diseased fruits sampled from Kochi, Agip and Bishishe market were found to be massively infected with eight genera of fungi namely *Aspergillus,Fusarium, Penicillium and Rhizopus,Mucor, Monilia , Byssochlamys* and *Cladosporium* spp. and four other uncharacterized spoilage moulds. The prevalence of fungi as the spoilage organism of fruits is due to a wide range of factors which are encountered at each stage of handling from pre-harvest to consumption and is related to the physiological and physical conditions of the produce as well as the extrinsic parameters to which they are subjected (Effiuvwevwere, 2000). Damage inflicted on produce at the time of harvest is a major cause of infection since most of the spoilage microorganisms invade the produce through such damage tissues; similarly, the extent of deterioration is influenced by the depth of the wound. Furthermore, the incidence of infection is worsened by poor sanitary practices such as cross-contamination, contact infection during the transportation of fruits (Effiuvwevwere, 2000).

Akinmusire (2011) also remarked that the contamination of fruits by fungi could also be as a result of poor handling practices in food supply chain, storage conditions, distribution, marketing practices and transportation. From personal observation at Agip, Kochi and Bishishe, the poor hygienic conditions of the vending house/store, the venders and the utensils used for keeping and weighing the fruits were responsible factors for mixing the deteriorating fruits with the healthier one and many other factors aggravate prevalence of fungi as the spoilage organism of fruits. In addition, the overall weather conditions of Jimma town, especially the high moisture content favours the growth of spoilage moulds on fruits and vegetables vended at Agip, Kochi and Bishishe. According to Alemu et al. (2011) from a climatic point of view, abundant rainfall makes this region one of the best watered of Ethiopian highland areas, conducive for the growth of post-harvest fruit spoilage moulds.

The overall analysis of the isolation and identification process implied various causes for the growth of the moulds on fruits. As clearly described by Korsten (2006), postharvest losses of fruits is a serious problem, because the values of fresh product significantly increase while passing from the farm to the consumers table and due to overpopulation the demand for fruits increases in the world. Fungal pathogens are mainly responsible for postharvest losses of fruits (Korsten 2006).

Conclusions:-

This study detected the profile of spoilage fungi involved in the deterioration of orange and tomato sold in main markets of Jimma town (Agip, Kochi and Bishishe*).* It showed that, fungi were involved in the spoilage of fruits. *Fungal flora of orange and toma*to samples found from February 2013 to May 2013 were dominated by moulds of *penicillium, Aspergillus, Mucor, Fusarium, Rhizopus, Monilia, Byssochlamys and Chladosporium.* Generally, of all isolated moulds*Penicillium*spp. was the dominant isolate 15(28.81%), followed by *Aspergillus* spp. 11(21.15). *Byssochlamys* and *Cladosporium* spp. were the least encountered 2(3.84%) and 1(1.92%), respectively.

The results of this study indicate that fruits sold at Agip, Kochi and Bishishe were massively infected with spoilage fungi due to several factors as poor hygienic conditions of the vending site/store, venders and vending utensils. In addition, the high humid content of the vending site also contributes for the spoilage. Mechanical injuries of fruits such as bruises or cuts was occur during harvesting or post harvesting, provide infection site for spoilage of fruits. The high moisture content of fruits and vending/storing site will be a serious limiting factor in their preservation. Since the fruits used in this study are produced in the neighboring woredas of Jimma town, thus, they are transported to the city in locally woven baskets and sacks under weather conditions that encourage the incubation of these contaminating fungi. Generally it can be concluded that, fruit spoilage fungi are caused due to the lack of occurrence of safety fruits within market sellers of Jimma town.

The high prevalence of the spoilage fungi demand that appropriate control measures against infection, should be employed if farmers expect good performance of their produce. Adequate mycological knowledge and handling practices of these produce would therefore help minimize wastes due to deterioration and unacceptability. It is therefore important that both the farmer who harvests the fruits into bags for transportation, the marketers and consumers take necessary precaution in preventing contamination and also try to create an environment that will not encourage the growth or multiplication of spoilage fungi. In addition, the hygienic conditions of vending/storing site, the venders and vending utensils should have to be improved in order to provide fresh and quality fruits for the consumers. This will help in providing fresh and quality fruits for the consumers as well as in preventing the consumption of contaminated fruits thereby reducing the risk of health problems which are produced by these fungi that have been isolated in this study.

This research pointed out the common fungi which spoil fruits of orange and tomato, which are sold in Jimma town market sellers. Thus proper handling of these fruits should be practiced to reduce the fungal genera.
The majority of analyzed samples showed the presence of different fungal genera in fruits of orange and tomato. This indicates that the lack of selling parameters of fruits found in Jimma town market sellers. So, appropriate measurement must be taken by responsible bodies to reduce the fungal load and to enhance the quality of fruits sold in Jimma town.

In most unorganized markets, fruits were available in local retain shops without appropriate temperature control and unsuitable storage. This is purchased by households, this leads to the disturbance of the health of the people. So, inhibition of fungal growth by lowering storage temperature through storage under refrigeration and use of fungicides must be applied.

The hygienic and sanitary conditions observed in fruit sellers were not very satisfactory. Thus, they should improve the sanitary condition of fruit markets. In order to improve the quality of fruits, it is better to give awareness for consumers as well as sellers and to all others who have access these fruits must understand about the following critical control points;

Since molecular characterization of the isolates reveals the true diversity of spoilage fungi associated with fruits than the traditional morphological characterization, molecular technique is recommended. This would be the delimitation/drawback of this study.

Conflict of interests:-
The author(s) have not declared any conflict of interests.

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References:-

1. Akinmusire OO (2011). Fungal species associated with the spoilage of some edible fruits in Maiduguri Northern Eastern Nigeria. Adv. Environ. Biol. 5: 157-161.
2. Alemu A, Tsegaye W, Golassa L, Abebe G (2011). Urban malaria and associated risk factors in Jimma town, south-west Ethiopia.
3. Al-Hindi RR, Al-Najada AR, Mohammed SA (2011). Isolation and identification of some fruit spoil age fungi: screening of plant cell wall degrading enzymes. Afr. J. Microibial Res., 5: 443-448.
4. Barth M, Thomas R,Hankinson, Zhuang H, Frederick B (2009). Microbiological Spoilage of Fruits and Vegetables, Compendium of the Microbiological Spoilage of Foods and Beverages, Food Microbiology and Food Safety, DOI 10.1007/978-1-4419-0826-1_6,
5. Bukar A, Mukhtar MD, Adamu S (2009). Isolation and identification of postharvest spoilage fungi Associated with sweet oranges (citrus sinensis) traded in kano metropolis. Bayero Journal of Pure and Applied Sciences, 2(1):122 – 124.
6. Domsch KH, Gams W, Anderson TH(1993). Compendium of soil fungi. Academic press. London, pp.860.
7. Effiuvwevwere BJ (2000). Microbial Spoilage Agents of Tropical and Assorted fruits and Vegetables (An Illustrated References Book). Paragraphics publishing company, Port Harcourt. Pp. 1-39. Factors in Jimma town, south-west Ethiopia. Malaria J. 10: 173
8. ElSayed IA, Nada O Edrees (2014). Using of plant growth promoting rhizobacteria as biocontrol agent for root-knot nematode under greenhouse. Nature and Science 12(12).
9. James GC, Natalie S (2001). Microbiology. A laboratory manual (Ed) pp. 211-223.
10. Janisiewicz WJ, Koresten L (2002). Biological control of postharvest disease of fruits. Ann. Rev. Phytopathol., 40; 411-444.
11. Jolt TG, Krief NR, Sneath PHA, Stanley JT, Williams, ST (1994). Bergey's manual of systematic bacteriology, 9th ed. Williams and Wilkins Co- Balti more, mary land, pp786.
12. Klich MA (2002). Identification of common Aspergillus species-CBS, Utrecht. pp. 116.
13. Korsten L (2006). Advances in control of postharvest diseases in tropical fresh produce. Int. J. Postharvest Technol. Innovat. 1: 48–61

14. Mbajiuka SC, Enya E, (2004). Isolation of Microrganisms associated with deterioration of Tomato (Lycopersicum esculentum) and Pawpaw (Carica papaya) fruits. International journal of Current Microbiology and Applied Sciences 3:501-512.
15. Miedes E, Lorences EP (2004). Apple (malusdomestica) and tomato (lycopersicum) fruits cell-wall hemicelluloses and xyloglucan degradation during penicilliumexpansum infection. Journal of Agricultural and Food Chemistry, 52, 7957–7963.
16. MitraS (1997). Post-harvest Physiology and Storage of Tropical and Sub-tropical Fruits. 2nd edition. Biddles Limited, Guildford and Kings hymn, United Kingdom. Pp 179-183.
17. Ogawa JM, Dehr EI, Bird GW, Ritchie DF, Kiyoto V, Uyemoto JK (1995). Compendium of stone fruit disease Aps pres, USA. pp 449.
18. Oyeleke SB, Manga SB (2008). Essentials of laboratory practical are in microbiology. To best publisher, minna, Nigeria, pp 36-75.
19. Oviasogie FE, Ogofure AG, Beshiru A, Ode JN, Omeje FI, (2015). Assessment of fungal pathogens associated with orange spoilage. African Journal of Microbiology Research, 9(29), 1758-1763.
20. Rickman JC, Barett DM, Bruhn CM (2007). Nutritional comparison of fresh, frozen and canned fruits and vegetables. Vitamin C, B and phenolic compounds. J. Sc. food agric., 14: 345-354.
21. Samson RA, Varga J (2007). Aspergillus systematic in the genomic era. CBS fungal Biodiversity centre, utreat, pp 2006.
22. Sherratt, T. N., Wilkinson, D.M. and Bain, R. S. (2006). Why fruits rot, seeds mold and meat spoils: area appraisal. Ecol. Model., 192: 618-626.
23. Singh H, Fairs G, Syarhbil M (2011). Anti-fungal activity of capsicum frutecence and zingiberofficiale against key post-harvest pathogens in citrus. Int. conf. Biomed – eng.Technol., 11: 327-332.
24. Sravanthi J, Gangadhar SR, (2015). Phytochemical and antioxidant composition in Lycopersicum esculentum. Journal of Medicinal Plants Studies 3(4): 107-110.
25. Tournas VH, Katsoudas E (2005). Mould and yeast flora in fresh berries, grapes and citrus fruits. Int. J. Food Microbiol. 105: 11-17.
26. Wada L, Ou B (2002). Antioxidant activity and phenolic content of Oregon cranberries. J. Agric. Food chem., 50: 3495-3500.
27. Wogu MD, Ofuase O (2014). Microorganisms responsible for the spoilage of tomato fruits, Lycopersicum esculentum, sold in markets in Benin City, southern Nigeria. Scholars Academic Journal of Bioscience 2(7): 459-466.