International Journal of
Life Sciences

(A peer reviewed International Journal)

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International Journal of Life Sciences, Vol. 1 No. 2. pp. 19-27 2277-193x. 2012

ISSN *2277 – 193x*

Article type *Full Length Research Article*

Submission date *22 January 2012*

Acceptance date *30 March 2012*

Publication date *15 April 2012*

Article URL *http://www.crdeep.org/category/ijls*

Authors *Belay Berza,[1*] Kebede Abegaz [2], Tameru Alemu [3], Fasil Assefa [4]*

CRDEEP Head Office:
315/10, Kaulagarh Road, Rajendranagar, Indervihar, Dehradun, 248001, Uttrakhand, India.

International Journal of Life Sciences Vol.1 No.2. 2012. Pp. 19-27

Full Length Research Paper

Isolation, Characterization and Identification of Post-havest Spoilage Fungi of Ginger *(Zingiber officinale)* at Hadaro-Tunto and Boloso-Bombae, Southern Ethiopia

Belay Berza,[1*] Kebede Abegaz [2], Tameru Alemu [3],Fasil Assefa [4]

1. Department of Biology, Debre-Markos University, P.O. Box 269 , Debre-Markos, Ethiopia.;
2. Department of Food Science and Post-harvest Technology, Hawassa University, P.O. Box 05 Hawassa, Ethiopia.
3 Department of Plant and Horticultural Sciences, Hawassa University, P.O Box 05 Hawassa, Ethiopia.
4. Faculty of Life Sciences, Addis Ababa University, P.O. Box 1176, Addis Ababa, Ethiopia

ABSTRACT

Post-harvest deterioration is the most important cause of loss of *Zingiber officinale* production and this is mainly due to fungal invasion. Therefore this research was conducted to identify major fungi responsible for post-harvest deterioration of ginger. A survey was conducted at Hadaro-Tunto and Boloso-Bombae to identify research priorities. Total of 115 ginger samples were collected from harvest, drying and stored ginger between August 2009 and January 2010. The moisture content of dried ginger was determined. Post-harvest ginger deteriorating fungi were isolated and identified to genera level, and the pathogenicity of the isolates was tested. Cleaning, washing, sorting and separating damaged and injured rhizomes from healthy ones was not practiced at the study sites. Six genera of fungi were identified from the spoiled ginger samples including Fusarium, Penicillium, Aspergillus, Rhizopus, Eurotium and Mucor. Fusarium, Penicillium and Aspergillus were isolated from both districts at harvest, drying and dried-stored ginger. Fusarium, penicillium and Aspergillus were found to be responsible for most of deterioration observed. Rhizopus was isolated from drying and dried-stored samples where as Eurotium and Mucor were isolated from dried-stored samples. The percentage incidence of identified ranged 2.2% (Mucor) to 34.7 %(Fusarium). All the fungi identified were pathogenic to fresh ginger with pathogenicity indices ranging 17.35 %(Rhizopus strain 3) to 27.35 %(Rhizopus strain 4). The cause of post-harvest spoilage was mainly the fungi. Post-harvest deterioration of ginger can be reduced by proper harvesting, post-harvest handling operations, and adoption of appropriate ginger harvesting and post-harvest handling technology is important.

Key Words: *Fungi, Ginger, Pathogenicity, Post-harvest spoilage*

INTRODUCTION

Ginger (*Zingiber officinale* Roscoe) belongs to a tropical and subtropical spice plant family *Zingiberaceae.* It is thought to be originated in South East Asia and introduced to many parts of the world; and has been cultivated for thousands of years as spice and medicinal purpose in India and China (Rafie and Olczyk, 2003; Shulka and Singh, 2007). Ginger rhizome is typically consumed as a fresh paste, dried powder, slices preserved in syrup, candy (crystallized ginger), as a beverage or as flavoring agent. In many countries, especially in India and China, fresh ginger is used to prepare vegetable and meat dishes and used in preparation of various foods for seasoning, flavoring and imparting aroma in all over the world (Shukla and Singh, 2007).

Ginger has been affected by many diseases. Ginger wilt is the most limiting factor in the production of culinary ginger in Hawaii (USA). That was responsible for a 45% loss in ginger production of the country in 1993 (Hepperly, *et al*., 2004). Various fungal and bacterial genera caused post-harvest losses of ginger. Some these include the genera Penicillium (soft rot and blue mould), *Fusarium* (dry rot), *Rhizopus* (watery rot), *Armillaria* (Armillaria rot), *Sclerotium* (rhizome rot), *Pythium* (cottony-watery rot), *Stachybotrys* (soft rot), *Erwinia* (soft rot) and *Pseudomonas* (rhizome wilt) (Pandey, *et al.*, 1997; Cherian.2002; Dohroo, 2005; Stirling, 2004).

According to Stirling (2002), in the years 1997 and 1998, some ginger growers in Queensland (Australia) experienced severe losses of newly planted 'seed pieces'. Kumar *et al*., (2004) also reported bacterial wilt of ginger caused by *Ralstonia solanacearum, that* is widely distributed in tropics, subtropics and temperate regions world wide. According to Rahaman *et al*. (2009), over the last few years, rhizome diseases have affected the crop in many states of India resulting in decline of rhizome yield from1:8 ratios (seed rhizome to harvested rhizome) to 1:4. In addition, Rahaman *et al* (2009) in their survey reported that more than 60% of farmers in Sikkim (India) feel that wilt and soft rot of ginger rhizomes were the major limiting factors in ginger cultivation.

In Assam (India), 30% yield loss was reported due to rhizome rot. It has been also indicated in Rahaman *et al* (2009) that in traditional agricultural system as well as organic farming, managing rhizome diseases is a great challenge. Consequently, many growers have given up ginger cultivation and others are still struggling to survive because of rhizome diseases in India. Stirling *et al.* (2009) indicated in their survey based observations on the etiology of rhizome rot of ginger that, the disease generally develops during hot and wet conditions, and often causes losses of more than 50% of seed crops in Fiji and Australia. Moreover, a review by Dohroo (2005) indicated that ginger crops in the Indian states of Kerala and Tamil Nadu are sometimes almost totally destroyed by rhizome rot. Rhizome rot is a serious spoilage of rhizome, causing considerable economic loss to growers in different countries (Ghosh and Purkayastha, 2003**).** According to FAO (2004) post-harvest spoilage in ginger is normally due to rough harvesting and handling practices which result in injury to the skin and flesh of the rhizome.

According to NARI (2004), infection by *Fusarium* is typically associated with wounds or insect and nematode damaged tissue. Small, brownish, irregular, water-soaked patches characterize the rot over the rhizomes and white mycelium is also observed over these areas (Cherian, 2002).Watery rot, caused by the fungus *Rhizopus*, is one of the most rapidly developing storage rots of ginger. Symptoms include a soft watery rot that progress rapidly and may rot an entire rhizome within a week. Infected tissue is mottled brown and soft. In a humid atmosphere the infected area is soon covered with large amounts of white moulds and the mould will eventually turn black. *Rhizopus* is a wound pathogen and is not effective in colonizing healthy tissue (NARI, 2004). Dry rot due to rhizopus is also reported by Cherian, (2002).

Kaneshiro and Shintaku (1996) reported that the major factor in dissemination of bacterial wilt of ginger on the island of Hawaii was the unintentional use of plant materials infected by *R. solanacaerum* . Cutting up an infected rhizome for planting divides one in to several infected seed pieces and further disseminates the fungi and/or bacteria via the contaminated cutting instrument. Kumar *et al.* (2004) indicated that primarily rhizome-borne pathogens are believed to be transmitted to many ginger growing areas through latently infected rhizomes and secondary spread within the field and the neighboring localities is through rain splashes and run-off water in the field. Agricultural tools also may contribute in disseminating the pathogens. Reduction of infection can be achieved by split-drying the rhizome to a moisture content of 10- 12%. Ginger diseases have been minimized further by cultural methods such as the use of clean seeds, crop sanitation, crop rotation, varying time of planting (early planting) to ensure disease escape, and seed treatment against nematodes (Bernard, 2008).

Ginger is known to have been introduced to Ethiopia as early as in the 13th century (Jansen, 1981). It is cultivated in South, Southwestern and Northwestern parts of Ethiopia as cash crop, and is among the important spices used in every Ethiopian kitchen for the preparation of pepper powder, stew, bread, etc. It has also some use in traditional medicine for the treatment of flu and stomach ache (Jansen, 1981; Girma Hailemichael and Digafie Tilahun, 2004).

Ginger is a multi-functional crop and its rhizome quality affects the economic return of the growers, establishment, growth and yield of the crop. At Hadaro-Tunto and Boloso Bombae the ginger crop is named as “the crop against poverty” or “the cash in the bank”. This multifunctional crop often spoiled, loses quality and deteriorated by microorganisms that caused rhizome product loss. Therefore, these problems can be solved by good management practices based on operational research. Although attempts have been made to conduct research on agronomic aspects and chemical constituents of ginger cultivated in Ethiopia, there has been no research conducted on ginger spoilage microorganisms in Ethiopia. Thus, this research was aimed at isolating, identifying and characterizing the major fungi associated with post-harvest deterioration and diseases.

MATERIALS AND METHODS

Post-harvest survey

Ginger samples were collected from Hadaro-Tunto and Boloso-Bombae Southern Ethiopia. The post-harvest field survey and sample collection was carried out between August 2009 and January 2010. The survey was aimed at identifying the indigenous harvesting, drying and storing practices and knowledge on microbiological post-harvest spoilage of ginger. It was carried out using closed-ended questionnaire. The growers were asked in their native language at their village or at the market place of each district. In addition, thirty-seven stores in both districts were visited in order to observe storage conditions and the atmospheric relative humidity during sample collection was recorded using Hygrometer(DIPLEX,HYGROMETER 409 GERMANY). The questionnaire was prepared to identify problems associated with indigenous practices while harvesting, transporting to the market, drying and storing and also knowledge about post-harvest microbial spoilage of ginger. The data from the survey were translated and analyzed using simple descriptive statistics to identify research priorities.

The sample collection

A total of 115 ginger rhizome samples (each sample about 250-500 g) were collected from both districts and transported to Hawassa University, College of Agriculture, plant pathology laboratory. The sampling method was purposive sampling in which the samples consisted of rhizomes that show symptoms of spoilage (rot development, discoloration, and fungal growth etc.) and some healthy rhizome were included in the sample. The ginger samples included 37

sample (21 from Hadaro-Tunto and 16 from Boloso-Bombae) from fresh harvest, 40 (23 from Hadaro-Tunto and 17 from Boloso-Bombae) from sun drying and 38 (22 from Hadaro-Tunto and 16 from Boloso-Bombae) from stored dry ginger. For fresh harvest samples, those harvested rhizomes before being exposed to sun drying were obtained from randomly selected small-holder farms. Similarly the drying ginger samples were those ginger rhizomes that were exposed to sun for drying for 5-15 days and in the case dried stored ginger samples, those ginger rhizomes stored for 1-3 years were included.

Moisture Content Determination

Moisture content of dry ginger was determined according to methods of Nwinuka *et a*l., (2005) and Phoungchandang *et al*., (2009). Dried ginger samples (5 - 6.9 g) were weighted in triplicates and placed in washed, dried and weighted crucibles. The samples on the crucibles were placed in an oven and dried at 105 $^{\circ}$C for three hours until constant weight is obtained. The samples were then cooled in a desiccator (Scheibler pattern Desiccator, Moncrieff-Perth-made in Scotland) and reweighed. The percentage moisture content was calculated by computing or expressing the loss in weight on drying as a fraction of initial weight of samples and multiplied by 100.

Isolation and Identification

Ginger rhizome samples (250 to 500g) were washed with sterile distilled water and surface sterilized by using 1 % sodium hypochlorite for 3-5 minutes and then washed with five successive changes of distilled and sterile water. Then the samples were crushed using sterile mortar and pestle. Slurry was prepared by placing 250 g powder in sterile beaker and adding 250 ml of sterile distilled water. The slurry was mixed well and 0.1 ml was spread plated on pre-dried sterile Potato Dextrose Agar (PDA) (pH, 5.6) (Oxoid), Sabourud Dextrose Agar (SDA) (pH, 5.6) (Oxoid) and Czapeck Dox Agar (CDA) (pH, 7.3) (Oxoid) using micropipette (Device Id., 05053258, Method Id., VQ1000EX, Var-100-1000 µl). The plates were incubated at 25 ^{0}C for 4-7 days. When the mycelia emerge, the organisms were sub-cultured on to fresh sterile respective agar media to obtain pure culture of each isolate.

Seven to fifteen day old fungal cultures grown on Malt Extract Agar (MEA) (Oxoid), PDA and CDA were aseptically taken using sterile inoculating needle, placed separately on to clean microscopic slide and were stained using cotton blue (aniline blue) (1g in 1000ml distilled water) and / or shear's mounting fluid [3 g potassium acetate, 150 ml distilled water, 60 ml glycerin, 90 ml ethanol (95 %)]. Identification of fungi was carried out to the genera level based on morphological features using compound microscope and colony characteristics on PDA, CDA and MEA and compared with structures in Samson *et al* (2009) and Dugan (2006) and colored plates in Samson *et al* (2009). The percentage incidence of each identified genera was determined as a percentage of total samples as follows (Nuamah *et al*., 2005):

$$Ip\ (\%) = \frac{NTG}{TNI} \times 100$$

Where; Ip = percentage incidence of the genera;
NTG = number of times the genera isolated;
TNI = total number of Isolates.

Pathogenicity Test

Pathogenicity of the identified genera was established according to the methods of Salami and Akintokun, (2008), and Yusuf and Okusanya (2008). Pathogenicity was determined by testing the ability of each genus to induce rot in healthy fresh rhizomes. The fresh healthy rhizomes of 9-12 months age were obtained from the sample collection site (Hadaro). Rhizomes were washed in sterile distilled water and surface sterilized using 1% sodium hypochlorite and then washed in five successive changes of sterile distilled water. Holes of 1mm deep were dug in the rhizome by using 1 mm diameter sterile cork-borer; the plug was pulled out and replaced with 1 mm diameter mycelia disc from PDA and placed at the bottom of the hole on fresh rhizome. A Small portion of the rhizome plug was cut off to compensate for the thickness of the mycelial disc introduced in to the hole. The plug was carefully placed and the wounded areas were sealed with about 5 mm thick sterile agar blocks to prevent extraneous infection. The rhizomes were weighted and incubated for 15 days at room temperature (25 ^{0}C). Five healthy rhizomes were used for pathogenicity test for each genus. The control experiment was done by using sterilized PDA discs of equal diameter made in to the holes of healthy rhizomes. At the end of 15th day, the rhizomes were reweighed to determine the pathogenicity index of each genus. The pathogenicity index was determined by using the expression (Salami and Akintokun, 2008):

$$\text{Pathogenicity index (PI)} = 100 - \left[\frac{WD}{WH} \times 100\right]$$

Where: WD=weight of the diseased rhizome and WH= weight of healthy rhizome.

The rhizomes were cut along the plane of inoculation by means of sterilized scalpel-blade and the type of rot developed was characterized. Decayed portion from each inoculated rhizome was surface sterilized and inoculated on to PDA, MEA and CZA to confirm that the decay was due to the test inoculants.

RESULTS

The survey

Harvesting ginger in both districts is carried out by digging around the rhizome using fork, spade and/or hoe to loosen the soil around the crown of the plant. It was observed at Hadaro that farmers put fresh harvested ginger with sack on unclean ground which initiates mould growth that further facilitates spoilage of fresh ginger. The survey also indicated that cleaning and washing are not common activities in both sites, where 93% of the respondents do not wash their ginger. The survey further revealed that 85% of respondents did not

separate the injured and damaged rhizomes from the healthy ones. All respondents dry their ginger by exposing in to sun light. Furthermore 81.7 % of the respondents dry their ginger simply on unclean soil ground. In both sites, the majority of ginger growers store dried ginger in sack on the ground (68.3%). The remaining the respondents, still store using sack but on the wooden structure arranged on the floor. The survey has also shown that the characteristics of spoiled ginger are similar at both districts. All respondents had experience about the characteristics spoiled ginger. The characteristics of spoiled ginger include rhizomes covered with moulds, rhizomes rotted in the injured and /or damaged places, rhizomes possess pungent smell etc.

Fungal load and diversity on harvest and drying ginger

Fungal load and diversity from fresh harvest and sun drying ginger samples at Hadaro-Tunto and Boloso-Bombae is shown (**Table 1**). At both sites, fresh harvest and drying ginger samples contained high fungal load and diversity .At both sites rhizopus was not isolated from fresh harvest samples.

Table 1. Sample type, fungal load and diversity at both sites.

Sites	Sample type	Number of samples	Fungal load	Fungal diversity
Hadaro-Tunto	Harvest	21	86	Aspergillus,Fusarium,Penicillium
	Drying	23	104	Aspergillus,Fusarium,Penicillium,Rhizopus
Boloso-Bombae	Harvest	16	68	Aspergillus,Fusarium,Penicillium
	Drying	17	93	Aspergillus,Fusarium,Penicillium,Rhizopus

Moisture Content of Dry Ginger

During sample collection of dried and stored ginger, the mean atmospheric relative humidity was generally high (79%) at sample collection sites (79.12 at Hadaro-Tunto and 78.82 at Boloso-Bombae). The moisture content of dried and stored ginger samples obtained from both districts ranged between 7.28 % and 24.94 % (**Table 2).**

Table 2 Moisture content, fungal load and diversity of stored ginger samples at Hadaro-Tunto and Boloso-Bombae

Sites	Number of samples	Moisture		Fungal load and diversity	
		Category (%)	Sample (%)	Fungal load	Fungal diversity
Hadaro-Tunto	11	7.28-12.00	50	28	Asp,Eur,Muc
	5	12.1-15.99	22.7	79	Pen,Rhi,Fus,Asp,Eur,Muc
	3	16.00-19.9	13.6	59	Pen,Rhi,Fus,Asp,Eur,Muc
	3	>20	13.6	48	Pen,Rhi,Fus,Asp,Eur,Muc
	Mean	**13.79**			
Boloso-Bombae	5	7.28-12.00	33.3	13	Asp,fus,Eur,Muc
	4	12.1-15.99	26.6	48	Pen,Rhi,Fus,Asp,Eur,Muc
	4	16.00-19.9	26.6	36	Pen,Rhi,Fus,Asp,Eur,Muc
	2	>20	13.3	15	Pen,Rhi,Fus,Asp,Eur,Muc
	Mean	**13.39**			

Asp=Aspergillus,Eur=Eurotium ;Muc=Mucor;Pen=Penicillium;Fus=Fusarium;Rhi=Rhizopus

The mean moisture content of dried and stored ginger at Haharo-Tunto was 13.39 %±5.11and at Boloso-Bombae was 13.79±4.94. About 50% of the samples were found to contain moisture in the range of 7.0% - 12.0% at Hadaro-Tunto while 33.3% of samples at Boloso-Bombae were found to contain in similar range. The rest 50% samples at Hadaro-Tunto and 66.7% at Boloso-Bombae were found containing moisture beyond the standard **(Table 2)**.In both sites, the fungal load and diversity is also numerous in samples containing moisture content above 12%. Fusarium, Penicillium and Rhizopus were isolated from samples with moisture content above 12% **(Table 2)**.

Occurrence of fungi

Microorganisms comprising six fungal genera and bacterial strains associated with the post-harvest spoilage of ginger from Hadaro-Tunto and Boloso Bombae were isolated and. The numbers of isolates were 154 from harvest (86 at Hadaro-Tunto and 68 at Boloso Bombae), 197 from drying on the ground (104 at Hadaro-Tunto and 93 at Boloso Bombae) and

326 (214 at Hadaro_Tunto and 112 at Boloso-Bombae) from stored ginger samples. The six genera of fungi were identified. These include *Aspergillus, Fusarium, Penicillium, Rhizopus, Eurotium and Mucor* (**Table 3**).

Table 3 Occurrence of fungi during harvest and post-harvest handling of ginger at Hadaro-Tunto and Boloso-Bombae.

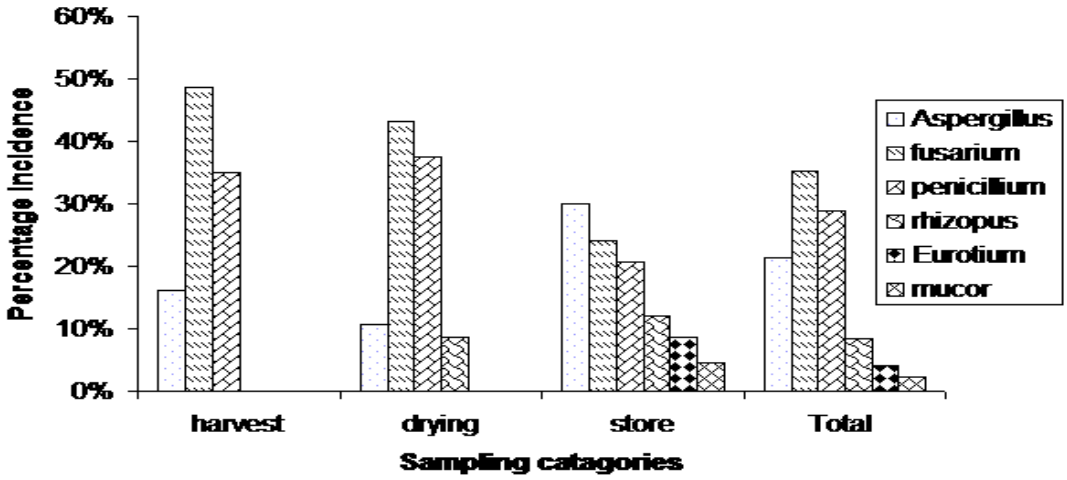
	Hadaro-Tunto				Boloso- Bombae			
Fungal genera	**Fresh harvest**	**Drying ginger**	**Stored ginger**	**Total**	**Fresh harvest**	**Drying ginger**	**Stored ginger**	Total
Aspergillus	+	+	+	3	+	+	+	3
Fusarium	+	+	+	3	+	+	+	3
Penicillium	+	+	+	3	+	+	+	3
Rhizopus	-	+	+	2	-	+	+	2
Eurotium	-	-	+	1	-	-	+	1
Mucor	-	-	+	1	-	-	+	1
Total	3	4	6	13	3	4	6	13

+; presence of corresponding fungal genera in the sample -; absence of corresponding fungal genera in the sample

The isolates of bacteria were not identified. However, the occurrence of different bacteria has been detected from fresh drying ginger samples using nutrient agar. The genera of *Fusarium, Penicillium*, and *Aspergillus* were isolated at both districts, from fresh harvest, during drying and stored dried ginger samples (**Table 3**). *Rhizopus* was isolated from drying and stored dry ginger samples at both districts whereas *Eurotium* and *Mucor* were isolated from dried stored ginger samples at both districts.

Percentage incidence of fungi

The percentage incidences of post-harvest spoilage fungi isolated from spoiled ginger vary among fresh harvest, during drying and dried stored ginger samples (**Fig.1)**.



The percentage incidence of fungi from spoiled ginger rhizomes ranged from 2.2 % (*Mucor*) to 35.3% (*Fusarium*). At fresh harvest condition, 48.7% of spoilage was due to *Fusarium* followed by *Penicillium* (35.1%) *and Aspergillus* (16.2%) (**Fig.1**). In a similar manner, the deterioration of drying ginger was mainly due to Fusarium and Penicillium. In drying ginger samples *Fusarium* and *Penicillium* account (43.1%) and (37.6%) respectively. It indicates that they were major causes of spoilage during slow sun drying conditions. *Aspergillus* was found to be the cause of 30% ginger deterioration during storage followed by *Fusarium* and *Penicillium* accounting for 24% and 20% of spoilage respectively (**Fig. 1)**. *Rhizopus* was also found to be the causative agent of ginger deterioration during drying. During storage, fungal load and diversity was increased with addition of Eurotium and Mucor. Deterioration of stored ginger at both sites was caused by diversified fungal groups. The increased fungal load and diversity seems to associated with the beyond standard moisture content, long time storage and the high relative humidty at the sample sites. In general, *Fusarium* was

found to be responsible for 35.3% of deterioration observed followed by *Penicillium* 28.8% and *Aspergillus* 21.3 %. *Rhizopus, Eurotium* and *Mucor* were found to be responsible for 8.3%, 4.1% and 2.2%, of the incidence in spoilage observed respectively.

Pathogenicity Test

All the fungal genera were found to be pathogenic to fresh ginger rhizomes with pathogenicity indices ranging from 17.35 % (*Rhizopus* 3) at Hadaro-Tunto to 27.35 % (*Rhizopus 4***)** at Boloso-Bombae **(Table 4).** *Rhizopus* and *Eurotium* had relatively the highest percentage of pathogenicity indices (27.35%) and (27.20%) respectively at Boloso-Bombae followed by *Fusarium* (25.78%) at Hadaro-Tunto. The pathogenicity tests revealed that all the fungal isolates from spoiled ginger rhizomes caused rot in fresh ginger rhizomes. *Rhizopus* caused relatively more ginger deterioration at drying and storage places while *Eurotium* caused high ginger deterioration at storage condition. Following Rhizopus and Eurotium, *Fusarium* and *Penicillium* caused more deterioration of ginger and *Mucor* caused relatively the least spoilage and rot.

Table 4 Pathogenicity test of fungi isolated from ginger rhizomes at Hadaro-Tunto and Boloso-Bombae.

	Fungal genera	Pathogenic index (%)	Characteristics of rhizome rot
Hadaro-Tunto	*Rhizopus*		
	R1*	17.85	Dry rot, black margin
	R2*	20.11	Dry rot, black brown color
	Eurotium		
	E1*	19.98	Dry rot, brown color
	Fusarium		
	F1*	22.17	Dry rot, dark brown
	F2*	21.02	Dry rot, brown at the margin
	F3*	25.78	Dry rot, pale to dark brown
	Penicillium		
	P1*	20.34	Dry rot covered with blue mold
	P2*	25.36	Dry rot covered with blue mold
	P3*	21.03	Dry rot with blue mold surrounding point of inoculation
	Aspergillus		
	A1*	20.70	Dry rot, brownish yellow
	A2*	21.62	Dry rot, brown margin
	Mucor	17.75	Dry rot , black margin
	Control	1.63	No rot induced
Boloso-Bombae	*Rhizopus*		
	R3*	17.35	Dry rot, black color
	R4*	27.35	Dry rot, black color
	Eurotium		
	E2*	27.20	Dry rot, brown
	Fusarium		
	F4*	20.83	Dry rot, yellow margin
			Wet rot, brown color at the margin
	F6*	23.65	Dry rot , pale brown
	Penicillium		
	P4*	22.73	Dry rot, brown to dark brown
	P5*	21.48	Dry rot, bluish margin
	Aspergillus		
	A3*	22.32	Dry rot, dark brown
	Mucor	17.75	Dry rot , black margin
	Control	1.63	No rot induced

**** Strains of respective genera***

Even though *Rhizopus* and *Eurotium* were found to be relatively the most pathogenic genera, they were found in relatively the least frequent. If these two genera were most frequent in percentage incidence, they would have caused severe deterioration of ginger at storage places. Although, the extent of infection and the characteristic color of rot developed

vary among ginger spoilage fungi **(Table 4)**, the type of rot induced was almost similar (dry rot) on fresh ginger rhizomes. *Fusarium* was associated with wet and dry rots accompanied by brown to dark brown characteristic color either through out the rot and/or at the margins. Similarly, *Penicillium* induced dry rot which is covered by blue mould surrounding the point of inoculation and Aspergillus induced brown to brownish yellow colored dry rots. *Rhizopus* and *Mucor* produced black dry rot.

DISCUSSION

It is predictable to obtain such high fungal load and diversity **(Table 1)** since there were less sanitary operations practiced (as indicated in this survey) during ginger drying as shown in sample collection seasons. The survey further showed that ginger rhizomes were exposed to sun on unsanitary ground (soil) and even some farmers polish the drying ground with animal dung.. The results of this investigation revealed that the average moisture content of dried and stored ginger was 13.56 %(13.39 at Hadaro-Tunto and 13.79 at Boloso-Bombae), which was higher than the recommended value (7-12%) (Jansen, 1981; Yiljep *et al*., 2005). It was not surprising to record such moisture content since ginger at both districts was sun dried and stored with sacks either simply on ground (soil), wooden structured floor or spread on the floor. Inappropriately prepared ginger storage places may increased the possibility of absorbing moisture from the surrounding condensation and increase the moisture content of dried ginger. During collection of dried and stored ginger samples, the mean atmospheric relative humidity was generally high (79%) at sample collection sites. This may be another possibility that might have caused moisture migration from the surrounding condensation to the stored dry ginger, which favored the growth of moulds. Moreover, such moisture content may have been recorded due to the fact that ginger sun drying practices carried out by growers were not effective to accomplish the standard moisture content (7-12%). The other possibility that might have contributed to the high mean moisture content may be the weight based marketing practices of dry ginger at both districts that might have motivated the growers to sell hardly dried ginger. According to Butt *et al*. (2004), fungal growth was retarded at lower moisture content, but at 14% or slightly above, fungal growth was initiated. Butt *et al*., (2004) also reported that moisture content of stored agricultural produces is affected by atmospheric relative humidity. The standard moisture content of dry ginger at which microbial growth inhibited was reported to be at 7-12% (Jansen, 1981; Yiljep *et al*., 2005). Some growers (43.3%) dry their ginger to the safer range (7.28-12.0%) and others dry with mean moisture content higher than standard range. It might be high moisture content and /or atmospheric relative humidity or their interaction that could have contributed to the growth of diverse mould groups and the deterioration of dry and stored ginger at both districts.

The fungal genera identified in this study were Aspergillus, *Fusarium, Penicillium, Rhizopus, Eurotium* and *Mucor.* These fungal genera were found to be associated with post-harvest deterioration of *Zingiber officinale* rhizomes at harvest, during drying and/or dried and stored at study sites. Results further showed that *Fusarium* was the predominant cause of ginger deterioration at harvest and during drying. This may be associated with the fact that *Fusarium* is soil-borne fungi (NARI, 2004), and since the soil was not removed from the rhizome surfaces by washing or using another mechanism. *Fusarium* was reported to be both field and storage fungi (Adams and Moss, 2000). Ginger harvesting and drying mechanisms at study sites might have also encouraged the entrance of spoilage fungi (*Fusarium*) to rhizomes through injuries, damages and natural openings. It was seen at study sites that ginger was not cured, that allows the skin to thicken/heal and the cut ends to suberize. It was well documented that ginger spoilage was mainly associated with injuries and damages induced during harvesting and post-harvest handling practices (FAO, 2004; NARI, 2004 and Yiljep *et al*., 2005). The injured, damaged and cut ends (if not carefully cured), may serve as starting point for mould growth and rot, and may also serve as inocula sources for the spread of deteriorating and pathogenic fungi to the whole/stock ginger and hence facilitate spoilage and deterioration. The survey results further indicated that growers and traders at study sites dry ginger by exposing to sun on unclean soil for long time (3 weeks and above**)**. Exposing fresh ginger on sun for long time may cause high rate of water loss and shriveling and this may create cracks which may serve as entry point for spoilage and pathogenic fungi and extended drying on such unclean ground would also make conducive environment for diversified fungal growth and may facilitate spoilage and rot development.

As the results indicated *Penicillium* was among the most predominant post-harvest deteriorating fungi at harvest, during drying and storage. It was well documented that *Penicillium* is a common storage fungus and commonly associated with high relative humidity and requires moisture condensation on the rhizome surface (Adams and Moss, 2000; FAO, 2004; NARI, 2004). Relatively high mean relative humidity (79%) at study sites and beyond standard mean moisture content (13.56%) might have also contributed. *Rhizopus* was isolated during drying and at storage. It was well established that *Rhizopus* is a wound pathogen and non-effective in colonizing the healthy rhizomes (FAO, 2004; NARI, 2004). Isolation of *Rhizopus* during drying may indicate the fact that some injured and damaged rhizomes during harvest and drying were infected by *Rhizopus* most probably from the soil and other sources.

Fungi fall practically in to two broad ecological categories: storage and field fungi (Subramanian, *et al*., 2007). The most important genera of the storage fungi are *Penicillium* and *Aspergillus.* Species of *Fusarium* are involved in spoilage at storage under moist condition (Adams and Moss, 2000). Isolation of *Fusarium*, *Penicillium* and *Rhizopus* from spoiled ginger rhizomes in this study is inline with (Pandey *et al*.,

1997; Cherian, 2002 and Dohroo, 2005) who reported these genera as a cause of post-harvest deterioration of ginger. Stirling (2002; 2004 and 2009), and Paull and Chen (2008) isolated *Fusarium* from diseased ginger rhizomes and plant materials. In addition, Mandeel (2005) isolated and considered *Aspergillus, Penicillium* and *Rhizopus* as the most frequently recovered as contaminant of ginger. *Eurotium* and *Mucor* were isolated from stored ginger samples. Isolation and identification of *Eurotium* and *Mucor* as causes of post-harvest deterioration of ginger, particularly at storage may be the first report. There were studies indicating that these fungal genera were causes of spoilage of many agricultural produces at store. Kacániová (2003) isolated *Eurotium* and *Mucor* from dried soybean, and considered them as problems of stored agricultural products.

The pathogenicity tests revealed that all the fungi isolated from spoiled rhizomes at study sites infected and caused rot in fresh rhizomes. In this regard *rhizopus* and *Eurotium* caused relatively more spoilage followed by *Fusarium and Penicillium.* Relatively low percentage pathogenicity indices of fungi may be resulted from the microbial activity of ginger, microaerobic environment created while sealing with agar blocks and fibrous nature of ginger rhizomes. The pathogenicity of genus *Fusaium* is inline with that of Stirling (2004) who performed pathogenicity test and reported *Fusarium oxysporum*, and *Fusarium sp.* pathogenic to ginger rhizomes. The pathogenicity test carried out by Yusuf and Okusanya (2008) on *Rhizopus stolonifer*, *Aspergillus niger* and *Penicillium oxalicum* isolated from rotten yam tubers indicated that these fungi were found to be pathogenic to yam tuber. Salami and Akintokum (2008) also showed that *Rhizopus*, *Fusarium sp.*, *Aspergillus sp.*, and *Penicillium* species were pathogenic to root crops such as yam and cassava tubers. Even though *Rhizopus* and *Eurotium* were found to be relatively the most pathogenic, they were relatively the least frequently isolated at study sites. Most spoilage fungi developed almost similar characteristic rot types (dry rot) which differ in color. *Fusarium* induced dry rot that is pale to dark brown whereas *Penicillium* developed dry rot covered with blue mould. *Aspergillus* produced dry rot which is brownish yellow and dark brown. *Eurotium* similarly caused dry rot which is brown in color. *Rhizopus* and *Mucor* induced similar rots (dry rots with black color). The rots induced by ginger spoilage fungi in the present study were comparable to results reported in Cherian (2002).

ACKNOWLEDGEMENTS

We are grateful to Wondwosen Tadesse,Yassin Hassen,and Alazar Ergena for assistance in laboratory works and also thanks to all people assisted during survey and sample collection.

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